Louis Bolk Instituut

Impact of regenerative practices on soil, plant, and crop

First-year results of the Soil Heroes regenerative experiment

Anne Hoek van Dijke, Marianne Hoogmoed

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Content

Summary

The Soil Heroes Foundation set up a field experiment in the south-western Netherlands to study if and how regenerative practices impact soil health, plant nutrient content, and crop nutritional value. The study investigates the effect of various practices including compost tea and biofertilizer, mulching, companion crops, and lasagne (alternating layers of rock dust, compost, and manure). These regenerative practices are tested on a till and no-till field, and they are compared with a conventional agricultural practise.

The year 2024 was the first full experimental year. In January, chemical and physical soil properties were determined to study the starting position. In summer, plant and crop nutrient content was analysed, and in autumn, soil chemical characteristics were analysed. In this report, we investigate the starting position of the experiment, and the soil, plant, and crop nutrient content after the first experimental year.

The soil analyses of January 2024 indicate that the regenerative field and conventional fields are different in their chemical and physical properties. These differences were expected, because the two fields have a very different management history, but they are relevant to consider for future analyses.
The analyses after the first experimental year show that, for phosphorus (P) and potassium (K), a high supply (with the mulch) increased soil P and K content, while soil P and K content decreased for a treatment with low supply (compost tea + biofertilizer). Soil organic matter content increased, also for the conventional treatment without the supply of organic matter. For most macro- and micronutrients, there were no or minor changes in soil nutrient densities, but no big change was expected after only one year.
The treatments did partly, but not always, impact the leaf nutrient content. With did find a high leaf K content for the mulch treatment with a high supply of K and high soil K content.
The yield was lowest for the conventional plots, followed by the regenerative till, and regenerative no-till plots. There was a ten-fold difference in the lowest yield (conventional) and highest yield (no-till, lasagne + compost tea + biofertilizer). The crop nutritional values for macronutrients and vitamins were highest for the conventional treatment. The conventional treatment had the lowest yield, and this might explain the overall high nutrient density.

The results mark the end of the first full experimental year of the regenerative experiment of the Soil Heroes Foundation. The coming years of data and analyses will be very interesting and relevant, to confirm, and further explain, the preliminary results that were presented here.

1 Introduction

The Soil Heroes Foundation set up a field experiment to test the impact of regenerative practices on soil and crops. The field experiment was set up at the Klompe Farm in the South-western Netherlands in 2023 and the final cropping season will be in 2026. The main objective of the field experiment is to test if regenerative agricultural practices increase the nutritional value of the harvested product of food crops, enhance the availability of nutrients in soil, and enhance the uptake of these nutrients by the plants, compared to conventional agriculture. Several regenerative practices are tested in a tilled and no-till system, and measurements are taken in soil, plant and harvested product. The regenerative practices are compared with a conventional system.
A first non-replicated pilot study of the field experiment was set-up and tested during the cropping season of 2023 by Soil Heroes Foundation and Edacious (USA). In 2024, the field experiment was extended with two replicates, and a full sampling campaign was carried out for soil, plant, and crop, by the Soil Heroes Foundation and the Louis Bolk Institute. The full project is foreseen for three cropping seasons, and this allows us to study during a full crop rotation, and study soil processes that evolve slowly.

In this report we describe the results of the first year of the experiment. The main research questions for this report are:

1. What were the baseline soil characteristics at the start of the field experiment? In other words, how heterogeneous is the soil, and do the treatments have a similar starting position in terms of soil characteristics?
2. How do different regenerative farming practices versus conventional agriculture impact the soil health and nutrient density?
3. How do different regenerative farming practices versus conventional agriculture impact plant and crop nutrient density?

2 Methodology

2.1 Field site

The study is carried out on the Klompe Farm in the Hoeksche Waard in the south-western Netherlands (51.81° N, 4.44° E). The region has a temperate climate, with a mean winter temperature of 3° C, a mean summer temperature of 17° C, and an annual precipitation of 863 mm. The Hoeksche Waard consists of reclaimed land with fertile sea clay soils, and since reclamation, the majority of the land has been used for agriculture. Klompe Farm is a large farm of about 300 ha with a mixture of conventional fields and regenerative fields. The treatments of regenerative farming practices (see experimental set up) are located on a historically regeneratively managed field, and the conventional treatments were located on a historically conventionally managed field. The regenerative field has been under no-till management since 2017. For the current experiment, a first pilot study was set up in 2023, and the study was extended to a full experiment with replicates in 2024.

In spring, soya was sown (and re-sown) on the experimental plots, but the soya seeds and sprouts were twice eaten by hares and pigeons. In July, when it was too late for a third re-sowing, it was decided to carry the study out with turnips, and these were sown in July. The different amendments were applied to the field once or twice (see experimental set up), and the plots were weeded manually. The organic natriummolybdaat was applied to the plots against flea beetles, and the plots were irrigated three times in August and September.

2.2 Experimental set up

The field experiment was set up to test the effect of several regenerative farming practices, such as the use of different soil amendments (compost tea + biofertilizer, and lasagne), mulching and companion crops. These practices are tested in a tilled and no-till system. For the till plots, a strip of the historically no-till field was ploughed to a depth of 30 cm in the spring of 2023 to establish the field. The till field was also ploughed in spring 2024. The various regenerative practices are compared with conventional agriculture. There are 11 different treatments with three replicates each, which brings the total number of plots to 33 (Figure 1).

The eleven different treatments are:

1. **Conventional**. The conventional field is tilled, has no cover crop, and is fertilised with NPK in spring and 250 mg urea in summer.
2. **Biofertilizer + compost tea, no-till.** The treatment has a soil amendment of biofertilizer + compost tea that is brewed on-farm. The biofertilizer and compost tea were applied four times (twice in July and twice in August). Per plot, 1.5 l of compost tea and 150 ml of biofertilizer was applied. Cover crops are sown in winter. The treatment is no-till.
3. **Biofertilizer + compost tea, till**. This treatment is comparable to number 2, with the difference that it is tilled.

4. **Companion crop, no-till**. A companion crop was sown for this treatment. The companion crop is alfalfa with white clover and crimson clover (focus on nitrogen fixing companion species). Furthermore, a cover crop is sown in winter. The treatment is no-till.
5. **Companion crop, till**. This treatment is comparable to number 4, with the difference that it is tilled.
6. **Mulch, no-till.** Alfalfa mulch was applied twice (108 kilo / plot in June and 30 kilo / plot in July). Furthermore, a cover crop is sown in winter. The treatment is no-till.
7. **Mulch, till**. This treatment is comparable to number 6, with the difference that it is tilled.
8. **Lasagne, no-till**. The lasagne treatment has alternating layers of rock dust + compost + manure. These organic amendments were applied in May, right before sowing. Furthermore, a cover crop is sown in winter. The treatment in no-till.
9. **Lasagne, till**. This treatment is comparable to number 8, with the difference that it is tilled.
10. **Lasagne + biofertilizer + compost tea, no-till (reference treatment).** This treat has application of lasagne (similar to treatment 8), and biofertilizer and compost tea (similar to treatment 2). Cover crops are sown in winter. The treatment is no-till.
11. **Lasagne + biofertilizer + compost tea, till**. This treatment is comparable to number 11, with the difference that it is tilled.

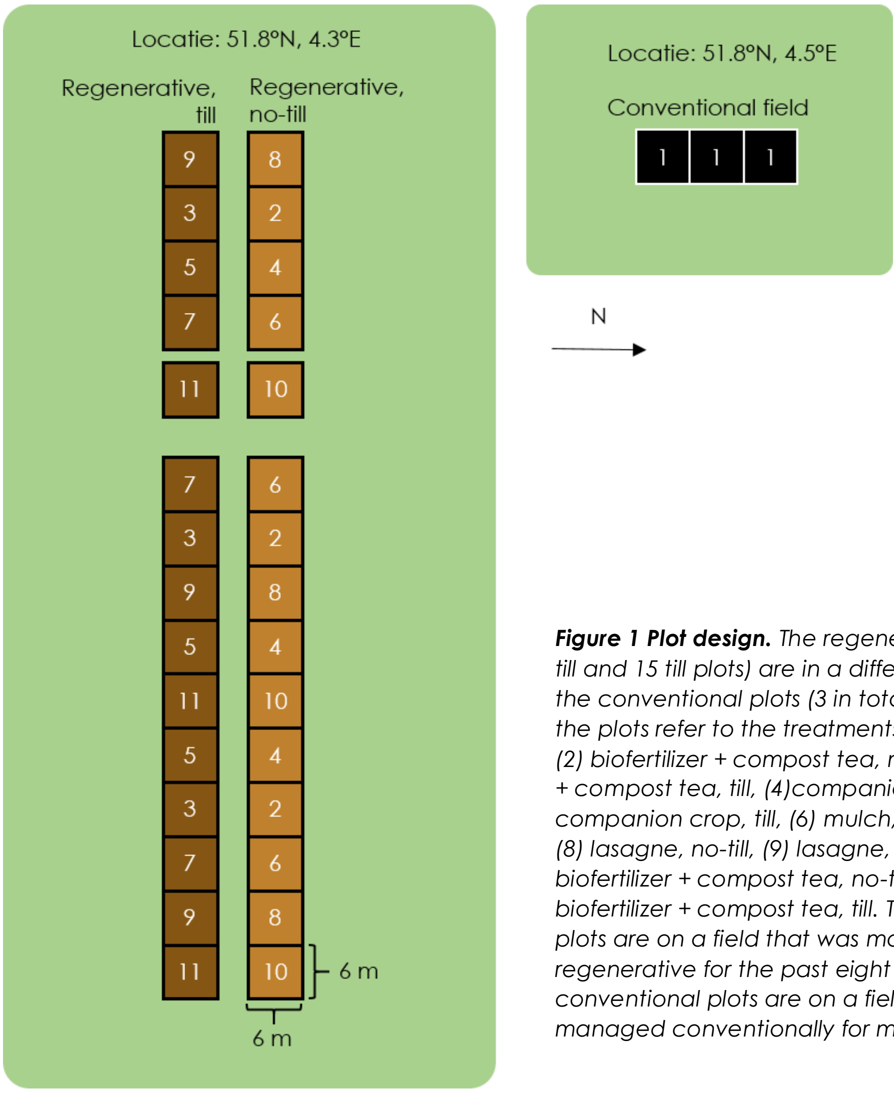

Figure 1 Plot design. *The regenerative plots (15 no-till and 15 till plots) are in a different location from the conventional plots (3 in total). The numbers in the plots refer to the treatments: (1) conventional, (2) biofertilizer + compost tea, no-till, (3) biofertilizer + compost tea, till, (4)companion crop, no-till, (5) companion crop, till, (6) mulch, no-till, (7) mulch, till, (8) lasagne, no-till, (9) lasagne, till, (10) lasagne + biofertilizer + compost tea, no-till (11) lasagne + biofertilizer + compost tea, till. The 30 regenerative plots are on a field that was managed regenerative for the past eight years, while the conventional plots are on a field that was managed conventionally for many years.*

2.3 Data collection and analyses

2.3.1 Soil

In January 2024, a first soil sampling campaign was done to determine the soil characteristics at time zero (T0). Soil samples were taken from the 0 – 10 and 10 – 30 cm layer. The soil samples were analysed on farm using the Bruker TRACER, and they were sent out to two laboratory: chemical and physical analyses were done by eurofins Agro, and Normec Groen Agri Control determined the hot water-extractable carbon. Bulk density was determined in triplicate for the upper (0 – 10 cm) and deeper layer (10 – 30 cm) from undisturbed Ø 53 millimetres soil sample rings. Penetration resistance was measured using an electronic penetrologger with a cone surface of 1.0 cm^2. The field was too wet for the measurements of water infiltration, so these were postponed.
In October 2024, a second soil sampling campaign was done to determine the soil characteristics after the first experimental season. Again, soil samples were taken from the 0 – 10 and 10 – 30 cm layer. These samples were analysed for soil biology on farm using the MicroBiometer, and by the eurofins Agro laboratory. The T0 water infiltration was measured in situ in triplicates using a Ø 15 cm, 15 cm high PVC pipes. Each pipe received 500 mL of tap water and the time needed for complete infiltration in the soil was recorded. The maximum waiting time was 15 minutes, and when this waiting time was exceeded, the remaining water level was measured.

