Environment. Technology. Resources. Rezekne, Latvia
Proceedings of the 14th International Scientific and Practical Conference. Volume 1, 101-107

Influence of Intercropping on Soil Microbial Activity and Strawberry Development

Valda Laugale
Unit of Agronomic Research and Variety Testing
Institute of Horticulture
Dobele, Latvia
valda.laugale@llu.lv

Līga Lepse
Unit of Agronomic Research and Variety Testing
Institute of Horticulture
Dobele, Latvia
liga.lepse@llu.lv

Solvita Zeipiņa
Unit of Agronomic Research and Variety Testing
Institute of Horticulture
Dobele, Latvia
solvita.zeipina@llu.lv

Abstract. **Strawberry is one of the most important berry crop grown around the world and their consumption increases every year. The introduction of new practices promoting farm sustainability and long-term soil health in strawberry production systems is very essential. Towards environment- and climate-friendly farming practices intercropping can be used to improve soil microbiological activity and biodiversity, and reduce the use of pesticides and mineral fertilizers, while the right choice of intercropped plants is of great importance to achieve these goals. The trials on strawberry intercropping were established in Latvia in 2021. Three treatments with different intercropping plant rotations, including crimson clover, pea, garlic, marigold, and winter rye mix with vetch, were compared to conventional strawberry growing using straw mulch. Trial was installed in three locations: two organic farms and the Institute of Horticulture (LatHort). Thus representing climatically different regions and different soil conditions. Strawberries were grown in 1.2 m distant rows, where in the intercropping treatments, each second interrow was occupied by companion plants. Soil microbial activity was evaluated during vegetation seasons by determining soil respiration rate (SRR) and dehydrogenase activity (DHA) several times per season in 2021 and 2022. Strawberry vegetative development was evaluated at the end of each vegetation season. During the investigation period, soil microbial activity fluctuated during vegetation seasons, depending on growing conditions. In 2021, SRR varied from 1.9 – 3.3 CO_2 mg L^{-1}, while in 2022, from 2.1 - 3.4 CO_2 mg L^{-1}. DHA varied from 46 – 134 INTF, $\mu L \times L-1 \times h$ in 2021 and 60 – 101 INTF, $\mu L \times L-1 \times h$ in 2022. Intercropping had low influence on microbial activity and results differed within each location. Strawberry plant biomass differed among locations and treatments with the highest above-ground biomass observed in LatHort during second growing season in conventional growing system (790 g $plant^{-1}$).**

Keywords: companion plants, DHA and SRR, Fragaria x ananassa Duch., plant biomass.

I. Introduction

Strawberry is one of the most widely grown and consumed small fruits in the world with a tendency to increase. In 2021, according to FAO data, the total world production exceeded 9175384 t [1].

The development of growing technologies, giving higher income per unit of area and/or reduce pest and disease problems as well as improve soil properties, is of great importance nowadays and are in line with producers' possibilities and needs in implementing environment-friendly and sustainable farming practices. One of growing practices towards sustainability is intercropping. Intercropped plants can serve as biological and physical barriers against pests and diseases [2]. They can suppress weed growth [3], improve soil nitrogen content [4], positively influence the physical condition of the soil as well as growth and crop characteristics of the plants, improve the quality of crops, and the organic substance introduced into the soil restores its aggregate structure and fertility [5]. The right choice of intercropping plants is very important to obtain a positive effect not only on the main crop, but also on soil properties. Previous research on strawberry was mostly concentrated on strawberry – legume intercropping [6]-[9], while less attention was paid on strawberry relationship with other plants. In Nordic countries, strawberries mostly are grown as perennial crops [10] that allow some plant rotation in interrows during growing period of several years. This research was carried out to evaluate some intercropping plant rotations in strawberry plantation to determine their impact on strawberry growth and soil microbial activity.

