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Activity of antioxidant enzymes and malondialdehyde content in sweet cherry fruits under living mulch conditions

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Abstract

Living mulch is the most natural soil cover in the orchard; however, competition with grasses creates stressful conditions for fruit crops. The reaction of plants to stress can be determined by the activity of antioxidant enzymes and the content of lipid peroxidation products in plant tissues. The reaction of trees to coexistence with living mulch (natural grasses that were mowed four times during the growing season) was studied in an organic sweet cherry (*Prunus avium* L.) orchard with chestnut, sandy soil in the warm and arid conditions of Southern Steppe of Ukraine. The aim of the research was to determine how the living mulch (compared to bare fallow) affects the activity of antioxidant enzymes and the content of malondialdehyde (MDA) in sweet cherry fruits at different stages of fruit development. It was determined that living mulch significantly increased the activity of ascorbate peroxidase (by 21–52%), polyphenol oxidase (by 22–42%), and peroxidase (26–34%) in the tissues of sweet cherry fruits. The MDA content was significantly higher in sweet cherry fruits produced under living mulch (compared to the fruits of a bare fallow management system) only at the stage of partial reddening in 2018 (by 41%) and at the stage of stone hardening in 2019 (by 58%). At the picking maturity stage, no significant difference between the treatments of the experiment was found, which indicates the successful overcoming of oxidative stress caused by competition with natural grasses.

Keywords: *Prunus avium*, catalase, ascorbate peroxidase, polyphenol oxidase, peroxidase.

Introduction

Modern progressive humanity seeks to live in a fairer and safer world; therefore, along with overcoming poverty and providing food, it sets itself the goal of leaving the earth suitable for the lives of future generations (United Nations, 2015). Living mulch is the optimal solution for preserving and improving soil fertility in the orchard for the development of sustainable agriculture (Holden et al., 2017). First, just as animals have fur, the soil must have a protective covering that acts as an insulator protecting the soil from overheating in summer (this is especially relevant in the Southern Steppe zone of Ukraine) and freezing in winter (Gu et al., 2016). Secondly, the grass cover acts as a shock absorber slowing down the wind speed and reducing the impact force of the raindrops (also, protects the soil from deflation and spraying) (Fidalski et al., 2010). The root system of grass slows down water erosion in the soil (Atucha et al., 2013). Turf is a nesting place for numerous pollinators and beneficial insects, which normalises the natural balance of agrobiocenosis (Carvell et al., 2022). The rhizosphere of grasses is the habitat for numerous beneficial soil biota (Zheng et al., 2018; Culumber et al., 2019). However, living mulch has a negative effect on the physiological status of fruit crops, as natural grasses compete with fruit trees for water, nutrients, and mycorrhizal symbionts (Xing et al., 2012).

The use of natural grasses as living mulch also has the drawbacks: in addition to the competition for water and nutrients, which is typical of any living mulch, they can be a reservoir of nematodes (Kalatur, Pylypenko, 2017) and increase rodent populations (Merwin et al., 1999). Therefore, separately selected plant species are often used in turf orchards (Mia et al., 2021). However, the use of native plants as living mulch is important to preserve local authentic flora (Radić Lakoš et al., 2014; Oldfield et al., 2019). Natural herbs (considered weeds)

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can also successfully perform the function of a flower strip, as they are more adapted to local soil and climatic conditions compared to sown cultivated plant species (Fountain, 2022; Kowalska et al., 2023).

Over time, trees overcome this competition by shading grasses with their crowns and growing their roots into deeper soil horizons (Merwin, 2010). However, the physiological reactions of trees accompanying by this overcoming of competition with grasses have not been fully determined yet. The level of stress in plants can be indirectly determined by the amount of lipid peroxidation and malondialdehyde (Kolupaev et al., 2022). The activity of antioxidant enzymes is an indicator of plant resistance to the harmful effect of stress (Cansev, Kesici, 2013; Jovičić et al., 2017; Kolupaev et al., 2022). Studying the physiological response of sweet cherry (*Prunus avium* L.) trees to stress requires a holistic approach. Antioxidant protection of sweet cherry leaves during the growing season under living mulch has been previously reported (Gerasko et al., 2022b). The current study focuses on the status of the antioxidant defence system of fruits. The state of the antioxidant defence system is an important physiological indicator of cherry fruits, which can be used to assess their physiological differences and evaluate their adaptability to growing conditions (Hegedűs et al., 2013; Skrzyński et al., 2016; El Baji et al., 2019). At the same time, short-term oxidative stress is important for maintaining normal growth and development of cherry trees (Cai et al., 2019). According to the state of the antioxidant defence system in the tissues of cherry fruits at different stages of development and ripening, it is possible to trace the formation of their consumer quality (Tahir et al., 2013).

