Responses of young cucumber plants to a diurnal temperature drop at different times of day and night

Tatjana G. SHIBAEVA1*, Elena G. SHERUDILO1, Elena N. IKKONEN1, Alexander F. TITOV1

Received January 24, 2018; accepted October 25, 2018.

ABSTRACT

In greenhouse production of a number of vegetable and ornamental plant species, a short diurnal temperature drop in the end of the night or in the morning is commonly used to reduce stem elongation as an alternative to chemical growth retardants. Experiments were carried out to quantify the effects of a temperature drop at different times of the day and night on growth and photosynthetic activity of young cucumber plants. During 6 days plants were exposed daily to a temperature of 10 °C for 2 h at the beginning, in the middle and at the end of the night and day periods. The results have shown that plant response to drop may be qualitatively different in the light and darkness. While strongest effects of drop are observed when it is given in the daytime, for practical application in greenhouses it is more appropriate to reduce temperature at night. However, it may not be strictly necessary for cucumber seedlings to apply drop at the end of the night as it was stated in the literature. Thus, our results may cast doubt on the following statements: (a) temperature drops are not effective when delivered at other times of the day or night (except before sunrise), (b) optimal time for drop effects depends on the daily dynamics of stem and petiole elongation rate. It is rather drop itself is capable of modifying the dynamics of plant growth in the daily cycle.

Key words: Cucumis sativus L.; growth; daily rhythms

1 INTRODUCTION

Plant growth retardants are widely used as tools for height control in order to obtain more compact plants. However, the use of growth retardants on vegetables is prohibited in many countries because of the potential danger of chemical residues to the environment and the consumer. At the same time, it has long been known that stem elongation can be controlled by temperature treatments rather than by the use of chemical growth retardants (Moe & Heins, 1990). In particular, a daily temperature drop or dip for 2-4 h is the practice of lowering the temperature in order to obtain compact transplants of vegetable crops, bedding plants and ornamentals (Myster & Moe, 1995; Berghage, 1998; Runcle, 2009). It is believed that the time during 24-h
period when the temperature decreases, has a certain effect on the degree of stem elongation inhibition (Myster & Moe, 1995; Grindal & Moe, 1995; Grimstad, 1995). In the earlier studies on *Lilium longiflorum* Wiebe, *Begonia x hiemalis* Fortsch and *Solanum lycopersicum* L., it was shown that the temperature drop effect is the greatest in the first period of the day (Myster & Moe, 1995). Later, it was found that the temperature drop in the last part of the night is even more effective in reducing plant height and internode length in *Begonia x chienantha* Everett (Moe & Mortensen, 1992; Grindal & Moe, 1994, 1995; Bakken & Moe, 1995), *Euphorbia pulcherrima* Willd. (Moe et al., 1992), tomato and cucumber (Grimstad, 1995). This turned out to be very convenient for practical application of this technique as the lowering temperature in the end of the night is far more energy efficient than at the start of the day, especially if the lights are switched on in the morning. The required decrease in temperature can be obtained by turning off heaters for a short time. If the greenhouse does not cool rapidly enough, venting may be necessary to achieve the desired low-temperature setpoint. It is generally believed that temperature drops are not effective when delivered at other times of the day or night (except before sunrise) (Runcle, 2009). However, the information about the effects of a temperature drop given at other times of the day or night (except for the end of the night and the beginning of the day) is extremely scarce. There is evidence that a temperature drop in the middle of the night had small effect on plant height and petiole length in *Euphorbia pulcherrima* (Moe et al., 1992) and *Begonia x hiemalis* (Moe & Mortensen, 1992; Grindal & Moe, 1994). In our previous studies we have shown that a temperature drop at the beginning, middle or end of the night was equally effective in reducing plant height and petiole length in cucumber (Sysoeva et al., 1997, 1999). It is also suggested that a temperature drop at the beginning of the day is much more effective than later during the day in *Euphorbia pulcherrima* (Ueber & Hendriks, 1992) and *Pelargonium* (Ueber & Hendriks, 1995), and that a temperature decrease at any time during photoperiod may be equally effective in inhibiting stem elongation as a temperature decrease at the beginning of the day in *Fuchsia, Antirrhinum majus* L., *Petunia* and *Salvia splendens* F. (Erwin & Heins, 1995).