Print ISSN 1691-5402
Online ISSN 2256-070X
https://doi.org/10.17770/etr2023vol1.7236

II. MATERIALS AND METHODS

The trial on strawberry intercropping was established in May of 2021 at the Institute of Horticulture (LatHort) Pūre Research Centre (57°05'25"N, 22°90'55"E) and on two organic farms - Atvases (56°80'27"N, 24°41'83"E), and LM Product (56°70'56"N, 24°89'68"E).

Experimental design and treatments. Strawberries (cultivar 'Malwina') were planted in rows with a planting distance 1.20 m between rows and 0.4 m between plants in rows. In the trial, three treatments with different intercropping plant rotations were compared to conventional growing with application of straw mulch. In treatments with intercropping, companion plants were grown in every second interrow, while other interrows were mulched by straw. Treatments: 1) 1st growing year – Crimson clover (*Trifolium repens* L.) in interrows; 2nd growing year – marigolds (*Calendula officinalis* L.) following by winter rye (*Secale cereale* L.) + winter vetch (*Vicia villosa* L.) in the autumn; 2) 1st growing year – Crimson clover; 2nd growing year – green peas (*Pisum sativum* L.) following by winter rye + vetch; 3) 1st growing year – Crimson clover following by winter garlic (*Allium sativum* L.) planted in autumn; 2nd growing year – winter garlic following by winter rye + vetch; 4) control, without intercrops, straw mulch applied in all interrows. The technological processes (sowing, weeding, watering, cultivation) were performed according to the local conditions and technologies according to organic farming principles.

Each plot included 4 rows of strawberry of 6 m length with 15 strawberry plants in row and 4 interrows. Plots were arranged randomly in the trial field in four replicates.

Soil characteristics and meteorological data. All three trial locations differed not only by different geographical location, but also by soil conditions (Table 1).

TABLE 1 THE SOIL TYPE AND CHARACTERISTIC IN TRIAL LOCATIONS BEFORE ESTABLISHMENT

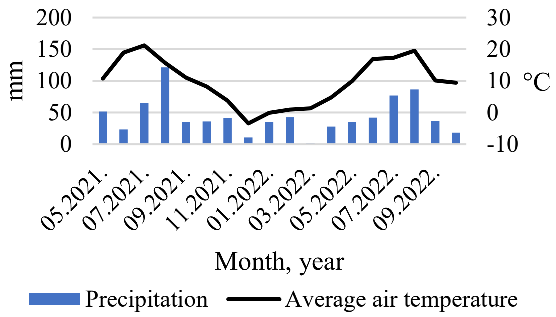
Location	**Soil type**	**pH KCl**	**Organic matter, %**	**P_2O_5, mg kg^{-1}**	**K_2O, mg kg^{-1}**
LatHort	Loamy sand	5.8	2.5	58	71
Atvases	Loamy sand	6.0	11.7	<14	82
LM Product	Clay loam	5.7	3.4	123	95

LM Product had the most heavy soil among trial locations, whereas Atvases is characterized by soil with high organic.

All three locations were characterized by low content of plant available mineral nutrients, therefore the basic fertilization was applied before establishment of trial, by using organically certified fertilizers. The fertilizer Physio Natur PKS 47 (0-13-15) with a dose 66 g m^{-2} was applied in farm LM Product. In the farm Atvases, fertilizers PHYSALG 25 with a dose 60 g m^{-2} and potassium magnesium (Patentkali) with a dose 50 g m^{-2} were applied. In LatHort, the organic fertilizer FERTIPLUS (4-3-3 65 OM) was applied with a dose 230 g m^{-2}. In the next year in LatHort and LM Product, strawberries were additionally fertilized by cattle slurry with a dose 1.2 L per strawberry row meter, which was diluted by water. No side-dressing was done in farm Atvases.

Meteorological data were recorded in two locations: LatHort in Pūre and LM Product, by using an automatic weather monitoring station (Davis Instruments Corp.).