2.3.2 Plant

Turnip plant leaves were analysed for essential nutrients by the Eurofins Agro laboratory. For each plot, four old and four new leaves were collected from ten randomly selected plants in the week before turnip harvest.

2.3.3 Crop

Crop yield was determined by harvesting all turnips of 50 cm of two rows from each plot. The turnips were weighed to determine the total crop yield for each treatment. Afterwards, the turnips were sent to the Nutricontrol laboratory for the nutritional analyses.

2.3.4 Amendments

The applied compost tea, biofertilizer, alfalfa and compost + manure were sent to the Eurofins Agro laboratory for a fertilizer analyses.

3 Results

3.1 Soil characteristics at time zero, T0

At the start of the replicated experiment, in the winter of 2024, before the application of the treatments, we took soil samples and conducted field measurements in all plots. This sampling is deemed time zero, T0. The conventional field and the regenerative field have a distinctly different history. The conventional field has been conventional for many years, while the regenerative field was used for regenerative agriculture for 8 years, including the application of compost tea and no-till. To establish the tilled regenerative treatments, a strip of the historically regenerative no-till field was ploughed in 2023, after several years of no-tillage.

Figures 2 till 6 show the results of the soil measurements at T0. Because the soil measurements were taken before the application of the various treatments (e.g. the amendments and companion crop), but after the ploughing of the no-till field to establish the tilled plots, the results are averages of the conventional, regenerative no-till and regenerative-till plots. The conventional and regenerative field have different soil characteristics, which is explained by the different management history of the fields. Within the regenerative field, the tilled and no-till plots show less differences. For example, nitrogen (N) soil stock and potassium (K) stock were substantially higher in the regenerative till and no-till plots compared with the conventional plots (Figure 2). For available N (the amount of N that can be directly absorbed by plants in the form of NO_3 and NH_4), there was no significant difference between the plots. Interestingly the phosphate (P) stock did not differ too much between the conventional and the regenerative no-till plots, but was substantially higher in the regenerative tilled plots. It is unclear what may have caused this.

Also for the macronutrients (calcium (Ca), magnesium (Mg), sodium (Na), and sulphur (S)), there are some differences in soil nutrient stock and nutrient availability between the fields at time zero (Figure 3). This difference is strongest for the soil stock and availability of magnesium; this is much higher in the regenerative fields, compared to the conventional field. The micronutrients are available in much smaller quantities (Figure 4), and for some micronutrients, the soil content was below the detection limit of the laboratory. Therefore, these nutrients are not included in the analyses. The plant availability of boron (B), selenium (Se), and cupper (Cu) is significantly lower in the conventional plots compared to the regenerative plots, while there is no difference between the regenerative tilled and regenerative no-till plots. On the other hand, the availability of zinc (Zn) is highest in the conventional plot.

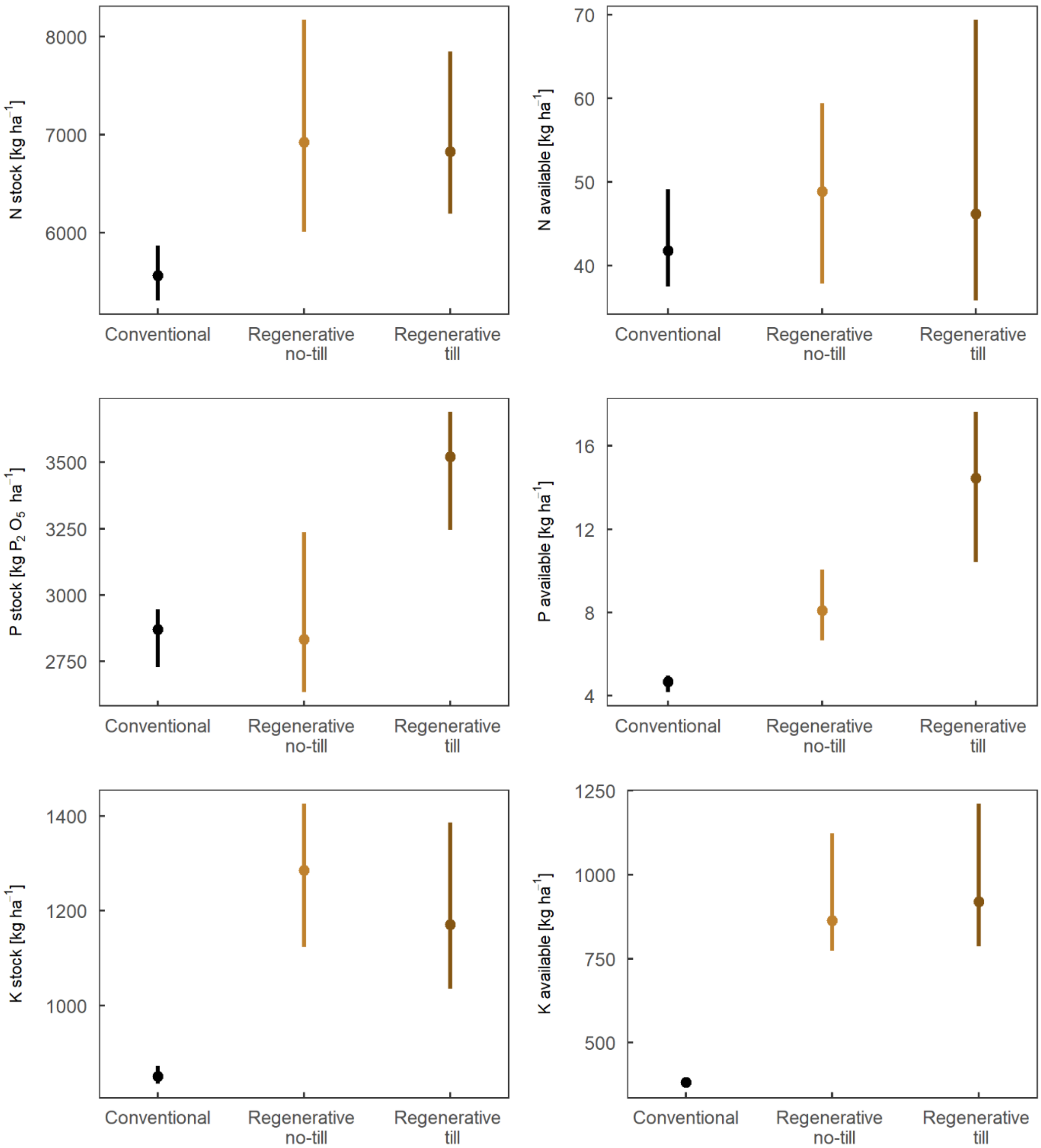

Figure 2 Soil NPK content at T0*. The total soil stock and availability of nitrogen (N), phosphorus (P), and potassium (K) in the root zone (0 – 30 cm depth). The point presents the mean value over 3 (conventional) and 15 (regenerative) plots, and the range shows the minimum to maximum value.*

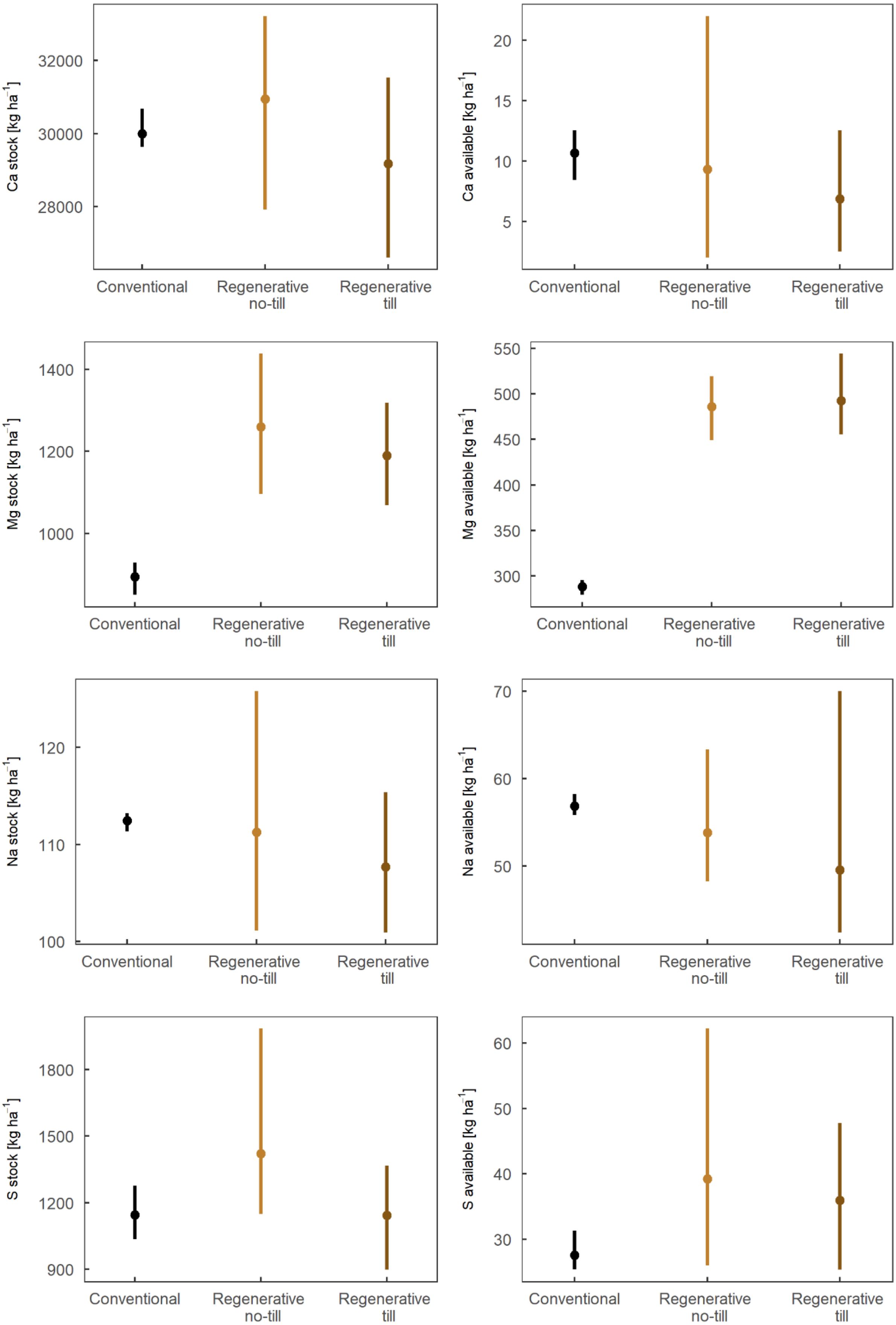

Figure 3 Soil macronutrients content at T0*. The total soil stock and availability of calcium (Ca), magnesium (Mg), sodium (Na), and sulphur (S) in the root zone (0 – 30 cm depth). The point presents the mean value over 3 (conventional) and 15 (regenerative) plots, and the range shows the minimum to maximum value.*