The aim of the study was to determine how living mulch affects the activity of antioxidant enzymes and the content of malondialdehyde in sweet cherry fruits at different stages of ripening. The obtained data will help to clarify how fruit trees react to stress caused by grass competition and what level of stress due to competition with grasses is critical for trees.

Material and methods

The site of the experiment was a research orchard of Dmytro Motornyi Tavria State Agrotechnological University in the Zelene village (46°46′ N, 35°17′ E), Melitopol district, Zaporizhzhia region, the Southern Steppe of Ukraine. The soil of the experimental site is chestnut, sandy, of light mechanical composition, and with a slightly alkaline reaction of soil solution (pH ranges from 7.1 to 7.4). No mineral nitrogen was detected; the content of P_2O_5 was 5.4 mg kg^{-1}, and K_2O 6.5 mg kg^{-1} of the soil. The upper layer of the soil contained little humus (0.6%), and total content of water-soluble salts was 0.015–0.024%. The upper layer of the soil was very poor in organic matter and the main elements of mineral nutrition, but sweet cherry (*Prunus avium* L.) on *P. mahaleb* L. rootstock had been successfully grown in such soils since the 19th century. Moreover, the trees were watered only during the first three years. Later, due to the deep root system of *P. mahaleb*, trees can grow without watering. The point is that the valley of the breakthrough of the fluvioglacial flow, which was formed during the melting of the Quaternary glacier, created unique soil conditions in the Melitopol district – under the upper (approximately 70–90 cm) layer of sand there is an about 1.5 m buried black soil (Chebanova, 2019).

The climate of the experimental area is warm and arid – long-term mean air temperature is +10.6°C, and mean annual precipitation for the last 10 years was 481 mm. The evaluation of weather conditions over the experimental years show that climate is warming – the average annual temperature was 1.2–1.6°C warmer relative to the long-term normal (Table 1). The April of 2018 was warm and very dry. The drought also lasted in May and June of 2018. April and May of 2019 were satisfactory in terms of moisture supply, but the June of 2019 was unusually hot and dry. Since picking maturity was on 1 June in 2018 and on 6 June in 2019, it can be stated that the conditions of fruit development in 2018 were unusually dry, and in 2019 this period was satisfactory in terms of moisture.

Table 1. Weather conditions during the period of sweet cherry fruit development (data of Melitopol Weather Station)

Year	April		May		June		Average annual	
	value	relative to long-term normal	value	relative to long-term normal	value	relative to long-term normal	value	relative to long-term normal
Mean monthly temperature °C								
2018	13.4	+3.0°C	19.8	+3.0°C	23.4	+2.2°C	11.8	+1.2°C
2019	11.4	+1.0°C	18.3	+1.5°C	25.3	+4.1°C	12.2	+1.6°C
Precipitation mm								
2018	5.5	−84%	22.4	−52%	32.4	−40%	528	+10%
2019	49.4	+44%	96.2	+107%	14.4	−73%	442	−8%

Sweet cherry cultivar ‘Dilemma’ with *P. mahaleb* L. rootstock was planted in 2011, at a distance of 7 × 5 metres. The cultivar ‘Dilemma’, grown in the Southern Steppe of Ukraine, ripens at the beginning of June, the fruits are convex heart-shaped, the skin and pulp are dark red in colour, with an excellent sweet and sour taste.

The most dominant natural grasses species present in the experimental plot were *Descurainia sophia* L., *Capsella bursa-pastoris* L., *Anthemis arvensis* L., *Papaver rhoeas* L., *Vicia villosa* Routh, *Elytrigia repens* L., *Cynodon dactylon* L., *Trifolium arvense* L., and *Avena fatua* L. Small curtains of *Achillea millefolium* L., *Echium vulgare* L., *Delphinium consolida* L., *Verbascum phlomoides* L., and *Lycopsis arvensis* L. were also observed. The amount of above-ground dry biomass of living mulch was 289 g m^{-2} in mid-May 2018 and 456 g m^{-2} in mid-May 2019, and the percentage of soil coverage by living mulch was 89% and 100%, respectively. All named species are weeds, but they are considered an important part of the natural biocenosis. It is interesting that all of them have medicinal properties (Ильина, 2015), and some of them (*A. millefolium* L., *C. bursa-pastoris* L., *E. repens* L., and *P. rhoeas* L.) are officially recognised as medicinal plants (State Pharmacopoeia of Ukraine, 2014).

