# Physiological Aspects of Olea europaea (Oleaceae) Attacked by Saissetia oleae (Hemiptera: Coccidae)

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# Physiological aspects of *Olea europaea* (Oleaceae) attacked by *Saissetia oleae* (Hemiptera: Coccidae)

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**Abstract**

Information on the occurrence and damage by *Saissetia oleae* (Olivier) (Hemiptera: Coccidae) on *Olea europaea* L. (Oleaceae) plants is scarce. *Saissetia oleae* is a sucking insect and its feeding on the phloem affects the photosynthetic apparatus of cells. The objective of this research was to evaluate the occurrence and to determine the effect of *S. oleae* on the physiological characteristics of *O. europaea* cultivars. The experiment was carried out in the field in Diamantina, Minas Gerais State, Brazil, in a randomized block design in a 2 × 4 factorial scheme (plants of the cultivars 'Arbequina,' 'Ascolano,' 'Grappolo,' and 'Koroneiki' attacked or not). The total number and population density of *S. oleae* on the abaxial and adaxial sides of the *O. europaea* leaves were evaluated by direct counting in summer, autumn, and winter. The emission of chlorophyll was evaluated by measuring the fluorescence parameters on leaves per treatment and season. The total number of *S. oleae* was higher on the cultivar Grappolo than on the Ascolano and Arbequina. Grappolo, Koroneiki, and Ascolano showed a population decline of this insect in winter. The initial chlorophyll "a" fluorescence was higher in *O. europaea* damaged by *S. oleae*. The maximum photochemical quantum yield of photosystem II/maximum fluorescence ratio of chlorophyll "a" was equal to or greater than 0.75 for all *O. europaea* cultivars not attacked by *S. oleae* in the summer, autumn, and winter. The electron transport rate was lower in plants attacked, except for those of the Grappolo in the winter. The increase of initial fluorescence, reduction of maximum photochemical quantum yield of photosystem II/maximum fluorescence, and electron transport rate in some cultivars of *O. europaea* attacked by *S. oleae* indicate damages to the photosynthetic apparatus, resulting in a possible decrease in growth and yield of the plants.

Key Words: Arbequina; Ascolano; chlorophyll fluorescence; Grappolo; insect pest; Koroneiki

**Resumo**

Informações sobre a ocorrência e danos de *Saissetia oleae* (Olivier) (Hemiptera: Coccidae) em plantas de *Olea europaea* L. (Oleaceae) são escassas. *Saissetia oleae* é um inseto sugador e, ao se alimentar do conteúdo do floema, afeta o aparelho fotossintético das células. O objetivo desta pesquisa foi avaliar a ocorrência e determinar o efeito de *S. oleae* nas características fisiológicas de cultivares de *O. europaea*. O experimento foi conduzido em campo em Diamantina, Minas Gerais, Brasil, em delineamento de blocos casualizados em esquema fatorial 2 × 4 (plantas atacadas ou não das cultivares 'Arbequina,' 'Ascolano,' 'Grappolo,' e 'Koroneiki'). O número total e a densidade populacional de *S. oleae* em *O. europaea* nas faces abaxial e adaxial das folhas foram avaliados no verão, outono e inverno. A emissão de clorofila foi obtida baseada nos parâmetros de fluorescência nas folhas por tratamento e estação. O número total de indivíduos de *S. oleae* foi maior na cultivar Grappolo que nas Ascolano e Arbequina e a população desse inseto diminuiu nas Grappolo, Koroneiki, e Ascolano no inverno. A fluorescência inicial da clorofila "a" foi maior em plantas de *O. europaea* atacadas por *S. oleae*. A relação do rendimento quântico fotoquímico máximo do fotossistema II/fluorescência máxima da clorofila "a" foi igual ou superior a 0,75 em todas as cultivares de *O. europaea* não atacadas por *S. oleae* no verão, outono e inverno. A taxa de transporte de elétrons foi menor em plantas atacadas, exceto para as de Grappolo no inverno. O aumento da fluorescência inicial e a redução do rendimento quântico fotoquímico máximo do fotossistema II/fluorescência máxima e transporte de elétrons em algumas cultivares de *O. europaea* atacadas por *S. oleae* indicam danos no aparato fotossintético e possível redução no crescimento e produção de plantas.

