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Improvement of grapevines rooting and growth of plants under stress conditions by Asahi SL

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ABSTRACT

The effects of grapevines cutting soaking in Asahi SL on their rooting and further plant development under optimal (20°C; 42% H_2O v/v), drought (20°C; 18, 25, 32% v/v) or temperature (10°C, 30°C; 42% v/v) stress conditions were investigated. Drought or temperature stress negatively affected subsequent development of grapevine plants in optimal conditions during the first growth season as well as in the year following winter storage. The influence of 10°C was the most unfavorable one among all the examined stress conditions. The growth of plants obtained from the rooted cuttings subjected to this temperature at the time when one developed primary root appeared was inhibited. Asahi SL application in 0.2% increased grapevine tolerance to temperature and drought stress. The positive reaction of this chemical was more prominent when cuttings with one developed primary root were exposed to the most severe drought stress (20°C, 18% H_2O v/v). This chemical agent decreased frost damages not only during winter storage but also positively affected further growth in the next vegetation season.

INTRODUCTION

Drought stress, accompanied with low or high temperatures, are the major agricultural problems due to the fact that these adverse environmental factors prevent plants from realizing their full genetic potential. In grapevine (*Vitis vinifera* L.) cultivation these stresses belong to the main constraints which decrease the quality of its products e.g. grape juices, jams, wines, raisins, vinegars or jellies (Flexas et al. 1999). The stress frequently appears to be the most dangerous at the first stage of plant development and directly after the planting of cuttings into the ground, when young plantlets are very sensitive to unfavourable weather conditions.

Application of biostimulators is one of the approaches to increase yield and quality of many crops yield. Asahi SL (ASAHI CHEMICAL MFG. CO., LTD., Japan), also known as Atonik is a biostimulator which has been developed from medicine and which perfectly fits into the required technologies for environment protection (Zraly 1999). Unlike other plant stimulators, instead of hormones it contains natural environmentally friendly compounds (0.3% sodium para-nitrophenolan + 0.2% sodium orto-nitrophenolan + 0.1% sodium 5-nitrogujakolan) which stimulate plant metabolism without causing malformation or toxicity in tomato and cotton (Djanaguiraman et al. 2004b). Asahi SL improves direct mineral nutrients uptake from the soil, accumulation of photosynthesis products (assimilates) in plant storage organs and development of soil bacteria. It also increases the activity of nitrate reductase (Sharma et al. 1984), concentration of pinitol and the flow of assimilates from a leaf into an ear in wheat (Kudrev 1969). The results of Djanaguiraman et al. (2004b) demonstrated that residual level of nitro phenolic compound in soil, cotton leaf, and fruit following its application was found to be below the threshold and it is safe for consumption. The chemical may also enhance the soil microorganism population by providing carbon and nitrogen sources.

In the light of the above, this study is an assessment of the sensitivity of young grapevine plants to drought, low and high temperature conditions, as well as a possibility of improving its resistance to these stresses by Asahi SL application.

MATERIAL AND METHODS

The research were carried out a French cultivar grape ‘Chrupka Zlota’, which is one of the most popular cultivars grown in Poland and is resistant to the low temperature (-20°C). The cane cuttings of 7 cm in length, containing one bud in the upper part in February 2005 were dipped for 24 hours at 25°C in Asahi SL solutions (Asahi Chemical Mfg. Co. Ltd., Japan), at concentration of 0, 0.1, 0.2 and 0.4%. Afterwards, they were rooted in the peat and sand medium (1:1) at 30°C for

20 days and then transferred to 20°C and 8 hour dark/16 hour light cycle (SON-T AGRO 400 W, 100 µmol m^{-2} s^{-1}). The experiment was designed in three repetitions, each including ten cuttings.

When 1, 3 or 6th primary root was developed the cuttings were transferred to:

– temperature stress, 10°C or 30°C, when moisture content of medium was 42% v/v,
– drought stress, moisture content of medium was reduced to 18, 25 and 32% v/v at 20°C,
– optimal condition for growth (control) at 20°C and moisture content of medium 42% v/v.

At the end of May, during intensive plant growth and after 75 days from the beginning of rooting, the top shoot over the third node was pinched off (removed) to improve better cuttings rooting.

In the middle of August i.e. after 156 days from the beginning of rooting the stressed and nonstressed cuttings were transferred to a greenhouse in which they were grown under growth-favorable condition and then stored over winter period during which the minimal temperature reached -20°C.

Drought stress was regulated by water content of growing media and was controlled by the Moisture Meter (Delta-T Devices Ltd) equipped with WET sensors. Drought stress was expressed as a percentage of volumetric water content in the growing media (% v/v).

