GROWTH OF THE NEW *Salvia officinalis* × *S. tomentosa* HYBRID UNDER REDUCED IRRIGATION IN GREENHOUSE AND GREEN ROOF CONDITIONS

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Abstract

Aiming to the exploitation of a new interspecific Mediterranean sage hybrid *Salvia officinalis* × *S. tomentosa*, along with its parental species *S. officinalis* and *S. tomentosa*, in extensive green roofs and xeriscaping, their resistance to xerothermic conditions was examined in a Mediterranean green roof and in a greenhouse under regular and reduced irrigation. In the green roof the hybrid and the parental species were grown on 10 cm deep substrate mixture of grape-marc compost: perlite: pumice 3:3:4, v/v, while in the greenhouse in 14 cm plastic pots, on peat: perlite 2:1, v/v. Planting took place in April 2021 and the experiment lasted 3 and 5 months, in the greenhouse and green roof, respectively. Two irrigation frequencies, a regular (every 2–3 days when substrate moisture 17–23% v/v) and a reduced (every 3–5 days when substrate moisture 7–13% v/v) were applied. In the greenhouse, the hybrid growth was very satisfactory under reduced irrigation, since only plant height was reduced from all growth parameters recorded (lateral shoot number and length, aboveground and root dry weight). In the green roof, *S. tomentosa* had higher plant height and diameter and aboveground dry weight than *S. officinalis* and their hybrid, while root dry weight was higher in the parental species compared to the hybrid. Reduced irrigation resulted in significant reduction of all growth parameters of all *Salvia* types. However, their growth remained satisfactory. So, both the hybrid and its parents are recommended for sustainable use in xeriscaping, including extensive green roofs.

Keywords: *drought resistance, Mediterranean sage, interspecific hybrid, Salvia hybrid, S. officinalis, Salvia tomentosa*

Introduction

Greek *Salvia* species being drought resistant could be ideal plants for use as ornaments, in arid and semi-arid regions, particularly in xeriscaping, which has gained worldwide acceptance in recent years due to the limited irrigation water needed. Native plants are ideal for use in xeriscaping, since they have low irrigation and cultivation requirements, a great adaptability to different soil and climatic conditions, high ornamental value, as well as a contribution to the preservation of the character of the landscape (Bayramoğlu *et al.*, 2015; Martinetti *et al.*, 2018; Tassoula *et al.*, 2021).

Following the trend of floriculture industry, which is constantly seeking to introduce new varieties to the market, a new interspecific sage hybrid *Salvia officinalis* × *S. tomentosa* was created through the crossbreeding between *S. officinalis*, as the seed parent, and *S. tomentosa*, as the pollen parent (Papafotiou *et al.*, 2021). *S. officinalis* is a strongly aromatic, rather grayish shrub up to 60 cm, with leaves oblong to elliptical, rough greenish above but white felted beneath and flowers violet-blue or pink. It is cultivated worldwide with many varieties as pharmaceutical and ornamental (Tutin *et al.*, 1972; Blamey & Grey-Wilson, 1993). *S.*

tomentosa is similar to *S. officinalis*, up to 80cm, but has leaves with a rounded or heart-shaped base and flowers usually violet with reddish-brown calyces (Blamey & Grey-Wilson, 1993). Both *Salvia* species flower in late spring to early summer. Their interspecific hybrid *S. officinalis* × *S. tomentosa* is a compact plant that produces numerous lateral shoots with grey-green leaves smaller than those of *S. officinalis*. It also forms many flowering stems with light pink flowers, while it has a lighter aroma than that of *S. officinalis* (Papafotiou *et al.*, 2021).
Aiming to exploit the new interspecific Mediterranean sage hybrid *Salvia officinalis* × *S. tomentosa*, along with its parental species *S. officinalis* and *S. tomentosa*, in extensive green roofs and xeriscaping, their resistance to xerothermic conditions was examined in a Mediterranean green roof and in a greenhouse under regular and reduced irrigation.