Palavras Chave: Arbequina; Ascolano; fluorescência da clorofila; Grappolo; inseto praga; Koroneiki

The olive tree, *Olea europaea* L. (Oleaceae), is cultivated extensively worldwide in climates apart from the Mediterranean, with growing oil and fruit production (Guex et al. 2018). This plant was introduced in the Americas and cultivated in Argentina, Chile, the USA (California, Florida, Hawaii, and Texas) and Brazil (Minas Gerais and Rio Grande do Sul) (Wrege et al. 2015; Allan & Gillett-Kaufman 2018; Lima et al. 2019).

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*Olea europaea* is produced most in small farms, but larger areas are being planted to serve the industry. Insect pests are the main biotic stressors reducing the development and production of olive.

*Saissetia oleae* (Oliver) (Hemiptera: Coccidae) is an important pest in the world and abundant in olive plantations (Ilias & Hammadi 2017). The nymphs and adult females of this insect suck the sap of branches and leaves and can cause direct damage at all stages of plant development and indirect by excreting honeydew. This substance increases the growth of sooty mold fungi that, by darkening branches, leaves, and fruits, reduce the plant's photosynthetic rate (Calvo-Agudo et al. 2022). *Saissetia oleae* also reduces the availability of nutrients and the growth of *O. europaea* plants, with symptoms such as chlorosis, leaf abscission, and decreased productivity (Velikova et al. 2010).

Determination of the stress level and prevention of losses by insects depend on the physiological plant processes (Ferreira et al. 2015). Calculation of fluorescence parameters is a non-destructive method to observe the fluorescence emission of chlorophyll. It allows the analysis of the absorption and use of light energy by photosystem II, and possible relationships with photosynthetic capacity (Netto et al. 2005). The initial fluorescence, maximum fluorescence, maximum photochemical quantum yield of photosystem II (maximum photochemical quantum yield of photosystem II/maximum fluorescence), and electron transport rate allow to evaluate physiological stress in attacked plants and to determinate the susceptibility of cultivars to pests (Madriaza et al. 2019). Physiological changes, such as those caused by *S. oleae*, can reduce plant productivity. Therefore, the damage caused by this insect increases the need for its management. The objective of this research was to evaluate the occurrence and the effect of *S. oleae* on the physiological characteristics initial fluorescence, maximum fluorescence, maximum photochemical quantum yield of photosystem II/maximum fluorescence, and electron transport rate of the olive cultivars Arbequina, Ascolano, Grappolo and Koroneiki.

## Materials and Methods

This research was carried out in an olive orchard on the Quinta do Campo Alegre rural property in Diamantina, Minas Gerais, Brazil (18.3216667°S, 43.6988889°W; 1,390 masl). The region's climate is Cwb, dry winter subtropical highland, according to the Köppen classification (Peel et al. 2007). Average minimum and maximum annual temperatures are 14.3 °C to 25.1 °C.

One-yr-old olive seedlings were purchased at the Experimental Farm of the Minas Gerais Agricultural Research Corporation in Maria da Fé, Minas Gerais, Brazil, and transplanted in the field in 2012 at 4 × 6 m spacing in alternating rows, totaling 10 rows. The experiment was evaluated between 2016 and 2017 when the olive trees were 5 yr old.

The experiment was conducted in a randomized complete block design in a 2 × 4 factorial scheme (plants attacked or not by *S. oleae* in 4 cultivars of *O. europaea*: 'Arbequina,' 'Ascolano,' 'Grappolo,' and 'Koroneiki' with 25 replicates per cultivar. The evaluation points were chosen randomly, and at these points, we searched for leaves with and without insect to carry out the physiological analyses. The leaves for the non-attacked plants were inspected thoroughly to ensure they were free of insects, but no control tactics were used to stop insect attacks. Each experimental unit consisted of 4 branches from the middle section of the plant's canopy, directed to the east, west, north, and south. The total number and population density of *S. oleae* were counted in the summer, autumn, and winter. The total number of *S. oleae* individuals in the abaxial and adaxial sides of the leaves per cultivar and replication was obtained over the 3 seasons. The population density was calculated by dividing the total number of individuals of *S. oleae* by the number of branches in the middle section of the canopy of each plant.