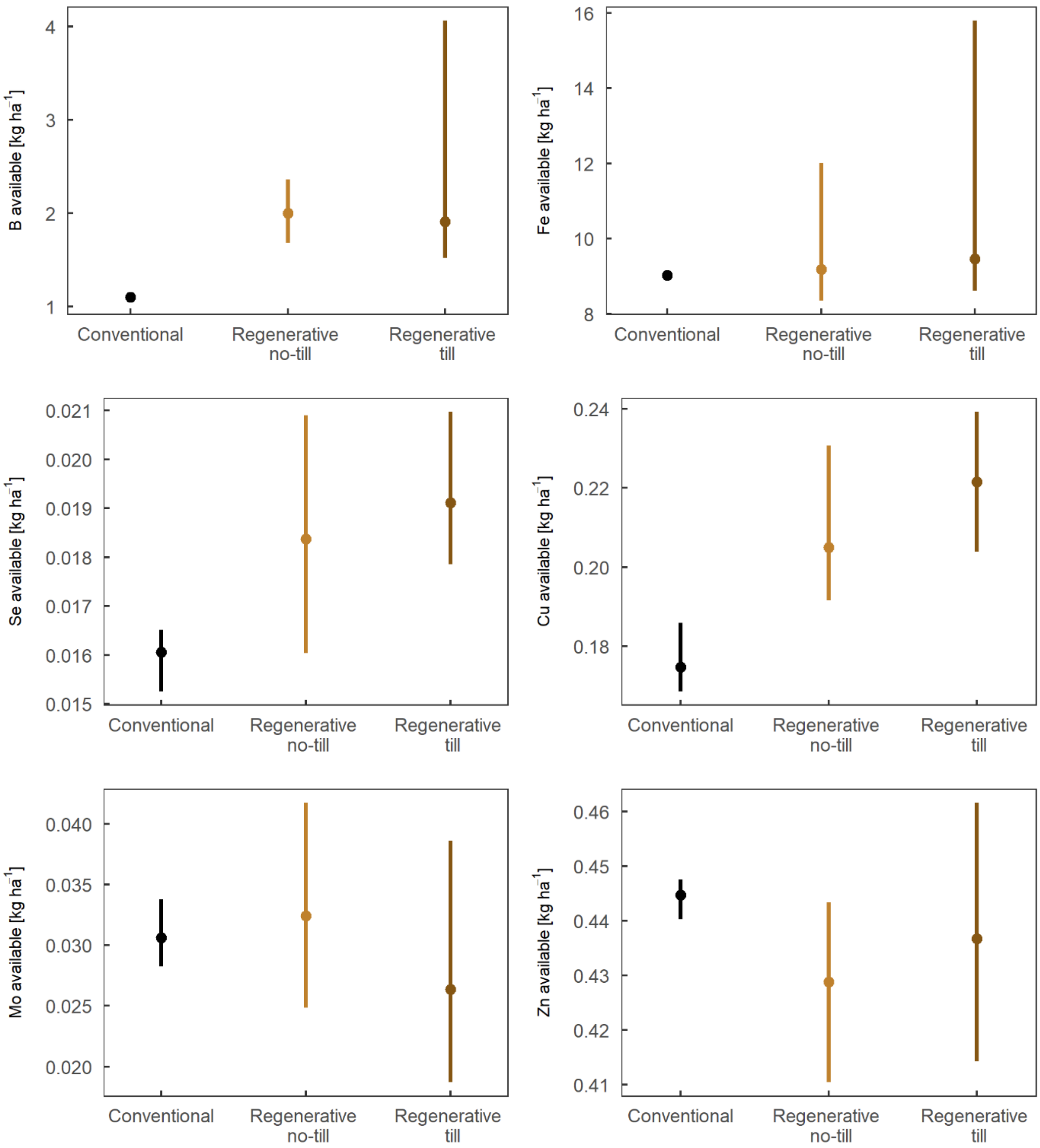

Figure 4 Soil micronutrient content at T0. *The total soil availability of boron (B), iron (Fe), selenium (Se), cupper (Cu), Molybdenum (Mo), and Zinc (Zn) in the root zone (0 – 30 cm depth). The point presents the mean value over 3 (conventional) and 15 (regenerative) plots, and the range shows the minimum to maximum value.*

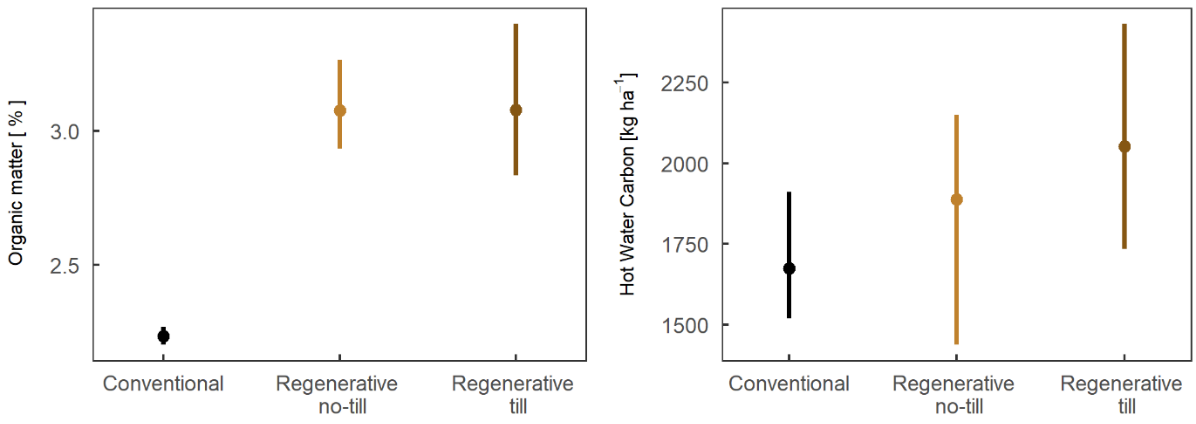

Figure 5 Soil carbon content at T0. *The mean organic matter content (OM) and hot water-extractable carbon (HWC) content in the root zone (0 – 30 cm depth). The point presents the mean value over 3 (conventional) and 15 (regenerative) plots, and the range shows the minimum to maximum value.*

The soil organic matter content is significantly lower for the conventional plots compared to the regenerative plots (Figure 5), which is most likely related to the different management practices with lower organic matter inputs in the conventional plots. The hot water-extractable carbon (HWC) is a measure of the labile soil carbon fraction. This fraction is easily decomposed by microorganisms, and can be removed from the soil by hot water. It is often used as a proxy for microbial activity and it is more sensitive to changes in carbon dynamics in the soil than other measures of organic carbon. This is why the HWC is frequently used to detect changes in the carbon pool. At time zero, we find the lowest HWC for the conventional plot, however, the difference in HWC for the regenerative and conventional field is much smaller than the difference in organic matter content.

The bulk density of the top layer (0 – 10 cm depth) is slightly higher for the conventional field, although the differences are not significant (Figure 6). The average bulk density increases towards the deeper layer of 10 – 30 cm, and in this deeper layer, bulk density is highest for the conventional plot. There is no significant difference in bulk density between the two regenerative fields, although the till field (that was tilled in 2023) has a slightly higher bulk density at 10 – 30 cm depth compared to the no-till field.

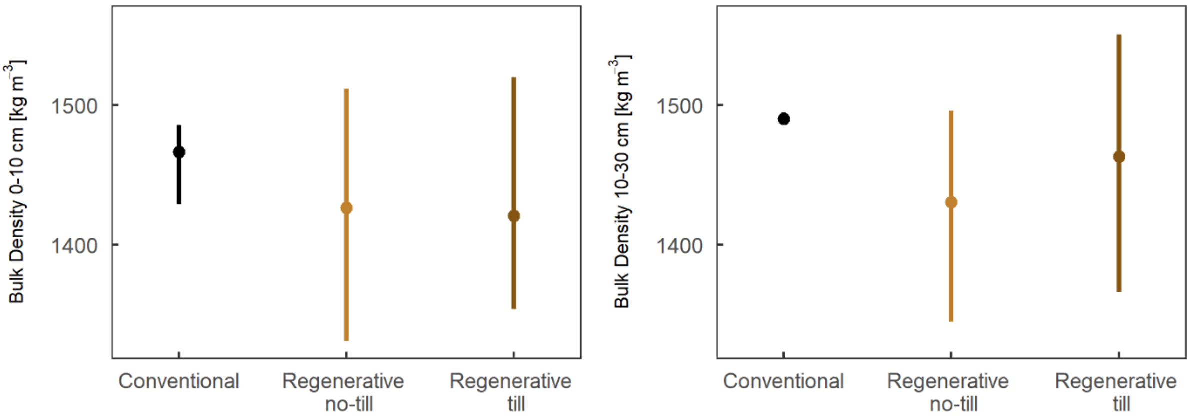

***Figure 6 Bulk density at T0**. The bulk density for the top soil (0 – 10 cm, left) and the deeper layer (10 – 30 cm, right. The point presents the mean value over 3 (conventional) and 15 (regenerative) plots, and the range shows the minimum to maximum value.*

The soil penetration resistance is an indicator for the ease with which plant roots can grow through the soil. Root development is hindered above a value of around 1.5 MPa, and roots can barely or not penetrate the soil above 3 MPa. For the rooting zone (0 – 30 cm), the penetration resistance is higher for the conventional field, compared to the regenerative fields (Figure 7). For the conventional field, the resistance is close to the 1.5 MPa above which root development is impeded by compaction. In the deeper soil (below 50 cm) the penetration resistance (and thus compaction) is higher for the regenerative fields, far above the threshold value of 3 MPa. A study by Wageningen Environmental Research showed that soil compaction decreased in the past years over the regenerative fields.

The water infiltration rate is a measure of how easily water enters the soil, and it depends on the soil texture and soil structure. The infiltration tests were carried out under relatively wet conditions, and this is why saturation of the soil could have played a role in the outcomes of the infiltration tests. The infiltration rate was lowest at the conventional plot, in fact, the maximum waiting time of 15 minutes was exceeded for all repetitions at the conventional plots (Figure 7). For the regenerative plots, the maximum waiting time was exceeded in less than half of the tests.

At time zero (T0), there is an overall differences between the soil health characteristics of the regenerative and conventional plots. This differences was expected, because the regenerative and conventional fields were managed significantly different during the past years. For a few soil nutrients, there is a difference between the regenerative tilled and regenerative no-till field. These fields have a similar management history, but the tilled field was tilled once, in spring 2023. It is unclear if this caused the difference in for example phosphorus and sulphur soil stock, or if this difference is related to spatial variability in the field.

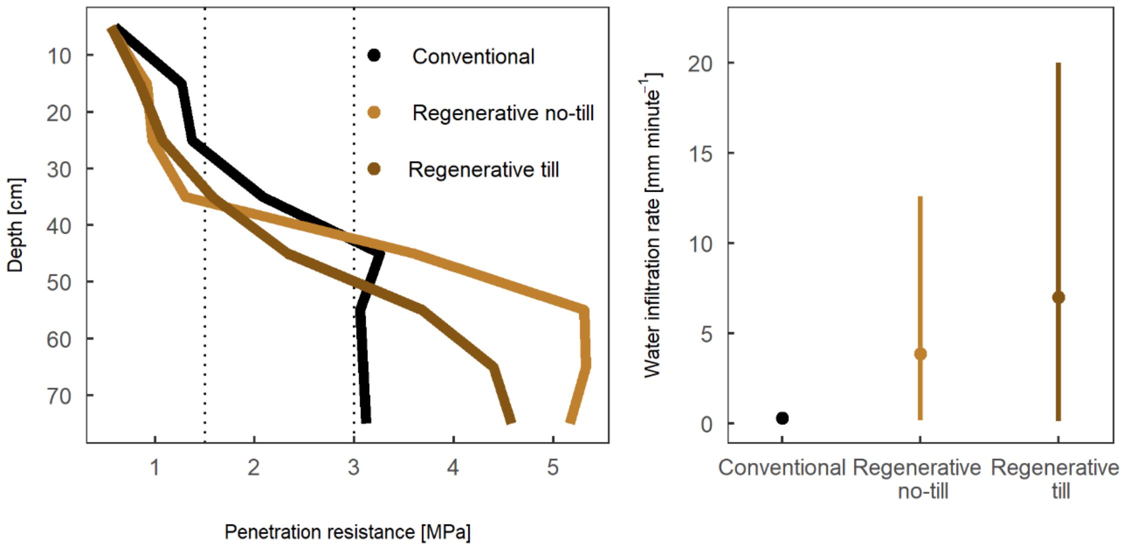

Figure 7 Penetration resistance and water infiltration at T0. *The soil penetration resistance for a soil depth of 0 – 80 cm (left) and the water infiltration speed of the top soil (right). Each line presents the mean value over 3 (conventional) and 15 (regenerative) plots. Root development is hindered above 1.5 MPa, and roots will barely or not penetrate into the soil above a resistance of 3.0 MPa. The water infiltration test were carried out in October, and are not a true T0 test.*

The difference in soil health at T0 will have implications for future analyses of the field experimental data. It means that differences between the conventional and regenerative fields could be related to both the historical agricultural practices and the studied regenerative versus conventional practices. Therefore, for the rest of this report, we focus on the change in soil characteristics compared to T0, rather than the absolute values itself. The different starting positions at T0 do not impact the analyses of different regenerative treatments, because the soil characteristics at T0 are not significantly different between different regenerative treatments. This indicates that future differences between regenerative treatments can be attributed to the studied regenerative agricultural practices.

