NARDI FUNDULEA, ROMANIA ROMANIAN AGRICULTURAL RESEARCH, NO. 40, 2023
www.incda-fundulea.ro; doi.org/10.59665/rar4031 Print ISSN 1222-4227; Online ISSN 2067-5720

THE IMPACT OF CONSERVATIVE SOIL MAINTENANCE PRACTICES IN VINEYARDS ON SOIL MICROBIAL DIVERSITY AND ON THE BIODIVERSITY OF USEFUL AND HARMFUL ENTOMOFAUNA

Adrian Șerdinescu*, Marian Ion, Lidia Fîciu, Elena Brînduşe

Research and Development Institute for Viticulture and Enology Valea Călugărească, Valea Mantei str., no. 1, 107620 Valea Călugărească, Prahova County, Romania

*Corresponding author. E-mail: serdinescuadrian@yahoo.com

ABSTRACT

In the context of actual climatic changes, which led to an increase of soil hydric stress, new agrotechnical solutions were experimented to ensure a moisture preservation in soil and the conservation of soil microbial activity. These included the use of mulching systems and the cover crops, in order to replace the use of the classical black furrow system. In this respect, the aim of our study was to evaluate the influence of the use of soil mulching systems and of the cover crops in vineyards on the microbial biodiversity of the soil and on the biodiversity of the useful and harmful entomofauna, in comparison with the classical maintenance system, represented by black furrow. The researches were performed during two particularly dry years in two experimental plots located in Valea Călugărescă viticultural center. In one of them were experimented, as soil maintenance systems, the total straw mulching and the partial mulching (interval between rows) with marc compost, in the other the artificial grassing of the interval between rows with ornamental clover Nano and with honey plants represented by *Melilotus officinalis*. The experimental data have shown the positive effect of mulching systems on soil microbial activity, highlighted by an increase of soil microbial load and of its diversity (represented by bacteria, fungi, algae and protozoa), especially in case of mulching with marc compost (15.94 million microorganisms/g of soil, as compared with 5.9 million in case of black furrow system). Also, the ratio between fungi and bacteria was higher in case of partial mulching, this aspect being a favorable factor for soil biological activity. The use of cover crops indicated a positive effect in the increasing of useful entomofauna, belonging to the orders *Ortoptera*, *Coleoptera* and *Arahnidae* (especially in case of the artificial grassing).

Keywords: biodiversity, mulching systems, cover crops, soil microbial diversity, useful and harmful entomofauna.

INTRODUCTION

In the context of actual climatic changes, which led to an increase of soil hydric stress in many wine regions of the world, new conservative soil maintenance practices were experimented, in order to ensure a good preservation of water in soil and of its fertility. These included the mulching systems (total or partial) using as mulch materials the cereal straws, the marc compost or the shredded wood wastes and also the cover crops, based on the artificial grassing of the interval between rows with small plants (such as ornamental clover) or honey plants (Watson, 2006; Chan et al., 2010; Șerdinescu et al., 2013; DeVetter et al., 2015; Fraga and Santos, 2018; Morisod, 2018).

The studies conducted worldwide have highlighted that the use of the new soil maintenance systems caused a change of the microbial activity in soil (in case of the mulching systems) and of the biodiversity of useful and harmful entomofauna present in vineyards (in case of the use of cover crops) as compared to the classical black furrow system. These modifications were the result of changes occurred in soil water regime, soil temperature and on soil fertility determined by the use of new soil maintenance systems (Holloway and Stork, 1991; Pankhurst et al., 1995; Mundy and Agnew, 2002; Șerdinescu and Brînduşe, 2014).

In this respect, our researches aimed to experiment some new soil maintenance systems suitable for viticulture in case of drought conditions, establishing the impact that these systems have on soil microbial activity (as microbial mass and biodiversity) and on the biodiversity of vineyard

Received 1 September 2022; accepted 28 September 2022. First Online: September, 2022. DII 2067-5720 RAR 2023-1

entomofauna, as an indicator of natural biological balance in viticulture ecosystem.

