

Antioxidant responses to drought and salinity in *Lavandula angustifolia* Mill.

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Abstract

Drought and salinity are amongst the most damaging environmental stressors that can affect a plant's life cycle, from germination to senescence. In the present study were analysed the responses to salinity and drought in greenhouse-controlled conditions of two varieties of *Lavandula angustifolia*. Three-month-old lavender seedlings were subjected to water deficit and salt stress (100, 200 and 300 mM NaCl) during a 30-day period. Complementing a previous analysis focused on stress tolerance mechanisms based on the regulation of ion transport and the synthesis of osmolytes, we have now evaluated the effects of the water deficit and salt treatments on the generation of secondary oxidative stress, by measuring malondialdehyde levels, and the activation of antioxidant systems, both non-enzymatic and enzymatic, determining total phenolic compounds and flavonoids contents and calculating superoxide dismutase, catalase, ascorbate peroxidase and glutathione reductase specific activities, respectively, in extracts of control and stressed plants. The results obtained confirm that both lavender varieties react in the same way to the applied stress treatments, activating the same antioxidant responses. However, some differences were observed when comparing the specific mechanisms triggered by each type of stress. Thus, the oxidative stress induced under drought conditions was counteracted by accumulation of phenolic compounds and flavonoids, without apparent involvement of antioxidant enzymes. Salt stress, on the other hand, in addition to an increase in flavonoid levels also induced superoxide dismutase and catalase activities. These antioxidant responses are likely to contribute to the relatively high tolerance (as compared to most crops) of lavender to drought and salinity.

Keywords: abiotic stress; antioxidant enzymes; oxidative stress; *Lamiaceae*; non-enzymatic antioxidants; reactive oxygen species

Introduction

Drought is becoming a major environmental challenge in many areas of the world, imposing the need for irrigation, which is eventually causing soil salinisation (Fahad *et al.*, 2017). Such environmental changes,

which pose a serious threat to agriculture, have also been reported in Romania (Croitoru *et al.*, 2016; Irimia *et al.*, 2018; Prăvălie *et al.*, 2020).

Plants are exposed to a wide range of environmental stress conditions (salinity, drought, extreme temperatures), which negatively affect their growth and may even prevent the completion of their life cycle (Kaur and Asthir, 2016; Forni *et al.*, 2016; Nareshkumar *et al.*, 2020). Under stress conditions, plants activate their defence mechanisms, including physiological, biochemical, and molecular responses (Bartels and Ramanjulu, 2005; Forni *et al.*, 2016; Acosta-Motos *et al.*, 2017; Mittal *et al.*, 2018; Rao *et al.*, 2019). Drought and salinity, as well as other abiotic and biotic stresses, alter the metabolic activity of the cell by producing an excessive amount of reactive oxygen species (ROS) (Aftab, 2019). Although ROS are toxic when in excess, they are important signalling molecules and have a role in regulating biological processes from germination to senescence (Miller *et al.*, 2008; Morales and Munné-Bosch, 2019). The accumulation of ROS triggers antioxidant mechanisms that include the accumulation of antioxidant compounds, such as phenols and flavonoids, amongst others, and the activation of antioxidant enzyme systems, such as catalase (CAT), superoxide dismutase (SOD), ascorbate peroxidase (APX) (and other peroxidases), and glutathione reductase (GR) (Sharma *et al.*, 2012).

Lavandula is a genus of the *Lamiaceae* family, native to the Mediterranean region. It includes over 45 species and some 400 varieties (Benabdelkader *et al.*, 2011; The Plant List, 2020), many of which are prized for their aromatic oils as medicinal and ornamental plants (Biesiada and Kucharska, 2008; Lis-Balchin 2012; Mokhtarzadeh *et al.*, 2013; Prusinowska and Śmigielski, 2014; Carrasco *et al.*, 2016; Matysiak and Nogowska, 2016). In terms of environmental requirements, most lavender species and varieties grow well in a wide range of climatic conditions (Lis-Balchin, 2002; Upson and Andrews, 2004; Adam, 2006; Wells *et al.*, 2018), but their ecological optimum is found in regions with warm, sunny summers and cold winters (Lis-Balchin, 2002).