Materials and methods

Aiming to test growth and drought resistance of the new interspecific sage hybrid *S. officinalis* × *S. tomentosa*, two experiments were set in early April 2021.
In the first experiment, that took place in a greenhouse, four-week-old rooted cuttings of this hybid were transplanted singly in plastic pots, 14 cm in diameter, which contained 1 L of peat-perlite mixture 2:1 (v/v). Climatic conditions during the experiment are shown in Figure 1 a-b. Plants were fertilized monthly with 2 g/L (100 ml fertilizer per pot) water soluble fertilizer (20-20-20 plus, HUMOFERT, Metamorfosi, Greece). Two irrigation frequencies were applied: (i) when the moisture content of the substrate was 20–23% v/v (normal irrigation) and (ii) when the moisture content of the substrate was 8–13% v/v (sparse irrigation). In the first month of cultivation, the plants under normal irrigation were irrigated every 3–4 days and under sparse irrigation every 5 days, while in the following months every 2 and 3–4 days, respectively. This experiment lasted for 3 months, until July 2021.

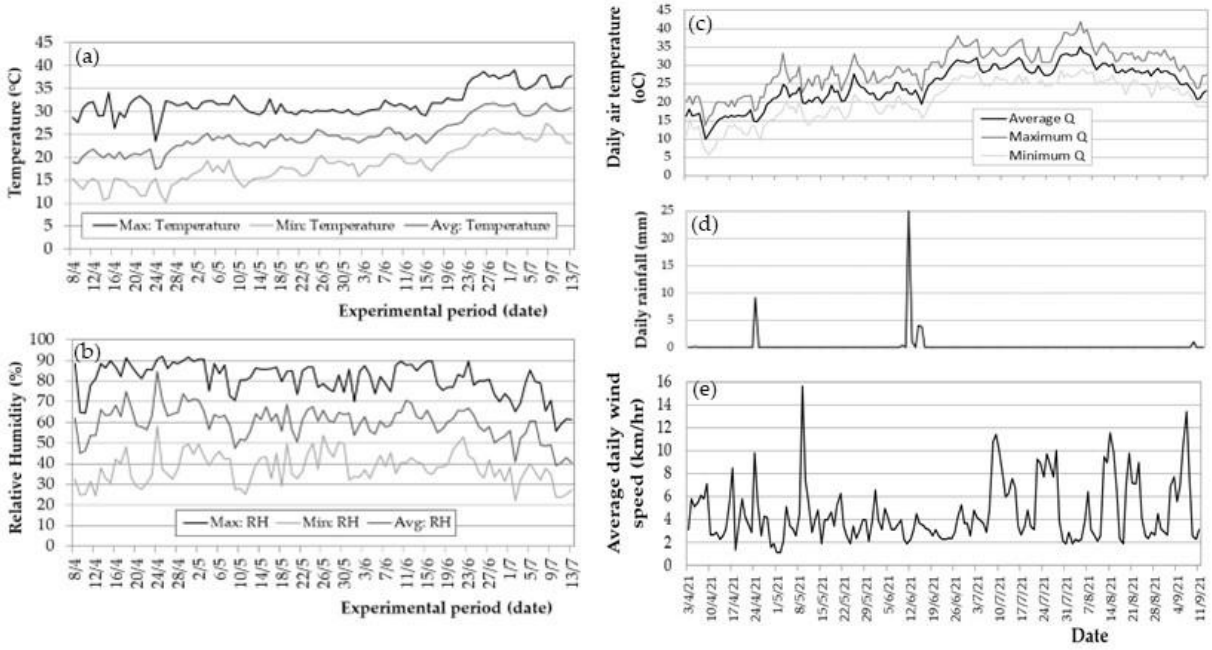

Figure 1. Climatic conditions, i.e., temperature (a) and relative humidity (b) inside the glass greenhouse where the experiment was conducted, during the 3-month experimental period (from April 2021 until July 2021), as well as temperature (c), daily rainfall (d) and average daily wind speed (e) in the green roof, during the 5-month experimental period (from April to September 2021).