MATERIAL AND METHODS

The researches were performed in the period 2020-2021, characterized by drought, in two experimental plots located in two vineyards from Valea Călugărească viticultural center. In the first experimental plot was studied the influence of soil maintenance systems on soil microbiological diversity. There were studied two variants of soil mulching, the total mulching (rows and interval between rows) with cereal straws applied in a layer of 10 cm and the partial mulching (interval between rows) with marc compost applied in a layer of 10 cm. As control was considered the classical black furrow system. In the second experimental plot was studied the impact of the cover crops (as soil maintenance system), on the biodiversity of useful and harmful entomofauna. There were studied two variants, the artificial grassing with ornamental clover Nano and the artificial grassing with *Melilotus officinalis* (a honey plant), applied on the interval between rows. As control was considered the classical black furrow system.

To establish the influence of the new soil maintenance systems on the biodiversity of soil microorganisms and on the the vineyard entomofauna were performed determinations concerning the soil water regime during the growing season of grapevine, the microbiological activity of the soil and the presence of useful and harmful entomofauna on vineyards.

The soil water regime was establish monthly, between April and September, on the depth of 0-100 cm, taken samples from 20 to 20 cm with an agrochemical bore.

To collect the soil samples for the determination of soil microorganisms, the topsoil consisting mainly of plant debris was removed and then the soil samples were collected from three different locations of the experimental plot in paper bags by using a tubular bore. All materials and instruments were sterilized before use by steaming at a temperature of 180ºC for one hour. The soil samples were collected at the end of August in sterile conditions from 0-20 and 20-40 cm depth, sieved through a 2 mm sieve mesh and then scattered and homogenized in a sterile mortar. The total number of soil viable microorganisms was achieved by inoculation on solid culture media using the method of serial dilution. Each serial dilution was inoculated in three Petri dishes. The number of microorganisms per gram of soil was calculated according to the following formula:

$$N = m \times c \times 10$$

where:

N = the number of living microorganisms per gram of soil;

m = the average number of the colonies developed on the three Petri dishes;

c = inverse of the dilution used for inoculation;

10 = the coefficient for reporting the results to 1 gram of soil.

In order to identify the microbial colonies, selective culture media for each systematic group of microorganisms were used: Potato dextrose agar medium for bacteria, Glycerol yeast agar medium for actinomycetes, Sabouraud dextrose agar with streptomycin for fungi and Yeast extract peptone dextrose medium (YEPD) for yeasts. The inoculation was performed by the distribution of 0.1 ml of the selected homogeneous dilutions on the surface of solidified medium, previously distributed in plates. The cultures were then incubated at 28ºC. The determination by microscopy of the number of bacterial colonies was achieved after 24 hours and the number of fungi after 5 days. The data obtained were submitted to one-way analysis of variance (ANOVA) and the means values were separated using the Tukey test (0.05%).

In the second experimental plot the determinations of the useful and harmful entomofauna were achieved in June and September. To collect the biological material were used different methods appropriate to the target groups, such as the Barber traps, the leaf samples and the typing technique. The identification of the insect species was performed in the laboratory using specialized instruments.

RESULTS AND DISCUSSION

The experimental data obtained in the first experimental plot highlighted that in comparison with the classical maintenance system (black furrow), where the microbial load had a value of 7.7x10^6 microorganisms/g of soil on the depth of 0-20 cm, a significant increase was observed in case of total and partial mulching, with values of 16.8x10^6 and 22.1x10^6 microorganisms/g of soil. The same trend can be mentioned also for the depth of 20-40 cm, but in this case the differences between variants were smaller (Figure 1).