The experiment was designed as a randomized complete block with two treatments, three replications.

Each experimental plot consisted of 36 trees spanning 3 rows with 12 trees per row. For measurements, 10 central trees of the middle row were used, and the other 26 were protective trees. Orchard rows were oriented from north to south. Trees were trained as central leader. Since 2013, the experimental orchard has been maintained with two different orchard floor management systems: bare fallow (disking to a depth of 15 cm, hand weeding as weeds re-emerged) and living mulch (natural grasses). Natural grasses were mowed four times during the growing season (in April, May, June, and August), and the clippings were left on the ground to decompose; any other management was the same for each treatment. Synthetic fertilisers, chemical plant protection products, and irrigation were not used.

Fruit sampling for analysis was carried out during May–June 2018 and 2019 at the growth stages: petal fall, stone hardening, partial reddening, and picking maturity. For laboratory analyses, 30 intact fruits in four replicates were selected from each experimental plot (in total 120 fruits per each experimental plot).

The collected data included the content of malondialdehyde (MDA, nmol g^{-1}) and the activity of antioxidant enzymes: catalase (CAT, μmol H_2O_2 g^{-1} min^{-1}), ascorbate peroxidase (APX, mg of oxidised ascorbic acid g^{-1}), polyphenol oxidase (PPO, U g^{-1} min^{-1}), and peroxidase (POD, mCAT g^{-1}), in sweet cherry fruits.

The content of MDA was determined spectrophotometrically with a scanning spectrophotometer UV-2800 (UNICO, USA), as described by Costa et al. (2002). The method is based on the formation of a pink trimethyl complex with an absorption maximum at 535 nm from the reaction of MDA with thiobarbituric acid (TBA) at 95°C in the acidic environment: 250 mg of plant material was homogenised with 4 ml of 20% trichloroacetic acid (TCA) and filtered. 4 mL of 0.5% TBA dissolved in 20% TCA was added to 1 mL of the filtrate and boiled for 30 min at 95°C in a water bath, then cooled. Optical density measurements were performed at wavelengths of 532 and 600 nm.

The content of MDA was calculated according to equation 1:

$$C = (E_{532} - E_{600})\, E^{-1} \quad (1),$$

where C is MDA concentration; E_{532} and E_{600} are optical density at wavelengths of 532 and 600 nm, respectively; E is the molar extinction coefficient of the trimethyl complex at a beam length of 1 cm ($E = 1.55 \times 10^5$ M^{-1} cm^{-1}). The result of the MDA content calculation was expressed in nm per g of fresh mass.

To determine the activity of catalase (CAT, EC 1.11.1.6), Korolyuk et al. (1988) method was used, based on the ability of hydrogen peroxide (H_2O_2) to form a stable-coloured complex with molybdenum salts. For this, the reaction was initiated by adding 0.1 ml of plant homogenate to 3 ml of 0.03% H_2O_2 solution. The reaction was stopped after 10 min by adding 4% ammonium molybdate, followed by photometry at a wavelength of 400 nm against the control sample, where instead of the homogenate 0.1 ml of distilled water was added. The CAT activity was calculated according to equation 2:

$$A = (Ec - Et)\, N\, (0.1\, t\, K) - 1 \quad (2),$$

where A is the activity of CAT, μmol H_2O_2 g^{-1} min^{-1}; Ec is extinction of the control sample; Et is extinction of the test sample; N is dilution factor; 0.1 is the volume of homogenate, ml; t is incubation time, min; K is the extinction coefficient of H_2O_2, equal to 22.2 × 103 mm^{-1} cm^{-1}.

The activity of ascorbate peroxidase (APX, EC 1.1.11.1) was determined as described by Gorodniy et al. (2006): by titration of the residual unoxidised ascorbic acid with a 0.001 n solution of 2,6-dichlorophenolindophenol to a light pink colour that does not disappear within 30 seconds. In the control, APX was deactivated with metaphosphoric acid.