3.2 Soil nutrients

To study the impact of regenerative practices on soil health, we compare the soil nutrient content at time zero (T0), with the contents after the first experimental season. From this comparison, it is evident that there is no, or only a minor, difference between soil characteristics at T0 and after the first experimental year (T1). This was also expected for this first year, because one year is a short period, especially for the relatively slow evolving soil processes. The plant available phosphorus (P) and plant available potassium (K) increased most strongly for both the tilled and no-till mulch treatments (Figure 8). Plant available P also increased strongly for the no-till treatment with lasagne + biofertilizer + compost tea, but the plant available K decreased slightly for most of the other regenerative treatments. The applied P and K was relatively high for the alfalfa mulch treatments, and this might explain the increase in these essential nutrients in the soil. The applied P and K was low for the treatments with biofertilizer + compost tea, and the soil P and K decreased slightly for these treatments. The soil nitrogen (N) stock increased (slightly) for all treatments, with the strongest increase in the conventional plots and regenerative mulch plots. For other macro- and micronutrients, the results differ per nutrient and per treatment. For example the magnesium stock, Na stock, and S stock increased, but the variability between different plots was almost as high as the variability between the treatments. The soil sodium (Na) stock increased most strongly for the conventional field, and available cupper (Cu) and selenium (Se) increased most strongly in the mulch field.

The soil organic matter content increased for most treatments, but it declined for the 10 – 30 cm layer of the regenerative no-till treatments with amendment of biofertilizer + compost tea, and lasagne + biofertilizer + compost tea (Figure 9). Generally, for this deeper layer, the OM increased more strongly for the tilled plots, compared to the no-till plots. Surprisingly, the conventional plots had a high increase in OM, comparable to the regenerative plots, while these plots received no additional input of OM. Also for several other treatments, there is no direct link between the added OM and soil OM content. For example, the biofertilizer and compost tea provided a low input of OM, while high increase in soil OM was found, and the OM content for the lasagne treatment is high, while a decrease in soil OM content was observed.

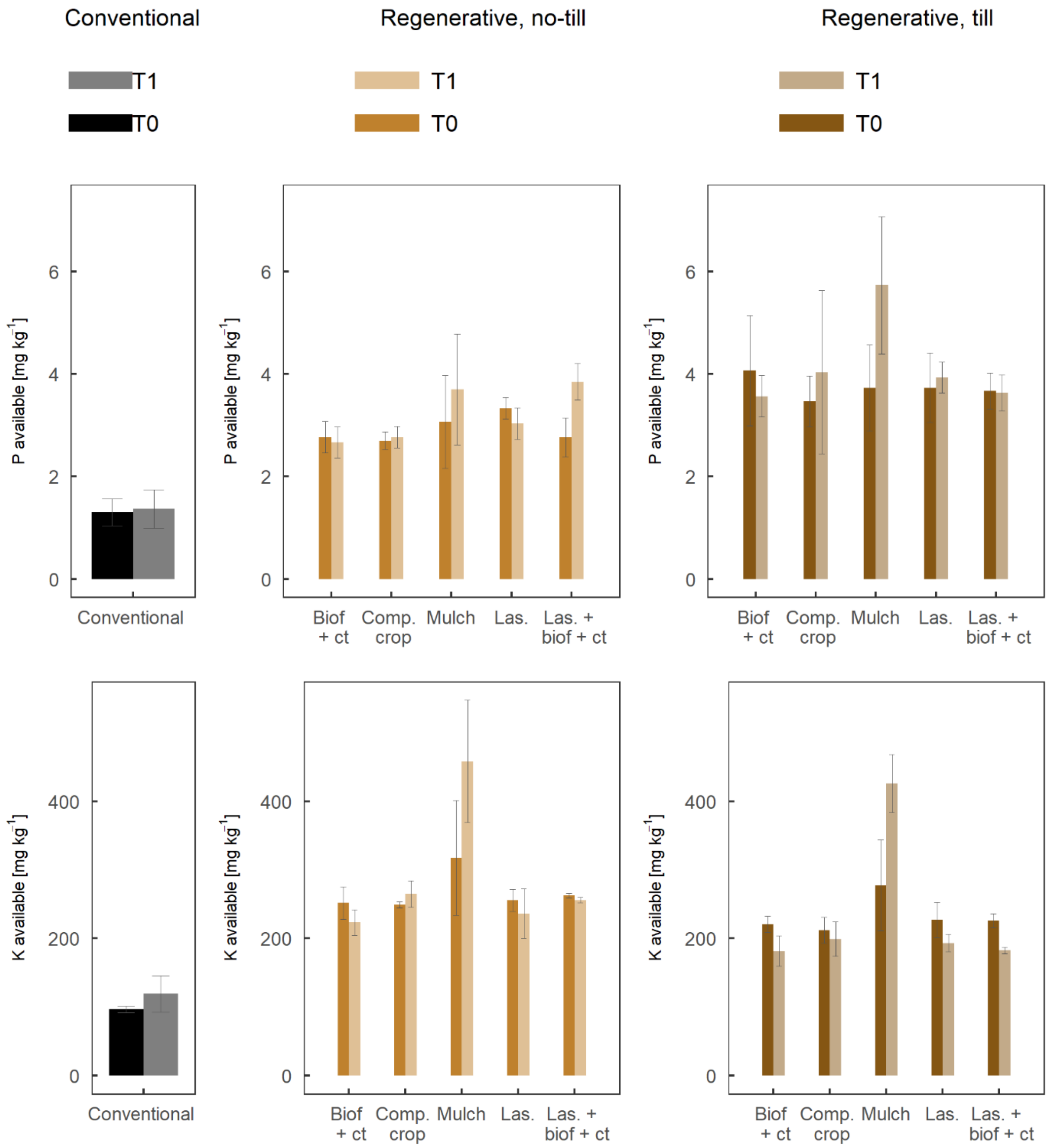

Figure 8 Change in soil PK availability*. The total soil availability of phosphorus (P) and potassium (K) for the top soil (0 – 10 cm depth) per treatment. Each bar represents the mean value for each treatment (three plots per treatment) for time zero (T0, left bar) and autumn measurement (T1, right bar). The grey errorbar gives the standard deviation over the three plots. N availability is not shown, because it was not determined at T1.*

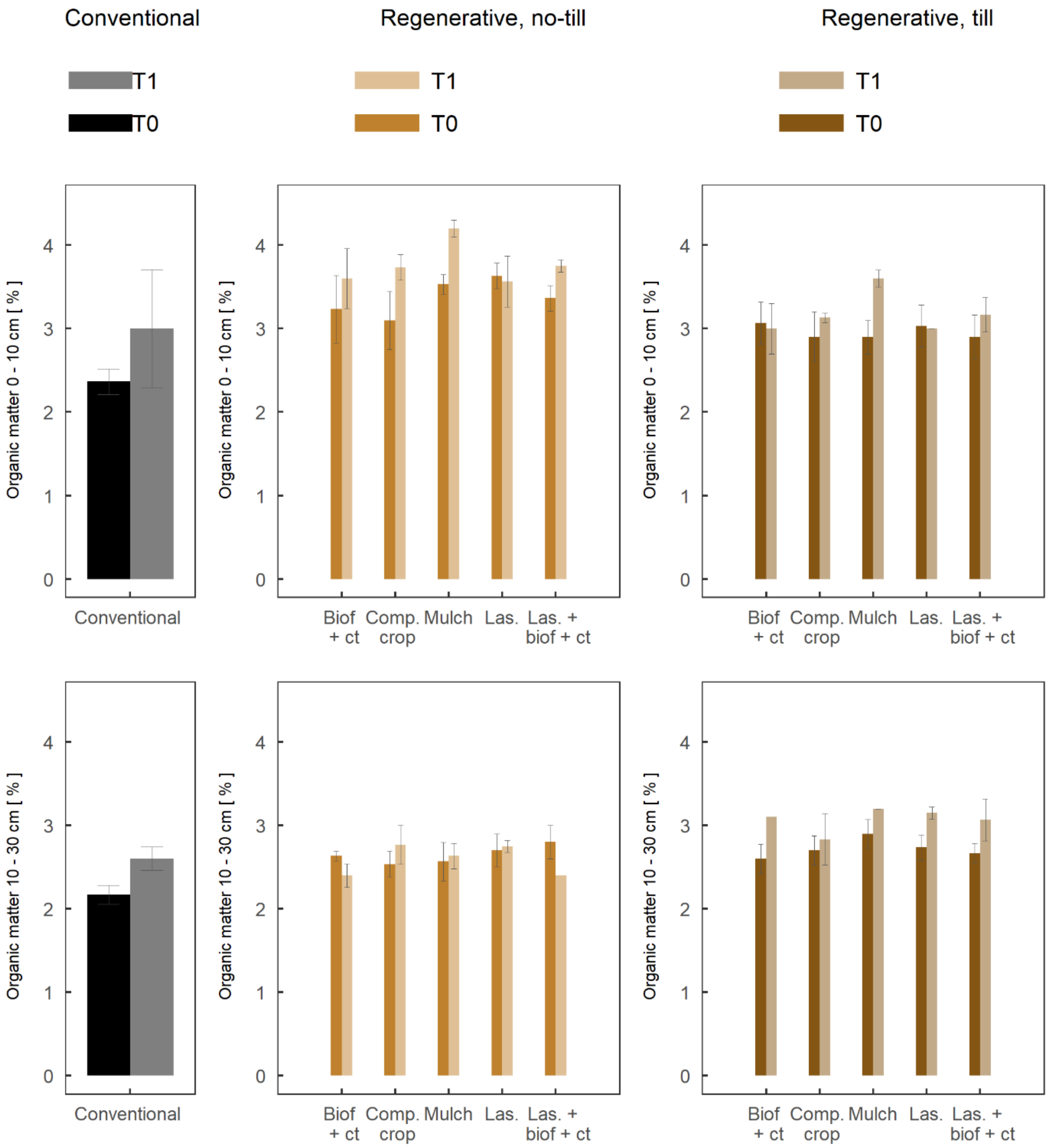

***Figure 9 Change in organic matter content**. The total soil organic matter (OM) content for the top soil (0 – 10 cm depth) and deeper layer (10 – 30 cm) per treatment. Each bar represents the mean value for each treatment (three plots per treatment) for time zero (T0, left bar) and the autumn measurement (T1, right bar). The grey errorbar gives the standard deviation over the three plots.*

3.3 Soil Biology

The microbial soil life is well developed in all fields. The microbial biomass was 326 mg kg^{-1} at T0, and this high in comparison to the reference for arable land on clay (107 – 417 mg C kg^{-1} [2]) (Figure 10). The microbial biomass is higher at the no-till plots (355 mg kg^{-1}) compared to the till plots (330 mg kg^{-1}), and highest in the mulch treatments (366 mg kg^{-1}). Also the fungal biomass (mean: 115 mg kg^{-1}) is high for arable land (reference values: 13 – 163 mg kg^{-1}), with the highest values for the regenerative, no-till field (125 mg kg^{-1}), followed by the regenerative till field (107 mg kg^{-1}), and the conventional field (99 mg kg^{-1}). The mean fungal-to-bacterial ratio is 0.91, which is a relatively high value for arable land. A high ratio (ratio > 1.0) indicates a relatively stable soil ecosystem with little disturbance. There is no difference between the different fields, which is surprising, because the no-till field is less disturbed in terms of tillage than the regenerative till, and conventional field.

