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Relationship Between Vegetation Succession and Earthworm Population in Vineyards

Erika Hurajová[1], Petra Martínez Barroso[2], Ladislav Havel[1], Igor Děkanovský[3], Jan Winkler[1*]

[1] Department of Plant Biology, Faculty of AgriSciences, Mendel University in Brno, Zemědělská 1, 613 00 Brno, Czech Republic

[2] Department of Applied and Landscape Ecology, Faculty of AgriSciences, Mendel University in Brno, Zemědělská 1, 613 00 Brno, Czech Republic

[3] University Hospital Brno, Jihlavská 20, Brno 625 00, Czech Republic

[*] Corresponding author's e-mail: jan.winkler@mendelu.cz

ABSTRACT

The aim of the study was to determine the effect of succession of vegetation on the population of earthworms in selected vineyards. Earthworms (*Annelida, Lumbricidae*) are an important group of soil invertebrates. The population of earthworms in vineyards is influenced by environmental conditions and human activities. The presence of earthworms is beneficial to the ecosystem of vineyards. Earthworms aerate the soil, improving the quality and structure of the soil in vineyards. They decompose organic matter, contribute to the formation of humus, and increase the soil fertility. Vegetation cover in vineyards affects earthworm populations. The vegetation species spectrum in the vineyard changes over time, as succession is controlled by human activity. The research took place between the years 2020 and 2023 in the wine-growing villages of Horní Dunajovice, Hostěradice, Miroslav and Miroslavská Knínice (Czech Republic). 4 species of earthworms have been recorded. *Aporrectodea caliginosa* and *A. rosea* occurre frequently in younger vineyards. Annual dicots supported the occurrence of *Aporrectodea caliginosa* and *A. rosea*. *Lumbricus terrestris* and *L. rubellus* are more common in older vineyards. Perennial species supported the occurrence of *Lumbricus terrestris* and *L. rubellus*. The annual grasse contributed to the occurrence of *Lumbricus rubellus*. Changes in the composition of the vineyard vegetation affect the occurrence of the observed species of earthworms.

Keywords: succession; earthworms; vineyards; vegetation; biodiversity change.

INTRODUCTION

Soil macrofauna, especially animal organisms larger than 2 mm (Lavelle et al., 2006) provide numerous ecosystem services (Bardgett and van der Putten 2014). Soil macrofauna comprises two main groups of animals that participate in many processes in ecosystems, namely ants (*Hexapoda, Formicidae*) and earthworms (*Annelida, Lumbricidae*), (Vepsäläinen et al., 2008; Pouyat et al., 2010; Blouin et al., 2013). Earthworms play a key role in supporting the functioning of arable soils in agroecosystems, as well as in other ecosystem services (Cenci and Jones 2009). Certain soil properties can be directly affected by earthworm activities (Eijsackers et al., 2005; Römbke et al., 2005). Earthworm populations affect soil structure by creating macropores and increasing the stability of soil aggregates. Thanks to their aktivity a favorable soil structure is maintained for longer (Li and Ghodrati 1995). They reduce soil bulk density and change soil moisture availability (Lavelle et al., 2004; Eijsackers et al., 2005).

The physico-chemical properties of soil influence the vitality of the grapevine plants substantially and differ according to the natural variability of soil conditions within vineyard ecosystems. The higher vitality of grapevines is mainly associated with clay soils, which are characterized by a better ability of retaining water and an amount of readily available nutrients. For example, the changes in soil structure affect shoot growth and grapevine yield considerably (Karn et al., 2024). The physico-chemical properties of the soil play a key role in the availability of water, which affects the vitality of grapevine especially in a semi-arid climate. In addition, excessive water and nitrogen levels can lead to harmful effects by promoting excessive vegetative growth of grapevines, creating a favorable environment for diseases and nutrient deficiencies (Leeuwen et al., 2009). Soil moisture and water availability is also affected by non-target vegetation in vineyards. The grapevine competes for water and nutrients with other vegetation. Grapevine growth can be reduced by up to 55%, which can be asscribed to the competition for soil moisture and nutrients (Giese et al., 2015; Hickey et al., 2016). To reduce competition for water and nutrients, management with bare soil (mainly in new vineyards) is used, as well as management with cover crops (CC) (mainly on slopes), however, the most employed management type in the Czech Republic (CZ) is the spontaneous greening of the inter-rows of vineyards. The type of inter-row vegetation varies between regions markedly (Ragasová et al., 2019).