Lavandula angustifolia Mill. is a perennial shrub, which can grow from 23 cm to 90 cm, and has a lifespan of 20-30 years, depending on the mode of cultivation (Koulivand *et al.*, 2013; Seidler-Āoīykowska *et al.*, 2014). Large-scale lavender cultivation flourished in the 17th century in Spain, France, and Italy, while in Central and Eastern Europe, in countries such as Romania, Bulgaria, Hungary, and Poland, cultivation began only in the 20th century (Pisulewska *et al.*, 2009).

Two varieties of lavender were subjected to water deficit (complete withholding of irrigation) and salt stress (watering the plants with increasing NaCl concentrations) in our experimental set-up: *Lavandula angustifolia* var. 'Codreanca' (LAC) from Romania, and var. 'Sevtopolis' (LAS) from Bulgaria. Their growth responses, together with an analysis of the uptake and transport of ions to the leaves and the accumulation of osmolytes have been published previously (Szekely-Varga *et al.*, 2020). The aim of the present study was to investigate the effects of the applied drought and salinity treatments on the generation of oxidative stress and the activation of antioxidant mechanisms in the two cultivars. The degree of oxidative stress suffered by the plants was evaluated by measuring the accumulation of malondialdehyde (MDA). As representative non-enzymatic antioxidants, the leaf contents of total phenolic compounds (TPC) and total flavonoids (TF) were determined, and the specific activities of the main antioxidant enzymes, superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX) and glutathione reductase (GR) were in turn measured.

Materials and Methods

Plant material and stress treatments

In this work, two varieties of *Lavandula angustifolia* were used as experimental material. *L. angustifolia* var. 'Codreanca' (LAC) is a Romanian variety approved in 1992; it reaches up to 60 cm high, and a width of 60-70 cm, with silvery blue leaves. It produces blue-purple flowers, sometimes deep blue, which bloom in May-June. LAC is considered one of the best varieties for Romania's climatic conditions and is resistant to low temperatures and even frost (Cantor *et al.*, 2018). The second analysed lavender variety was *L. angustifolia* var.

'Sevtopolis' (LAS), native to Bulgaria. It reaches a height of 40-60 cm, and a width of 50-60 cm. It has blue flowers and greyish green leaves and a great commercial interest due to its high content in essential oils (Kivrak, 2018). Both varieties are grown mainly for the extraction of essential oils, but they also have ornamental value, and can be used as cut flowers.

Seedlings of both lavender varieties were grown individually in 0.3 L plastic pots with a mixture of 50% peat, 25% perlite and 25% vermiculite. Throughout the experiment, the plants were kept in a climate-controlled chamber, under artificial light with a long daytime photoperiod (16 hours of light and 8 hours of darkness), and light intensity of $130 \mu\text{E m}^{-2} \text{s}^{-1}$, at a constant temperature of 23 °C and 60-80% relative humidity. For each treatment, five pots of each cultivar were placed separately in a plastic tray. Until the treatments began, the lavender seedlings were watered every three days with Hoagland nutrient solution (Hoagland and Arnon, 1950). The treatments were started after 3 months from planting, when the development of the seedlings of the two lavender varieties corresponded to the main growth stage 4 according to the BBCH coding system (Meier, 2001). Treatments were applied by watering the plants twice a week with NaCl solutions of final concentrations of 0 (for the controls), 100, 200 and 300 mM, adding 1 L of solution per tray. For water stress treatments (WS), irrigation was completely withheld. The treatments were stopped after 30 days. Plant samples (roots, stems and leaves) were collected. Five individual lavender plants of each variety per treatment were used as biological replicates.