Data on the effects of a temperature drop at different times of the day on plant characteristics other than stem elongation rate are even more fragmentary and contradictory. It was shown that plant dry mass was decreased by low temperature pulse treatment in cucumber and tomato and the largest effect was achieved at the end of the night in cucumber, but at the beginning of the day in tomato (Grimstad, 1995). Begonia plants affected by a temperature drop at the end of the night or in the morning had smaller dry mass compared to the control plants, but a temperature drop at the beginning of the night increased plant dry mass. Moreover, a temperature drop given in the night increased leaf weight ratio (LWR), while a temperature drop given in the morning did not affect LWR (Bakken & Moe, 1995). There are also data on the effect of a temperature drop on the chlorophyll content. In experiments with *Ocimum basilicum* L., there was a tendency that plants treated by a temperature drop at the beginning and in the end of the day had lower concentrations of chlorophyll $a$ and $b$, but in the cold-resistant species *Viola x wittrockiana* Gams., a temperature drop in the early morning increased total chlorophyll content (Vägen et al., 2003).

The objective of the present experiment was to quantify the effects of a temperature drop at different times of the day and night on growth and photosynthetic activity of young cucumber plants.

**2 MATERIALS AND METHODS**

Experimental facilities for this study were offered by the Shared Equipment Center of the Institute of Biology, Karelian Research Center, Russian Academy of Sciences. Imbibed seeds of cucumber (*Cucumis sativus* ‘Zozulya F1’) were sown in 7 cm pots containing sand and seedlings were watered daily with a complete nutrient solution (based on 1 g l$^{-1}$ Ca(NO$_3$)$_2$, 0.25 g l$^{-1}$ KH$_2$PO$_4$, 0.25 g l$^{-1}$ MgSO$_4$ 7H$_2$O, 0.25 g l$^{-1}$ KNO$_3$, trace quantity of FeSO$_4$ and pH 6.2-6.4, EC 2.0 mS cm$^{-1}$). Plants were grown under air temperature 23 °C, air humidity 70%, a photoperiod of 12 h (from 9:00 to 21:00), under controlled environmental conditions (*Vötsch* growth chamber, Germany) with a photosynthetic photon flux density of 200 μmol m$^{-2}$ s$^{-1}$.

Starting from the 7th day from seed soaking, different groups of plants were exposed daily for 6 days to a temperature of 10 °C for 2 hours at the beginning (21:00-23:00, N1), in the middle (2:00-4:00, N2) and at the end of the night period (7:00-9:00, N3), as well as at the beginning (9:00-11:00, D1), in the middle (14:00-16:00, D2) and at the end of the day period (19:00-21:00, D3). Control plants were not treated by a temperature drop (DROP). At the end of DROP treatments all plants were grown under air temperature.
of 23 °C. All measurements were carried out on the next day after the last DROP treatment (on the 14th day). Plant height, petiole length of the first true leaf, leaf area (n ≥ 10), dry mass of leaves, stems and roots (n = 5) were recorded. The total chlorophyll content was determined non-destructively by using a SPAD 502 Plus chlorophyll meter (Konica Minolta, Japan) (n ≥ 10). The chlorophyll fluorescence was measured using a portable chlorophyll fluorometer MINI-PAM (Walz, Germany) on the 1st true leaf. The maximal quantum yield of PSII photochemistry was calculated as $F_{v}/F_{m} = (F_{m} - F_{0})/F_{m}$ after 20 min of dark adaptation of leaves (n = 5) (Maxwell & Johnson, 2000).

All results are presented as means ± SE (n ≥ 5). Data were tested for normality and homogeneity of variance using Chi-Square test and Levene’s test in Statistica (v.8.0.550.0, StatSoft, Inc). Differences between the treatment means were tested with one-way ANOVA followed the Bonferroni test with P < 0.05 level of significance. Two similar trials were run.

3 RESULTS

The obtained results showed that in general DROP treatments reduced the plant height by 14-16 %, compared with the control (Fig. a). The leaf petiole length in all plants treated by DROP was also less than in the control (Fig. b). The greatest retardation of the leaf petiole growth was noted when DROP had been given in the middle and at the end of the day (by 46-47 %) (treatments D2 and D3), while the smallest inhibition (by 20 %) occurred when it had been given in the middle and at the end of the night (N2 and N3).

The leaf area was reduced by all DROP treatments compared with the control. In case of DROP given in the daytime the reduction was by 47-49 %, but in the night – by 25-29 % (Fig. c). There were no significant differences between leaf area of plants treated by DROP at the beginning, in the middle or at the end of the day as well as between treatments at different times in the night. The similar effect was observed for the plant dry mass, which was decreased by 51-53 % compared with the control by DROP given in the daytime and by 21-30 % by DROP given in the night (Fig. d). However, no effect of DROP on biomass allocation to leaves, stems and root (data not shown) was observed.