Years of study differed in weather conditions (Figure 1).



a)
b)

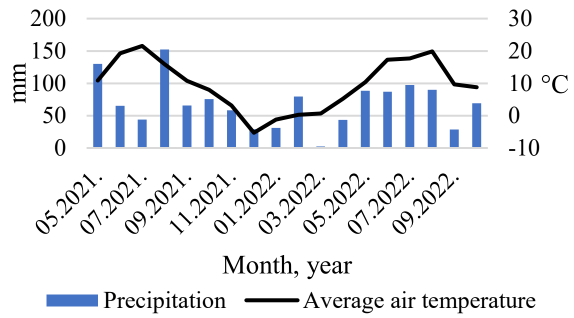


Fig. 1. Mean air temperature and amount of precipitation from May, 2021 to October, 2022 in two trial places: a) LatHort; b) LM Product.

In total, the vegetation season of 2021 had a higher average air temperature and higher amount of precipitation than the season of 2022. Comparing both places, a higher amount of precipitation was observed at LM Product in both years, while air temperature was similar in both places.

In 2022, soil temperature and volumetric moisture was measured at 15 cm depth in all three locations using Soil Scout™ wireless sensors. During vegetation season the highest average soil temperature was observed in August in all trial places with the highest value at farm Atvases and the lowest at LatHort (Table 2).

TABLE 2 AVERAGE SOIL TEMPERATURE AND VOLUMETRIC MOISTURE AT 15 CM DEPTH IN 2022 IN TRIAL LOCATIONS DURING VEGETATION SEASON

Month	Soil temperature, °C			Soil moisture, %		
	Atvases	*LM Product*	*LatHort*	*Atvases*	*LM Product*	*LatHort*
April	5.2	4.7	4.5	50.2	44.3	24.1
May	10.6	9.5	9.5	43.3	42.0	22.6
June	16.8	15.5	15.6	50.4	41.9	22.9
July	18.0	17.3	16.7	42.5	36.6	22.9
August	18.1	17.7	17.1	46.3	35.4	21.0
September	12.3	11.8	11.8	44.9	31.4	23.7
October	10.1	9.5	9.5	49.1	40.1	22.3

At Atvases, the highest average soil moisture was observed in June, while at LM Product and LatHort, the highest was in April. At Atvases, the lowest average soil moisture was observed in July, at LM Product – in September and at LatHort - in August.

Measurements and analysis. Soil dehydrogenase activity (DHA) and soil respiration rate (SRR) were evaluated during both seasons as indicators of soil microbial activity. The soil samples were collected during the vegetation season in all three locations from every treatment in four replicates. In 2021, soil samples were collected from the end of May to end of September and, in 2022, from the middle of May till the middle of October several times per season. Soil analyses were performed in the Soil Laboratory at LatHort.

DHA activity was detected according to Kumar et al. method [11] as modified by Dane and Šterne [12]. One gram of soil sample was exposed to 0.2 mL of 0.4% INT (2-p-iodophenyl-3-p-nitrophenyl-5-phenyltetrazolium chloride) and 0.05 mL of 1% glucose in 1 mL distilled water for at least 6 hours. The formed INTF (p-iodonitrotetrazolium formazan) was extracted by adding 10 mL methanol and actively shaking for 1 min. INTF was measured spectrophotometrically at wave length 485 nm.

Soil respiration was evaluated by a closed container method, where a soil sample (50 g) was placed in a jar where a low container with 5 mL of 0.1 M KOH was placed inside. After exposing it for 24 hours at 28 °C in the dark, the liquid was titrated with 0.1 M HCl [9].

In all three trial locations, every year at the end of growing season the strawberry plant development was accessed by weighting of aboveground plant biomass. The measurement was performed for 4 plants per every plot.

Descriptive statistics, analysis of variance, followed by Fisher`s *LSD* (least significant difference) test ($P \leq 0.05$) and Pearson`s correlation were used for data analysis. The statistical analyses were performed using the MS Excel 2013.

III. Results And Discussion

Soil microbial activity. Soil dehydrogenase enzymes are one of the main components of soil enzymatic activities participating in and assuring the correct sequence of all the biochemical routes in soil biogeochemical cycles [13] and measurement of changes in soil enzyme activities may provide a useful index of changes in soil quality [14]. In our trial, DHA significantly varied during season and among locations.