Malondialdehyde (MDA)

Malondialdehyde (MDA) concentration was determined in aqueous methanol extracts (80%, v/v), prepared by grinding 0.05–0.10 g of fresh lavender leaves, and centrifuging the samples at 12,000 rpm for 15 min. MDA was quantified in the supernatants, as previously described (Hodges, 1999). Each sample was mixed with 0.5% (w/v) thiobarbituric acid (TBA) prepared in 20% (w/v) trichloroacetic acid (TCA)-or with 20% TCA without TBA for the controls-and then incubated at 95 °C for 15 min, in a water bath. The reactions were stopped on ice, the samples were centrifuged at 12,000 rpm for 10 min, at 4 °C, and the absorbance of the supernatants was measured at 532 nm. After subtracting the non-specific absorbance at 600 and 440 nm, the MDA concentration was calculated applying the equations described by Hodges (Hodges, 1999) based on the molar extinction coefficient of the MDA-TBA adduct at 532 nm ($\epsilon_{532} = 155 \text{ mM}^{-1} \text{ cm}^{-1}$).

Non-enzymatic antioxidants

Total phenolic compounds (TPC) and total flavonoids (TF) concentrations were determined in the same 80% methanol extracts used for MDA measurements. TPC were determined according to the protocol of Blainski (Blainski *et al.*, 2013), which is based on the reaction with the Folin-Ciocalteu reagent, in the presence of NaHCO_3 ; the reaction mixtures were incubated at room temperature, in the dark, for 90 min, and the absorbance was then recorded at 765 nm. TPC concentration was expressed as equivalents of the standard, gallic acid ($\text{mg eq. GA g}^{-1} \text{ DW}$).

TF were determined by nitration of catechol groups with NaNO_2 , followed by reaction with AlCl_3 under alkaline conditions (Zhishen *et al.*, 1999). The absorbance of the samples was read at 510 nm, using catechin as the standard. TF concentration was expressed as equivalents of catechin ($\text{mg eq. C g}^{-1} \text{ DW}$).

Antioxidant enzyme activities

The specific activity of four major antioxidant enzymes, namely, superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), and glutathione reductase (GR), was determined at room temperature ($\sim 25 \text{ }^\circ\text{C}$) in crude protein extracts prepared from lavender leaves, as previously described (Gil *et al.*, 2014). The plant material was ground in liquid N_2 and mixed with extraction buffer [20 mM HEPES, pH = 7.5, 50 mM KCl, 1 mM EDTA, 0.1% (v/v) Triton X-100, 0.2% (w/v) polyvinylpyrrolidone, 0.2% (w/v) polyvinylpolypyrrolidone and 5% (v/v) glycerol]; to improve protein extraction, 1/10 volume of 'high salt buffer' (225 mM HEPES, pH 7.5, 1.5 M KCl and 22.5 mM MgCl_2) was added to the samples, mixed well by

vortexing and kept 15 min on ice. After centrifugation at 13,500 rpm for 15 min at 4 °C, the supernatants were collected, concentrated in U-Tube concentrators (Novagen, Madison, WI, USA), and centrifuged again to remove precipitated material. The supernatants, referred to as 'protein extracts' were frozen in liquid N₂, and stored in aliquots at -75 °C. Protein concentration in the extracts was measured by the method of Bradford (Bradford, 1976), using the Bio-Rad reagent and bovine serum albumin (BSA) as standard.

SOD activity in the protein extracts was determined at 560 nm following the inhibition of nitroblue tetrazolium (NBT) photoreduction in reaction mixtures containing riboflavin as the source of superoxide radicals (Beyer, 1987). A SOD unit was defined as the amount of enzyme that causes 50% inhibition of NBT photoreduction under the assay conditions.

Catalase (CAT) activity was assessed by the decrease in absorbance at 240 nm, which parallels the consumption of H₂O₂ added to the extracts (Aebi, 1984). A CAT unit was defined as the amount of enzyme that will decompose one mmol of H₂O₂ per minute at 25 °C.