The chlorophyll content in the leaves of plants exposed to DROP-treatments did not differ from the control, except for the plants treated by DROP at the beginning of the day (D1), when the chlorophyll content was reduced by 23 % (Fig. e).

The values of $F_{v}/F_{m}$ in leaves treated by DROP in the night time were not significantly different from the control ones, but DROP given in the daytime lowered the values of $F_{v}/F_{m}$ (Fig. f).
4 DISCUSSION

The results of this work showed that the DROP treatments applied at any time of the day or night results in growth retardation (reduced plant height, leaf petiole length) of cucumber plants, which unequivocally indicates that there are no periods in the daily cycle when DROP treatments are absolutely ineffective for obtaining compact plants. The strongest morphogenetic responses to DROP treatments were observed when DROP had been given in the middle or at the end of the day, which from the practical point of view is of little use. DROP given at the end of the night or at the beginning of the day similarly reduced stem and petiole elongation, as it was earlier reported by Grimstad (1995) for young cucumber plants. Unlike in case with *Euphorbia pulcherrima* (Moe et al., 1992) and *Begonia x hiemalis* (Moe & Mortensen, 1992; Grindal & Moe, 1994), when DROP given in the middle of the night had only slight effect on plant height and leaf petiole length, in our work with cucumber plants there were no significant differences revealed between plant morphogenetic response to DROP given at different times during the night period. The same results we reported earlier (Sysoeva et al., 1997, 1999). Already in the early studies on the potential of fluctuating

**Figure 1**: Effect of a temperature drop (DROP) given at the beginning (N1), in the middle (N2), in the end (N3) of the night and in the beginning (D1), in the middle (D2) or in the end (D3) of the day.
temperatures as alternative to growth retardants it was pointed out that plant responses are very much crop dependent (Cuypers & Vogleezang, 1992). Therefore, it is not surprising that the results obtained in experiments with different species may differ significantly.

Pronounced morphogenetic effect of DROP given in the daytime also indicates that although it is known that stem elongation is not constant during a 24-h period and that the major stem extension occurs during the night (Lecharny et al., 1985; Sweeny, 1987; Erwin & Heins, 1988; Bertram & Karlsen, 1994; Tutty et al., 1994; Luna-Maldonado et al., 2017), however, the optimal time for DROP treatments in order to obtain compact plants may not coincide with the periods of the greatest stem elongation rate. This has been also shown in several experiments with tomato (Gertsson, 1992; Grimstad, 1995). There are data that an increase in a temperature drop duration from 2 to 4 h (Grimstad, 1995; Sysoeva et al., 2008) or from 1.5 to 3 h (Mortensen & Moe, 1992) decreased or did not change the effect of DROP on stem length. These data are also formally in contradiction with the opinion that the optimal time for DROP effects depends on the daily dynamics of stem and petiole elongation rate. In this case it would seem logical to expect stronger DROP effects. Explanations for this fact have not yet been found, but there are some interesting results worth taking into consideration. In experiments with Chenopodium rubrum L., DROP stimulated stem elongation within 10 h after the termination of DROP treatment (Lecharny et al., 1985). In experiments with Dendranthema grandiflorum (Ramat.) Kitam., short-term temperature changes did not rephase the rhythm of stem growth, but significantly affected the amplitude for the remainder of the diurnal cycle (Tutty et al., 1994). These results let us to suggest that the DROP itself is capable of modifying the dynamics of plant growth in the daily cycle. Perhaps, sometimes this may be the reason of mismatch between the results obtained and those expected.

Our results demonstrate significant decrease in the leaf area of plants treated by DROP in the daytime (by 47-49 %) and in the night (by 25-29 %). Previously it was reported that DROP does not affect leaf length or width in cucumber, tomato (Grimstad, 1995), poinsettia (Moe et al., 1992). We believe that the reason for this disagreement may be related to the fact that in our experiments the temperature during DROP treatments was lowered to 10 °C, but in the experiments carried by Grimstad (1995) and Moe et al. (1992) the setpoint for low temperature was 12 ° and 13 °C, correspondingly. It is well established that at the temperature critical for chilling injury (10 °C for species of tropical and subtropical origin) cellular membranes in sensitive plants undergo a physical-phase transition from a normal flexible liquid-crystalline to a solid gel structure and other changes occur in cells that lead to numerous physiological dysfunctions (Theocharis et al., 2012).

