

**SELECTIVITY OF SOIL HERBICIDES ON VIGOUR
AND PHYSIOLOGICAL RESPONSES OF GRAFTED
CUTTINGS OF RYAHOVO**

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Abstract

Between 2018 and 2021, at the IASS “Obraztsov Chiflik” experimental vine nursery – Ruse, trials were conducted to investigate the effect of pendimethalin (alone and in combination with clomazone) on the vegetative and physiological parameters of grafted vine cuttings. The study was carried out on leached chernozem using the table-grape cultivar Ryahovo grafted onto rootstock SO4 (*Berlandieri* × *Riparia*), and compared with an untreated control. Results show that pendimethalin applied alone or combined with clomazone had no statistically significant effect on the percentage of cuttings which sprouted, with values remaining from 92.5 to 96.3%. A slight suppression of shoot length was observed with the combined treatment, whereas pendimethalin alone did not produce meaningful deviations from the control. Analysis of growth rate indicates that the primary significant factor with proven influence was the year (climatic conditions), whereas the herbicides and their interaction were not statistically significant. Chlorophyll and carotenoid contents remained stable under pendimethalin treatment, which indicates lack of adverse effect on the photosynthetic apparatus. The pendimethalin + clomazone combination produced a mild to moderate phytotoxic effect manifested as a chlorotic halo on the first emerged leaves of the cuttings; this symptom resolved later and did not affect further development. The tested soil herbicides exhibited high selectivity towards grafted vine cuttings and can be determined as safe at the

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doses used and under the conditions. Climatic factors had a dominant influence on growth dynamics, while herbicide treatment did not cause substantial physiological disturbances.

Key words: vine, pendimethalin, clomazone, vegetative development, phytotoxicity, photosynthetic pigments

Introduction. The rooting stage is particularly important, as the physiological sensitivity of the grafted vine cuttings can lead to variations in growth, photosynthetic activity and the success rate of the grafting.

According to [1,2], the application of selective soil herbicides at recommended rates generally does not induce significant alterations in pigment composition; however, transient physiological disruptions may occur under certain conditions.

LIN et al. [3] and SREEKANTH et al. [4] established that herbicides can affect both shoot development and the physiological status of plants, varying with the active substance and environmental factors.

Climatic factors also play a major role in grapevine responses to herbicide treatment. Temperature and water regime can modify both herbicide activity and plant physiological responses [5]. It is therefore necessary to carry out trials under specific soil and climatic conditions in order to obtain reliable and applicable results.

The primary aim of this study was to evaluate the effects of two soil herbicide formulations containing pendimethalin alone and combined with clomazone on vegetative performance and physiological state of grafted vine cuttings.

Materials and methods. From 2018 to 2021, at the experimental vine nursery of the IASS “Obraztsov Chiflik” – Ruse, on leached chernozem soil, the effects of two soil herbicides were tested: 330 g l⁻¹ pendimethalin (Sharpen 330 EC) and a combination of 298 g l⁻¹ pendimethalin + 43 g l⁻¹ clomazone (Alcance Sync Tech), on vegetative traits of grafted vine cuttings. The trial used the table-grape cultivar Ryahovo grafted on SO4 rootstock (*Berlandieri* × *Riparia*).

Prior to rooting, stratified grafted cuttings were paraffined to two-thirds of their length after which they were rooted using the open-field technology adopted by IASS “Obraztsov Chiflik” [6].

Herbicides were applied with a knapsack sprayer fitted with flat-fan nozzle, at a spray volume of 200 l ha⁻¹. Application was carried out at the following active ingredient (a.i.) rates: 1.98 kg ha⁻¹ for pendimethalin and 0.85 kg ha⁻¹ for the pendimethalin and clomazone combination. The control plots were kept weed-free by hand-weeding. Selectivity was assessed on the 9-point EWRS scale [7].

To determine the herbicides effects on grafted cuttings, the following indicators were monitored: budburst dynamics; main shoot length; main shoot growth rate in dynamics; photosynthetic pigment content in leaves [8].

Statistical processing used one- and two-factor ANOVA with SPSS 19.0 [9]. Post-hoc Least Significant Difference (LSD) test was applied to determine signif-

icance among means while reliability of results was verified at appropriate significance levels.

Results and discussion. Application of pendimethalin alone did not cause phytotoxicity throughout the vegetation season and showed good selectivity to emerging grafted cuttings. Visual EWRS assessments 30 days after treatment revealed a chlorotic halo on the first leaves of the cuttings in the pendimethalin + clomazone variant. These symptoms subsided by day 60 and did not affect subsequent development. The chlorotic halo is attributable to the active substance clomazone, which under certain environmental stress conditions inhibits the biosynthesis of photosynthetic pigments.