The activity of polyphenol oxidase (PPO, EC 1.10.3.1) was determined by Ermakov et al. (1987) method: the optical density of the reaction products formed during the oxidation of pyrocatechin was measured at a wavelength of 420 nm. PPO activity was expressed in units per 1 g of fresh mass per 1 min.

The activity of peroxidase (POD, EC 1.11.1.7) was determined by Frew et al. (1983) method: peroxidase decomposes H_2O_2, releasing oxygen. Oxygen oxidises indigo carmine. As a result, indigo carmine changes colour from blue-green to yellow-pink. The sample of plant tissue (1 g) was homogenised with acetate buffer (6 ml, pH = 4.9), and 2 ml of indigo carmine were added. After that, 0.5 ml of 0.03 M H_2O_2 solution was added to 1 ml of homogenate. After 2 min, sulphuric acid (20% H_2SO_4) was added to stop the reaction, then photometered at a wavelength of 610 nm against distilled water. The control sample contained 0.5 ml of distilled water instead of H_2O_2. The POD activity was calculated according to equation 3:

$$A = (E_c - E_t)\, N\, (\varepsilon\, d\, t)^{-1} \quad (3),$$

where A is the activity of POD, mCAT g^{-1}; E_c is the optical density of the control sample; E_t is the optical density of the test sample; N is the final dilution factor; ε is the coefficient of millimolar extinction of indigo carmine, $\varepsilon = 10.5 \times 10^{-3}$; d is the thickness of the cuvette, cm; t is incubation time, s.

Statistical analysis. All analyses were repeated three times (biological replicates). The results were compared by the Tukey's HSD (honestly significant difference) test at a significance level of $P \leq 0.05$ and were processed by Pearson's correlation analysis using software Minitab 19 (Minitab Inc., USA). The tables present mean values with standard deviations (±SD).

Results and discussion

Sweet cherry trees experience stress from coexistence with natural grasses, but this does not necessarily lead to a reduction in fruit size and quality. Thus, in the current study (as shown previously), fruit size and weight tended to decrease under living mulch conditions, but the difference compared to bare fallow treatment was insignificant (Gerasko, 2020). At the same time, living mulch contributed to the accumulation of tissue antioxidants in fruits, which increases their consumption quality (Gerasko et al., 2022a). For a deeper understanding of the physiological response of sweet cherry trees to the stress of competition with natural grasses, it was necessary to analyse the state of the antioxidant defence system.

MDA content in sweet cherry fruits gradually increased as they matured (Tables 2 and 3). There is a tendency to the increase of MDA content under living

Table 2. Malondialdehyde (MDA) content and the activity of antioxidant enzymes in sweet cherry fruits in 2018

Treatment	MDA nmol g^{-1}	CAT µmol H_2O_2 g^{-1} min^{-1}	APX mg of oxidised ascorbic acid g^{-1}	PPO U g^{-1} min^{-1}	POD mCAT g^{-1}
Petal fall					
Bare fallow	20.6 ± 2.15 e	6.3 ± 0.55 d	7.2 ± 0.33 g	3.7 ± 0.22 e	7.8 ± 0.54 f
Living mulch	24.5 ± 1.85 e	6.1 ± 0.42 d	10.6 ± 0.35 f	3.9 ± 0.27 e	10.1 ± 0.55 e
Stone hardening					
Bare fallow	36.4 ± 2.17 c	7.6 ± 0.33 c	20.3 ± 0.42 e	10.8 ± 0.34 d	20.2 ± 0.78 b
Living mulch	38.5 ± 2.24 c	7.5 ± 0.26 c	25.4 ± 0.47 d	15.2 ± 0.33 c	25.4 ± 0.81 a
Partial reddening					
Bare fallow	32.2 ± 3.35 d	9.2 ± 0.33 b	35.9 ± 0.55 c	15.4 ± 0.49 c	14.5 ± 0.67 d
Living mulch	45.5 ± 3.65 b	8.9 ± 0.25 b	45.6 ± 0.58 b	20.6 ± 0.55 b	18.9 ± 0.69 c
Picking maturity					
Bare fallow	50.8 ± 4.35 a	10.7 ± 0.40 a	46.1 ± 0.64 b	20.9 ± 0.65 b	15.2 ± 0.54 d
Living mulch	49.4 ± 3.14 a	11.4 ± 0.41 a	55.8 ± 0.65 a	25.5 ± 0.67 a	20.4 ± 0.57 b