[2] M., Elsen, E. van den, Haan, J. de, Visser, S., Elsen, E. van den, Haan, J. de, & Visser, S. (2019). Bodemkwaliteitsbeoordeling van landbouwgronden in Nederland : indicatorset en sys-tematiek (Versie 1.0, Ser. Stichting Wageningen research rapport, wpr-795). Stichting Wageningen Research.

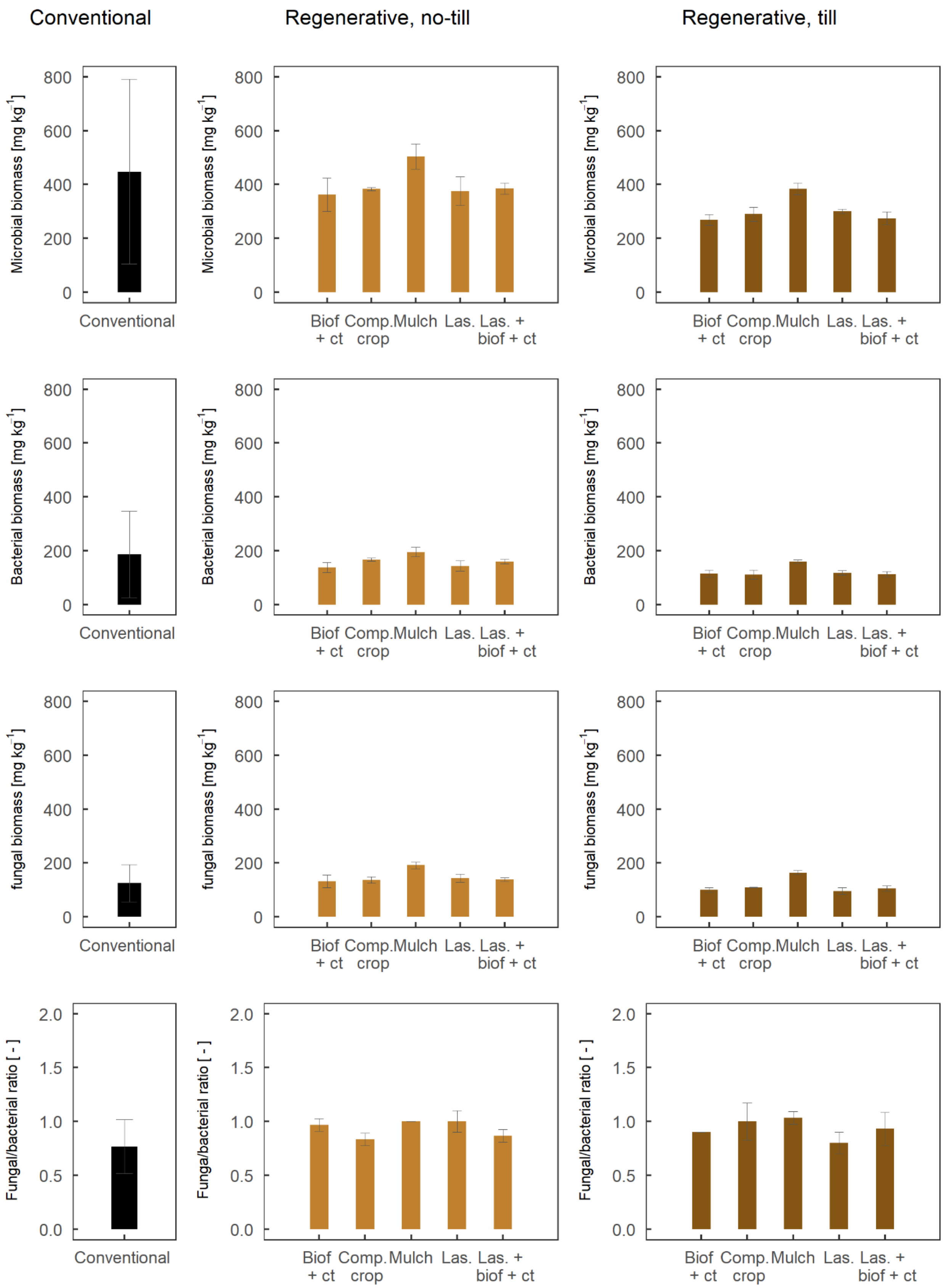

Figure 10 Soil biology. *The soil microbial biomass, bacterial biomass, fungal biomass, and fungal/bacterial ratio for 0 – 10 cm depth per treatment. Each bar represents the mean value for three plots per treatment. The grey errorbar gives the standard deviation over the three replicates.*

3.4 Plant leaf nutrients

The nutrient content of leaves gives insight in the uptake of nutrients by the plant during the whole growing season. The leaf content of total nitrogen (N), nitrate (NO_3^-), phosphorus (P), and potassium (K) per treatment is given in Figure 11. The high K content for the companion crop and mulch treatments immediately stands out. The alfalfa mulch applied relatively high amounts of K, and the soil K content was high. This might explain the high K uptake by the turnip plants. The leaf content of N and NO_3^-, is low for the companion crop treatment, and this suggests that N limitation (because of the competition with the companion crop) could have limited crop growth (see paragraph 4.3). The total leaf N content was highest for the conventional treatments, and NO_3^- was highest for the mulch treatment and lowest for the companion crop treatment. In summary, there are a few differences in plant NPK content between the treatments, and these differences can partly, but not fully, be explained by soil nutrient content.

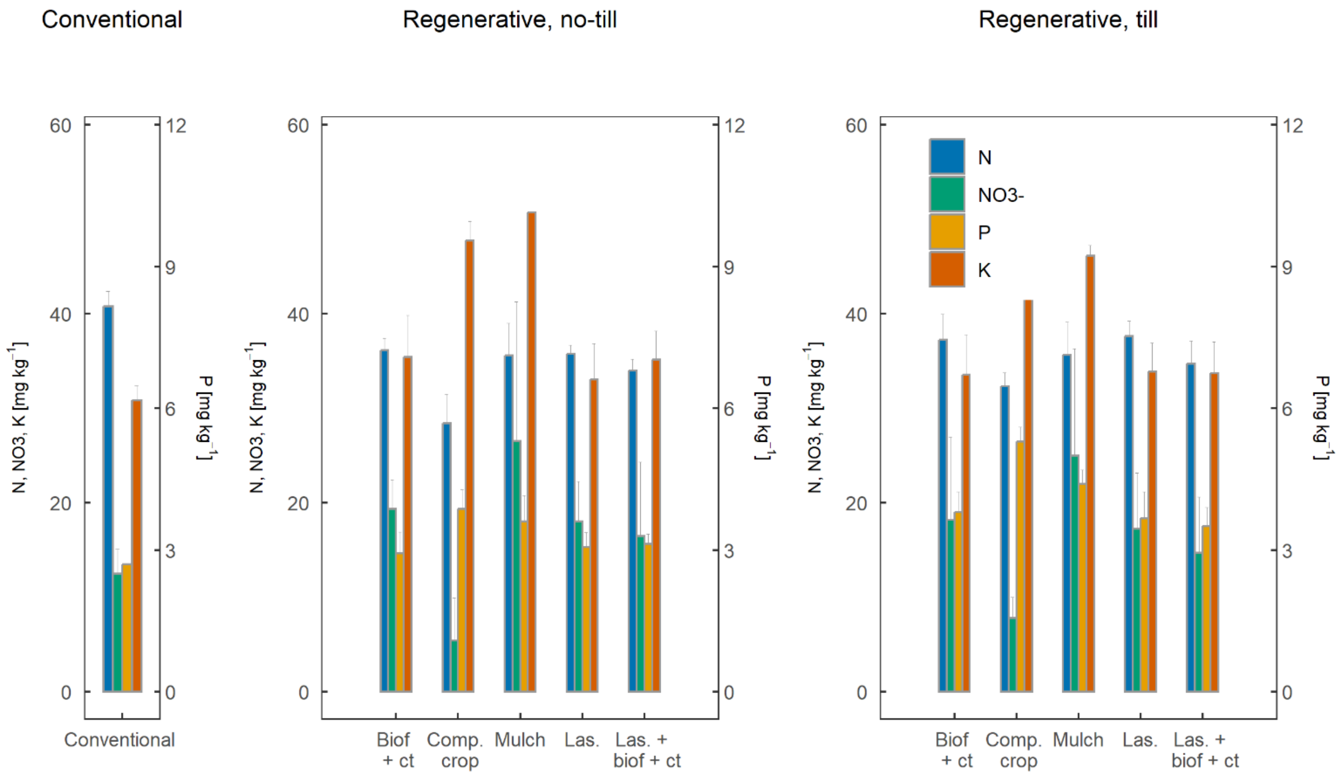

***Figure 11 Plant nutrient content**. The plant leaf content of nitrogen (N), nitrate (NO3-), phosphorus (P), and potassium (K) per treatment. Each bar represents the mean value for each treatment (three plots per treatment), and the grey errorbar gives the standard deviation over the three plots.*

3.5 Crop yield

Figure 12 High and low turnip harvest. *For the yield measurements, for each plot, two rows of 0,5 m length were harvested and weighted. The two pictures reflect a plot with high and low yield: (above)a regenerative no-till plot with lasagne, compost tea, and biofertilizer, and (below) a conventional plot.*

The turnip yield ranges from as low as 7.7 tons ha^{-1} for the conventional treatment to 84 tons ha^{-1} for the regenerative no-till treatment with amendment of lasagne, compost tea, and biofertilizer (Figure 12, Figure 13). Comparing the regenerative till and no-till reveals that the mean yield was 35% higher for the no-till plots than the till plots. Within the regenerative treatments, the input of lasagne + biofertilizer + compost tea gave the highest yield, followed by the separate inputs of lasagne or the input of biofertilizer + compost. The yield for the treatment with companion crops was significantly lower than all other regenerative treatments, and this was probably related to the late sowing and therefore high competition with the companion crops.

It should also be noted that because of the difficulty of sowing the fine seeds by hand, the sowing/plant density was not the same throughout the plot, and the turnips for the conventional field were sown and harvested a few days later than the regenerative plots. This makes it harder to make a fair comparison of the yields and the results should be interpreted with care.