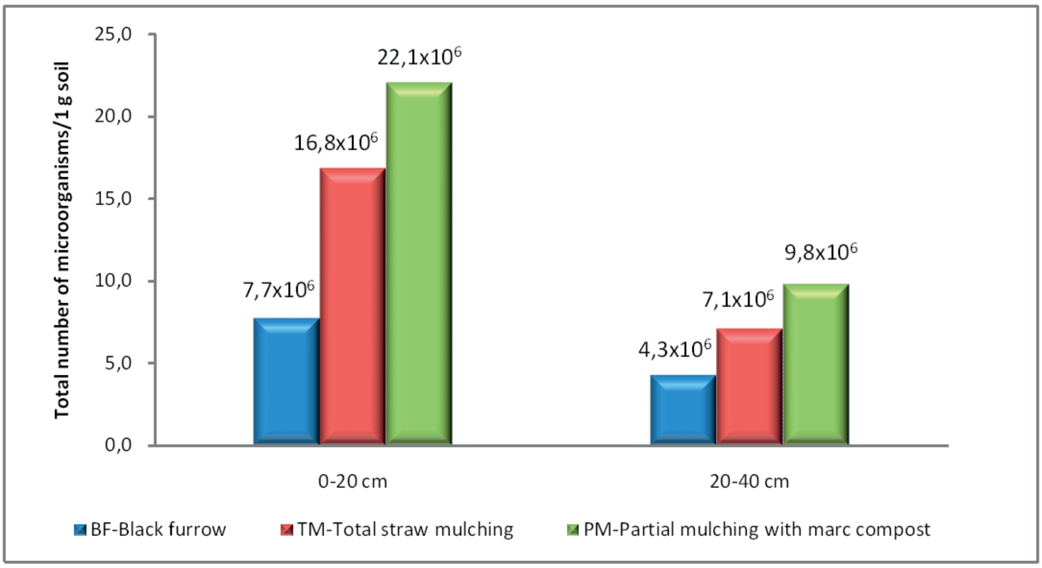

Figure 1. Influence of soil maintenance systems on soil microbial load

The statistical analysis showed that these differences had a good statistical significance (Table 1). The microbial load was correlated with the soil humidity which was different depending on the soil maintenance systems. The soil moisture showed normal levels at the beginning of the bud burst, than it gradually decreased during the growing season of grapevine due to the excessive thermal regime. In case of black furrow system during August and September the soil moisture registered values closed to the wilting coefficient, especially in the surface horizon (0-20 cm).

Table 1. Analysis of variance regarding the average values of soil microbial load

Variant	Depth 0-20 cm		Depth 20-40 cm	
	Mean value x 10^6	Significance	Mean value x 10^6	Significance
Black furrow	7.71		4.26	
Total straw mulching	16.83	A	7.11	a
Partial mulching with marc compost	22.06	A	9.82	A

A, B: $p<0.01$; a, b: $p<0.05$.

Compared with the classical method of soil maintenance, the systems based on soil mulching (total or partial) ensured a higher moisture, due to the reduction of soil surface water evaporation, this being higher with 17% in case of total mulching and with 12% in case of partial mulching with marc compost (Figure 2).

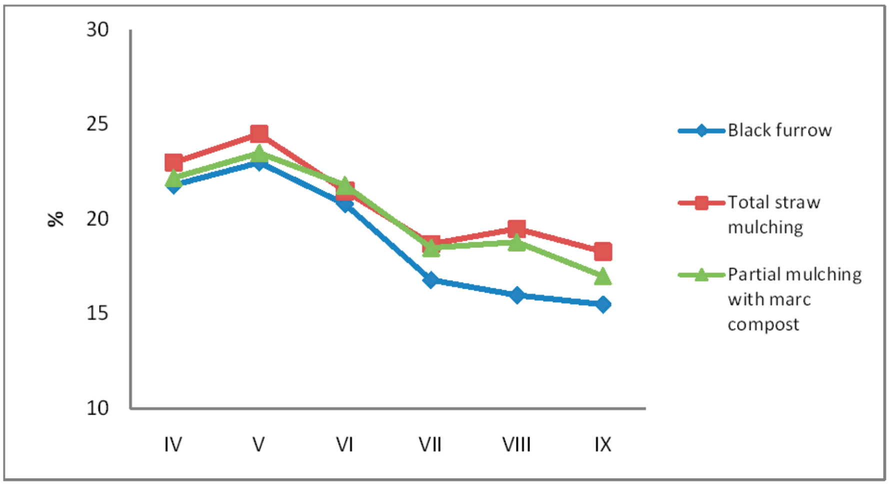

Figure 2. The dynamic of soil moisture during the growing season of grapevine under different soil maintenance systems

