



[DOI: 10.22620/agrisci.2021.30.001](https://doi.org/10.22620/agrisci.2021.30.001)

## THE CHANGE IN THE BIOMETRIC AND PHYSIOLOGICAL PARAMETERS OF ASTER (*CALLISTEPHUS CHINENSIS*), HELICHRYSUM (*HELICHRYSUM BRACTEATUM*) AND ECHINACEA (*ECHINACEAE PURPUREA*) UNDER CONDITIONS OF INDUCED WATER DEFICIT

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### Abstract

The global climate models predict changed precipitation patterns with frequent episodes of drought. Scarcity of water is a severe environmental constraint to plant productivity. Plants display a variety of physiological and biochemical responses at cellular and whole-organism levels towards prevailing drought stress. This study included container experiments with three flower crops aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinaceae purpurea*). Drought was simulated by reducing the number of watering's to field capacity from 25–30 to 85–90 %. Three levels of watering were studied - three times, twice and once a week. The results showed that the cultivation of ornamental plants in containers under reduced watering conditions (twice or once a week) for a period of 3 months led to considerable inhibition of growth and even to plants death. The relative water content and the level of electrolyte leakage vary depending on the weekly number of waterings. The highest values of electrolyte leakage were reported in a single watering for aster -5107.1  $\mu\text{S} / \text{g}$ , for helichrysum -8314.9  $\mu\text{S} / \text{g}$  and for echinacea -3722.8  $\mu\text{S} / \text{g}$ . The high rates of conductivity, especially with one-time weekly watering, are evidence of the damage caused by the simulated water stress. This corresponds to the reported low percentages of *RWC* % and the low values for the height and diameter of the plants. The relative water content in plant tissues decreases depending on the irrigation regime. The lowest values for aster, helichrysum and echinacea were again observed in the variant with a single weekly watering, respectively 15%, 11.5% and 15.8%.

**Key words:** *Callistephus chinensis*, *Helichrysum bracteatum*, *Echinaceae purpurea*, water deficit, relative water content, conductivity

### INTRODUCTION

Climate change affects water resources - directly, through changes in natural factors (rainfall, temperature, evaporation) and indirectly - caused by the changes in land use (forests, agricultural land, etc.) The increasing frequency of the extreme events - floods and drought, exacerbates this problem. The reported

results of the international project "Mitigation of Water Vulnerability to Climate Change" (CC-WARE) confirm that these trends will continue, affecting water resources and water use (Čenčur Curk et al 2014) When the tendency of reduced rainfall and runoff coincides with the tendency of increased water consumption, then a region or a river basin may be vulnerable. In such regions the role of adaptation measures becomes



increasingly important. According to the results of CC-WARE and RBMP 2016-2020, there is a lasting trend towards drought in the Balkan Peninsula (Popova, Z. 2012, Ilcheva, I., et al. 2015, Spiridonov, V. et al. 2014). The forecasts of the National Institute of Meteorology and Hydrology at the Bulgarian Academy of Sciences show an expected increase in air temperatures in Bulgaria by 1.6 °C to 3.1 °C by 2050. Regarding precipitation, the models forecast a decrease in precipitation during the summer to 15% by 2050 and up to 30 - 40% by 2080 (MoEW, 2019, 3). To reduce or prevent damage from droughts, adaptation measures should be developed, including the following: establishing the vulnerability of the agricultural crops, including ornamental plants, to water scarcity and its impact on the quantity and quality of production, the use of new crop varieties that have increased tolerance to drought and a change in technologies for growing irrigated crops in different agro-climatic regions under the conditions of water scarcity. A number of studies have identified specific species and a variety of response to a decrease in moisture retention and an increase in the impact of water deficit. Some crops are characterized by a better adaptation due to increasing the level of adaptive water exchange of plants, better quality of production, while others show inhibition of development and deterioration of the quantity and quality of production (Valchev, et al. 2005, Bozhanova et al. 2009, Zapryanova and Ivanova, 2017, Lozanova et al. 2012, Mohamed et al., 2014, Spiridonov, et al., 2014, Stoyanov, 2005, Zapryanova, Nencheva 2015). The aim of the present study is to investigate the effect of the regulated water deficit on the development and the physiological state of aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinacea purpurea*).