Permanent vegetation cover of inter-rows in vineyards is considered the most effective measure for reducing soil erosion (Capello et al., 2020; Telak and Bogunovic, 2020). The long-term impact of erosion processes was manifested in a high concentration of soil carbonates and changes in pH. An increased content of soil organic carbon (SOC) and a higher volumetric weight and porosity of the soil can be observed in grassed inter-rows or on abandoned vineyards (Lieskovský et al., 2024). Vegetation and its biomass in vineyards ensures also other benefits such as weed control, improved rainfall infiltration and stimulation of beneficial predators and other animals (Marques et al., 2020; Blanco-Canqui et al., 2015).

Annual crops, used as CC and as inter-row crops in vineyards, are increasingly used because of the benefits they provide to the soil. Incorporating CC into agricultural practices is one of the ways to increase SOC content (Poeplau and Don 2015), which leads to improving soil structure, biological activity and the amount of available mineral nutrients (Blanco-Canqui et al., 2015). The stability of soil organic matter also determines the dynamics and ecotoxicity of Cu in the vineyard ecosystem (Ouédraogo et al., 2024). The accumulation of biomass on the soil surface leads to the stabilization and regeneration of soil properties. In grassed vineyards, most of the SOC was accumulated in the underground biomass of grasses (Lieskovský et al., 2024). According to Sciubbo et al. (2021) dead vegetation biomass accumulated during succession in abandoned vineyards increases the biological activity of soil organisms. Earthworms play an important role in decomposing plant remnants by ingesting them and mixing them with minerals in the soil (Bot and Benites 2005). Earthworms affect soil organic matter content directly or indirectly, its transformation and dynamics, nutrient cycles and soil fertility (Lavelle et al., 2004; Bhadauria and Saxena, 2010). Soil is a natural environment for earthworm communities that is affected by agricultural practices such as tillage, crop residue management, use of organic and mineral fertilizers and pesticide application (Chan and Munro 2001; Eijsackers et al., 2005). Earthworms are sensitive to chemical and physical changes in soil properties (Coll et al., 2011).

Agricultural intensification has reduced soil biodiversity in cultivated fields (Bengtsson et al., 2005; Doran and Zeiss 2000). Organic systems and low-input crops have been proposed as alternatives to intensive agricultural practices to reduce the impact of chemicals on human health and the environment (Bengtsson et al., 2005; Hole et al., 2005). However, the effects of pesticides application restrictions on biodiversity and especially on soil organisms require further investigation (Hole et al., 2005). Soil is a living non-renewable resource that changes its properties based on the effects of climate change, human activities including agricultural practices, crop type, tillage and the use of chemical compounds (Floch et al., 2009). Understanding the impact of agricultural management on soil ecosystems is essential for a development of sustainable viticulture (Fonte et al., 2009). Grapevines belong to perennial crops with a very specific method of cultivation. Vineyards are an agroecosystems composed of grapevine plants and other non-target vegetation. Long-term management of vineyards affects the succession of non-target vegetation. According to our hypothesis, changes in the vegetation of vineyards cause a response in other organisms that depend on plants as a source of food, including the population of earthworms. The aims of our study were to: (i) establish trends in earthworm population, (ii) establish trends in the succession of non-target vineyard vegetation, and (iii) clarify the relationship between vegetation succession and earthworm population in vineyard ecosystem.