In the second experiment, that was established in an extensive green roof, rooted cuttings, about 8 weeks old, of the hybrid *S. officinalis* × *S. ringens*, alongside with its parental species *S. officinalis* and *S. ringens*, were planted in plastic containers 40 cm × 60 cm × 22 cm in size. Each container had a green roof infrastructure fitted, i.e., moisture retention and protection of the insulation mat FLW-500, a drainage layer Diadrain-25H and a filter sheet VLF-150 (Landco Ltd., Diadem Green Roof Systems, Athens, Greece). Two plants of the same type per container with six containers per treatment were used. The containers were arranged following a completely randomized design on a second-floor flat roof at the Agricultural University of Athens (37°59′ N, 23° 42′ E). The substrate used was grape marc compost: perlite: pumice (3: 3: 4, v/v) and had a 10 cm depth. Climatic conditions during this experiment are shown in Figure 1 c-e. Irrigation frequencies similar to those of the first experiment were applied, i.e., when substrate moisture 17–23% v/v (regular) and when substrate moisture 7–13% v/v (reduced). In the first month of cultivation, the plants were irrigated normally every 3 days and sparsely every 5 days, while in the following months every 2 and 4 days, respectively. Automatic drip irrigation on the substrate surface was applied before sunrise by two drippers placed at equal distances from the center of the container and the plants (dripper supply 4 L h^{-1}, irrigation period: 60 min). This experiment lasted for 5 months, until September 2021.

Plant growth was evaluated at the end of the experiments. The completely randomized design was used. The significance of the results was tested by either one- or two- way analysis of variance (ANOVA) and the means of the treatments were compared by Student's *t* test at $p< 0.05$ (JMP 11.0 software, SAS Institute Inc., Cary, NC, 2013, USA).

Results and Discussion

In the first experiment, in the greenhouse, all plants survived and the hybrid grew very satisfactorily under reduced irrigation, since only plant height was reduced from all growth parameters recorded (Table 1, Figure 2).

In the second experiment, in the green roof, survival percentage was affected neither by *Salvia* type nor by irrigation frequency and was ranging between 66.7% and 91.7% (Table 2). *S. tomentosa* had higher plant height and diameter and aboveground dry weight than *S. officinalis* and their hybrid, while root dry weight was higher in the parental species compared to the hybrid. Reduced irrigation resulted in significant reduction of all growth parameters of all *Salvia* types. However, their growth remained satisfactory (Table 2, Figure 3).

In other *Salvia* species, such as *S. fruticosa* (Chrysargyris *et al.*, 2016) and *S. officinalis* (Sabry *et al.*, 2016; Soltanbeigi *et al.*, 2021), water deficiency was also reported to reduce plant height and plants yield components (above ground and leaf fresh and dry weight). Regarding other Labiatae species, in *Lavandula latifolia*, *Mentha piperita* and *Thymus capitatus*, the above ground fresh weight was reduced by drought stress, whereas in *Salvia sclarea*, *Salvia lavandutifolia* and *Thymus mastichina* it remained unaffected. As regards the above ground dry weight, only in *L. latifolia* there was a significant reduction under water deficit conditions (García-Caparrós *et al.*, 2019).

The root system was found to play a key role in plant drought resistance in *S. officinalis* (Abate *et al*. 2021), a result that was also supported by our research since *S. officinalis* showed the largest root/aboveground fresh and dry matter under both normal and sparse irrigation. In order to obtain drought tolerant genotypes, it is important to enlarge root-to-shoot ratios, since enhanced root growth in plants is fundamental to improve drought tolerance and yield under water stress in various crops (Idrissi *et al.*, 2015; Mathew *et al.*, 2018; Mwenye *et al.*, 2018).

Table 1. Growth of the new interspecific hybrid *S. officinalis* × *S. tomentosa* for three months (April–July 2021) in a greenhouse under normal and reduced irrigation (data per plant).