Not only the microbial load was influenced by the soil maintenance systems, but also the development of the main groups of microorganisms in soil (Table 2).

Table 2. Soil microbial load and its biodiversity in relation with the soil maintenance systems

Variant	Depth cm	Total number of microrg./ 1 gr. soil	Bacteria		Actinomycetes		Fungi		Yeasts	
			No.	%	No.	%	No.	%	No.	%
Black furrow	0-20	$7.71x10^6$	$5.90x10^6$	76.5	$3.50x10^5$	4.5	$1.46x10^6$	18.9	$0.13x10^3$	0.002
	20-40	$4.26x10^6$	$2.70x10^6$	63.4	$2.00x10^5$	4.7	$1.36x10^6$	31.9	-	-
Total straw mulching	0-20	$16.80x10^6$	$8.73x10^6$	51.9	$3.70x10^5$	2.2	$7.73x10^6$	45.9	$2.10x10^3$	0.012
	20-40	$7.11x10^6$	$4.43x10^6$	62.3	$1.80x10^5$	2.5	$2.50x10^6$	35.2	-	-
Partial mulching with marc compost	0-20	$22.10x10^6$	$9.63x10^6$	43.7	$4.00x10^5$	1.8	$12.00x10^6$	54.5	$3.50x10^3$	0.016
	20-40	$9.82x10^6$	$5.26x10^6$	53.6	$2.30x10^5$	2.3	$4.33x10^6$	44.1	-	-

Bacteria was the largest group of microorganisms, representing an average from the total viable microorganisms of 60.1% for the depth of 0-20 cm and of 58.5% for the depth of 20-40 cm, fallowed by *Fungi* with 36.7% and 37.0%, respectively, *Actinomycetes* with 3.2% and 3.5%, respectively, and *Yeasts* with 0.01%, highlighted only in the 0-20 cm depth (Figure 3).

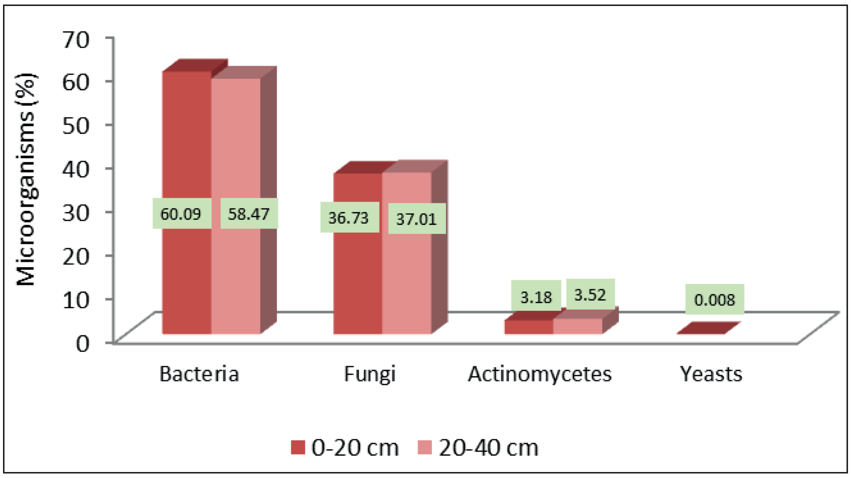

Figure 3. The percentage of the number of microorganisms in relation with systematic groups

The highest values were recorded for all the microorganisms groups in case of partial mulching with marc compost and the lowest values in case of black furrow maintenance system. Regarding the *Actynomicetes* and *Fungi* the development was more intense for 20-40 cm depth.