CAT – catalase, APX – ascorbate peroxidase, PPO – polyphenol oxidase, POD – peroxidase; significant at $p < 0.05$

Table 3. Malondialdehyde (MDA) content and the activity of antioxidant enzymes in sweet cherry fruits in 2019

Treatment	MDA nmol g^{-1}	CAT µmol H_2O_2 g^{-1} min^{-1}	APX mg of oxidised ascorbic acid g^{-1}	PPO U g^{-1} min^{-1}	POD mCAT g^{-1}
Petal fall					
Bare fallow	42.0 ± 3.51 b	6.1 ± 0.21 g	8.8 ± 0.25 g	4.5 ± 0.24 f	9.4 ± 0.45 e
Living mulch	40.5 ± 3.55 b	7.2 ± 0.25 f	13.4 ± 0.29 f	4.8 ± 0.25 f	10.3 ± 0.49 e
Stone hardening					
Bare fallow	35.6 ± 4.61 c	10.7 ± 0.30 e	24.6 ± 0.33 e	13.2 ± 0.31 e	14.6 ± 0.89 d
Living mulch	56.4 ± 3.45 a	12.1 ± 0.20 d	31.4 ± 0.35 d	18.8 ± 0.35 d	25.3 ± 0.86 a
Partial reddening					
Bare fallow	41.5 ± 2.45 b	19.3 ± 0.30 c	44.1 ± 0.47 c	18.9 ± 0.49 d	28.5 ± 0.76 a
Living mulch	35.5 ± 2.65 c	23.5 ± 0.34 b	56.4 ± 0.45 b	23.5 ± 0.44 c	20.5 ± 0.74 b
Picking maturity					
Bare fallow	49.5 ± 3.21 a	45.4 ± 0.51 a	56.7 ± 0.59 b	25.7 ± 0.52 b	17.8 ± 0.65 c
Living mulch	52.4 ± 3.25 a	47.3 ± 0.67 a	69.9 ± 0.62 a	32.6 ± 0.58 a	23.3 ± 0.59 b

CAT – catalase, APX – ascorbate peroxidase, PPO – polyphenol oxidase, POD – peroxidase; significant at $p < 0.05$

mulch and a significant increase of MDA content in the fruits grown under living mulch compared to bare fallow treatment, at the stages of partial reddening in 2018 (by 41%) and of stone hardening in 2019 (by 58%). But at the stage of partial reddening of fruits in 2019, MDA content in fruits under living mulch was significantly lower (by 15%) compared to that under bare fallow. The increase of MDA content in sweet cherry fruits under living mulches indicates a higher level of lipid peroxidation and is an indicator of oxidative damage to cell membranes (Dos Santos et al., 2017). According to a significant increase in MDA content, it can be stated that sweet cherry trees suffered from the oxidative stress more at the stage of partial reddening in 2018 and at the stage of stone hardening in 2019. At the picking maturity stage, no significant difference was observed between the treatments of the experiment both in 2018 and in 2019. This means the successful overcoming of oxidative stress caused by competition for water and nutrients with grasses by sweet cherry trees.

The activity of CAT showed a tendency to decrease under living mulch compared to bare fallow at the stages of petal fall, stone hardening, and partial reddening in 2018, but the difference was insignificant. At the stages of petal fall, stone hardening, and partial reddening in 2019, CAT activity was higher (by 18, 13, and 22 %, respectively) in fruit tissues under living mulch compared to bare fallow. During two experimental years, CAT activity in sweet cherry fruits gradually increased as they matured. In 2019, CAT activity was significantly higher in both treatments of the experiment and at the picking maturity stage was 4.1–4.2 times higher compared to 2018.

The APX is an important part of the AsA-GSH cycle: it uses two AsA molecules to reduce H_2O_2 to water (Ahmad et al., 2010). The APX activity increased in the tissues of the fruits as they matured and was significantly higher under living mulch, both in 2018 (by 21–47%) and in 2019 (by 23–52%). In 2019, APX activity in the fruits was higher compared to 2018: by 23% under bare fallow and by 25% under living mulch.

The activity of PPO also increased as the fruits ripened and was significantly higher under living mulch, except for the petal fall stage. Increased PPO activity during maturation is associated with the softening of cell walls in the tissues of the fruits (Richter, 2001). It should be noted that the difference between the treatments gradually smoothed out as the fruits ripened: it reached 41–42% at the stone hardening stage, 24–33% at the partial reddening stage, and 22–27% at the picking maturity stage.