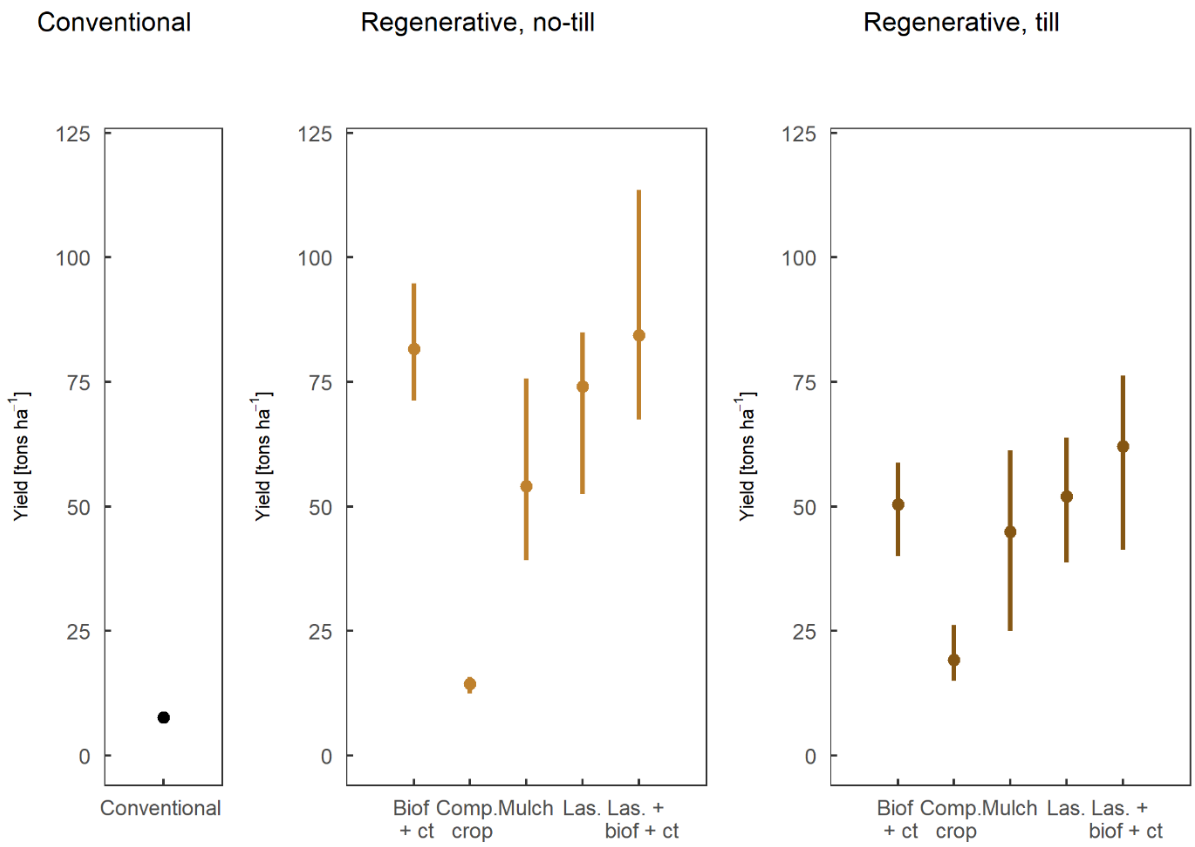

Figure 13 Crop yield. *The total crop yield for the conventional (left), regenerative no-till (middle) and regenerative till (right) plots. The point presents the mean value over 3 plots per treatment, and the range shows the minimum to maximum value.*

3.6 Crop nutrients

The nutritional analyses of the turnips are available in Table 1. For several nutrients, especially most amino acids, turnip nutrient values were below the detection limit. This is not related to the experiment; turnips usually contain low quantities of amino acids and specific vitamins. Overall, we find the highest nutritional value for the conventional plots. For the main nutrient groups, we find the following results:

The *macronutrients* carbohydrates and proteins are highest in turnips from the conventional treatment. While proteins are lowest for the companion crop treatment (both till and no-till), carbohydrates and sugars are relatively high for this treatment. Also several other macronutrients (calcium, phosphorus, sodium and magnesium) have the highest density in the conventional plots. Only the mean potassium (K) content is higher in the regenerative plots.

For the *micronutrients* the zinc, copper, and manganese, nutrient contents were below the detection limit for all treatments. The iron content could be measured, and iron was significantly higher for the conventional treatment compared to the regenerative treatments. The regenerative till turnips has significantly higher iron content compared to the regenerative no-till plots.

For most of the amino acids, the crop content was below the detection limit, except for tryptophan, one of the essential amino acids. Tryptophan values were highest for the conventional plots, and there was no difference between the regenerative till and regenerative no-till field. Vitamins B4, B11, and C are highest for the conventional plots, and there is no significant difference in vitamin content between the regenerative plots (Figure 14). For all plots, the vitamin B1 and B2 content were 0,2 mg kg-1, and the contents for vitamin A, B3, B5, and E were below the detection limit of 0,1 mg kg-1 (vit. A), 5 mg kg-1 (vit. B3), and 10 mg kg-1 (vit. B5), and 1 mg kg-1 (vit. E).

Table 1 Crop nutrition. *Crop content for various macro- (in red) and micronutrients (in orange), amino acids (in blue), and vitamins (in green). Nutrients that have non-detectable low values are not included in the table. The macronutrients and amino acids are measured in g 100 g^{-1}, iron, vitamin B4 and vitamin C are measured in mg kg^{-1}, and vitamin B11 is measured in μg kg^{-1}.*

	Conventional	**Regenerative, no-till**					**Regenerative, till**				
Treatment	Conven-tional	Bio-fertilizer + compost tea	Com-panion crop	Mulch	Lasagne	Lasagne + Biofertilizer + compost tea	Bio-fertilizer + compost tea	Com-panion crop	Mulch	Lasagne	Lasagne + Bio-fertilizer + compost tea
Proteins	1.1	0.8	0.6	0.7	0.9	0.80	0.90	0.67	0.8	0.9	0.9
Carbohydrates	3.3	2.4	2.8	2.1	2.5	2.3	2.4	2.9	2.3	2.4	2.6
Fructose	1.3	1.0	1.0	0.9	1.1	1.0	1.0	1.2	0.9	1.0	1.1
Glucose	1.7	1.3	1.6	1.2	1.4	1.2	1.2	1.6	1.3	1.2	1.4
Total sugars	3.0	2.3	2.6	2.0	2.4	2.2	2.2	2.8	2.2	2.2	2.5
Calcium	0.051	0.037	0.043	0.045	0.041	0.040	0.039	0.034	0.041	0.038	0.038
Phosphorus	0.033	0.022	0.028	0.024	0.025	0.024	0.030	0.035	0.034	0.032	0.031
Potassium	0.30	0.28	0.27	0.31	0.32	0.32	0.32	0.27	0.34	0.35	0.34
Sodium	0.012	0.0077	0.0050	0.0067	0.0073	0.0063	0.0050	0.0050	0.0063	0.0067	0.0053
Magnesium	0.012	0.0077	0.0083	0.0080	0.0083	0.0080	0.0087	0.0083	0.0093	0.0093	0.0087
Iron	21	11	11	10	12	13	16	14	18	18	14
Tryptophan	0.010	0.0063	0.005	0.0053	0.0073	0.0063	0.0067	0.0060	0.0057	0.0067	0.0070
Vit B4	199	110	96	77.7	108	89	96	90	99	103	103
Vit B11	370	263	244	173	226	204	172	323	184	114	139
Vit C	199	91	125	83	83	82	80	133	102	85	96

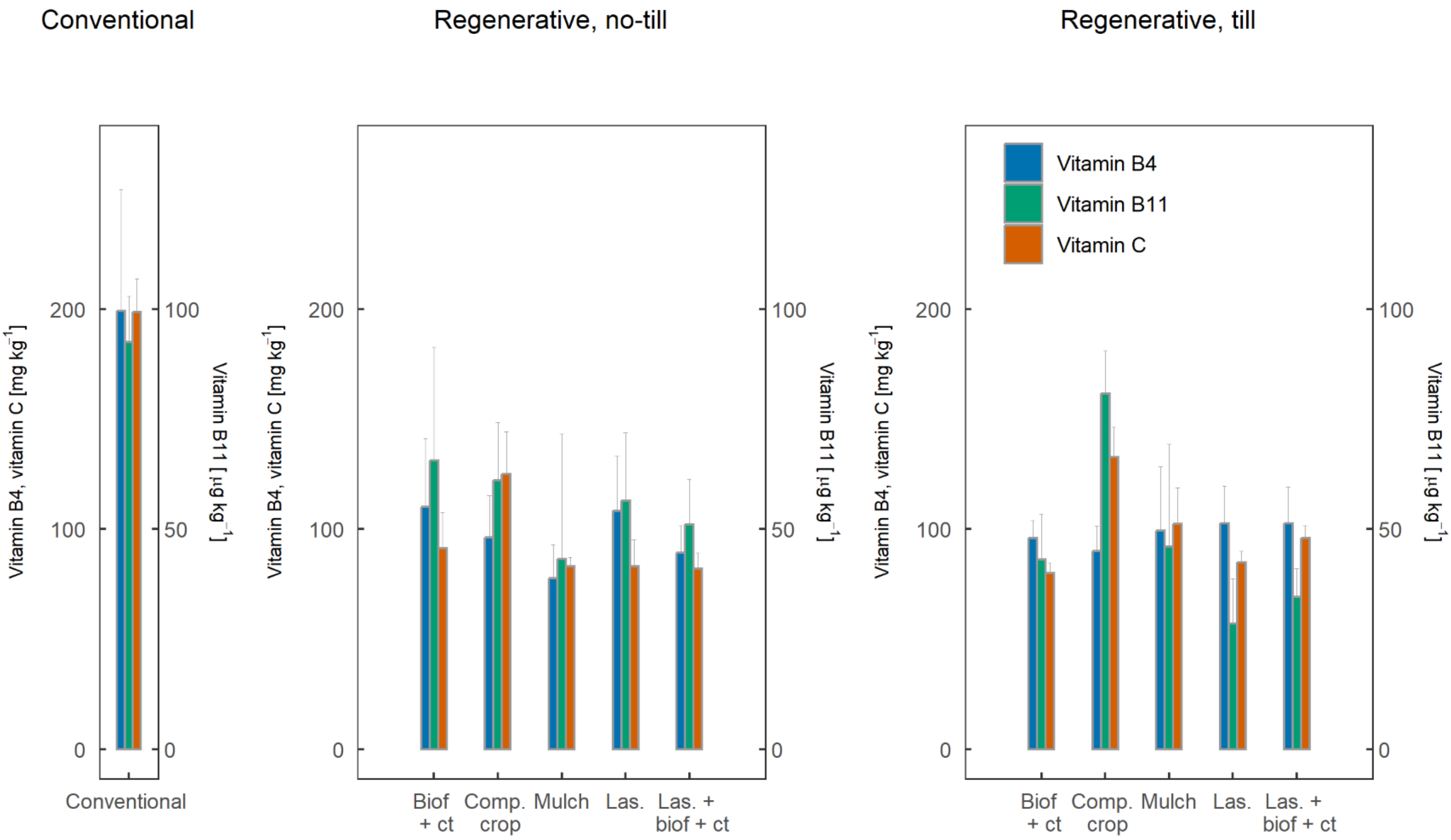

Figure 14 Crop vitamin content. *The crop content of vitamin B4, vitamin B11, and vitamin C per treatment. Each bar represents the mean value for each treatment (three plots per treatment), and the grey errorbar gives the standard deviation over the three triplets.*

Treatments with a low yield generally had a high nutrient density. For example, the yield is lowest for the conventional plot, followed by the regenerative treatments with companion crop. The crop nutritional value is highest for the conventional plot, and above average for the companion crop treatments. The nutrients might be more condensed in the small turnips, compared to bigger turnips, and this hypothesis is supported by the lower dry matter content for regenerative plots (around 10%), compared to 15.5% for the conventional plots.

The turnip nutrient density does therefore not provide any information about the difference in total amount of nutrients delivered by the soil. Multiplication of the yield with nutrient density provides information about the total amount of nutrients that were harvested. For vitamins B4, B11, and C, the total harvested weight is lowest for the conventional treatment and regenerative treatment with companion crop (Figure 15). The regenerative no-till treatments biofertilizer + compost tea, lasagne, and lasagne + biofertilizer + compost tea had a high total yield of vitamins.