In the farm Atvases in 2021, the highest DHA was observed in the middle of August, while the lowest was at the beginning of June, when the first soil sample was collected (Fig. 2). DHA significantly varied also among treatments (p=0.020) with the highest average activity observed in the treatment 2, where Crimson clover was grown all season in interrows, and the lowest it was in the treatment 3, where Crimson clover was cut and incorporated in soil at the end of September and later garlic planted.

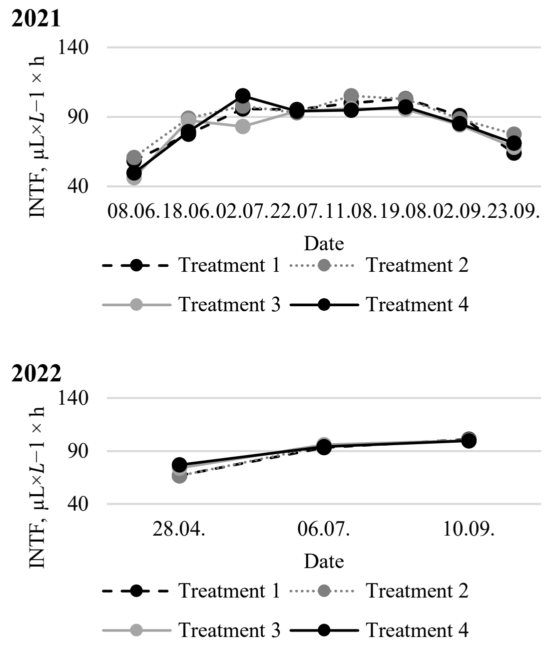


Fig. 2. DHA activity in soil at Atvases in 2021 and 2022 in different intercropping treatments.

In 2022, soil samples at Atvases were collected only three times. The highest DHA was observed in September, while the lowest was in April, when soil was still cold and wet (Fig. 2). Significant difference among treatments was not observed (p=0.102).

In the farm LM Product in 2021, DHA significantly varied among sampling dates (p=0.000) and treatments (p=0.016). The highest DHA was observed in the middle of July, while the lowest was in the middle of September, when the last soil sample was collected (Fig. 3). Among treatments, the highest average activity was observed in treatment 1, where Crimson clover was grown all season in interrows. The lowest DHA was in the treatment 3, as in farm Atvases.

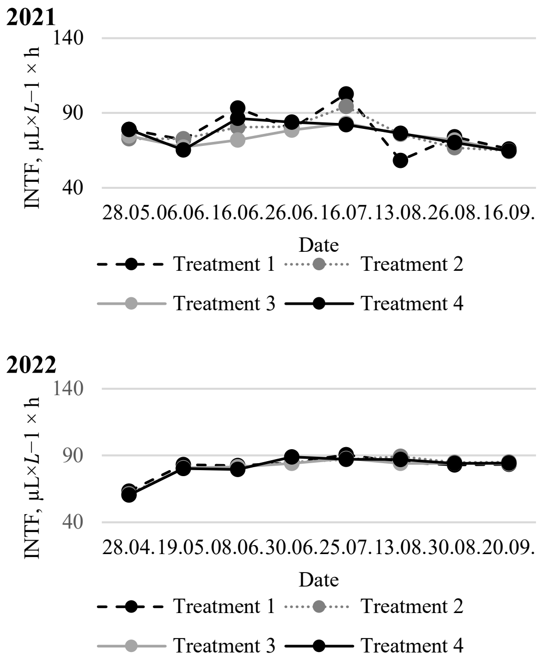


Fig. 3. DHA activity in soil at LM Product in 2021 and 2022 in different intercropping treatments.

In 2022, the highest DHA was observed at the end of July, while the lowest was in April (Fig. 3). Significant difference among treatments was not observed (p=0.726).