The enzymatic activity of ascorbate peroxidase (APX) was determined by the decrease in absorbance observed at 290 nm as ascorbate becomes oxidised in the reaction (Nakano and Asada, 1981). One APX unit was defined as the amount of enzyme required to consume one mmol of ascorbate per minute, at 25 °C.

Glutathione reductase (GR) activity was quantified according to Conell and Mullet (Conell and Mullet, 1986), following the decrease in absorbance at 340 nm due to oxidation of NADPH – the cofactor of the GR-catalysed reduction of oxidised glutathione (GSSG). One GR unit was defined as the amount of enzyme that will oxidise one mmol of NADPH per minute, at 25 °C.

Statistical analysis

Data were analysed using Statgraphics Centurion XVI (Statgraphics Technologies, The Plains, VA, USA). Before the analysis of variance, a Shapiro–Wilk test was used to check for the validity of normality assumption and a Levene's test was used for the homogeneity of variance. If ANOVA requirements were met, significant differences among treatments were tested by one-way ANOVA at the 95% confidence level, and post hoc comparisons were made using a Tukey HSD test. All mean values throughout the text, followed by their SE, are based on five biological replicas per variety and per treatment.

Results

Oxidative stress

Malondialdehyde (MDA) concentrations were determined in leaf extracts of the lavender seedlings subjected to water and salt stress treatments (Figure 1). Under our experimental conditions, after 30 days without watering the plants, average MDA content decreased with respect to the control in the LAC variety, whereas an increase was observed in LAS seedlings (Figure 1). On the other hand, the salt treatments induced a considerable increase in MDA contents in both varieties. Average MDA values in salt-stressed plants of both varieties were significantly higher than in those from control (Figure 1).

Non-enzymatic antioxidants

Total phenolic compounds (TPC) (Figure 2a) and total flavonoids (TF) (Figure 2b) were quantified in all leaf samples collected from the two lavender varieties in the presence of the indicated NaCl concentrations (100, 200 and 300 mM NaCl) or under water deficit stress (WS) for 30 days. No significant differences were observed between the two varieties for any of the treatments, neither in TPC (Figure 2a) nor in TF (Figure 2b) contents.

In both lavender varieties, a significant increase in TPC contents was detected in the water-stressed plants. In response to the salt stress treatments, a relatively smaller, but still statistically significant increase in

TPC was found in LAC, but no significant changes with respect to the control were observed in the LAS (Figure 2a).

Under our experimental conditions, TF contents recorded significant increases in water deficit and salt stress compared to the control in the two lavender varieties. In both, the greatest increase was observed in plants subjected to water stress treatments (Figure 2b).

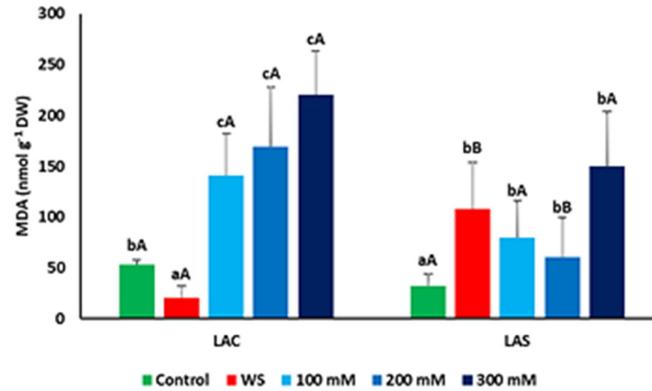


Figure 1. Malondialdehyde (MDA) levels in leaves of *L. angustifolia* var. 'Codreanca' (LAC) and var. 'Sevtopolis' (LAS) seedlings after 30 days of growth in the presence of the indicated NaCl concentrations or subjected to water deficit stress (WS)

Bars represent means \pm SE ($n = 5$). Different lowercase letters above the bars indicate significant differences between treatments for each variety, and different uppercase letters indicate significant differences between the two varieties for plants undergoing the same treatment, according to the Tukey test ($\alpha = 0.05$).