## MATERIALS AND METHODS

The study was conducted at the Institute of Ornamental Plants - Sofia for one growing season in an unshaded compartment of a steel glass greenhouse, at high atmospheric temperature, low humidity and reduced watering. Drought simulation is performed by adjusting the number of waterings - as the range of PPV ranges from 30-90% humidity. The plant material used is aster (*Callistephus chinensis*), *Helichrysum bracteatum* and *Echinacea purpurea*. The aster and helichrysum plants were sown in March in greenhouses under greenhouse conditions. The seedlings laid in the experimental setup are in phase 5-6 leaves. Of the species *Echinacea purpurea* 'Pow Wow Wild Berry', adapted in vitro material was used. The plants are planted in early June in pots №15 s (volume 1.5 l) in a mixture of soil, peat and perlite in a ratio of 1: 1: 0.5. The experiment is carried out in an unshaded compartment of a steel glass greenhouse, at high atmospheric temperature, low humidity. Drought simulation starts after catching the plants - (2 weeks after planting them in pots №15) and is done by adjusting the number of waterings. The experiments are set in 3 variants of 10 repetitions: Var.1 / Control / - three waterings per week (maintenance of optimal maximum field moisture content 80% PPV); Var.2 - two waterings per week (maintenance of optimal field moisture content 50-53% PPV); Var.3 - one watering per week (maintenance of optimal maximum field moisture content 27-30% PPV). The amount of water for one watering was 200 ml of water for a pot. Temperature and humidity were recorded daily with a thermometer and hygrometer. The water content of the soil mixture was determined by the drying method by calculation and calculated by the corresponding formula:  $w = \frac{A-B}{B-C} \times 100, \%$ , where A is the mass of the wet sample together with the tare, c (g); B - the mass of the dried sample is occupied by the tare, c (g); The C-



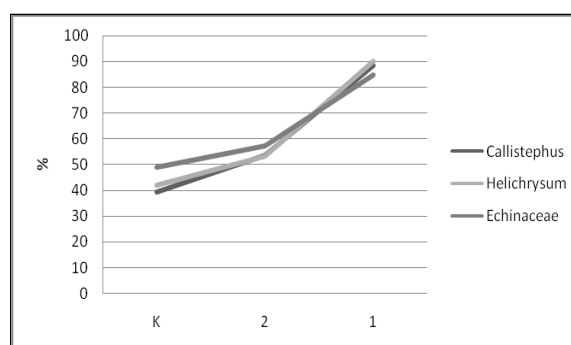
mass of the tare, in (gr). At the beginning of August, the influence of the reduced water regime on the development of the plants was monitored, taking into account the following indicators: the height and diameter of the plant (cm). The change in the following indicators was reported: - the relative water content (% OVS) and the degree of damage to the cell membranes. The relative water content (RWC) is measured simultaneously with the leakage of electrolytes and is calculated by the formula:  $OVS\% = (sv.t. - s.t.) / (t.t. - s.t.) \times 100$  methodology of Turner (Turner, 1981) The water deficit (VD,%) is expressed by the formula:  $VD\% = 1 - OVS$ . The degree of membrane damage is determined by the leakage of electrolytes from the leaves, taking into account the conductivity only after stress and is expressed in  $\mu S / g$  fresh weight. The first upper fully developed leaf of the crops is used for the analysis of the physiological indicators. The data were analyzed using the t-test of the GraphPad Prizm software. The results were statistically proven at  $P < 0.05$  (\*),  $P < 0.01$  (\*\*),  $P < 0.0001$  (\*\*\*), respectively, compared to the control.

## RESULTS AND DISCUSSION

Reducing the number of waterings significantly affects the development of the three flower crops - aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinacea purpurea*). The obtained results show that when applying 3 times watering the plants are subjected to moderate stress and this is due to the action of other factors: high atmospheric temperature and low humidity. The measured air temperature during the growing season is ranged on average in the range of 25-35 °C, and on the soil 20-25 °C. The air humidity in the greenhouse was on average about 60%, according to studies by Cornic G. and Fresneau B. (2002) taken as mild or moderate stress.

When maintaining an optimal field moisture content of 27-30% PPV (single watering), the water deficit in plants reaches up to 90% in annual crops - helichrysum and aster, and in echinacea by almost 5% lower - 85% (Fig. 1).

The reported high percentage of water deficit indicates that the plants are under stress and this is complemented by the obtained biometric and physiological results. Lack of water causes destruction of membrane systems, loss of turgor and inhibition of growth, and the manifestation of these disorders depend on the effective and protective mechanisms of plant species.