At LatHort in 2021, similar to both farms, DHA significantly varied among sampling dates (p=0.000) and treatments (p=0.024). During season the highest DHA was observed at the beginning of July, when the weather was warm and wet, while the lowest it was in June and at the end of season (Fig. 4). Among treatments the highest average activity observed in the treatment 2, where Crimson clover was grown all season in interrows, and the lowest it was in treatment 4 (control).

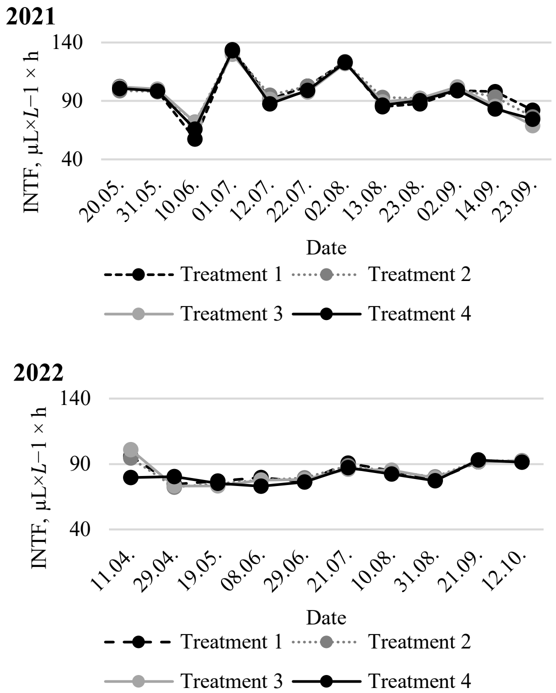


Fig. 4. DHA activity in soil at LatHort in 2021 and 2022 in different intercropping treatments.

In 2022, the highest DHA was observed at the beginning and at the end of season, while the lowest was at the end of April and May. Similar to other locations, significant difference among treatments was not observed (p=0.052).

In total, the highest average DHA was observed in the soil of Atvases, probably because of higher content of organic matter in the soil and higher soil moisture than in other locations (Table 1, 2). Among the most important functions performed by DHA is the biological oxidation of soil organic matter [15]. It is also stated that *DHA* is strongly influenced by water content and its activity reduced with the decrease of soil moisture [16, 17].

In our trial, also a significant positive correlation among DHA and average air temperature was found in the soil of LM Product (r= 0.60; n= 64) and LatHort Pūre (r= 0.25; n= 84). In Atvases, air temperature was not recorded and correlation was not calculated.

Similar to DHA, SRR significantly varied throughout the season and among locations. In the farm Atvases in 2021, SRR significantly varied among sampling dates (p=0.000). The highest SRR was observed at the end of September, when the last soil sample was collected, while the lowest was at the beginning of July (Fig. 5).

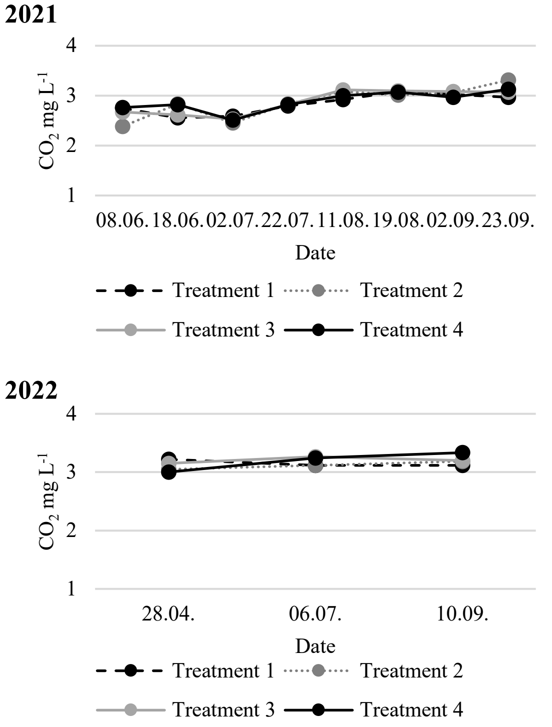


Fig. 5. Soil respiration rate at Atvases in 2021 and 2022 in different intercropping treatments.