MATERIAL AND METHODS

Study area

The selected vineyards are located on the edge of the Dyjskosvratecký valley in the cadastral territories of the municipalities of Horní Dunajovice, Hostěradice, Miroslav and Miroslavské Knínice (South Moravian Region, CZ). All vineyards belong to the Moravian wine region, to Znojmo subregion. The name of the vineyards, the year of establishment of the vineyards and the prevailing soil type are described in Table 1. The altitude is between 240 m and 320 m. The average annual temperature is 8.5 °C, the annual rainfall is 470 mm. These data come from the nearest meteorological station of the Czech Hydrometeorological Institute in Kuchařovice (Culek et al., 1996, CGS 2017, CGS 2018).

Plots for vegetation and earthworm population evaluation were established in 37 vineyards of different ages. The areas for evaluation were located in the inter-rows of vineyards. The vegetation of the inter-rows in the vineyards was created by spontaneous greening and was regulated by mulching.

Methodology of earthworm population assessment

The assessment of the earthworm population was carried out between the years 2020 to 2023. In each year, three dates of observation were chosen. These were during June, August and October. Earthworm monitoring took place in 3 repetitions. The vegetation in the closest proximity of the phytocenological images was removed so that the earthworms could be better observable. The size of the area was 0.30 × 0.30 m.

The earthworm assessment method was based on a combination of the AITC method and collection from soil plot. The AITC method uses an AITC solution which is composed of mustard and isopropanol diluted in water to a concentration of 0.01%. The solution was poured into the soil. Subsequently, earthworm activity was observed. The method of collection from soil excavations/plots is not dependant on the activity of earthworms. Earthworms were collected on an area of 0.30×0.30 m from a depth of 0.30 m. Earthworm species were identified at the species level using the identification key according to Cuendet (2009).

Method of vegetation assessment

Vegetation assessment took place at the same sites where the earthworm population

Table 1. A general characteristics of selected vineyards

Municipality	Vine lines	GPS	Year	Soil type
Horní Dunajovice	Frédy	48°56’41.153”N, 16°10’41.676”E	1995, 2002, 2009, 2016, 2018	Cambisols, Chernozems
Horní Dunajovice	Stará hora	48°57’6.969”N 16°10’45.489”E	1995, 2000, 2020, 2021	Cambisols, Chernozems
Hostěradice	Volné pole	48°57’17.479”N, 16°17’12.790”E	1972, 2003, 2014, 2015, 2016, 2017, 2018, 2020, 2021	Cambisols, Chernozems
Miroslav	U vinohradu	48°56’37.223”N, 16°17’59.253”E	2003, 2004, 2007, 2014, 2019	Chernozems
Miroslav	Weinperky I	48°56’14.871”N, 16°19’9.292”E	1996, 2011, 2014, 2015, 2017	Cambisols, Chernozems
Miroslav	Weinperky II	48°56’17.996”N, 16°18’22.325”E	1996, 1998, 1999, 2001, 2002, 2003, 2004, 2008, 2009	Cambisols, Chernozems
Miroslavské Knínice	Stará hora	48°58’16.344”N, 16°19’50.606”E	2001	Cambisols, Chernozems
Miroslavské Knínice	Zolos	48°58’36.594”N, 16°20’0.307”E	2011	Cambisols

was assessed. Vegetation was assessed using the standard method of phytocenological relevés. The coverage of all plant species was estimated and recorded in percents. Observations took place between 2020 and 2023, in three dates each year (in June, August and October). In each vineyard, 3 images were recorded, each measuring 2×4 meters. Taxonomic nomenclature of plants followed Kaplan et al. (2019). The identified plant species were divided into functional groups according to their biological properties.

Statistical data evaluation

The results of the assessment of the number of earthworms and the coverage of plant species groups were processed using multivariate analyses of ecological data. The selection of the optimal analysis was factored by the length of the gradient (lengths of gradient), determined by the Principal Component Analysis (PCA). Furthermore, Redundancy Analysis (RDA) was used. Statistical significance was determined using the Monte Carlo test where 999 permutations were calculated. All multivariate analyses and necessary calculations were performed in the Canoco 5.0 program (ter Braak and Šmilauer, 2012).