Concerning the spectrum of fungi, there were reported pathogenic species of the following genera: *Verticillium, Mucor, Rhizopus, Penicillium, Cladosporium, Phoma, Trichoderma, Aspergillus, Gliocladium, Fusarium* and *Alternaria* (Table 3).

Table 3. Fungal spectrum in relation with soil maintenance systems

Variants	Depth cm	Genera of fungi
Black furrow	0-20	*Penicillium, Verticillium, Rhizopus, Cladosporium, Aspergillus, Fusarium, Alternaria*
	20-40	*Verticillium, Mucor, Penicillium, Cladosporium, Aspergillus, Fusarium*
Total straw mulching	0-20	*Verticillium, Penicillium, Cladosporium, Phoma, Trichoderma, Aspergillus, Gliocladium, Fusarium, Alternaria*
	20-40	*Penicillium, Cladosporium, Phoma, Trichoderma, Aspergillus, Fusarium, Alternaria*
Partial mulching with marc compost	0-20	*Verticillium, Mucor, Rhizopus, Penicillium, Cladosporium, Phoma, Trichoderma, Aspergillus, Gliocladium, Fusarium, Alternaria*
	20-40	*Penicillium, Cladosporium, Phoma, Trichoderma, Aspergillus, Gliocladium, Fusarium, Alternaria*

Concerning the frequency of the fungal genera, we can mention a little number of genera in case of the black furrow system and a higher number (11 genera) in case of partial mulching with marc compost, especially for the depth of 0-20 cm. The presence of the genera *Penicillium* and *Aspergillus* was observed for all the studied variants.

The ratio between *Fungi* and *Bacteria* was higher in case of the mulching systems, as compared to the black furrow system, this being a favorable factor for soil biological activity and for soil health (Figure 4).

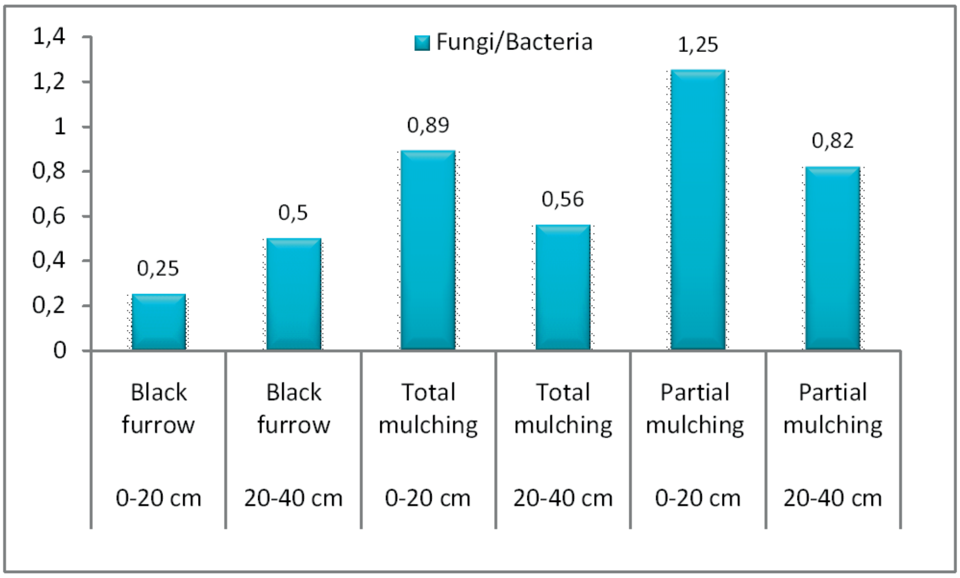

Figure 4. The ratio between fungi and bacteria in soil depending on soil maintenance system

The data obtained in the second experimental plot (where the impact of cover crops on the entomofauna biodiversity was studied) highlighted that the number of insects collected had the highest value in case of the artificial grassing of soil with ornamental clover Nano and the lowest value in case of black furrow maintenance system, for all the studied groups of insects (Table 4).