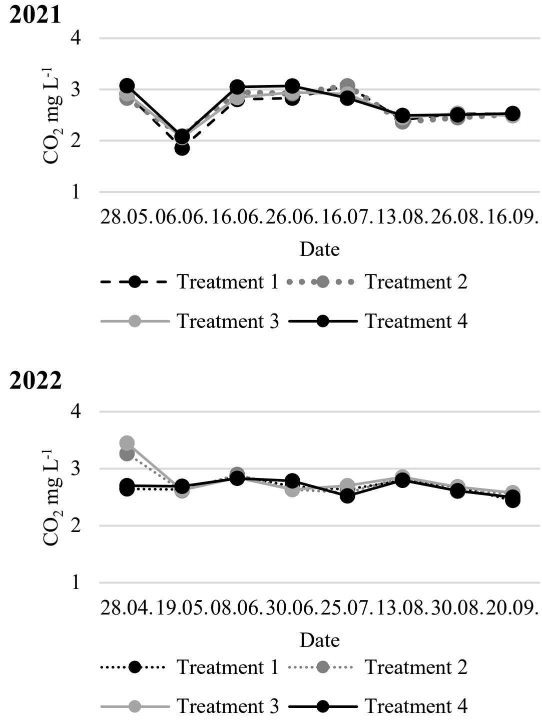


Fig. 6. Soil respiration rate at LM Product in 2021 and 2022 in different intercropping treatments.

In 2022, significant difference during vegetation season and treatments was not found ($p>0.05$).

In the farm LM Product, SRR significantly varied among sampling dates in both years ($p=0.000$ in 2021; $p=0.012$ in 2022). In 2021, the highest SRR was observed in July, while the lowest was at the beginning of June (Fig. 6).

In 2022, the highest SRR was observed at the end of April, while the lowest was at the end of September. A significant difference among treatments was observed only at the end of April, where the highest SRR was in the treatment 2, where pea was grown in interrrows, and treatment 3, where garlic was grown in interrows.

At LatHort in 2021, SRR significantly varied among sampling dates ($p=0.000$) and treatments ($p=0.016$). During season, the highest SRR was observed at the end of May, while the lowest was at the beginning of June and the end of September (Fig. 7). Among treatments, the lowest season`s average SRR was observed in the treatment 2, where Crimson clover was grown in interrows. At the same time in other treatments, where also Crimson clover was grown, SRR was similar to control.

In 2022, the highest SRR was observed in August and October, while the lowest was at the beginning of April. Significant difference among treatments was not observed ($p=0.644$).

In total, the highest average SRR was observed in soil of Atvases like for DHA. SSR significantly correlated with content of organic matter in soil ($r= 0.23$; $n= 192$).

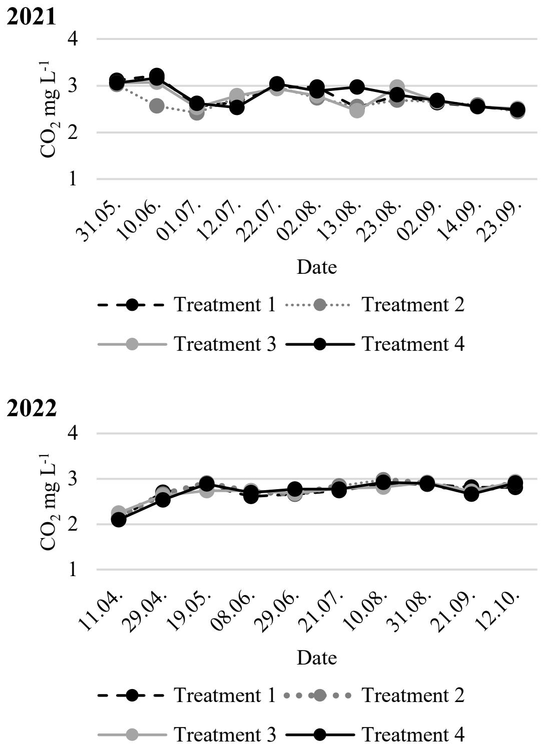


Fig. 7. Soil respiration rate at LatHort in 2021 and 2022 in different intercropping treatments.