RESULTS AND DISCUSSION

Four species of earthworms were captured in the monitored vineyards during the four-year period. Two species were characterized by endogeic movement (*Aporrectodea caliginosa*, *A. rosea*) and two species by anectic movement (*Lumbricus terrestris*, *L. rubellus*). The average numbers of earthworm species found in vineyards of different ages are demonstrated in Figure 1. The species *Aporrectodea caliginosa* was the most numerous, the occurrence of this species was higher especially in younger vineyards, established between 2019 and 2021. The numbers of the species *Aporrectodea rosea* were also higher in young vineyards, established between 2019 and 2021, as well as in vineyards established in 2007 and 2008. The species *Lumbricus terrestris* was captured mainly in older vineyards established in 1972, 1995, 1996 and 1998. Earthworms of the species *Lumbricus rubellus* were registered mainly in vineyards established in 1998, 1999, 2007, 2019 and 2020. Trends in the occurrence of earthworms in the vineyards of different ages can be deduced from the obtained results. The occurrence of endogeic earthworm species (*Aporrectodea caliginosa*, *A. rosea*) had a decreasing trend with the growing age of the vineyard. The occurrence of *Lumbricus terrestris* increased with the growing age of the vineyard and the occurrence

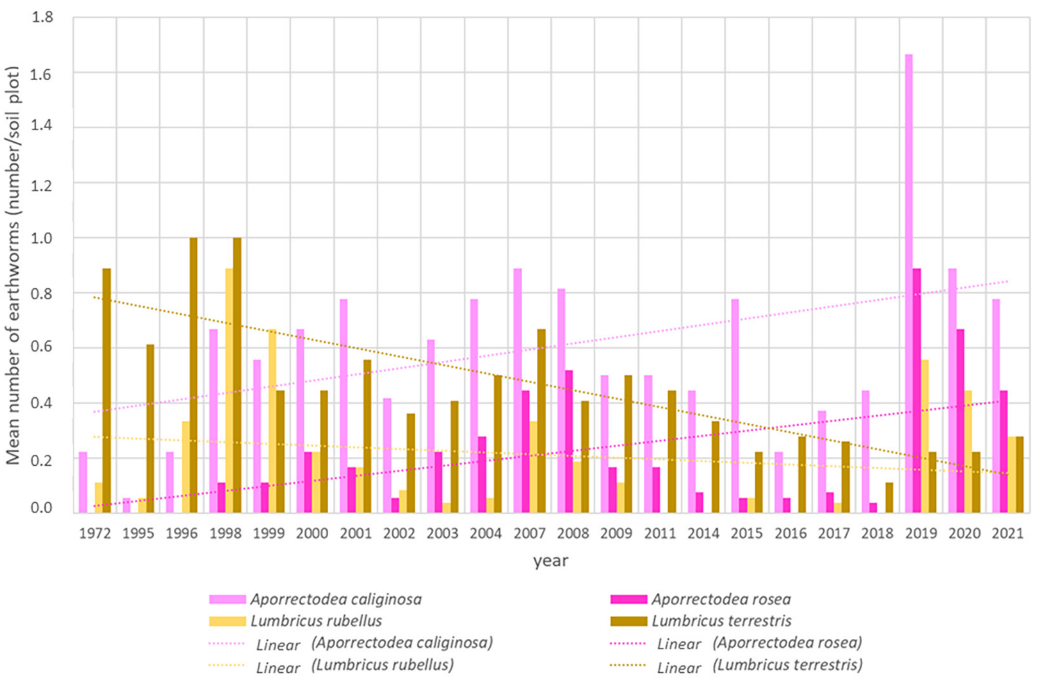

Figure 1. Earthworms populations in the conditions of vineyards of different ages

of the species *Lumbricus rubellus* increased only slightly with the growing age of the vineyard.