The percentage of sprouted grafted cuttings in treated variants was lower or close to the one in the control (Table 1).

T a b l e 1

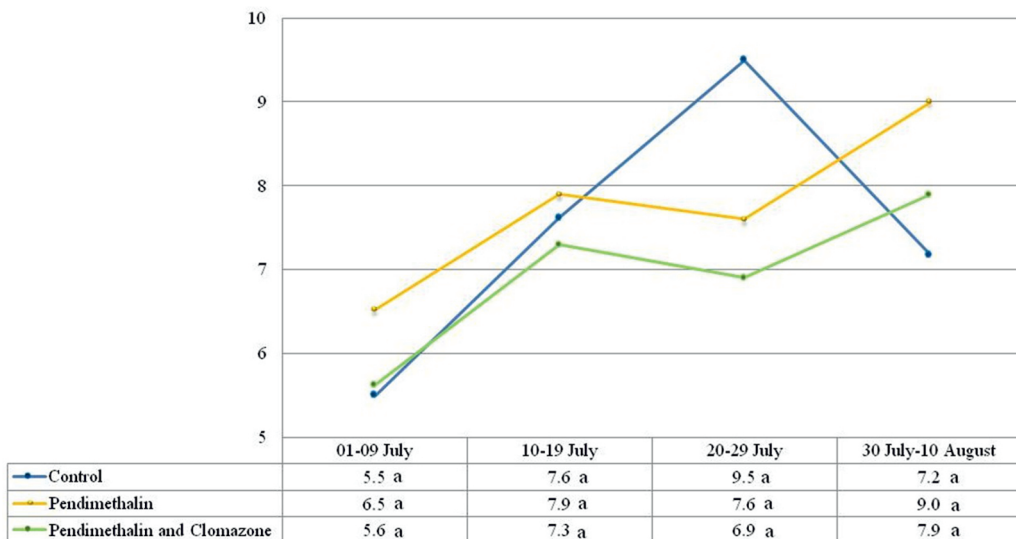
Percentage of sprouted grafted cuttings and main shoot length (cm) by ten-day periods, mean for 2018–2021

Variants	30th day	Differ- ence	40th day	Differ- ence	50th day	Differ- ence	60th day	Differ- ence	70th day	Differ- ence
Percentage sprouted cuttings										
Control	93.8 ^a	–	93.5 ^a	–	94.4 ^a	–	95.1 ^a	–	96.3 ^a	–
Pendimethalin	93.1 ^a	–0.8	93.3 ^a	–0.2	94.1 ^a	–0.3	94.9 ^a	–0.3	95.8 ^a	–0.5
Pendimethalin + clomazone	92.5 ^a	–1.3	92.8 ^a	–0.7	93.6 ^a	–0.8	94.1 ^a	–1.0	95.4 ^a	–0.9
Main shoot length (cm)										
Control	6.4 ^a	–	11.9 ^a	–	19.5 ^a	–	29.0 ^a	–	36.2 ^a	–
Pendimethalin	6.1 ^a	–0.3	11.7 ^a	–0.2	19.6 ^a	0.1	27.2 ^a	–1.8	36.3 ^a	0.1
Pendimethalin + clomazone	5.2 ^a	–1.2	11.7 ^a	–0.2	19.0 ^a	–0.5	25.9 ^a	–3.1	33.8 ^a	–2.4

Legend: Means followed by the same letters are not significantly different at $\alpha < 0.05$

Results indicate that application of pendimethalin (alone or combined with clomazone) had no statistically significant effect on percentage budburst. Throughout the observation period (days 30–70) values ranged narrowly (92.5–96.3%), with minimal differences versus the control (up to –1.3%). This confirms high tolerance of grafted vine cuttings to the herbicide treatments regarding their grafting and initial development.

Regarding main shoot length, a more noticeable tendency to growth suppression was seen with combined pendimethalin + clomazone. The greatest lag compared with control occurred on day 60 (–3.1 cm) and day 70 (–2.4 cm), suggesting a possible inhibitory effect on vegetative growth. Pendimethalin alone produced a weaker effect, differences being minimal; in some periods (day 50 and day 70) slight exceedance of control values was observed. Acceleration of growth inten-



Legend: Means of the same letters are not significantly different at $\alpha < 0.05$.

Fig. 1. Growth rate (mm day^{-1}), mean for 2018–2021

sity mid-season likely reflects root system strengthening and forming of increased assimilatory surface. Differences among treatments fell within experimental error and did not exceed $\text{LSD}_{0.05}$ critical values.

Figure 1 illustrates shoot growth rate, reflecting dynamics of cutting development. All variants showed initial acceleration followed by fluctuations later. The control maintained relatively high rates until mid-period, then declined. Combined treatment (pendimethalin + clomazone) produced lower, more even growth rates, indicating some inhibiting effect. Pendimethalin alone showed a stable trend with a gradual increase, reaching highest values at the end of the observation period – probably due to compensatory growth or better adaptation of the rooted grafts.