Strawberry plant development. In 2021 at the end of vegetation season, significant difference in strawberry above-ground biomass among treatments was not observed in all locations (Table 3).

TABLE 3 AVERAGE WEIGHT OF STRAWBERRY ABOVE-GROUND BIOMASS AT THE END OF SEASON IN THREE LOCATIONS, G PLANT^{-1}

Treatment	Atvases	LM Product	LatHort
2021			
1	84	35	104
2	81	36	94
3	67	45	93
4	87	35	127
$LSD_{0,05}$	44	17	41
P-value	0.738	0.531	0.280
2022			
1	239	294	663
2	298	204	627
3	323	141	629
4	256	184	790
$LSD_{0,05}$	131	64	140
P-value	0.486	0.003	0.031

Better plant development was observed in LatHort, followed by Atvases and the weakest growth was in farm LM Product, probably because of harsh soil conditions.

In 2022, the strawberry above-ground biomass was significantly increased compared to 2021 in all locations. Similar to 2021, the best plant development was observed in LatHort and the weakest growth was in farm LM Product.

Comparing the strawberry growth in different treatments, at LatHort the highest plant biomass was observed in control treatment, following by treatment 1, where Crimson clover was grown during 1st growing year in interrows and marigolds were grown during 2nd growing year. In farm LM Product in treatment 1, plant above-ground biomass was the highest among treatments and it was significantly higher than in control treatment. In farm Atvases, significant difference among treatments was not observed.

IV. CONCLUSIONS

Soil microbial activity significantly fluctuated during the vegetation season depending on meteorological conditions and soil characteristics. Positive influence of soil organic matter content on soil microbial activity was observed. Intercropping, using different plant rotations in strawberry interrows, had lower influence on soil microbial activity and showed different results within each location.

Strawberry plant above-ground biomass increased with plant age. It was not influenced by intercropping during the first growing year, while in the second growing year the biomass differed among growing locations and intercropping treatments. In two of three trial places, the most positive impact of intercropping on strawberry above-ground biomass was observed, when Crimson clover was grown during 1st growing year and marigolds were grown during 2nd growing year in interrows.

V. ACKNOWLEDGEMENTS

This work was supported by ERDF funded project "Elaboration of environment-friendly crop growing technologies identified by the Green Deal and their implementation in horticultural production in Latvia (GreenHort)" (No. 1.1.1.1/20/A/169).

REFERENCES

[1] FAOSTAT, FAO 2023. [Online]. Available: https://fenix.fao.org/faostat/internal/en/#data/QCL. [Accessed: March 23, 2023].

[2] J.A. LaMondia, W.H. Elmer, T.L. Mervosh, and R.S. Cowles, "Integrated management of strawberry pests by rotation and intercropping," Crop Prot., vol. 21 (9), pp. 837–846, 2002. http://dx.doi.org/10.1016/S0261-2194(02)00050-9

[3] S.L. Poggio, "Structure of weed communities occurring in monoculture and intercropping of field pea and barley," Agric. Ecosyst. Environ., vol. 109 (1-2), pp. 48–58, 2005. http://dx.doi.org/10.1016/j.agee.2005.02.019

[4] R.J. Haynes, R.J. Martin, and K.M. Goh, "Nitrogen fixation, accumulation of soil nitrogen and nitrogen balance for some field-grown legume crops," Field Crop Research, vol. 35, pp. 85 – 92, 1993. https://doi.org/10.1016/0378-4290(93)90141-9

[5] M. Konopiński, T. Kęsik, and M. Błażewicz-Woźniak, "The effect of intercrops and ploughing term on the structure of yield and some qualities of salsify (*Tragopogon porrifolius* L.) roots," Acta Scientiarum Polonorum Hortorum Cultus, vol. 12(3), pp. 35-45, 2013.