131 plant species were identified in the monitored vineyards. The identified species were divided into 5 groups. Species included in the group of annual dicots were *Amaranthus powellii, Amaranthus retroflexus, Anagallis arvensis, Anthemis arvensis, Atriplex sagittata, Brassicca napus* subsp. *napus, Camelina microcarpa, Camelina sativa, Capsella bursa-pastoris, Consolida regalis, Conyza canadensis, Datura stramonium, Erigeron annuus, Erodium cicutarium, Euphorbia helioscopia, Fagopyrum esculentum, Fumaria officinalis, Galium aparine, Geranium pusillum, Holosteum umbellatum, Chenopodium album, Chenopodium hybridum, Chenopodium polyspermum, Lactuca serriola, Lamium amplexicaule, Lamium purpureum, Linaria vulgaris, Linum usitatissimum, Matricaria discoidea, Mercurialis annua, Myosotis arvensis, Papaver rhoeas, Phacelia tanacetifolia, Pisum sativum, Polygonum aviculare, Portulaca oleracea, Raphanus raphanistrum, Raphanus sativus, Scleranthus annuus, Senecio vulgaris, Silene noctiflora, Sinapis alba, Solanum nigrum, Sonchus oleraceus, Stellaria media, Urtica urens, Veronica hederifolia, Vicia pannonica,* and *Viola arvensis*.

Species included in the group of legumes (family *Fabaceae*) were *Anthyllis vulneraria, Lotus corniculatus, Melilotus albus, Medicago lupulina, Medicago sativa, Melilotus officinalis, Onobrychis viciifolia, Securigera varia, Robinia pseudoacacia, Trifolium alexandrinum, Trifolium incarnatum, Trifolium pratense* and *Trifolium repens*. Species included in the group of perennial dicots were *Agrimonia eupatoria, Achillea millefolium, Arctium lappa, Arctium tomentosum, Artemisia absinthium, Berteroa incana, Carduus acanthoides, Cichorium intybus, Cirsium arvense, Convolvulus arvensis, Crepis biennis, Daucus carota, Echium vulgare, Eryngium campestre, Euphorbia esula, Falcaria vulgaris, Fragaria vesca, Galeopsis tetrahit, Galium album, Galium verum, Geranium pyrenaicum, Geum urbanum, Hypericum perforatum, Lamium album, Lathyrus tuberosus, Lepidium draba, Malva neglecta, Nonea pulla, Onopordum acanthium, Petrorhagia prolifera, Pilosella aurantiaca, Pilosella officinarum, Plantago lanceolata, Plantago major, Potentilla argentea, Potentilla reptans, Reseda lutea, Rosa canina, Rubus* sect. *Rubus, Rumex crispus, Rumex obtusifolius, Scabiosa ochroleuca, Senecio jacobaea, Silene latifolia* subsp. *alba, Symphytum officinale, Tanacetum vulgare, Taraxacum* sect. *Taraxacum, Tragopogon dubius, Tragopogon orientalis* and *Urtica dioica*.

Species included in the group of annual grasses were *Avena fatua, Bromus hordeaceus, Digitaria sanguinalis, Echinochloa crus-galli, Hordeum murinum, Secale cereale, Setaria pumila, Setaria viridis,* and *Triticum aestivum*. Species included in the group of perennial grasses were *Arrhenatherum elatius, Calamagrostis epigejos, Dactylis glomerata, Festuca arundinacea, Festuca pratensis, Festuca rubra, Lolium multiflorum, Lolium perenne, Poa pratensis,* and *Stipa pennata*.