Irrigation frequency	Plant height (cm)	Lateral shoot number	Lateral shoot mean length (cm)	Lateral shoot total length (cm)	Above ground d.w. (g)	Root d.w. (g)	Root d.w./ Above ground d.w.
Normal	30.1 a[†]	10.3 a	10.2 a	105.1 a	9.4 a	2.6 a	0.3 a
Reduced	24.6 b	8.1 a	9.0 a	73.5 a	9.0 a	2.6 a	0.3 a
Significance [§]							
$F_{\text{one-way ANOVA}}$	*	NS	NS	NS	NS	NS	NS

[†] Mean values (n = 10) in each column followed by the same lowercase letter (a-b) did not differ significantly at $p \leq 0.05$ using Student's t-test.
[§] NS or * or **, non-significant at $p \leq 0.05$ or significant at $p \leq 0.05$ or $p \leq 0.01$, respectively.

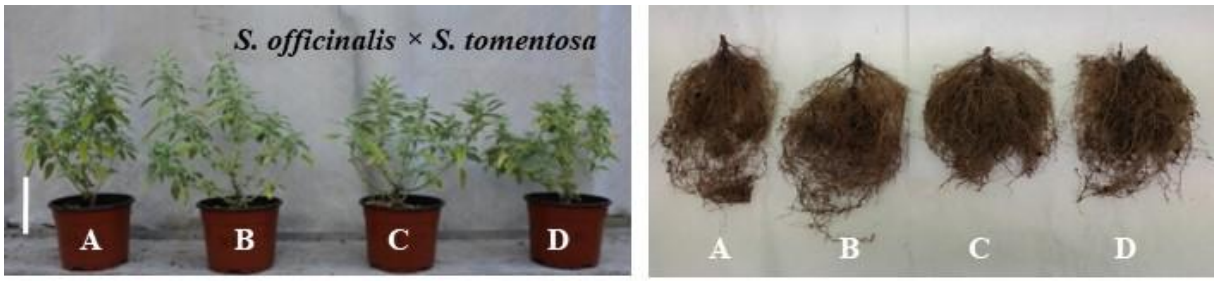

Figure 2. Typical above ground and root system of the new interspecific hybrid *S. officinalis* × *S. tomentosa*, after having grown for three months in greenhouse conditions (A: without and B: with 25 g/L attapulgite/normal irrigation, C: without and D: with 25 g/L attapulgite/sparse irrigation). Size bar = 10.0 cm.

Conclusions

Despite the reduction of all growth parameters of all *Salvia* types under reduced irrigation in the extensive green roof, their growth remained satisfactory. So, both the hybrid *S. officinalis* × *S. tomentosa* and its parents, *S. officinalis* and *S. tomentosa*, are recommended for sustainable use in xeriscaping, including extensive green roofs.

Acknowledgements

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Table 2. Comparative evaluation of growth of two Greek sage species (*S. officinalis* and *S. tomentosa*) and their interspecific hybrid (*S. officinalis* × *S. tomentosa*) after five-month cultivation (April–September 2021) in an urban Mediterranean green roof under normal and reduced irrigation.

Data		Per container	Per plant		Per container (2 plants)		
Salvia type	Irrigation Frequency	Survival (%)	Foliage height (cm)	Plant diameter (cm)	Above ground d.w. (g)	Root d.w. (g)	Root d.w./ Above ground d.w.
S. officinalis	Normal	83.3 a[†]	23.9 c	29.2 bc	70.3 b	95.9 a	1.4 a
	Reduced	75.0 a	20.8 c	25.4 c	50.4 c	74.9 ab	1.5 a
S. tomentosa	Normal	66.7 a	37.0 a	43.2 a	109.4 a	93.1 a	0.8 a
	Reduced	91.7 a	32.0 b	32.8 b	72.0 b	38.0 bc	0.5 b
S. officinalis × *S. tomentosa*	Normal	91.7 a	29.9 b	30.2 bc	85.1 b	61.8 abc	0.7 b
	Reduced	66.7 a	24.0 c	26.0 c	52.5 c	32.2 c	0.6 b
Significance [§]							
$F_{Salvia\ type}$		NS	**	**	**	*	**
$F_{irrigation\ frequency}$		NS	**	**	**	**	NS
$F_{ineraction}$		NS	NS	NS	NS	NS	NS
$F_{one\text{-}way\ ANOVA}$		NS	**	**	**	**	**

[†] Mean values (n = 8-12) in each column using Student's t test; means followed by the same letter are not significantly different at $p \leq 0.05$

[§] NS or * or **, non-significant at $p \leq 0.05$ or significant at $p \leq 0.05$ or $p \leq 0.01$, respectively.