The number of internodes was evaluated after 75 and 105 days from the beginning of the rooting. The visual estimation of the root system in six-grade scale and the number of canes were investigated.

The following year, in May, after winter storage the number and length of grapevine canes and number of plants damaged by frost were additionally measured.

The least significance differences (LSD) were calculated at the significance level $p = 0.01$ and 0.05 for all experimental data.

RESULTS AND DISCUSSION

The observations of grapevine cuttings and plant growth showed that both drought and temperature stress applied at the beginning of rooting and plant development negatively affected the number of developed roots and cane internodes (Figs 1 and 2). The extent of reaction depended on thermal stress conditions, stage of rooting during which the plants were subjected to drought stress and the moisture content of the growing media. The influence of 10°C was the most unfavourable one among all examined stress conditions. The growth of plants obtained from the rooted cuttings subjected to this temperature when one developed primary root appeared was inhibited (Fig. 1). It was especially visible after 75 and 105 days from the beginning of the rooting and in the case of the number of internodes and root development. The results stay in agreement with the ones obtained by Hendrickson et al. (2004), who reported 34-63% reduction in growth rates due to temperature decreased by 2°C. Chilling temperatures, similarly to drought stress, apart from limitation of growth (Buttrose 1969) also decrease leaf water potential (Báló et al. 1991). Low soil water temperature decreases root function and water transport because hydraulic resistance, stomatal conductance, and leaf transpiration are decreased (Flexas et al. 1999). Reduced plant growth due to the chilling temperature is probably indirectly caused by photosynthesis inhibition. It is more pronounced in the light condition, because it may impair activation of the carbon reduction cycle and lead to photoinhibition. However, long-term exposure to combine high and low temperature is needed to photoinhibit grapevines (Chaumont et al. 1997). In our experiments, grapevine was stored in these stress conditions for 156 days, therefore photoinhibition phenomenon may have occurred. The negative influence of 10°C during rooting of cuttings was significant even after greenhouse storage over winter (Fig. 2). After that period, the plants possessed 3 internodes less and were 10 cm shorter than the control ones.

In the case of 30°C, the earlier the plants were transferred to these conditions the better developed was the root system and the higher was the number of internodes after 75 or 105 days from appearance of the first root (Fig. 1). However, after winter storage the grapevine cutting soaked in water and subjected to 30°C for 156 days were in worse condition than the control soaked in water and exposed to 20°C (non-stressed) (Fig. 2).

Drought stress at 20°C also remarkably slowed down the development of roots and cane internodes (Fig. 1). It was especially evident in the case of rooted cuttings subjected to these conditions when one or three primary roots were developed and when water content of growing media was the lowest (18% H_2O v/v). Lower water content of growing media and earlier subjection of the cuttings to these conditions resulted in the less developed root system. The most adverse effect of drought conditions on the number of internodes was observed after 156 days of exposure to drought stress (20°C, 18% H_2O) and then after storage period in a greenhouse over winter time (Fig. 2). The rooted and stressed cuttings had on average almost two internodes fewer than those kept at optimal temperatures. However, stressful conditions hardly influenced the number of canes (data not shown). It is commonly known that drought stresses affect virtually every aspect of plant physiology and metabolism. Numerous changes that occur under these stresses have been widely documented. The effect of drought stress, similarly to chilling stress, is usually perceived as a decrease in photosynthesis and growth, and is associated with alterations in C and N metabolism (Osmond 1981).