Differences in plant response to DROP in the daytime or in the night, recorded in our experiments, can partly be explained by the involvement of a mechanism of the DROP effect on gibberellins (GA) metabolism, which is different in light and dark. It was shown in pea that DROP in the light increases expression of GA-deactivation gene PsGA2ox2 (Stavang et al., 2007). By contrast, DROP in darkness does not affect steady-state expression of this gene, but instead slightly stimulate the GA-biosynthesis genes PsGA2ox1, PsGA3ox1 and NA. Therefore, plant responses to DROP in light and darkness can differ not only quantitatively, but also qualitatively. The fact that DROP given in the dark also reduces petiole length and leaf area (although to a lesser extent) indicates that this also involves other, not GA-dependent mechanisms. The reduced leaf area in DROP-treated plants can be considered as very important in practical terms, since undesirable elongation of stems when growing seedlings of vegetable or ornamental plants occurs mainly at the time when the leaves of neighboring plants begin to overlap and shade each other, leading to competition for light in dense monoculture crops.

Data on the plant dry mass also indicate a significant difference between plants treated by DROP in the light and in the dark (DROP given in the daytime decreased the plant dry mass the most) and no differences related to the timing of DROP within the dark or light periods. Earlier, we found that DROP in the light period results in greater decrease in the apparent quantum yield of photosynthesis in cucumber plants than DROP in the dark (Ikkonen et al., 2016). At the same time, DROP in the light reduces not only light use efficiency, but also the rate of photosynthesis, which does not occur in plants treated by DROP in darkness. It is to presume that a greater decrease of biomass in plants treated by DROP in the light is associated with temporal photoinhibition, which is likely to occur, as indicated by lower values of the intrinsic photochemical efficiency ($F_v/F_m$).

In our experiments DROP had no effect on the chlorophyll content, except for its decrease in the leaves of plants treated by DROP at the beginning of the day, which was also observed in the experiments with Ocimum basilicum (Vägen et al., 2003). The loss of chlorophyll in the leaves chilled in the light, as opposite to the leaves chilled in the dark, may occur as a result of the degradation of PSII and PSI and during photoinhibition (Hetherington et al., 1989). PSI, which had been believed to be tolerant to environmental stresses, was shown to be photoinhibited when chilling-
sensitive plants were exposed to a chilling temperature under moderate light (Kudoh, Sonoike, 2002; Gururani et al., 2015). Why this does not happen when the leaves are short-term chilled in the middle or at the end of the day, remains unclear so far. It is worth noting that we observed very quick (within 24 h under optimal temperature conditions) increase in chlorophyll content to the control level in cucumber leaves after the cessation of DROP treatments (12 °C) (Shibaeva et al., 2018).

The results of our experiments with cucumber plants treated by short-term temperature drop at different times of the day and night have shown that light conditions during chilling treatment (light or darkness) rather than diurnal variability in growth rate are responsible for plant response to short-term temperature changes. Plant response to a temperature drop may not only quantitatively, but also qualitatively different in light and darkness. While the strongest effects of a temperature drop are observed when it is given in the daytime, for practical application in greenhouses it is more appropriate to reduce temperature at night. However, it may not be strictly necessary for cucumber seedlings to apply a temperature drop at the end of the night, as it was stated earlier. In fact, the same effect may be obtained by a temperature drop given at any time within the night period as the timing of a temperature drop within the dark or light periods has little effect on plant response. It might be of use in plant factories, where the interaction with the exterior climate is minimized (Graamans et al., 2018). Thus, our results may cast doubt on the following statements: (a) temperature drops are not effective when delivered at other times of the day or night (except before sunrise), (b) optimal time for a temperature drop effects depends on the daily dynamics of stem and petiole elongation rate. It is rather DROP itself is capable of modifying the dynamics of plant growth in the daily cycle.

5 ACKNOWLEDGEMENTS

The reported study was partially supported by Russian Foundation for Basic Research, research project № 14-04-00840a and Russian Federal Agency for Scientific Organizations (№ 0221-2017-0051).

6 REFERENCES


Bertram, L. & Karlsen, P. (1994) Patterns in stem elongation rate in chrysanthemum and tomato plants in relation to irradiance and day/night temperature. Scientia Horticulturae, 58, 139-150. doi:10.1016/0304-4238(94)90134-1


Responses of young cucumber plants to a diurnal temperature drop at different times of day and night