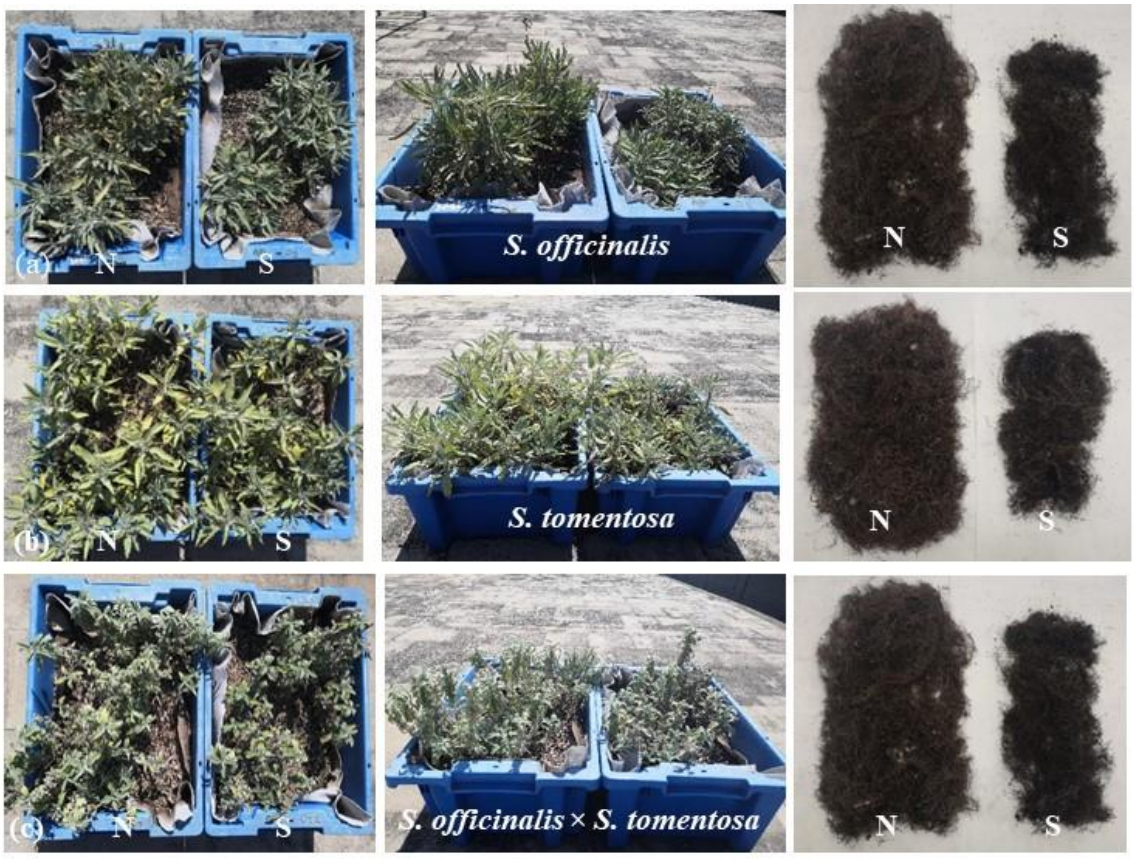

Figure 3. Typical growth of aboveground and root system of *S. officinalis* (a), *S. tomentosa* (b), and *S. officinalis* × *S. tomentosa* (c), after five months cultivation (April - September 2021) in an extensive green roof, under normal (N) and sparse (S) irrigation.