The initial fluorescence (electron quantum$^{-1}$), maximum fluorescence (electron quantum$^{-1}$), the ratio between the variable and maximum fluorescence (maximum photochemical quantum yield of photosystem II/maximum fluorescence), and the electron transport rate (electron transport rate, µmols electrons m$^{-2}$ s$^{-1}$) were measured in the summer, autumn, and winter. These measurements were made in the median adaxial region of 4 leaves of the middle section of the canopy per treatment, with a fluorometer (MINI-PAM-2014, Heinz-Walz GmbH, Effeltrich, Germany), with emission of pulses of 0.3 s saturating light at 0.6 kHz frequency (Maxwell & Johnson 2000). Leaves were dark-adapted for 30 min before the measurements to ensure that all capable photosystem II reaction centers would oxidize fully.

The number of individuals of *S. oleae* was analyzed using the Kruskal-Wallis test, and the means compared by the Student-Newman-Keuls method ($P \leq 0.05$). Data of the population density of *S. oleae* in different cultivars and seasons were analyzed by trend estimation constructed with means and standard errors. Initial fluorescence and maximum fluorescence data were transformed into percentages per cultivar with and without *S. oleae*, submitted to analysis of variance, and when significant, analyzed by the Tukey's honestly significant difference test ($P \leq 0.05$). The maximum photochemical quantum yield of photosystem II/maximum fluorescence data were submitted to analysis of variance and analyzed by Tukey's honestly significant difference test ($P \leq 0.05$). The averages and standard error of all treatments were analyzed.

## Results

The total number of *S. oleae* was higher on the Grappolo olive cultivar than in the Ascolano and Arbequina ($H = 8.6115$; df = 3; $P = 0.0068$ and 0.0421) (Fig. 1) and its population density was highest on the Grappolo and Koroneiki cultivars in all seasons (Fig. 1). The population of *S. oleae* declined on the Grappolo, Koroneiki, and Ascolano in the winter, but increased on the Arbequina in this season (Fig. 2).

The initial chlorophyll "a" fluorescence was consistently higher on *O. europaea* attacked by *S. oleae* than on the non-attacked. Initial fluo-

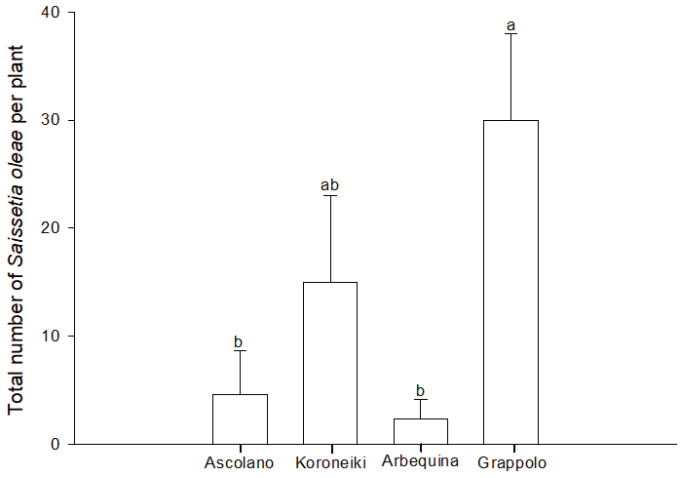


**Fig. 1.** Total number of individuals (mean ± standard error) of *Saissetia oleae* (Hemiptera: Coccidae) per plant of olive cultivars in Diamantina, Minas Gerais, Brazil. Means followed by the same letter do not differ by the Kruskal-Wallis test ($P \leq 0.05$).