The mean coverages of plant groups in vineyards of different ages are illustrated in Figure 2. The trends in the coverage of plant groups and the age of vineyards are evident from the obtained results. The group of annual dicotyledonous species had a higher coverage mainly in younger vineyards established between 2019 and 2021, their coverage was lower in older vineyards. The trend was decreasing coverage with increasing age of the vineyard. The group of perennial grasses had higher coverage in older vineyards and there was a noticeable trend of increasing coverage with increasing vineyard age. Similar to perennial grasses, this was also the case with the group of perennial dicotyledonous plant species. The coverage of annual grasses and clovers was similar and the coverage had only a slightly increasing trend with increasing vineyard age. The species composition of the vegetation corresponded to the grassy vegetation of inter-rows used in vineyards (Ragasová et al., 2021) and in orchards (Winkler et al., 2023) in the observed region.

The numbers of captured earthworms and values of coverage of plant groups obtained during monitoring were first processed by PCA. The adjusted explained variance was calculated and its value was 34.31. Subsequently, the data were processed by the RDA method in order to determine the relationship between earthworm species and plant groups in the conditions of vineyards of different ages. The RDA analysis defines a spatial arrangement of individual earthworm species, plant groups and the age of vineyards. The result of the analysis is graphically expressed in an ordination diagram (Figure 3). The results of the RDA analysis, evaluating the number of earthworms and the coverage of plant groups, was significant at the $\alpha = 0.001$ significance level for all canonical axes. Based on the RDA analysis, it can be seen that a higher occurrence of earthworms of the species

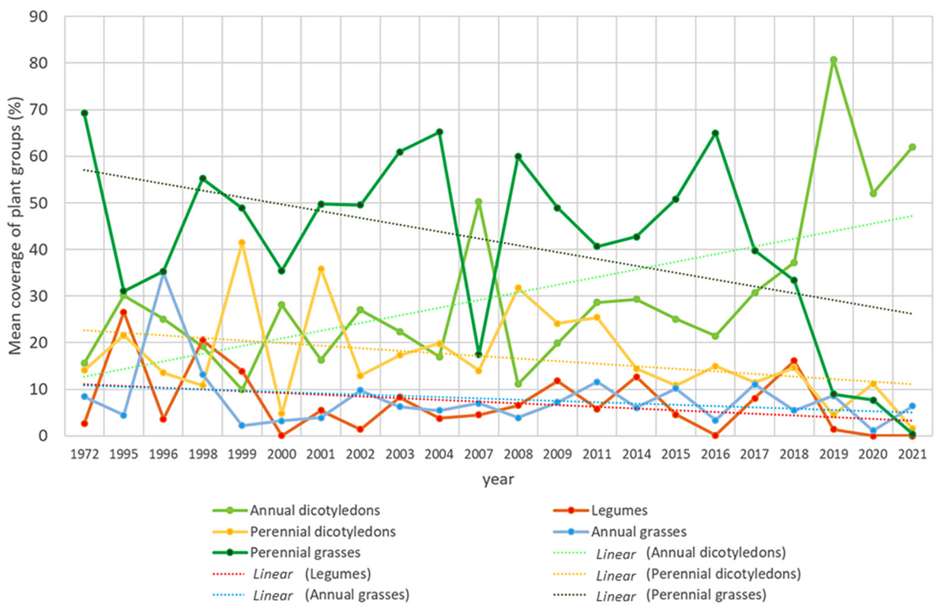

Figure 2. Coverage of plant groups in the conditions of vineyards of different ages

