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RESEARCH ARTICLE

Morphogenesis of mustard white under the action of the antigibberellic preparation chlormequat chloride

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Abstract

The research in question reveals the influence of the antigibberellic preparation-chlormequat chloride-on growth processes, morphogenesis, content of pigments, leaf mesostructure and functioning of the leaf apparatus of white mustard plants (*Sinapis alba* L.). It is established that CCS-750 reduces plant height by 4.09% overall. Inhibition of growth processes at the start of the growing season by the retardant led to the formation of more leaves throughout the whole period of vegetation. Similarly, leaf area increased significantly in the experimental version. The basis of such changes was the enlarged stem branching under the action of the preparation. Treatment of mustard plants in the budding phase caused optimization of the leaf anatomical structure; there was thickening of the assimilation parenchyma due to the expansion of its cells. The use of the quaternary ammonium salt of chloroquate chloride led to an increase in the amount of chlorophyll in the mustard leaves.

Keywords: Mustard white (*Sinapis alba* L.), retardant, chlormequat chloride, morphogenesis, leaf mesostructure

Introduction

The use of synthetic growth regulators of different physiological directions of action aimed at regulating morphogenesis and optimizing the production process is a principal modern direction of phytophysiology. Among the external plant growth regulators, the most effective is the use of growth inhibitors-retardants. The physiological action of these substances is based on blocking the synthesis or use of gibberellin in the plant (Rademacher 2016). These are synthetic preparations used to inhibit linear growth (Rogach et al. 2016; Kuryata and Polyvanyi 2018; Carvalho et al. 2016), accelerating the transition to the resting period (Kuryata 2009), and plant resistance to adverse environmental conditions (Barányiová and Klem 2016).

Among the exogenous growth inhibitors, the quaternary ammonium salt– chlormequat chlorid-is the most commonly used one. It has no carcinogenic properties, does not accumulate in the body, and is completely eliminated in two days. Therefore, it is widely used in agriculture. The impact of this and other growth inhibitors on the morphogenesis and productivity of agricultural crops still remains open to investigation (Kuryata and Golunova 2018; Kuryata and Kravets 2018; Kuryata and Khodanitska 2018, Kuryata et al. 2019).

In view of this, the purpose of this study was to investigate the effect of chlormequat chloride on the morphogenesis of white mustard plants.

Materials and Methods

Plants of mustard white (*Sinapis alba* L.) cv. Oslava were treated during the budding period with a solution of 0.5% chloromethochloride (by active ingredient) using an OP-2 sprayer, while the control plants were sprayed with tap water. The studies were conducted in Vinnytsia region in 2018-2019. The plots were randomly located, the size of the plots was 10 m^2, the repetition was five times.

Chlormequat chloride (CCC, α-chloroethyltrimethylammonium chloride)-[Cl-CH_2-$CH_2N(CH_3)^3]^+Cl^-$ is a white crystalline substance that decomposes at 245°C, insoluble in hydrocarbons, but soluble in water: the solubility is 74% at 20°C. The antigibberellic action of this preparation is associated with the inhibition of the activity of enta-cauren synthase in the formation of copolylyl pyrophosphate from geranylgeranyldiphosphate during the synthesis of gibberellins.

Morphometric parameters were determined every 10 days. The peculiarities of the leaf mesostructure of the control and experimental variants were studied at the end of the growing season on the leaves that had finished growing. The material for anatomical studies was preserved in a mixture consisting of equal parts of ethyl alcohol, water and glycerol and 1% formalin. The measurements of the leaf individual tissue were established under the microscope on cross sections using an eyepiece micrometer. The cell size measurements of columnar and spongy chlorenchyma were performed in macerated leaf tissues. A 5% solution of CH_3COOH in 2N HCl. Was used as a macerate. The quantitative content of the amount of chlorophylls (a+b) was fixed on a SF-16 spectrophotometer (AOAC, 2010).

The repetition of analytical studies is fivefold. The tables and charts present the average data over a two-year research. Statistical processing was carried out using the STATISTICA-6 computer program StatSoft Inc. The significance of the difference between the control and experiment indicators was determined by Student's t-test.

Results

According to the results obtained, the preparation chlormequat chloride caused a typical antagonizing effect (Kuryata and Golunova 2018; Kuryata and Polyvanyi 2018). During the growing season, the linear dimensions of the mustard plants were smaller than those of the retardant. In the waxy maturity phase, the mustard plants treated with the growth inhibitor were below the control by 4.09%. Nonetheless, there was a significant stem thickening, which increased the plant's resistance to lodging (Tab. 1.).

The assimilation apparatus plays the main role in the production process. It is determined by the area of the leaf surface, the number and duration of leaf life, the leaf mesostructural organization. The obtained experimental results reveal that the treatment with a solution of chloromethochloride led to an increase in the number of leaves on the mustard plants (Tab. 1.).

Table 1. Morphometric parameters of plants of *Sinapis alba* L. cv. Oslava under the action of chlormequat chloride (M ± m, n = 20).