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Introduction

The new Mediterranean sage hybrid *Salvia officinalis* × *S. tomentosa was* produced through interspecific crossbreeding between *S. officinalis* and *S. tomentosa* native to Greece (Papafotiou *et al*., 2021). Aiming to exploit this hybrid along with its parental species, in extensive green roofs and xeriscaping, their resistance to xerothermic conditions was examined in a Mediterranean green roof and in a greenhouse under normal and sparse irrigation frequency.

Figure 1. Typical aboveground and root system of the new interspecific hybrid *S. officinalis* × *S. tomentosa*, after growing for three months in greenhouse conditions, under normal (N) and sparse (S) irrigation frequency.

Table 1. Growth of the new interspecific hybrid *S. officinalis* × *S. tomentosa* for three months (April–July 2021) in a greenhouse under normal and sparse irrigation (data per plant).

Irrigation frequency	Plant height (cm)	Lateral shoot number	Lateral shoot mean length (cm)	Lateral shoot total length (cm)	Above ground dr.wt. (g)	Root dr.wt. (g)	Root dr.wt./ Above ground dr.wt.
Normal	30.1 a†	10.3 a	10.2 a	105.1 a	9.4 a	2.6 a	0.3 a
Sparse	24.6 b	8.1 a	9.0 a	73.5 a	9.0 a	2.6 a	0.3 a
Significance §							
$F_{\text{one-way ANOVA}}$	*	NS	NS	NS	NS	NS	NS

† Mean values (n = 10) in each column followed by the same lowercase letter (a-b) did not differ significantly at $p \leq 0.05$ using Student's t-test.
§ NS or *, non-significant at $p \leq 0.05$ or significant at $p \leq 0.05$, respectively.

Table 2. Comparative evaluation of growth of two Greek sage species (*S. officinalis* and *S. tomentosa*) and their interspecific hybrid (*S. officinalis* × *S. tomentosa*) after five-month cultivation (April–September 2021) in an urban Mediterranean green roof under normal and sparse irrigation frequency.

Data		Per container	Per plant		Per container (2 plants)		
Salvia type	Irrigation Frequency	Survival (%)	Foliage height (cm)	Plant diameter (cm)	Above ground dr.wt. (g)	Root dr.wt. (g)	Root dr.wt./ Above ground dr.wt.
S. officinalis	Normal	83.3 a†	23.9 c	29.2 bc	70.3 b	95.9 a	1.4 a
	Sparse	75.0 a	20.8 c	25.4 c	50.4 c	74.9 ab	1.5 a
S. tomentosa	Normal	66.7 a	37.0 a	43.2 a	109.4 a	93.1 a	0.8 a
	Sparse	91.7 a	32.0 b	32.8 b	72.0 b	38.0 bc	0.5 b
S. officinalis × *S. tomentosa*	Normal	91.7 a	29.9 b	30.2 bc	85.1 b	61.8 abc	0.7 b
	Sparse	66.7 a	24.0 c	26.0 c	52.5 c	32.2 c	0.6 b
Significance §							
$F_{\textit{Salvia}\text{ type}}$		NS	**	**	**	*	**
$F_{\text{irrigation frequency}}$		NS	**	**	**	**	NS
$F_{\text{interaction}}$		NS	NS	NS	NS	NS	NS
$F_{\text{one-way ANOVA}}$		NS	**	**	**	**	**

† Mean values (n = 8-12) in each column using Student's t test; means followed by the same letter are not significantly different at $p \leq 0.05$
§ NS or * or **, non-significant at $p \leq 0.05$ or significant at $p \leq 0.05$ or $p \leq 0.01$, respectively.

Conclusions

The new Mediterranean sage hybrid *Salvia officinalis* × *S. tomentosa* grew very satisfactorily in a green house under sparse irrigation.
Despite the reduction of all growth parameters of all three *Salvia* types under sparse irrigation in the extensive green roof, their growth remained satisfactory and are recommended for sustainable use in xeriscaping, including extensive green roofs.