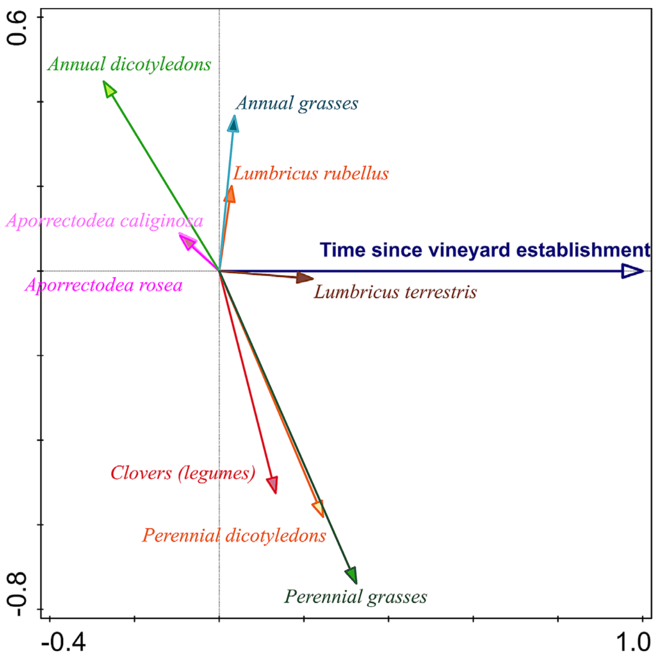

Figure 3. The relationship between the population of earthworms and groups of plants in the conditions of vineyards of different age (RDA result; total explained variance = 4.8 %; F-ratio = 20.1; P-value = 0.001)

Aporrectodea caliginosa and *A. rosea* was associated with a higher coverage of annual dicotyledonous plants. The occurrence of *Lumbricus rubellus* species was higher in vineyards with a higher coverage of annual grasses. A higher occurrence of *Lumbricus terrestris* species was recorded mainly in older vineyards. According to Schreck et al. (2012) soil management in vineyards has direct and indirect impacts on earthworm ecology and physiology. Both mechanical and chemical regulation of vineyard vegetation affect the earthworm populations in vineyards negatively. Nevertheless, the information on the influence of other agricultural practices in vineyards is lacking, such as grass biomass mulching, nitrogen fertilization, irrigation regimes and pesticide applications. It is evident from the results of this study that the fate of the earthworm population is linked to the dynamics and changes of non-target vineyard vegetation. Vegetation of the inter-rows grown in vineyards also supports the occurrence of other invertebrates species, such as *Collembola* and *Acari* (Möth et al., 2023).

Weeds provide food and shelter for a variety of animals (Marshall et al., 2003). Frequent soil cultivation, used in vineyards for weed control, induces low values of observed earthworm parameters (Chan and Munro 2001; Pommeresche and Loes 2009). Chemical weed control reduced the number of earthworms and had a negative effect on their activity (Pérès et al., 2010). Herbicides are the most commonly used pesticides, they have an important role in weed control strategies in vineyards (El Titi 2003; Kudsk and Streibig 2003), but they also affect the biodiversity of agroecosystem fauna (Freemark and Boutin 1995; Wardle et al., 1995). The direct harmfulness of herbicides on earthworms has not been proven (Gorzerino et al., 2009; Marwitz et al., 2012), but the application of herbicides causes a sharp decrease in vegetation biomass and a lack of food for earthworms, which can be considered an adverse consequence of weed regulation (Farenhorst et al., 2003). This is also supported by our results, according to which the occurrence of earthworms of the species *Aporrectodea caliginosa* is enhanced by a higher coverage of annual dicots, a group of plants that is often regulated by soil cultivation in vineyards. The availability of organic material increases the reproductive capacity of earthworms (Ivask et al., 2007; Capowiez et al., 2009). Limiting the application of herbicides, however, leads to a change in the species spectrum of weeds (Winkler et al. 2023b), which can trigger a response in the earthworm population.

Grassed zones in vineyards can become a large source of organic matter and so protect living organisms from their predators (Lacas et al., 2005). Grassy cover in vineyards generally has positive effects on earthworms (Eisenhauer et al., 2009). Long-term and stable management of grass communities leads to the creation of species-rich vegetation. Changes in the use of grasslands cause a decrease in the number of species and a change in the species composition of plants (Winkler et al., 2021; Winkler et al., 2022). Vršič (2011) found that soil managed with mulch increased the number of earthworms. However, according to our results, this only applies to the species *Lumbricus terrestris* and *Lumbricus rubellus*. Changes in the species composition of the earthworm population may reflect a different distribution of SOC in the soil profile. In the case of annual plant species, biomass can accumulate on the surface of the soil or in the soil to a depth of 0.1 m, which creates a more favorable living space for *Aporrectodea caliginosa* and *A. rosea*. In perennial plant species, SOC can accumulate even at depths below 0.1 m, thanks to which *Lumbricus rubellus* and *L. terrestris* can thrive better. As pointed out by Capowiez et al. (2009), the number of earthworms can be influenced by the date of observation, the course of precipitation and temperatures; the numbers of earthworms in individual locations can be considerably different.