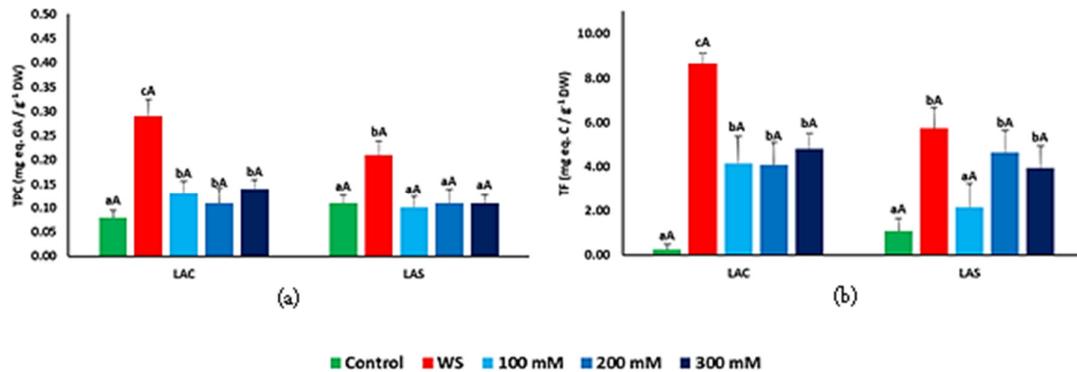


Figure 2. Total phenolic compounds (TPC) (a) and total flavonoids (TF) (b) leaf contents of *L. angustifolia* var. 'Codreanca' (LAC) and var. 'Sevtopolis' (LAS) seedlings, after 30 days of growth in the presence of the indicated NaCl concentrations or subjected to water deficit stress (WS)

Bars represent means \pm SE ($n = 5$). Different lowercase letters above the bars indicate significant differences between treatments for each variety, and different uppercase letters indicate significant differences between the two varieties for plants undergoing the same treatment, according to the Tukey test ($\alpha = 0.05$).

Antioxidant enzyme activities

The specific activities of some of the most relevant antioxidant enzyme systems: superoxide dismutase (SOD), catalase (CAT), ascorbate peroxide (APX) and glutathione reductase (GR), were determined by spectrophotometric assays, using protein extracts prepared from leaves of the control and stressed lavender seedlings.

A significant increase in SOD specific activity was observed in the lavender seedlings subjected to salt stress, but not to water deficit; SOD variation patterns were similar in the two varieties, which did not show

significant differences in any of the treatments (Figure 3a). Similarly, for both varieties, CAT activity did not change in water-stressed plants, with respect to the corresponding controls, whereas it increased significantly in response to the salt treatments (Figure 3b). It should be notice that the average CAT activity was generally lower in LAS plants than in the LAC variety, for each specific treatment, although with statistically significant differences between varieties observed only in the controls and in the 300 mM NaCl treatment (Figure 3b). In the variety LAS, the APX specific activity did not vary significantly, as compared to the control, in any of the salt treatments, or under water deficit conditions (Figure 3c). On the other hand, plants of the LAC variety subjected to salt stress showed a strong decrease in APX activity, whereas it did not vary in water-stressed plants (Figure 3c). Regarding GR specific activity, both varieties showed a significant decrease in response to watter stress; on the contrary, GR activity increased with respect to the control in the presence of high salt concentrations (300 mM NaCl), but only in the LAS variety (Figure 3d).