ANOVA results show that the factor year (Y) had a statistically significant effect on growth rate ($p \leq 0.01$), accounting for 91.76% of variation. Herbicide factor (B) and the $Y \times B$ interaction were not statistically significant. This confirms that differences in growth dynamics were driven by climatic conditions rather than the herbicides. Pendimethalin alone and combined with clomazone did not affect shoot growth. Observed differences were within experimental error and mainly attributable to year (Table 2).

These results are consistent with the research of other authors, who indicated that pendimethalin and S-metolachlor do not suppress main shoot development in vine cuttings [10–13]. PRODANOVA-MARINOVA [14] and HILL and MORETTI [15] report that pendimethalin effectively controls weeds in vine nurseries without adversely affecting development of rooted vine grafts.

T a b l e 2

Percentage contribution (%) of factors “year”, “herbicides” and their interaction to variation in shoot growth rate

Source of variation	Growth rate	Contribution
Year (Y)	91.76	***
Herbicides (B)	0.39	n.s.
Year $\times$ Herbicides (Y $\times$ B)	0.26	n.s.
Error	7.59	n.s.

Legend: *** – significant at $\alpha = 0.001$, n.s. – non-significant

Grape leaves contain a range of secondary metabolites, notably phenolics, chlorophylls and carotenoids. Their study is important as they serve as indicators of vigour, senescence, and stress caused by drought or herbicide use. Chlorophylls, located in chloroplasts of green organs, underpin photosynthesis by converting solar energy into chemical energy. Levels vary among cultivars under identical environmental conditions. The chlorophylls/carotenoids ratio is a sensitive marker to distinguish natural senescence from stress-induced changes.

Table 3 presents photosynthetic pigment content (chlorophyll *a*, chlorophyll *b*, and carotenoids *c*) in leaves of grafted cuttings of cv. Ryahovo. Averaged over the period, data show similar values between control and treated variants. Pendimethalin treatments, alone and in combination with clomazone, displayed a slight tendency toward increased chlorophyll *a*, chlorophyll *b*, and carotenoids *c* compared to the control.

The chlorophyll *a* : *b* ratio remained nearly unchanged across all variants, indicating maintenance of pigment balance and stability of the photosynthetic apparatus. Similarly, the (*a* + *b*) : *c* ratio showed no substantial shifts, confirming that treatments did not induce significant physiological disturbances.

T a b l e 3

Content and ratios of photosynthetic pigments (chlorophyll *a*, *b* and carotenoids) in leaves of grafted cuttings of cultivar Siyana (mg·100 g⁻¹), mean for 2018–2021

Variants	Photosynthetic pigments				
	<i>a</i>	<i>b</i>	<i>c</i>	<i>a</i> : <i>b</i>	(<i>a</i> + <i>b</i>) : <i>c</i>
Control	0.089 ^a	0.031 ^a	0.142 ^a	2.871 ^a	0.845 ^a
Pendimethalin	0.091 ^a	0.032 ^a	0.154 ^a	2.844 ^a	0.792 ^a
Pendimethalin + clomazone	0.097 ^a	0.034 ^a	0.160 ^a	2.853 ^a	0.819 ^a
$\alpha < 0.05$	0.023	0.008	0.042	1.00	0.269

Legend: Means followed by the same letters are not significantly different at $\alpha < 0.05$

These results are consistent with those of many authors reporting that soil herbicide application, including pendimethalin, does not lead to substantial changes in photosynthetic pigment content in crops when used at recommended rates. SHANER [16] and JING et al. [17] note that such herbicides primarily target emerging weeds and have limited impact on the photosynthetic apparatus. LI et al. [18] similarly report that chlorophyll *a* and *b* levels remain relatively stable in treated plants except at high doses or under stressful conditions. Studies in grapevine and other fruit crops by STANEVA and RANKOVA [19] also emphasise lack of significant change in chlorophyll *a* : *b* ratio, which indicates preserved photosynthetic activity and physiological resilience.

Conclusions. The climatic conditions throughout the study period determine the biometric parameters and growth dynamics of the grafted vine cuttings. Pendimethalin application (individually and combined with clomazone) demonstrated no statistically significant impact on the budburst percentage of grafted grapevine cuttings.

The pendimethalin-clomazone combination caused minor and temporary vegetative growth inhibition, which did not compromise the ultimate development of the grapevine nursery stock.

The evaluated herbicides did not provoke significant alterations in the photosynthetic apparatus. The retention of stable chlorophyll and carotenoid concentrations, along with their respective ratios, indicates sustained physiological activity and functional state of the foliar biomass.

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