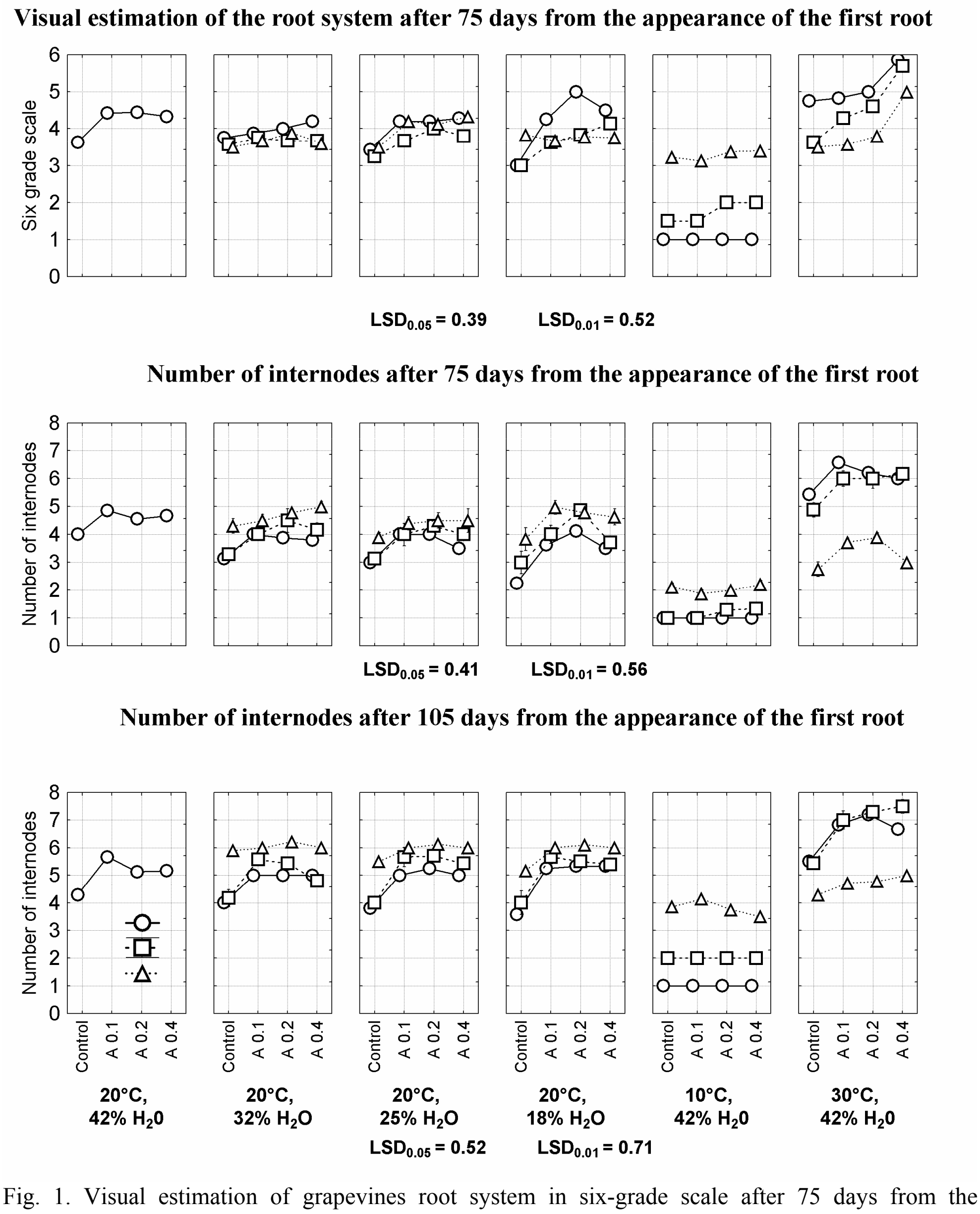

Fig. 1. Visual estimation of grapevines root system in six-grade scale after 75 days from the appearance of the first root and the number of internodes of rooted grapevine cuttings under optimal, drought and temperature stress conditions after 75 and 105 days from the appearance of the first root. The cuttings were soaked at 25°C for 24 h in Asahi SL at concentrations of 0 (control); 0.1; 0.2 and 0.4% and then kept under optimal (20°C; 42% H_2O), drought (20°C; 32, 25 and 18% H_2O) and temperature stress (10 and 30°C; 42% H_2O) conditions when first (-O-), third (-□-) or sixth (-Δ-) root appeared.