[6] S. Dane, V. Laugale, D. Šterne, and G. Bimšteine, "Spreading of diseases in strawberry - legume intercropping," Acta Horticulturae, vol. 1156, pp. 627-634, 2017. https://doi.org/10.17660/ActaHortic.2017.1156.92

[7] S. Dane, V. L augale, L. Lepse, D. Sterne, "Possibility of strawberry cultivation in intercropping with legumes: a review," Acta Horticulturae, vol. 1137, pp. 83-86, 2016. https://doi.org/10.17660/ActaHortic.2016.1137.12

[8] S. Dane, V. Laugale, and D. Šterne, "Strawberry yield and quality in intercrop with legumes," Acta Horticulturae, vol. 1242, pp. 177-182, 2019. https://doi.org/10.17660/ActaHortic.2019.1242.25

[9] S. Dane, V. Laugale, L. Lepse, D. Siliņa "Influence of legumes on soil fertility in strawberry – legume intercropping," in Research for rural development 2017 : annual 23rd international scientific conference proceedings, LLU, Jelgava, 2017, vol. 2, pp. 26-32, 2017. Available online: http://www2.llu.lv/research_conf/proceedings2017_vol_2/docs/LatviaResRuralDev_23rd_2017_vol2.pdf

[10] J. Davik, H. Daugaard, and B. Svensson, "Strawberry production in the Nordic countries," Adv. Strawb. Prod., vol. 19, pp. 13-18, 2000. Available online: https://www.researchgate.net/profile/Jahn-Davik/publication/266907385_Strawberry_Production_in_the_Nordic_Countries/links/546b30f50cf2f5eb1809042c/Strawberry-Production-in-the-Nordic-Countries.pdf

[11] S. Kumar, S. Chaudhuri, and S.K. Maiti, "Soil Dehydrogenase Enzyme Activity in Natural and Mine Soil – A Review," Middle-East Journal of Sciantific Research, vol. 13(7), pp. 898 – 906, 2013. doi: 10.5829/idosi.mejsr.2013.13.7.2801. Available online: https://www.researchgate.net/profile/Sanjoy-Kumar-2/publication/278017226_Soil_Dehydrogenase_Enzyme_Activity_in_Natural_and_Mine_Soil_-A_Review/links/55784f8208ae752158703381/Soil-Dehydrogenase-Enzyme-Activity-in-Natural-and-Mine-Soil-A-Review.pdf

[12] S. Dane, and D. Šterne, "Soil fertility in strawberry – legume intercrop," In Ražas svētki "Vecauce – 201". Lauksaimniecības zinātne nozares attīstībai, LLU, Jelgava, pp. 16–19, 2016 (in Latvian).

[13] S. Kumar, S. Chaudhuri, and S. K. Maiti, "Soil dehydrogenase enzyme activity in natural and mine soil - a review," Middle-East Journal of Scientific Research, vol. 13(7), pp. 898-906, 2013.

[14] S. Visser, and D. Parkinson, "Soil biological criteria as indicators of soil quality: Soil microorganisms," American Journal of Alternative Agriculture, vol. 7, pp. 3-37, 1992.

[15] A. Wolinska, and Z. Stepniewska, "Microorganisms abundance and dehydrogenase activity as a consequence of soil reoxidation process," Research Signpost: Kerala, India, pp. 111-143, 2011.

[16] D.R. Nayak Babu J. and T.K. Adhya, "Long-term application of compost influences microbial biomass and enzyme activities in a tropical Aeric Endoaquept planted to rice under flooded condition," Soil Biology and Biochemistry, vol. 39, pp. 1897-1906, 2007.

[17] I. Pascual, M.C. Antolin, C. Garcia, A. Polo, M. Sanchez-Diaz, "Effect of water deficit on microbial characteristics in soil amended with sewage sludge or inorganic fertilizer under laboratory conditions," Bioresource Technology, vol. 98, pp. 29-37, 2007.