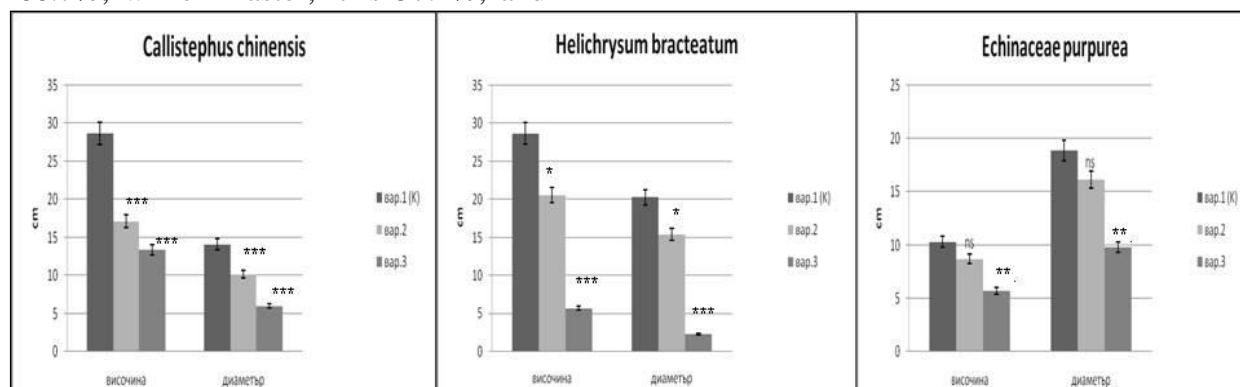
**Fig. 1** Water deficit in aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinacea purpurea*).

The three species - aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinacea purpurea*) show differences in the reaction to the applied drought. Echinacea reacts with drying and death of the leaf mass. On the above-ground mass in echinacea appears as a protective reaction of the species, which in terms of the development cycle belongs to the perennial flowering species, while in aster and helichrysum, which have a one-year cycle, the process is irreversible and the plants wither. Minimizing the number of waterings has a very strong negative effect on the growth of the three crops aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinacea purpurea*). The decrease



in plant height at 1 watering per week compared to that at the control (3rd watering per week) is most pronounced in helichrysum - respectively by 88.7%, while in aster, it is 57.4%, and in

echinacea - the difference is 45% (Fig. 2) The same trend is observed when considering the diameter of the plants (Fig. 2)

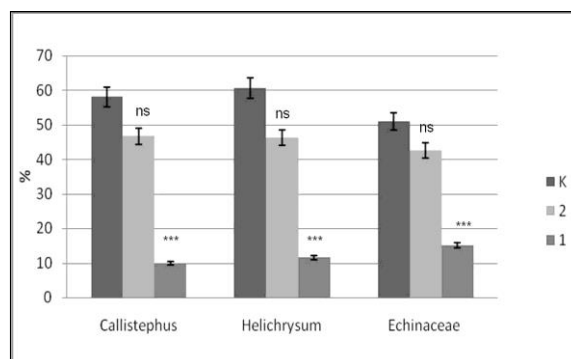


**Fig. 2** Biometric indicators in aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinaceae purpurea*)

When studying the reaction of other flower crops to drought, it was also found that reducing the number of waterings significantly affects the plant growth. In the case of gladioli, the reported difference in the height of the plants in phase 7-8 leaves at 1 watering per week compared to the control (3rd watering per week) is more pronounced in the variety Ekaterina - respectively by 48.5%, while in the variety Iva, is with 35.2%. (Zapryanova, Ivanova, 2017). In the case of the chrysanthemum, the reduction in the number of waterings for 3 months leads to the cessation of growth and the death of a large part of the plants watered once. The increase in height with 2 waterings was also very weak in all three varieties, as it was found that the least dead plants of the variety Milka (20%) and it was relatively less inhibited growth - 35.4% compared to the control, while in Gipsy the dead plants were 80% and the average height was 10.7% compared to that of the control plants. (Zapryanova, Nencheva, 2015). This shows that in addition to species, there is also a varietal reaction to the adverse environmental factors. Throughout the reporting period, the relative water content (% OVS) in the control plants in the three crops was around 50-60% (Fig. 3).

The relative water content in plant

tissues decreases depending on the irrigation regime. The lowest values for aster, helichrysum and echinacea were again observed in the variant with a single weekly watering, respectively 15%, 11.5% and 15.8% (Fig. 3). Similar results - (the highest percentage of OVS in control plants was about 80%) were obtained with rosemary grown with reduced watering (Hassan, et al.2013). In a study of the reaction of a chrysanthemum subjected to a reduced water regime, the lowest values were noted when maintaining a low 27-30% PPV (Zapryanova, Nencheva, 2015).