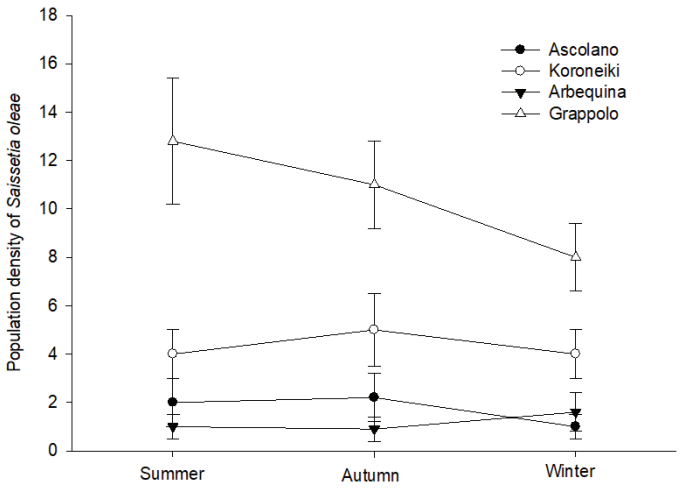


**Fig. 2.** Population density (mean ± standard error) of *Saissetia oleae* (Hemiptera: Coccidae) in olive cultivars in the summer, autumn, and winter in Dimantina, Minas Gerais, Brazil.

rescence on *O. europaea* attacked or not by *S. oleae* varied between 574.50 ± 82.75 and 914.25 ± 413.79 and 483.00 ± 102.03 and 583.75 ± 29.56, respectively with lower values in the Grappolo cultivar in autumn. The maximum chlorophyll "a" fluorescence was similar between attacked and non-attacked plants for all cultivars (Table 1).

The chlorophyll "a" maximum photochemical quantum yield of photosystem II/maximum fluorescence ratio was equal to or greater than 0.75 for all *O. europaea* cultivars not attacked by *S. oleae* in the summer, autumn, and winter. The chlorophyll maximum photochemical quantum yield of photosystem II/maximum fluorescence for the Ascolano, Koroneiki and Grappolo attacked in summer and winter was 0.75 and below 0.75 in the winter. The value for this parameter for the Koroneiki, Arbequina, and Grappolo attacked was below 0.75 for summer and winter (Table 1).

The electron transport rate in plants attacked by *S. oleae* was lower than in non-attacked ones, except for the Grappolo in the winter (Table 1). The electron transport rate of the non-attacked Ascolano cultivars was lower in the autumn and winter.

## Discussion

The high number of individuals and population density of *S. oleae* in the Grappolo cultivar may be related to a lower concentration of phenolic compounds. Damage by herbivorous insects break down the compartments of cellular organelles, and enzymes β-glucosidase and polyphenol oxidase activate oleuropein, reducing lysine and, con-

**Table 1.** Initial fluorescence (electron quantum$^{-1}$), maximum fluorescence (maximum fluorescence electron quantum$^{-1}$), variable fluorescence/maximum fluorescence ratio (maximum photochemical quantum yield of photosystem II/maximum fluorescence) and electron transport rate (µmols electrons m$^{-2}$ s$^{-1}$) in olive plants attacked or not attacked by *Saissetia oleae* (Hemiptera: Coccidae) during summer, autumn, and winter periods.