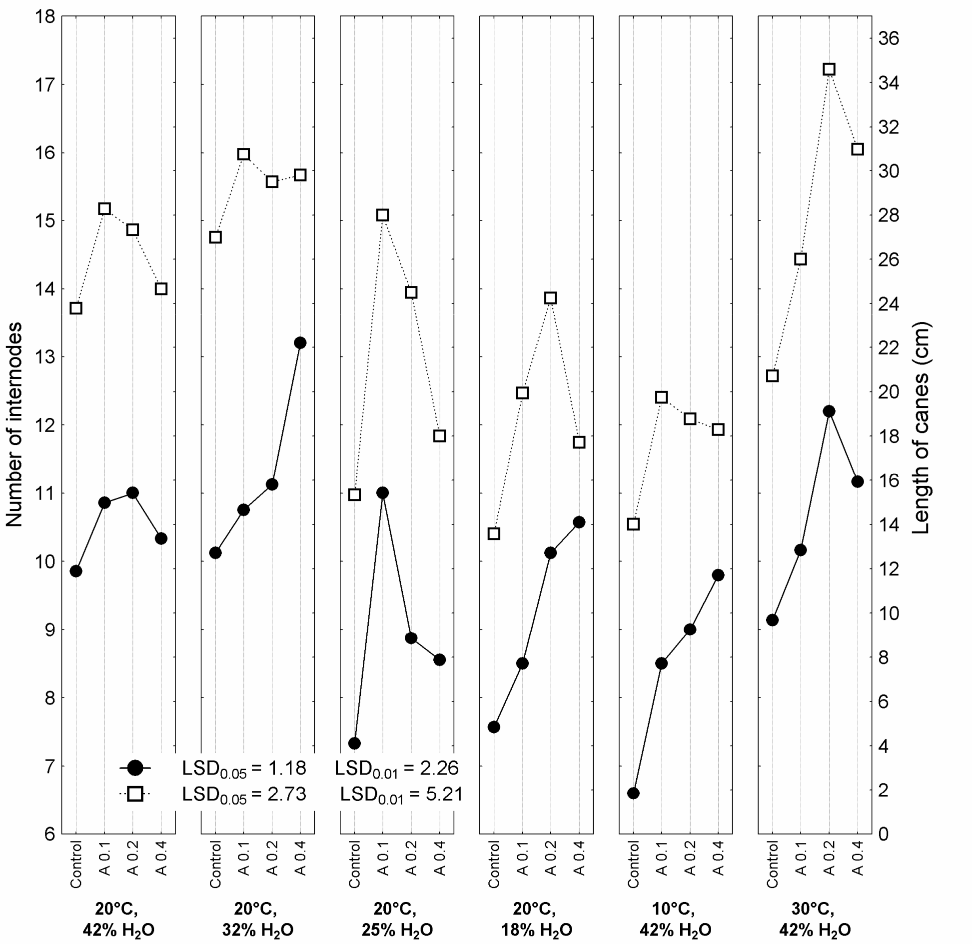

Fig. 2. Number of internodes (-●-) and length of rooted grapevine cuttings canes (-□-) under drought and temperature stress conditions applied when first root appeared after soaking of hardwood cuttings in Asahi SL at concentrations of 0 (control); 0.1; 0.2 and 0.4%. After 156 days from the beginning of rooting the cuttings were transferred to a greenhouse and stored over winter period.

Drought-related physiological changes such as decrease in leaf water content and stomatal closure result in limited CO_2 availability and the channeling of reducing equivalents to the production of active oxygen species rather than to CO_2 fixation (Osmond 1981). Oxidative damage of important molecules occurs as a result of the imbalance between production of reactive oxygen species (ROS) of reduced O_2 (i.e. superoxide radical [$^{\cdot}O^{2-}$], hydrogen peroxide [H_2O_2], and hydroxyl radical [$^{\cdot}OH^{-}$] and antioxidant defenses (Iturbe-Ormaetxe et al. 1998).

The experiment carried out showed that resistance of the rooted cuttings and developing plants to the drought and thermal stress can be increased by Asahi SL application. Its effectiveness depended on the temperature, stage of rooting and plant development during which the plants were exposed to drought stress as well as the concentration of the biostimulator. The most profitable effects of Asahi SL application were observed when grapevine possessing one developed primary root was exposed to the most severe drought stress (20°C, 18% H_2O – the growing media moisture content under which plants die). The cuttings soaked in Asahi SL at the concentration of 0.2% and then stressed in these conditions, developed two internodes more 75 days after the appearance of the first primary root than the control one (soaked in water) (Fig. 1). This treatment was especially beneficial because it entirely eliminated the adverse effect of drought stress, increasing the number of internodes above the level obtained in non-stressed cuttings, soaked in water (Fig. 1). These results also suggest that due to Asahi SL application the plants conferred tolerance to severe drought stress conditions. These findings are in line with the experiments of Vasudeva et al. (1981) on coffee (*Coffea arabica* L.) and of Badawy et al. (1984) on chamomile (*Matricaria chamomilla* L.) in which this chemical increased both plant height and the number of branches. It has also been found that Asahi SL positively affected photosynthetic efficiency of cucumber and potato leaves (Mikos-Bielak and Michałek 1999). Positive Asahi SL treatment may also be attributed to the increased internal auxin (IAA) pool by inhibiting its decarboxylation or due to modulation in the turgor of cell wall altered by cell elasticity, as it is the mode of action of auxin (Djanaguiraman et al. 2004a). Djanaguiraman et al. (2005) have also indicated that the application of this chemical significantly increased antioxidant enzymes activity which may favour increased fruit set and yield. The advantageous effect of Asahi SL application was also analyzed in our previous experiments conducted on China aster plants (Górnik and Grzesik 2002). Asahi SL improved seed yield and quality which was mainly caused by the stimulation of flower pollination, followed by improved fertilization (data not published). Higher seed yield due to Asahi SL treatments was also observed in mung bean (Gurbaksh et al. 1981), cucumber (Camargo and Passos 1976), pea (Yadav et al. 1992) and winter maize (Singh et al. 1987). Blum et al. (1988) have reported that when the phenols (the major compound of Asahi SL) were added to soil, the stimulation of bacterial and fungal populations growth was observed, as well as total soil respiration. This is because of microorganisms which are capable of polymerizing phenolic acids through the activity of the enzyme polyphenol oxidases and peroxidases.