Table 4. The load of the main groups of insects during the growing season of grapevine depending on soil maintenance system (mean values)

Groups of insects	Month	Variant		
		Artificial grassing with ornamental clover Nano (no)	Artificial grassing with honey plants (no)	Black furrow (no)
Enchitreide	June	222	187	71
	September	154	107	56
	Mean value	**188**	**147**	**64**
Nematode	June	298	227	202
	September	298	192	110
	Mean value	**298**	**210**	**156**
Colembole	June	255	209	120
	September	234	175	64
	Mean value	**245**	**192**	**92**

The most representative group of insects was *Nematode*, followed by *Colembole* for all the variants. We can mention also that the number of insects was higher in June compared to September. Analysing the structure of the fauna captured by using the typing technique we can notice the existence of a larger number of useful insects species compared to the harmful insects in case of all the variants. From the useful species the high number was registered for the orders *Ortoptera*, *Coleoptera* and *Arahnidae*. A low number of harmful species has been recorded among those that belong to the orders *Homoptera*, *Hymenoptera*, *Diptera* and *Heteroptera* (Figure 5).

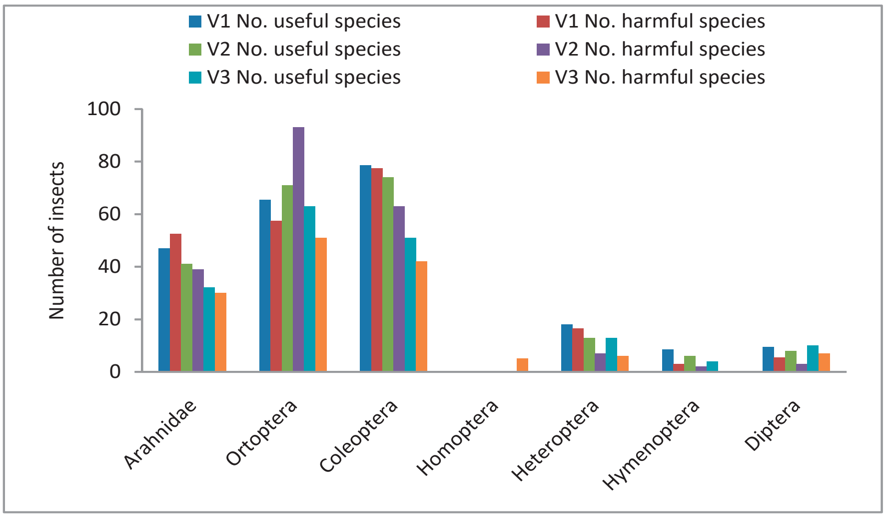

Figure 5. The structure of the entomofauna captured by using the typing technique (mean values)

Concerning the influence of the cover crops on the ratio between the useful and harmful insects we can mention that the number of useful species was higher in case of the artificial grassing of soil with ornamental clover Nano, this aspect being favorable for grapevine protection against the harmful insects. The total number of *Coleoptera* species captured by using the Barber traps during the two years of experiment was higher in case of the artificial grassing of soil with ornamental clover Nano, namely 208 insects/trap (Table 5).

Table 5. The main species of *Coleoptera* captured by Barber traps (mean values)

Species	Artificial grassing with ornamental clover Nano	Artificial grassing with honey plants	Black furrow	**Total**
	No. of insects/trap			
Carabus sp.	71	53	61	**185**
Amara aenea Deg	13	10	5	**28**
Blitophagaundata Mull.	11	4	6	**21**
Calosomaauropunctata L.	1	1	3	**5**
Harpaluspubescens L.	2	-	1	**3**
Cotinisnitida	11	2	-	**13**
Melolonthamelolontha	33	23	14	**70**
Coccinellaseptempunctata	35	27	27	**89**
Epicometishirta	8	5	8	**21**
Lebiahumeralis	23	15	11	**49**
Total	**208**	**140**	**136**	**484**

It was followed by the variant with artificial grassing of soil with honey plants, the lowest value being registered in case of the black furrow system.