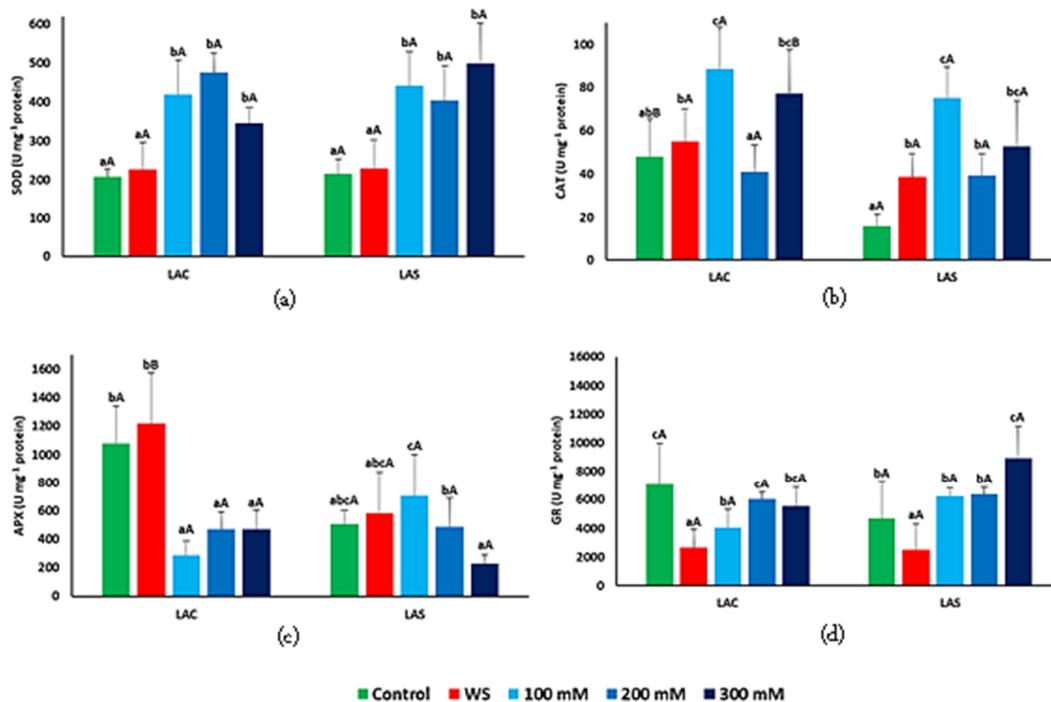


Figure 3. Antioxidant enzyme activities: superoxide dismutase (SOD) (a), catalase (CAT) (b), ascorbate peroxidase (APX) (c) and glutathione reductase (GR) (d) in leaves of *L. angustifolia* var. 'Codreanca' (LAC) and var. 'Sevtopolis' (LAS) seedlings after 30 days of growth in the presence of the indicated NaCl concentrations or subjected to water deficit stress (WS)

Bars represent means \pm SE (n = 5). Different lowercase letters above the bars indicate significant differences between treatments for each variety, and different uppercase letters indicate significant differences between the two varieties for plants undergoing the same treatment, according to the Tukey test ($\alpha = 0.05$).

Discussion

The results of the present study confirm previous data (Szekely-Varga *et al.*, 2020), clearly indicating that the responses to water deficit and salt stress of the two selected varieties of *L. angustifolia*, 'Codreanca' (LAC) and 'Sevtopolis' (LAS), were very similar. Lavender growth was significantly reduced with increased salt concentration and under water deficit conditions, which influenced the plant biomass; however, all plants survived the strong stress conditions applied, indicating that lavender is relatively tolerant to both, drought and

salinity. Growth inhibition is a very general response to stress, as plants under stressful conditions reallocate their growth and primary metabolism resources to the activation of defence mechanisms (Munns and Tester, 2008; Hendawy and Khalid, 2005). The study mentioned above (Szekely-Varga *et al.*, 2020), focused on the effects of the stress treatments on growth, ion transport and osmolyte accumulation in plants of the two lavender varieties. We reported some variations in the mean values of several growth parameters; however, the differences were too small to allow considering any of the two varieties as more stress-tolerant than the other. In the two varieties, the most relevant mechanisms activated in response to salt stress, which are likely to be involved in salt tolerance in this species, were the maintenance of foliar K⁺ levels and the accumulation in leaves of Ca²⁺, and of proline as a functional osmolyte; in the case of water stress, an osmotic adjustment with significant increases in root Na⁺ and K⁺ concentrations is presumably limiting root dehydration. The present work aimed at extending and complementing those previous results, investigating the possible contribution of antioxidant responses to the mechanisms of drought and salt tolerance in lavender.