In 2018, POD activity in sweet cherry fruits increased rapidly from the stage of petal fall to the stone hardening one in the tissues of the fruits of both treatments, then decreased at the partial reddening stage and slightly restored (insignificantly increased) at the

picking maturity stage. The dynamics of POD activity can be explained by the participation of this enzyme in the process of lignification of the endocarp during the stone hardening stage (Richter, 2001), which requires sufficient production of this enzyme. During the fruit ripening period in 2018, under living mulch, POD activity was significantly higher by 32, 26, 30, and 34 % at the stages of petal fall, stone hardening, and partial reddening, respectively. In 2019, POD activity in the fruits, similar to 2018, increased rapidly before the stone hardening stage in both treatments and was significantly higher (by 73%) under living mulch compared to bare fallow at this stage. However, later POD activity continued to increase in the fruits under bare fallow; under living mulch it decreased and at the partial reddening stage was significantly lower (by 28%). Before the picking maturity stage, POD activity in the tissues of the fruits decreased under bare fallow, while under living mulch it increased and was significantly higher (by 31%). The dynamics of POD activity in sweet cherry fruits in the present study did not coincide with the dynamics of PPO activity. This can be explained by the dynamics of organic acid accumulation: in the study of Gerasko et al. (2022a), the content of titrated acids decreased during fruit ripening, and the optimal substrate for POD activity is pH = 4.6–4.8, and for PPO it is pH = 6.0 (Richter, 2001).

Correlation analysis showed that MDA content in sweet cherry fruits in 2018 correlated mostly with the activity of PPO, APX, and CAT (Table 4).

In 2019, only an average correlation of MDA content with CAT activity was found.

It is known that environmental stresses (such as drought) increase the activity of antioxidant enzymes (Mirfattahi et al., 2017). Moreover, more stress-resistant plants have a higher activity of antioxidant enzymes and a higher amount of protective substances (Ahmad et al., 2010). In the current study, sweet cherry fruits had a higher activity of APX, PPO, and POD under living mulch. At the same time, in the content of MDA, although it increased under living mulch at the picking maturity stage, the differences between the treatments evened out.

According to the results of the research, it can be stated that the level of stress from competition with living mulch was not critical for sweet cherry trees. The scientific significance of the present study is that it shows for the first time the temporal patterns of the content of MDA and the activity of PPO, APX, CAT, and POD in sweet cherry fruits at different stages of fruit development under living mulch. The practical significance is that it can contribute to the introduction of living mulch in the arid conditions of the Southern Steppe of Ukraine.

Table 4. Correlation coefficients (r^2) of malondialdehyde (MDA) content with the activity of antioxidant enzymes in sweet cherry fruits

Year	CAT	APX	PPO	POD
2018	0.76**	0.83*	0.89*	0.42*
2019	0.29*	0.19 ns	0.16 ns	0.18 ns

CAT – catalase, APX – ascorbate peroxidase, PPO – polyphenol oxidase, POD – peroxidase; *, ** – significant at 0.01 and 0.05, ns – not significant

Conclusions

1. The content of malondialdehyde (MDA) in sweet cherry fruits increased as the fruit ripened, and at the picking maturity stage there was no difference between the treatments.

2. Under the conditions of living mulch, catalase (CAT) activity in sweet cherry fruits fluctuated compared to the bare fallow management system, in a smaller or larger extent, depending on the year. However, there was no difference between both orchard floor management systems at the picking maturity stage.

3. The activity of ascorbate peroxidase (APX) in sweet cherry fruits was significantly higher (by 21–52%) under living mulch, and the difference between the treatments remained until the picking maturity stage, both in 2018 and in 2019.

4. The activity of polyphenol oxidase (PPO) in sweet cherry fruits was significantly higher (by 22–42%) under living mulch.

5. In 2018, the activity of peroxidase (POD) in sweet cherry fruits was significantly higher (by 26–34%) under living mulch. In 2019, this indicator varied widely, but at the picking maturity stage, it was higher (by 31%) under living mulch.

6. The MDA content in sweet cherry fruits in the dry year 2018 had a strong correlation with the activity of PPO, APX, and CAT, and in 2019 (more favourable in terms of moisture supply) there was only an average correlation with the activity of CAT.

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