CONCLUSIONS

The experimental data obtained in our study showed that the use of conservative soil maintenance systems, respectively the mulching systems and the cover crops, caused changes (quantitatively and qualitatively) on the biodiversity of soil and vineyard organisms, as a result of the modifications occurred in soil water regime, soil temperature and also on soil fertility induced by these systems.

Concerning the soil microbial biodiversity it was found a significant increase of the total number of microorganisms in case of the mulching systems (especially in case of partial mulching with marc compost) as compared to the classical black furrow system. Bacteria was the largest group of microorganisms, followed by fungi, actinomycetes and yeasts (present only in the layer of 0-20 cm). There was a good correlation between the microbial load and the soil humidity. The ratio between fungi and bacteria was higher in case of the use of mulching systems, this being a favorable factor for soil health and for its biological activity.

The use of the artificial grassing of soil between the rows with ornamental clover or honey plants caused a change of the number of insects present in vineyard and of their diversity. The useful species of insects, especially from the orders *Ortoptera*, *Coleoptera* and *Arahnidae* registered the highest values in case of the artificial grassing of soil with ornamental clover Nano, this being a favorable factor for grapevine protection against harmful insects.

REFERENCES

Chan, K.Y., Fahey, D.J., Newell, M., Barchia, I., 2010. *Using composted mulch in vineyards - Effects on grape yield and quality*. Int. Journal of Fruit Science, 10(4): 441-453.

DeVetter, L.W., Dilley, C., Nonnecke, G., 2015. *Mulches reduce weeds, maintain yield and promote soil quality in a continental-climate vineyard*. Am. J. Enol. Viticult., 66(1): 54-64.

Fraga, H., and Santos, J.A., 2018. *Vineyard mulching as a climate change adaptation measure: Future simulations for Alentejo, Portugal*. Agricultural Systems, 164: 107-115.

Holloway, J.D., and Stork, N.E., 1991. *The dimensions of biodiversity: the use of invertebrates as indicators of human impact*. In: Hawksworth, D.I. (eds.), The Biodiversity of Microorganisms and Invertebrates: Its Role in Sustainable Agriculture. C.A.B. International, Wallingford, UK.

Morisod, T., 2018. *Entretien du sol en viticulture*. Ed. Agridea, Lausanne.

Mundy, D., and Agnew, A., 2002. *Effects of mulching with vineyard and winery waste on soil fungi and botrytis bunch rot in Malborough vineyards*. New Zealand Plant Protection, 55: 135-138.

Pankhurst, C.E., Hawke, B.G., MacDonald, H.J., Kirkby, C.A., Buckerfield, J.C., Michelsen, P., O'Brien, K.A., Gupta, S.R., Doube, B.M., 1995. *Evaluation of soil biological properties as potential bioindicators of soil health*. Aus. J. Exp. Agric., 35: 1015-1028.

Șerdinescu, A., Pîrcălabu, L., Enache, V., Ranca, A., Bosoi, M., Dumitru, E., Rățoi, I., 2013. *Technological solutions for the diminution of the disturbing effect of climatic changes in viticulture*. Proc. of the 36th World Congress of Vine and Wine, Bucharest.

Șerdinescu, A., and Brînduşe, E., 2014. *Influence of climatic changes and soil maintenance systems in vineyards on soil microbial diversity and hydrological properties*. Global Journal on Advances in Pure & Applied Sciences, 7: 28-34.

Watson, C., 2006. *Mulch softens the big dry*. Australian Viticulture, 10: 74-75.