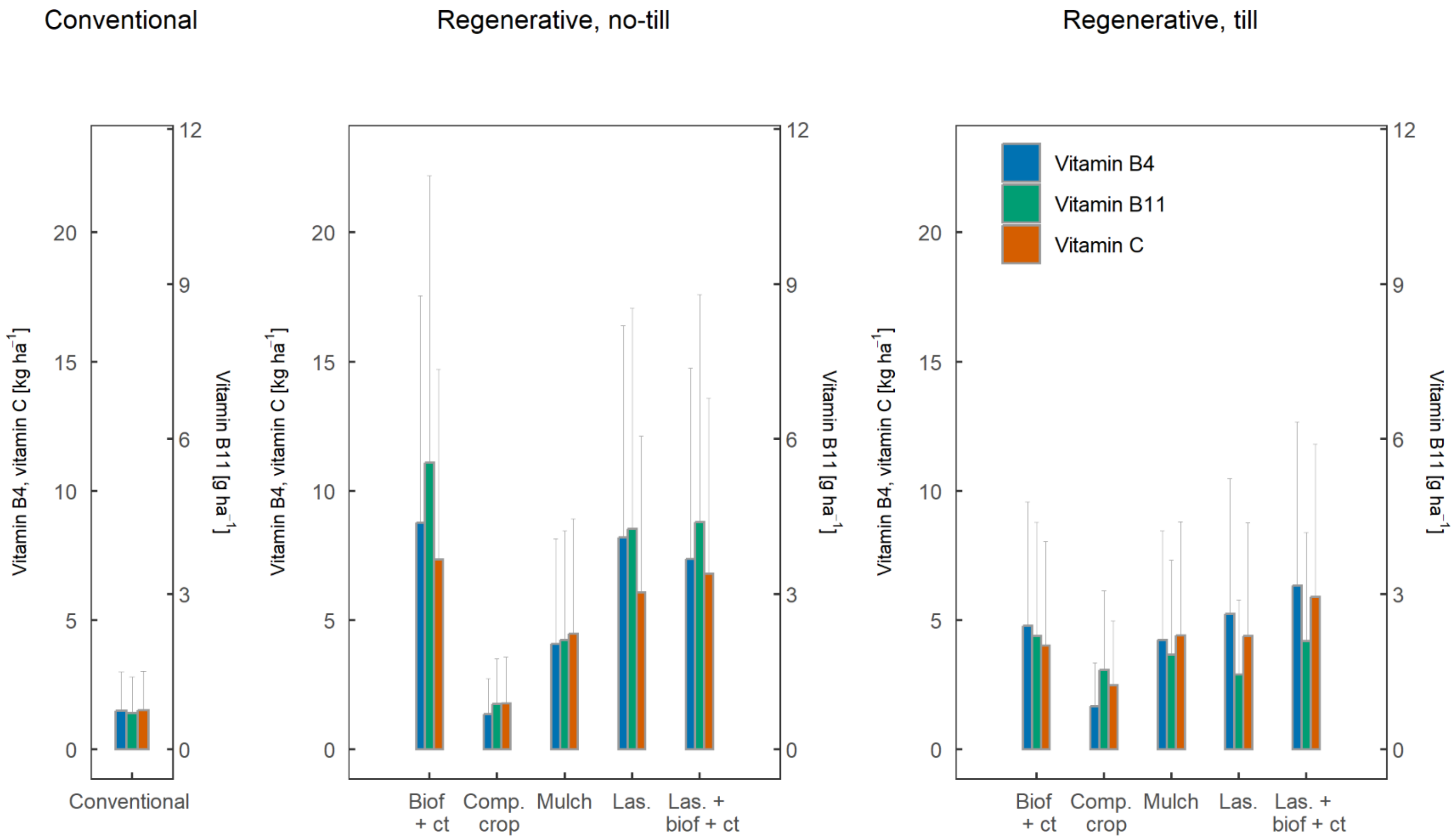

Figure 15 Total harvested vitamin weight *(crop yield * nutrient density). The total harvested amount of vitamin B4, vitamin B11, and vitamin C per treatment. Each bar represents the mean value for each treatment (three plots per treatment), and the grey errorbar gives the standard deviation over the three triplets.*

There is no relation between the soil nutrient levels and turnip nutritional value. For example, soil available phosphorus (P) is lowest for the conventional treatment, while turnip P content is high for the conventional plots. On the other hand, soil magnesium (Mg, both stock and available) is lowest for the conventional fields, while turnip Mg content is highest for the conventional field.

4 Comparison Eurofins, Bruker TRACER, MicroBiometer

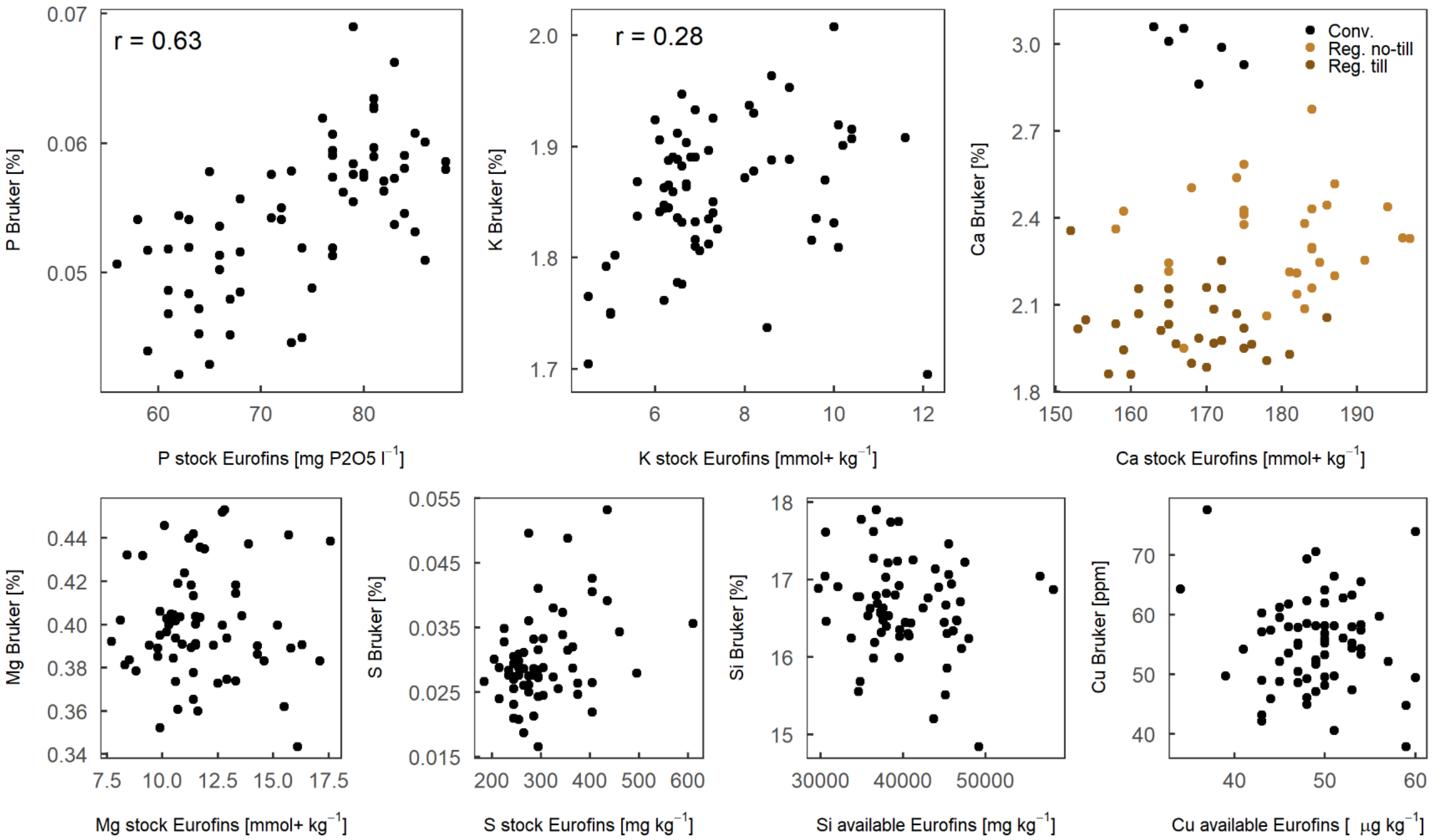

Figure 16 Comparison Eurofins and Bruker data. *Scatter plots of the lab analyses by Eurofins and the on-site Bruker TRACER analyses for phosphorus (P), potassium (K), Calcium (Ca), magnesium (Mg), sulphur (S), silicon (Si), and cupper (Cu). For P and K, there is a significant correlation between these two measurements, and the correlation coefficient is in the scatter plot. For the other elements, there is no correlation between the Eurofins and Bruker TRACER analyses. The Eurofins soil stock is given, except for Si and Cu, because the total soil stock was not measured.*

For this report, the Eurofins laboratory analyses were analysed and discussed. On the side, the Bruker TRACER and MicroBiometer were tested within the experiment. The Bruker TRACER allows for on-site determination of soil nutrients, which makes this a cheap and fast methodology. The MicroBiometer allows for on-site determination of microbial biomass and fungal to bacterial ratio, that allows for a quick determination of the soil health.

For phosphorus (P) and potassium (K), there is a correlation between the Eurofins and Bruker value, but for other elements calcium (Ca), magnesium (Mg), sulphur (S), silicon (Si), and cupper (Cu), there is no significant correlation (Figure 16) This indicates that the Bruker TRACER method cannot simply replace the Eurofins laboratory analyses. For Ca, the Bruker TRACER detects a strong difference between the conventional and regenerative field, which is not found by Eurofins. This suggests that other factors, for example soil texture, could play a role in the Bruker TRACER estimation of soil nutrients. The Bruker methodology does have one strong advantage over Eurofins data: the Eurofins analyses for this experiment cannot be used to detect any differences in soil

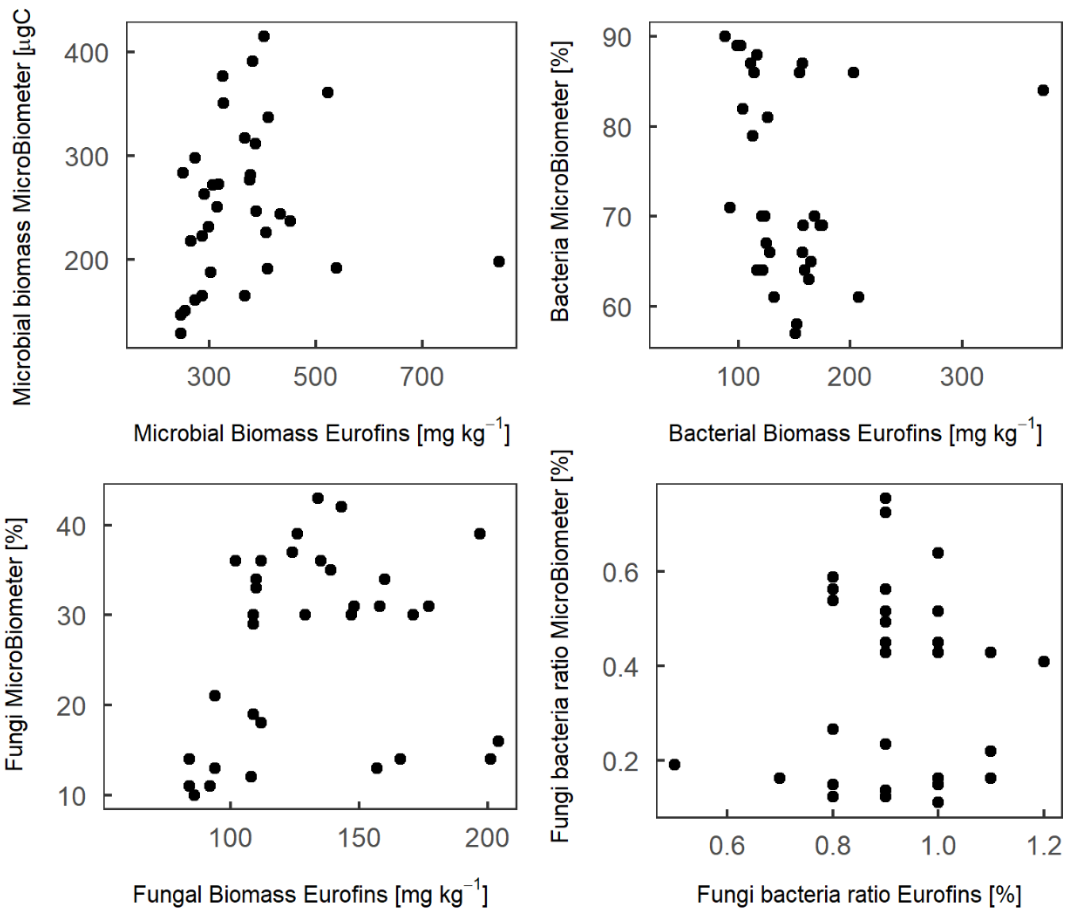

Figure 17 Comparison Eurofins and MicroBiometer data. *Scatter plots of the lab analyses by Eurofins and the on-site MicroBiometer analyses for microbial biomass, bacterial biomass, fungal biomass, and fungi bacteria ratio. For the biological indicators, there is no correlation between the Eurofins and MicroBiometer data.*

Manganese (Mn), Cobalt (Co), Iron (Fe) and Zinc (Zn), because all values are below the detection limit. However, the Bruker methodology is able to distinguish between nutrient values at these low contents.