Earthworms play indisputably a key role in soil biology. They influence soil structure through their decomposing activities and bioturbulence (Römbke et al., 2005). Soil has a special meaning in viticulture (Giffard et al., 2022), especially in the context of the term terroir (Van Leeuwen et al., 2004). Soil properties influence grape development and ripening as well as several grape quality parameters such as sugar content, total acidity and grape pH (Van Leeuwen et al., 2004), to which inter-row management in vineyards contributes (Griesser et al., 2022). The influence of earthworms on soil properties and quality varies according to their species and functional groups of earthworms (Ernst et la. 2009). Anectic species can increase turnover of organic residues and microbial activity. Therefore, they also contribute to increased mineralization and availability of nutrients in the soil, endogeic species change the structure of the soil by their activity (Langmaack et al., 1999).

According to Chalkia et al. (2021), there are large differences in earthworm populations between ecosystems, but also between habitats within ecosystems. In cultivated fields, good agricultural practices support high earthworm populations. Based on our results, it is clear that young vineyards with a higher proportion of annual vegetation support earthworm populations that are more similar to those from arable land. Older vineyards with a higher proportion of grasses have a population more similar to natural habitats. A similar earthworm species composition was also reported by Simon et al. (2022), but with higher abundance and more earthworm species. This is probably due to the different soil characteristics, especially the drier conditions and lower organic matter content of the vineyards studied.

The vegetation management of the vineyards is unique in its character, creating a unique environment for earthworm populations. The combination of the presence of diverse vegetation subject to natural succession also triggers a response in the earthworm population.

CONCLUSIONS

Based on four-year monitoring, it was found that the population of eartworms was influenced by the age of the vineyards and by the presence of different plant groups. Earthworm species with predominantly horizontal movement were present in the soil horizon of younger vineyards, and their number was lower in older vineyards. Species with predominantly vertical movement were present mainly in the soils of medium-aged and old vineyards. The vegetation of the vineyards undergoes succession. Annual dicotyledonous species recede over time. The coverage of perennial dicotyledonous species and grasses increase with the growing age of vineyards. The results showed that the earthworm species *Aporrectodea caliginosa* and *A. rosea* responded positively to a higher coverage of annual dicotyledonous plants. Higher coverage of annual grasses in vineyards increases the occurrence of *Lumbricus rubellus* species.

Our results indicate a closer relationship between the biomass of annual dicotyledonous plants and earthworm populations of the genus *Aporrectodea*. Therefore, it is necessary to create conditions for this vegetation and to change the perception of annual vegetation in vineyards as an undesirable weed and to realize its ecosystem functions. The use of sandwich management (alternating cultivated and herbaceous intercropping) will fragment vineyard areas and create favorable conditions for annual plants. Future research should focus on adapting the management of vegetation biomass to maintain and increase earthworm populations. Maintaining and increasing earthworm populations will allow us to benefit longer from their ecosystem functions, such as promoting rainfall infiltration, reducing erosion, increasing soil organic matter, and increasing soil carbon storage as part of carbon neutral agriculture.

Vegetation in vineyards creates living spaces for earthworms. Annual dicotyledonous species are food sources for earthworms. Biomass production is an ecosystem function that supports the growth of soil edaphones. This knowledge increases the biological relevance of annual dicotyledonous species, which are often considered undesirable weeds. Changes in vegetation due to succession will also be reflected in the species composition of the earthworm population.

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