Vegetation period	Parameters	Control variant	CCC 0,5%
blossom	Plant height, cm	66,63 ± 1,48	**58,61 ± 1,93
	Stem diameter, mm	4,81 ± 0,19	**5,97 ± 0,25
	Number of leaves, pcs	14,42 ± 0,46	**16,70 ± 0,52
	Leaf area, cm^2	368,96 ± 9,15	***496,28 ± 10,33
milk maturity	Plant height, cm	95,33 ± 1,39	*91,40 ± 1,21
	Stem diameter, mm	5,53 ± 0,26	***7,28 ± 0,31
	Number of leaves, pcs	19,07 ± 0,62	***22,78 ± ,56
	Leaf area, cm^2	622,68 ± 12,37	***830,45 ± 14,33
wax maturity	Plant height, cm	109,90 ± 1,26	*105,40 ± 1,56
	Stem diameter, mm	5,97 ± 0,22	***7,98 ± 0,34
	Number of leaves, pcs	26,63 ± 1,19	*30,58 ± 1,15
	Leaf area, cm^2	740,69 ± 17,07	***1003,97 ± 18,33

Note: * significant difference at p<0.05, **p<0.01, ***p<0.001

The number of leaves under the action of the preparation was greater than in the control variant throughout the whole growing season. Mustard culture is characterized by a short period of development; in the process of vegetation there is a rapid dieback of the lower leaves, which can affect the crop productivity. At the end of the growing season, the number of live leaves in the experimental variant was greater than in the control one. The establishment of the total area of leaves on a mustard plant indicates its increase relatively to the control variant throughout the whole observation period. Consequently, the increase in the number of leaves per plant inder the action of the antigibberellic preparation of chlormequat chloride caused an increase in the total leaf area. Such changes in the plants of the experimental variant occur due to the increase in the number of shoots of the second order as a result of srem branching and are -7,35 ± 0,28*** compared with the control variant, where their number is 5,20 ± 0,21 (the difference is significant at p<0.001). Enlarged stem branching under the action of antigibberellic preparations is a typical reaction of plants to the effect of retardants, similar changes have been observed by other researchers (Kuryata, 2009; Sousa Lima et al., 2016).

The photosynthetic process is determined by leaf anatomical and morphological features. The analysis of the mesostructural organization of white mustard leaves treated with a growth inhibitor, indicates a significant increase in leaf thickness because owing to the expansion of the assimilation parenchyma. The thickening of the layer of chlorenchyma under the action of chloromethchloride was due to the increase in the volume of the columnar cells and the size of the spongy parenchyma cells (Tab. 2). Accordingly, in the control variant, the thickness of the leaf was 206.34 ± 4.84 μm, when treated with 0.5% chloromethane chloride it turned out to be *286.59 ± 7.06 μm.

Table 2. Effect of chlormequat chloride on the functioning of the leaf apparatus of plants *Sinapis alba* L. cv. Oslava (M ± m, n=20).

Parameters	Control variant	CCC (05%)
Leaf thickness, μm	206,34 ± 4,84	***286,59 ± 7,06
Upper epidermis thickness μm	18,79 ± 0,49	***21,85 ± 0,56
Assimilation parenchyma thicknes, μm	167,31 ± 3,89	***245,44 ± 3,99
Lower epidermis thickness, μm	20,24 ± 0,46	19,30 ± 1,31
The cell length of spongy parenchyma, μm	23,03 ± 0,81	***37,04 ± 0,84
The cell width of spongy parenchyma, μm	16,48 ± 0,39	***22,49 ± 0,48
The volume of cells of columnar parenchyma, μm^3	2038,49 ± 101,92	**2489,86 ± 120,66
Amount of chlorophylls (a+b),%	0,52 ± 0,002	***0,67 ± 0,003

Note: *significant difference at p<0.05, **p<0.01, ***p<0.001

The content and ratio of photosynthetic pigments is an indicator of the effectiveness of the assimilation apparatus. The analysis of the experimental results suggests that the amount of chlorophylls in the mustard leaves increased remarkably under the action of chloromethochloride (Tab. 1 .). A similar effect of CCS-750 and EW-250 on *Solanum tuberosum* L. plants was found in the research paper (Rogach et al. 2016) and paclobutrazole on *Beta vulgaris* L. plants (Shevchuk et al. 2019).

Given the increase in the number and area of eaves in the experimental variant, as well as the increase in the content of the amount of chlorophylls, the optimization of mesostructure it can be stated that under the influence of this retardant the most optimal prerequisites for improving the production process of white mustard plants are formed.

Discussion

The results obtained demonstrate that the use of chlormequat chloride causes changes in anatomical and morphological parameters. The use of the retardant contributed to the formation of a more powerful leaf apparatus-an increase in the number of leaves and the area of leaf surface of white mustard plants was observed. The reason for such morphological changes was the increased stem branching under the action of the antigibberellic preparation. A stronger layer of photosynthetic tissue-chlorenchyma was formed in the mustard-white plants which were treated with chloride. Thickening of the assimilation parenchyma layer in the experimental version compared to the control one occurred due to the increase in the linear cell sizes of the columnar and spongy parenchyma.

Conclusion

Treatment of plants of mustard white with the retardant chlormequat chloride during the budding period caused changes in morphogenesis, leaf apparatus and leaf mesostructural organization. The use of this exogenous growth inhibitor led to an increase in the number of leaves forming a larger leaf surface. At the same time, chlorine chloride increased the thickness of the leaf blade due to the growth of chlorophyll cells and caused an increase in the content of chlorophylls in its cells.

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