ACKNOWLEDGEMENTS
This research has been co-financed by the European Regional Development Fund of the European Union and Greek national funds through the Operational Program Competitiveness, Entrepreneurship and Innovation, under the call RESEARCH – CREATE – INNOVATE (project code:T1EDK-04923). https://www.salvia-breed-gr.com/el/

Materials and Methods

In a greenhouse experiment, 4-week-old rooted cuttings of the new interspecific sage hybrid *S. officinalis* × *S. tomentosa*, were grown singly in 14 cm pots, on a peat-perlite mixture 2:1 (v/v). Plants were fertilized monthly with 2 g/L water soluble fertilizer 20-20-20 (100 ml fertilizer/ pot).
In an extensive green roof experiment, 8-week-old rooted cuttings of the hybrid *S. officinalis* × *S. tomentosa*, alongside with its parental species *S. officinalis* and *S. tomentosa*, were cultivated in plastic containers with a green roof infrastructure fitted, on a second-floor flat roof at the Agricultural University of Athens. The substrate used was grape marc compost: perlite: pumice (3: 3: 4, v/v) and had a 10 cm depth. Both experiments started in April 2021 and lasted 3 and 5 months, respectively. Two irrigation frequencies, a normal (every 2–3 days, when substrate moisture 17–23% v/v) and a sparse (every 3–5 days, when substrate moisture 7–13% v/v) were applied.

Results and Discussion

In the greenhouse experiment, all plants survived and the hybrid grew very satisfactorily under sparse irrigation, since only plant height was reduced from all growth parameters recorded (Table 1, Figure 1).
In the green roof experiment, survival rate was affected neither by *Salvia* type nor by irrigation frequency (Table 2). *S. tomentosa* had higher plant height and diameter and aboveground dry weight than *S. officinalis* and their hybrid, while root dry weight was higher in the parental species compared to the hybrid. Sparse irrigation resulted in significant reduction of all growth parameters in all *Salvia* types. However, their growth remained satisfactory (Table 2, Figure 2).
The root system was found to play a key role in plant drought resistance in *S. officinalis* (Abate *et al*., 2021), a result that was also supported by our research since *S. officinalis* showed the largest root/aboveground fresh and dry matter under both normal and sparse irrigation.

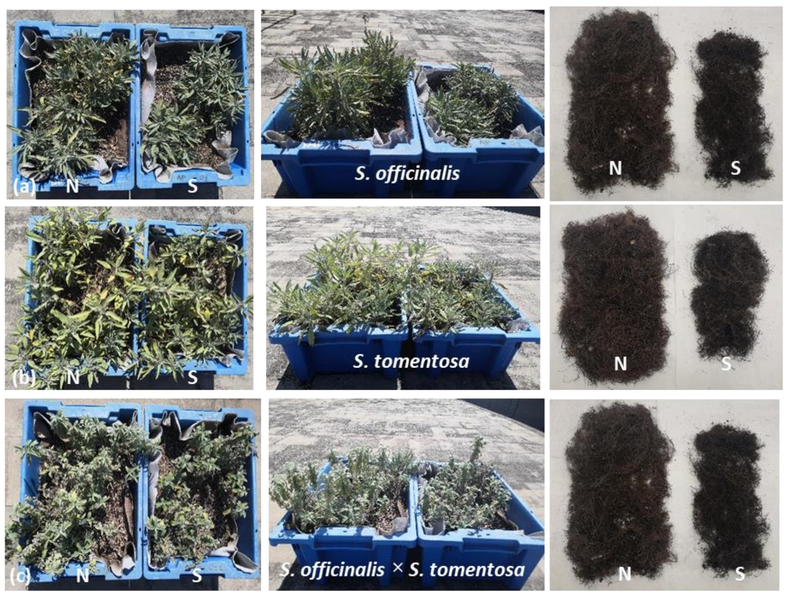

Figure 2. Typical growth of aboveground and root system of *S. officinalis* (a), *S. tomentosa* (b), and *S. officinalis* × *S. tomentosa* (c), after five months cultivation (April - September 2021) in an extensive green roof, under normal (N) and sparse (S) irrigation.

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