For biological parameters, there is no correlation between the Eurofins results (analysed by the NIRS methodology) and the on-site analyses with the MicroBiometer the (Figure 17). The estimation of microbial biomass, bacterial biomass, fungal biomass, and fungi bacteria ratio by the Eurofins laboratory and MicroBiometer are not positively correlated.

5 Issues

5.1 Wet field conditions

The autumn, winter, and spring of 2023 – 2024 were very wet; cumulative rainfall from October to May was 915 mm, which is significantly higher than the mean rainfall of 562 mm (Figure 18). October 2023 and May 2024 were even the wettest October and May since the start of measurements in 1974. The wet conditions did not allow a T0 field campaign in November 2023, and therefore, the T0 measurements were postponed until January 2024. In January, the field was too wet to perform infiltration tests, so these were conducted after the first experimental season in autumn 2024. Thus, the T0 field campaign is from a slightly different season than all subsequent field campaigns, and the infiltration test is not a true T0 measurement.

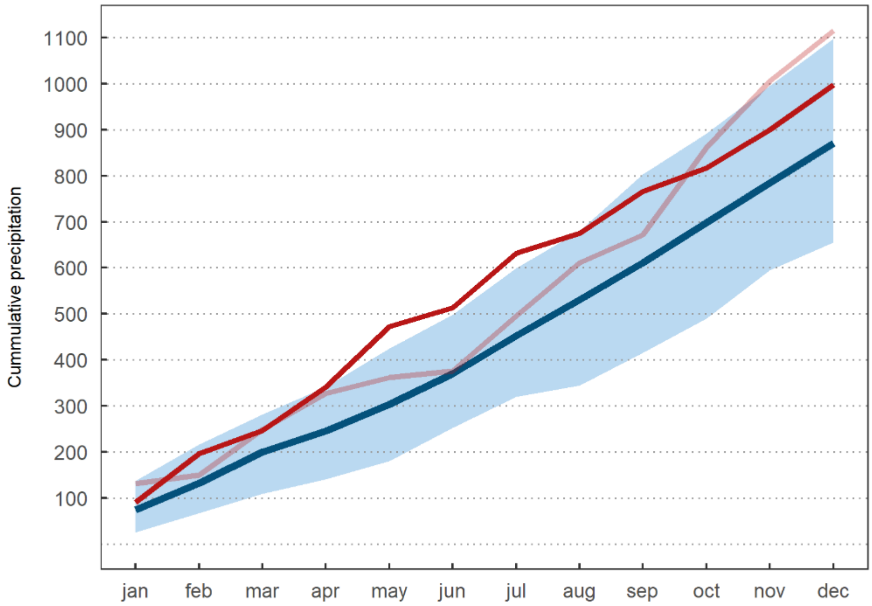

Figure 18 Cumulative precipitation. *Cumulative yearly precipitation since 1974. The blue range indicates the 5 - 95% driest to wettest years, and the blue line is the mean precipitation from 1974 - 2024. The year 2023 (light red) and 2024 (dark red) show that autumn 2023 and spring 2024 were very wet. The rainfall data is from the PPO station of WUR in Westmaas, 2 km from study site.*

5.2 Turnips replaced soya

The experiment was set-up with a crop rotation of wheat, soya, potato, and wheat, with soya in 2024. However, after sowing, and after sowing the second time, all soya beans and sprouts were eaten by hares and pigeons. The short growing season did not allow for a third reseeding with soya. Instead, turnip was sown for the experiment because it has a short growing season. The turnips for the companion crop and mulch treatments were sown by hand and hey have an irregular seeding distance. This could have impacted the yield determination. Also, the field was bare for much longer than planned (and longer than common for regenerative agriculture), and there was more movement of machinery in the field, which could have compacted the soil.

5.3 Change in conventional field

The conventional plots were located on a conventional field with similar soil conditions and similar crop rotation as the regenerative field. Unfortunately, from 2025 onwards, the conventional plot can no longer be used by Klompe Agri and the Soil Heroes foundation for the regenerative experiment. A new field was selected for the continuation of the experiment, but this field is different in several soil characteristics (Figure 19). The soil pH is very similar, but the new conventional plots are more sandy, they have a lower soil stock of N, K, S, and Mg, but a higher P stock. The new conventional fields were sampled in autumn (for chemical analyses at the Eurofins laboratory), and these data are considered the T0 measurement for the new conventional plots.

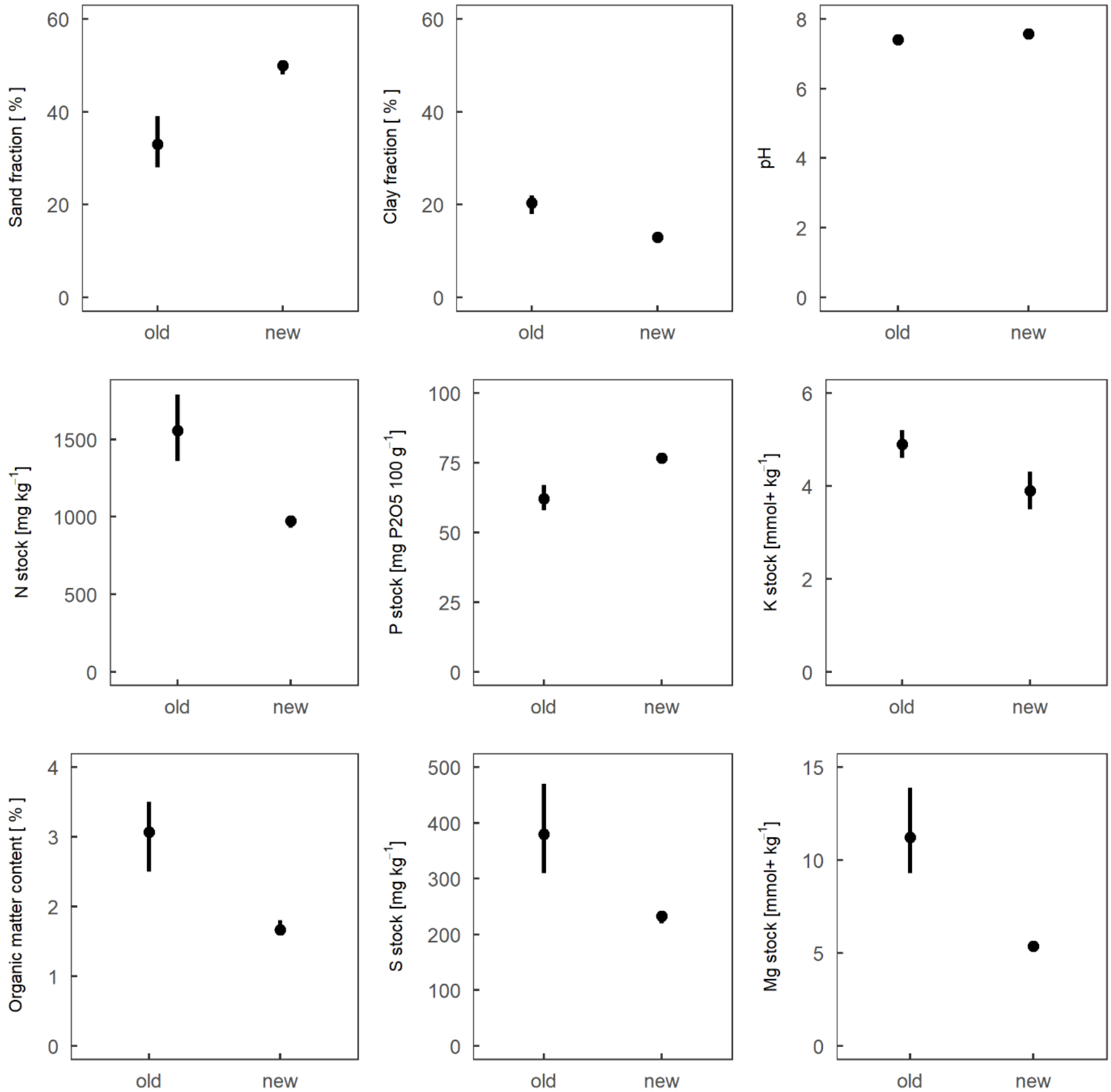

Figure 19 Comparison old and new regenerative plots. *The sand fraction, clay fraction, pH, total soil stock nitrogen (N), phosphorus (P), potassium (K), organic matter content, and total stock of sulphur (S) and magnesium (Mg). The point presents the mean value over 3 old and 3 new conventional plots, and the range shows the minimum to maximum value.*

6 Conclusion

After the first full experimental season of the regenerative agricultural experiment of the Soil Heroes Foundation, we can draw the following first conclusions:

1. The baseline soil characteristics for the regenerative field and conventional fields are different. The natural spatial variability and / or the different management history resulted in a different starting position for the treatments, and this should be considered in future analyses. There are no or only minor differences between the different regenerative treatments.
2. For phosphorus (P) and potassium (K), supply through (regenerative) amendments increased soil P and K content. For most other macro- and micronutrients, no impact of regenerative management can be observed. But no or only few changes were expected after only the first experimental year.
3. The regenerative farming practices had some impact on leaf nutrient density. For example, the mulch and companion crop treatments had a very high leaf K content. These observed differences can partly, but not fully, be explained by soil nutrient density. The yield was lowest in the conventional field, followed by the regenerative till, and regenerative no-till field. The conventional turnips, with a low yield, had the highest nutrient density for macro-nutrients and vitamins. The crop yield might (partly) explain the difference in nutrient density. The total amount of nutrients harvested from the soil is highest for the no-till treatments with lasagne and/or compost tea and biofertilizer.
4. The on-site determination of soil nutrients and soil biology by the Bruker Tracer and MicroBiometer show little agreement with the data of the Eurofins laboratory. Future analyses will focus on the Eurofins data, because most of their methods have been accredited based on the KWALIBO-regulation. Many more on-site, quick, and / or cheap methodologies exist, and it is very interesting to test those within the Soil Heroes experiment. For the next years, we recommend to focus on those methods that communicate transparently about their methodology, and that been tested for agricultural soils in the Netherlands.

The first full experimental year got hindered by too much rain and a last-minute switch from soya to turnips. Nevertheless, this first year provided very relevant insights in the (differences in) baseline soil characteristics for the different fields. Furthermore, interesting first results suggest that (regenerative) soil amendments might impact soil and plant nutrient density, and that different regenerative treatments can have a big impact on crop yield. The coming experimental years will be used to study the relationships between regenerative practices and soil health and soil, plant, and crop nutrient density in more depth. Future analyses will also be very interesting to confirm and explain the effects that were found in this first full experimental year.