| | Ascolano | Koroneiki | Arbequina | Grappolo |
|---|---|---|---|---|
| | | Initial fluorescence, electron quantum$^{-1}$ | | |
| Attacked | 718.50 ± 138.29 aA | 718.75 ± 96.42 aA | 914.25 ± 413.79 aA | 665.00 ± 42.64 aA |
| Not attacked | 583.75 ± 29.56 bA | 516.75 ± 84.69 bA | 565.25 ± 29.12 bA | 532.00 ± 39.66 bA |
| Attacked | 665.50 ± 33.21 aA | 701.50 ± 47.12 aA | 648.25 ± 80.75 aA | 574.50 ± 82.75 aB |
| Not attacked | 562.50 ± 66.79 bA | 572.75 ± 52.21 bA | 571.25 ± 36.64 aA | 483.00 ± 102.03 aB |
| Attacked | 621.00 ± 46.91 aA | 603.50 ± 33.49 aA | 623.25 ± 32.29 aA | 607.75 ± 31.98 aA |
| Not attacked | 558.50 ± 185.06 bA | 504.25 ± 74.31 bA | 523.75 ± 90.57 bA | 517.25 ± 64.50 bA |
| | | Maximum fluorescence electron quantum$^{-1}$ | | |
| Attacked | 2,040.75 ± 26.60 bA | 2,206.50 ± 338.65 aA | 2,295.25 ± 206.14 aA | 2,594.75 ± 214.05 aA |
| Not attacked | 2,875.50 ± 148.05 aA | 2,336.25 ± 500.61 aA | 2,342.75 ± 502.13 aA | 3,089.25 ± 363.12 aA |
| Attacked | 3,129.00 ± 313.73 aA | 2,349.45 ± 1560.06 aA | 2,051.53 ± 1403.88 aA | 2,542.00 ± 400.11 aA |
| Not attacked | 3,278.75 ± 204.67 aA | 2,728.25 ± 369.45 aA | 2,934.50 ± 202.87 aA | 2,585.75 ± 554.81 aA |
| Attacked | 2,391.50 ± 931.74 aA | 1,752.75 ± 950.28 aA | 1,944.00 ± 522.39 aA | 1,827.00 ± 640.76 aA |
| Not attacked | 2,683.50 ± 292.90 aA | 2,187.00 ± 364.16 aA | 2,143.75 ± 279.76 aA | 2,082.00 ± 156.67 aA |
| | | Maximum photochemical quantum yield of photosystem II/maximum fluorescence | | |
| Attacked | 0.75 ± 0.03 aA | 0.69 ± 0.08 aA | 0.61 ± 0.15 bA | 0.71 ± 0.02 aA |
| Not attacked | 0.79 ± 0.01 aA | 0.78 ± 0.02aA | 0.77 ± 0.02 aA | 0.79 ± 0.03a A |
| Attacked | 0.74 ± 0.01 aA | 0.73 ± 0.02 aA | 0.75 ± 0.02 aA | 0.68 ± 0.03 aA |
| Not attacked | 0.82 ± 0.01 aA | 0.79 ± 0.03 aA | 0.78 ± 0.03 aA | 0.75 ± 0.25 aA |
| Attacked | 0.75 ± 0.02 aA | 0.67 ± 0.14 aA | 0.71 ± 0.06 aA | 0.71 ± 0.04 aA |
| Not attacked | 0.76 ± 0.02 aA | 0.75 ± 0.01 aA | 0.75 ± 0.01 aA | 0.75 ± 0.01 aA |
| | | Electron transport rate, µmols electrons m$^{-2}$ s$^{-1}$ | | |
| Attacked | 33.42 ± 4.93 bA | 30.75 ± 3.50 bA | 29.50 ± 9.75 bA | 31.25 ± 3.50 bA |
| Not attacked | 43.95 ± 3.69 aA | 45.25 ± 4.57 aA | 44.25 ± 0.96 aA | 41.22 ± 1.56 aA |
| Attacked | 20.70 ± 7.11 bA | 25.07 ± 3.67 bA | 27.70 ± 10.69 bA | 23.40 ± 4.21 bA |
| Not attacked | 33.52 ± 4.43 aB | 37.40 ± 2.13 aA | 37.50 ± 1.73 aA | 39.10 ± 4.08 aA |
| Attacked | 21.17 ± 3.64 bA | 23.70 ± 2.14 bA | 23.82 ± 5.82 bA | 29.02 ± 11.56 aA |
| Not attacked | 29.57 ± 2.39 aB | 33.57 ± 4.23 aA | 40.05 ± 2.32 aA | 34.60 ± 5.9 aA |

*Standard error. Means followed by the same lowercase letter per column or uppercase letter per line do not differ by Tukey's honestly significant difference test ($P \leq 0.05$).

sequently, the nutritional value of food for other herbivorous insects (Konno et al. 1999). The denaturing effect of oleuropein inhibited the oviposition of *Dacus oleae* (G.) (Diptera: Tephritidae) in olive trees (Girolami et al. 1981). Herbivory can trigger the production of phenolic compounds protecting plants, and cultivars with lower production of these compounds could be detected and preferred by insects.