The treatment with Asahi SL in concentrations of 0.1, 0.2 and 0.4% also improves grapevine rooting at 20°C and 30°C and under drought stress conditions (Fig. 1). Less advantageous influence of Asahi SL was observed when stress was applied later i.e. when rooted cuttings possessed 3 or 6 primary roots. Also, Asahi SL did not improve grapevine rooting at 10°C.

Soaking of cuttings in solutions of Asahi SL also positively affected grapevine growth during the growth in greenhouse in favorable conditions and after winter storage (Fig. 2). The observations indicated that no damage to grapevines occurred. Moreover, positive effects of Asahi SL application expressed as higher number of internodes and longer canes remained visible after this period. It has been reported that phenolic compounds of Asahi SL enhanced nitrate reductase activity in the leaves of chickpea (*Cider arietinum* L.) (Sharma et al. 1984). Nitrate reductase is one of the most important enzymes in the assimilation of nitrate, a predominant form of nitrogen available to the plant (Sharma et al. 1984). Therefore it seems that in the present study an increase in the number of internodes, length of canes and better rooting, in result of Asahi SL treatments during cuttings soaking, might have been caused by enhanced reductase activity.

Positive responses of grapevine grown under optimal or drought and temperature stress after Asahi SL application indicated that this biostimulator can be widely used in order to improve cuttings rooting and their further growth.

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CONCLUSIONS

- The results of the experiment demonstrated that Asahi SL positively affected the development and rooting of grapevine cuttings both under optimal and drought and temperature stress conditions.
- Asahi SL was more effective when cuttings at the beginning of rooting were exposed to the severe drought stress.
- Asahi SL decreased frost damages during winter storage and positively affected further growth in the following vegetation season.

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POPRAWA UKORZENIANIA ORAZ WZROSTU WINOROŚLI POD WPŁYWEM ASAHI SL W WARUNKACH OPTYMALNYCH ORAZ STRESU

Streszczenie: Celem badań była ocena wpływu preparatu Asahi SL (5-nitroguajakolan oraz orto i para nitrofenolany sodu) na ukorzenianie sadzonek winorośli w warunkach optymalnych (20°C; 42% H_2O), stresu suszy (20°C; 18, 25, 32% H_2O) oraz temperatury (10°C, 30°C; 42% H_2O). Sadzonki o długości 7 cm moczono w 25°C przez 24 godziny w roztworze preparatu Asahi SL (ASAHI) o stężeniu 0,1, 0,2 i 0,4% i ukorzeniano w wilgotnym podłożu (torf:piasek – 1:1) przez 20 dni w 30°C, a potem do chwili uformowania się pierwszych korzeni w 20°C. Następnie, ukorzeniające się sadzonki przetrzymywano w warunkach optymalnych lub poddano działaniu stresu termicznego lub suszy przez 156 dni od chwili pojawienia się korzeni. Stres suszy i temperatury niekorzystnie wpłynęły na ukorzenianie sadzonek oraz wzrost sadzonek po 75 i 105 dniach od chwili pojawienia się pierwszego korzenia podstawowego jak również w następnym roku po ich przetrzymywaniu w nieogrzewanej szklarni podczas zimy. Wpływ temperatury 10°C był najbardziej niekorzystny spośród wszystkich badanych warunków stresowych. Wzrost sadzonek został zahamowany, gdy poddano je oddziaływaniu 10°C w chwili pojawienia się jednego korzenia podstawowego. Asahi SL zastosowany w stężeniu 0,2% zwiększył tolerancję sadzonek winorośli na stres temperaturowy oraz suszy. Najbardziej korzystne było traktowanie preparatem, gdy sadzonki były poddane największemu stresowi suszy (20°C; 18% H_2O) w chwili pojawienia się pierwszego korzenia podstawowego. Korzystny efekt stosowania preparatu utrzymywał się po przechowywaniu sadzonek w czasie zimy, co wskazuje, że preparat ten nie tylko łagodzi uszkodzenia spowodowane podczas zimy, ale także stymuluje dalszy wzrost w następnym okresie wegetacyjnym.

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