Reactive oxygen species (ROS) are ubiquitous in plants under stress (Abogadallah, 2010; Nareshkumar *et al.*, 2020). They are produced by plants as by-products during various metabolic reactions, for example, photosynthesis and respiration, and play essential roles in cell signalling and homeostasis (Van Breusegem *et al.*, 2001; Ahmad *et al.*, 2010). ROS are produced in excess in response to different stresses, and overproduction of ROS can lead to membrane lipid peroxidation, which is often quantified by measuring the levels of malondialdehyde (MDA), a product of this reaction and a reliable marker of oxidative stress (Del Rio *et al.*, 1996).

In the present study, in general, no significant differences in MDA levels were observed between the two selected varieties in any of the treatments, indicating a similar degree of drought- or salt-induced oxidative stress in both of them. However, analysing the mean values of MDA contents in the different treatments, some quantitative differences were observed. For example, high levels of MDA were found in the Bulgarian variety 'Sevtopolis', both under water and salt stress conditions, whereas in the 'Codreanca' variety, MDA increased only in salt-stressed plants. The low MDA concentration observed under water stress in plants of the LAC variety suggests that, concerning specifically antioxidant responses, it probably adapts slightly better to drought than the LAS variety, since it has a better defence against the harmful effect of oxidative stress associated with drought (Zhou *et al.*, 2017). In contrast, its higher MDA concentrations measured under low and moderate (100 and 200 mM NaCl) salt stress conditions, may indicate that 'Codreanca' is somewhat more susceptible to salt-associated oxidative stress than the 'Sevtopolis' variety. Similar results underlying the reliability of MDA as a marker of oxidative stress have been obtained by many different authors in plants subjected to various stress factors, for example, by Xia *et al.* (2020) (kiwi); Goharrizi *et al.* (2020) (*Lepidium draba*); Nareshkumar *et al.* (2020) (cucumbers, tobacco and rice); or Arora *et al.* (2020) (*Artemisia annua*).

Phenolic compounds, and especially the subgroup of flavonoids, play multiple roles in plants, including their adaptation to stressful conditions (Shah and Smith, 2020). Since most flavonoids and many other phenolics are strong antioxidants, their accumulation can be related to the reduction of oxidative damage and thus to an increased tolerance to stress (Hussain *et al.*, 2013; Al Hassan *et al.*, 2017). Both, total phenolic compounds (TPC) and total flavonoids (TF) contents increased significantly under water stress conditions in both varieties, although the relative increase over the corresponding controls, and the absolute mean values reached at the end of the water deficit treatment, were higher in the LAC variety. Accumulation of these antioxidant compounds to higher concentrations may explain the (slightly) stronger antioxidant response of the LAC variety suggested by the MDA concentration patterns. Regarding the salt treatments, an increase in TF was observed in the two varieties, LAC and LAS, albeit to lower levels than those reached in the water stress treatments, indicating a significant contribution of flavonoids to the antioxidant responses of lavender to salinity. This effect, however, seems to be masked by other phenolic compounds since TPC contents increased only slightly in response to salt stress in the LAC variety, and not at all in LAS seedlings. These results are in agreement with many published reports supporting the functional role of antioxidant flavonoids and other phenolics in the responses of plants to drought, salinity and other environmental stress conditions generating

oxidative stress, either directly or as a secondary effect (e.g., Sharma *et al.*, 2012; Hussain *et al.*, 2013; Al Hassan *et al.*, 2017). In fact, several authors have suggested that the accumulation of phenolic compounds and flavonoids could have been a key step in the evolution of plants towards tolerance to different stress factors (Bautista *et al.*, 2016; Sharma *et al.*, 2016; Rodríguez-Calzada *et al.*, 2019; Arora *et al.*, 2018, 2020).