Higher densities of *S. oleae* population during the summer in the cultivars Grappolo, Ascolano, and Koroneiki may be correlated with increased precipitation, humidity, and temperature as found for *S. oleae* in Algeria (Ilias & Hammadi 2017) and Spain (Tena et al. 2007) where the population peak reached its maximum in summer. The hotter and more humid summer in Brazil can favor the population increase of *S. oleae* on *O. europaea* cultivars. The number of individuals and population density of this insect varies with the olive cultivar, season of the yr, and the proximity to alternative hosts. This pest also damaged citrus in Florida, so olives near citrus orchards tend to have higher populations of this pest and need monitoring (Gillett-Kaufman et al. 2021).

The higher initial fluorescence on *O. europaea* cultivars attacked by *S. oleae* in the summer, autumn, and winter are due to damage to the reaction centers of the photosystem II or impairment in the excitation energy transport by the antenna complexes (Bolhàr-Nordenkampf et al. 1989). This was also found in varieties of wheat, *Triticum aestivum* (L.) (Poaceae) attacked by *Diuraphis noxia* (Kurdjumov) (Hemiptera: Aphididae), indicating a reduction in electron transfer through photosystem II and a possible decrease in D1 protein synthesis, increasing the initial fluorescence (Burd & Elliott 1996; Cavalheiro et al. 2015). The lower initial fluorescence of Grappolo in autumn may be due to abiotic factors, with reduced rainfall and temperatures in Brazil. Or due to better use of nitrogen in this cultivar, a component of chlorophyll, which could have consequently decreased the damage to the photosynthetic apparatus (Golan et al. 2015).

The damage by *S. oleae* in the summer seems to compromise and inactivate the photosystem II by reducing the maximum fluorescence in the Ascolano cultivar, but with no effect on the others. This may be due to lower plasticity of the Ascolano cultivar at high temperatures, and interruption of photo assimilated transport due to the increased attack of *S. oleae* in the summer. Increases in air temperature directly damage the olive trees' physiological processes (Tanasijevic et al. 2014). The reduction of the maximum fluorescence in Ascolano characterizes deficiency in the photoreduction of the quinone molecule (QA), the primary electron acceptor of the photosystem II (Silva et al. 2006). This deficiency may be associated with the inactivation of photosystem II in the tilacoidal membranes, directly affecting the electron flow between the photosystem II and photosystem I (Zabelin et al. 2016). This may be due to the decrease in the quantum yield of the photochemical process during this pulse when the reaction centers of the photosynthetic system are closed and the fluorescence emission reached its maximum (Maxwell & Johnson 2000). Damage by the herbivore *Spodoptera frugiperda* (J.E Smith) (Lepidoptera: Noctuidae) in photosynthetically active tissues interrupted the transport of photoassimilates, nutrients, and water, reducing the photosynthetic rate and the maximum fluorescence values in maize (De Souza et al. 2020).

Maximum photochemical quantum yield of photosystem II/maximum fluorescence values equal to or greater than 0.75 in the cultivars attacked or not are within the indicative range of non-stressed plants (0.75-0.85) (Björkman & Powles 1984). The lower maximum photochemical quantum yield of photosystem II/maximum fluorescence values than 0.75 of the cultivar Arbequina, Grappolo, and Koroneik indicate a reduction in the maximum quantum efficiency of photosystem II, and consequently, in the photosynthetic potential of the plant. This indicates susceptibility of these cultivars in the summer and winter and, depending on the cultivar, also in the autumn.

Lower electron transport rate values in *O. europaea* attacked by *S. oleae* indicate the effect of herbivory on the photosynthetic apparatus, as reported in tomato plants highly infested by *Phenacoccus solenopsis* (Tinsley) (Hemiptera: Pseudococcidae) (Huang et al. 2013). A lower electron transport in Ascolano not attacked in autumn and winter is possibly due to abiotic factors related to the season.

*Saissetia oleae* is a sucking insect and its feeding on the phloem content affects the photosynthetic apparatus of the cell and gas exchange. Feeding by this insect also reduces the transport of photoassimilates, nutrients and water, the photosynthetic rate, and leaf growth, causing premature leaf losses and reduced productivity. The increase in the initial fluorescence and the reduction in the maximum photochemical quantum yield of photosystem II/maximum fluorescence and electron transport rate in some cultivars of *O. europaea* attacked by *S. oleae* indicate damages to the photosynthetic apparatus, resulting in a possible decrease in growth and yield of the plants.

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