**Fig. 3** Relative water content (% OVS) in aster (*Callistephus chinensis*), *Helichrysum bracteatum* and *Echinaceae purpurea*

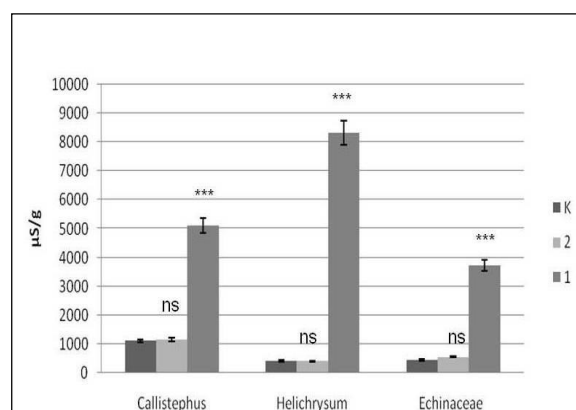
The same trend was observed with gladiolus, with reported percentages of about



14%. The results obtained for the change in the relative water content of both cultivars Ekaterina and Iva show insignificant differences, statistically unprovable (Zapryanova, Ivanova, 2017). The determination of the relative water content in the leaves (OVS) is an indicator of the water status of the plants (Georgieva et al., 2004). The percentage of OVS is a parameter showing the relationship between the physiological properties and the level of drought resistance of the plant species Farooq, (2009). In combination with additional parameters (proline content) it can be used to evaluate new species suitable for urban cultivation (Chyliński et al., 2007). Simulated drought by reducing watering causes changes in the cell membranes. Electrolyte leakage was also observed in control plants, ranging from about 450  $\mu\text{S} / \text{g}$  in helichrysum and echinacea to 1112  $\mu\text{S} / \text{g}$  in aster. The results are close to those obtained with double watering and have no statistical evidence. The highest values of electrolyte leakage were reported in a single watering, as in the case of aster it was - 5107.1  $\mu\text{S} / \text{g}$ , in the case of helichrysum - 8314.9  $\mu\text{S} / \text{g}$  and in the case of echinacea - 3722.8  $\mu\text{S} / \text{g}$ . The high conductivity rates are evidence of the damage caused by the simulated water stress. This corresponds to the reported low percentages of OVS and low values for the height and diameter of the plants (Fig. 4).

In the studies of Zapryanova and Ivanova (2017), electrolyte leakage was also observed to a low degree in the control plants, as the values for Ekaterina variety were 96.7  $\mu\text{S} / \text{g}$ , and for Iva variety - 101.11  $\mu\text{S} / \text{g}$ . In the variants with reduced waterings, the values increase, which signals a higher degree of damage to the membranes. The highest values were noted in the variant with one watering for Ekaterina variety - 156.46  $\mu\text{S} / \text{g}$ , which is 61% above the control indicators and for Iva variety 179.269  $\mu\text{S} / \text{g}$  where the increase is 77%. The simulated drought caused changes in the cell membranes of the chrysanthemum, leading to a

change in the physiological parameters in all three tested varieties "Milka", "Gypsy" and a candidate variety "№ 509" which has shown the highest values of 584.25  $\mu\text{S} / \text{g}$  in a single watering (Zapryanova, Nencheva, 2015).



**Fig.4** Electrolyte leakage from plant tissues in aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinaceae purpurea*)

## CONCLUSIONS

Reducing the number of waterings affected the development of flower crops aster (*Callistephus chinensis*), helichrysum (*Helichrysum bracteatum*) and echinacea (*Echinaceae purpurea*). With double weekly watering, the plants, although under slight stress, developed normally and the results obtained for growth and their physiological parameters did not differ statistically from those shown by the control plants. The negative effects of drought caused by the reduction of watering up to once a week have led to inhibition of the development of the tested flower species. The reported high percentage of water deficit - 85-90% indicated that the plants were under stress and this was complemented by the obtained biometric and physiological results. The decrease in the plant height in a single watering per week compared to that in the control (3rd watering per week) was most pronounced in helichrysum - respectively by



88.7%, while in aster, it was 57.4%, and in echinacea- the difference was 45%. The relative water content in the plant tissues decreased and the lowest values were observed in helichrysum - 11.5%. The changes in the cell membrane stability were proved by the high value of the electrolyte leakage from the plant tissues, as in the case of the aster it was  $-5107.1 \mu\text{S} / \text{g}$ , in the case of helichrysum -  $8314.9 \mu\text{S} / \text{g}$  and in the case of echinacea  $-3722.8 \mu\text{S} / \text{g}$ .

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