The increase in the activity of antioxidant enzymes (SOD, CAT, APX and GR) is generally triggered when an excess of harmful ROS, generated under stress conditions, is detected (Shi and Zhu, 2008; Ashraf, 2009; Ahmad *et al.*, 2010; Chan *et al.*, 2016; Gharsallah *et al.*, 2016). Plant species differ greatly in their responses to environmental stresses, and their ability to adapt to different abiotic stresses also depends, in many cases, on their ability to activate antioxidant enzyme systems (Kusvuran *et al.*, 2016).

Superoxide dismutase is considered a primary line of defence against oxidative stress, as it eliminates highly reactive superoxide radicals (Huang *et al.*, 2019). Increased SOD activity has been reported for both, water stress (Zgallai *et al.*, 2006; Wang and Li, 2008; Zhu *et al.*, 2020) and salt stress in many different plant species (Carrasco-Ríos and Pinto, 2014; Evelin and Kapoor, 2014; Al Kharusi *et al.*, 2019), as SOD is considered the most effective enzymatic antioxidant, ubiquitous in all aerobic organisms (Alscher *et al.*, 2002; Gill and Tuteja, 2010). Catalase and ascorbate peroxidase catalyse the breakdown of H₂O₂ in water and oxygen, and both play an essential role in the antioxidant system of plants (Ben-Amor *et al.*, 2005; Omamit *et al.*, 2006; Kangasjärvi *et al.*, 2008). Increases in CAT activity have been described in different species (Yang *et al.*, 2008; Chutipaijit, 2016; Plesa *et al.*, 2018). Glutathione reductase is an essential enzyme, highly conserved in nature, responsible for the recycling of glutathione, catalysing the reduction of its oxidised form, using NADPH as cofactor (Couto *et al.*, 2016). Increased GR activity has been reported in response to various stresses in many different plant species, e.g., in apple (Wang *et al.*, 2013), wheat (Shan *et al.*, 2015) or tomato (Ding *et al.*, 2018).

The results presented here indicate that the patterns of stress-induced changes in antioxidant enzyme activities were the same for the two selected lavender cultivars since, with very few exceptions, no significant differences between LAC and LAS plants were observed in any of the control or stress treatments. Also, no significant increase in any of the enzyme activities has been detected in response to water deficit; in fact, the GR activity even decreased significantly in both lavender varieties, as reported in other species, for example in spruce seedlings (Todea *et al.*, 2020). These data suggest that the antioxidant response to drought in lavender is primary based on the accumulation of phenolic compounds, in general, and of flavonoids, in particular, without involvement of the assayed antioxidant enzyme systems. Regarding salt stress, in addition to the accumulation of flavonoids, significant increases in SOD and CAT activities have been detected in salt-stressed plants, supporting the participation of these specific enzymes in the antioxidant defence against salt stress in *L. angustifolia*.

Conclusions

Comparative studies on the antioxidant responses of *Lavandula angustifolia* var. 'Codreanca' (LAC) and var. 'Sevtopolis' (LAS) to water and salt stress revealed that both varieties possess effective antioxidant mechanisms to mitigate oxidative stress associated to drought and salinity, without significant differences being observed between the two genotypes. The response to drought-induced oxidative stress is essentially based on the accumulation of antioxidant phenolics, including the subgroup of flavonoids, apparently without contribution of antioxidant enzymes. Oxidative stress caused by high salinity is partly counteracted by the synthesis of flavonoids but, in addition, by the increase of SOD and CAT specific activities. These antioxidant responses, together with previously described mechanisms based on the control of ion transport and the accumulation of the osmolyte proline, explain the relative tolerance of *Lavandula angustifolia* to drought and salinity.

Authors' Contributions

Conceptualization, M.B. and M.C.; Formal analysis, M.B., O.V. and M.C.; Investigation, Z.S.-V. and S.G.-O.; Resources, O.V.; Supervision, M.B., O.V. and M.C.; Visualization, O.V.; Writing-original draft, Z.S.-V.; Writing-review and editing, O.V. All authors have read and agreed to the published version of the manuscript.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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