# The effect of diethylamine derivative (3-DEC) on modification of barley and rape stem growth

## L. Miliuvienė,V. Gavelienė

Institute of Botany, Žaliųjų ežerų 49, LT-2021 Vilnius, Lithuania The present study is related to data on physiological activity of the quaternary ammonium salt analogue, diethylamine compound (3-DEC), the mechanism of its activity in modifying the growth of monocotyledonous (barley) and dicotylendonous (rape) plant stems. It has been detected that the use of the original compound 3-DEC, changing the amount of phytohormones and their relationship in the plant, results in a modification of barley and rape stem growth, structure, induction of mechanical tissue formation; the sclerenchyma and parenchyma ring width as well as vascular bundle number in them increase, rape primary cortex ring and stele width as well as vascular bundle number in it undergo changes.

**Key words**: rape, barley, diethylamine compound, 3-DEC, sclerenchyma, parenchyma, phytohormones

#### INTRODUCTION

The physiology of stem growth is closely related to the activity of the phytohormones auxin and gibberellin, while growth and development of the whole plant in individual stages of organogenesis are determined by a certain balance of phytohormones and their relationship both in the plant and its organs [1–4].

Spring barley, as well as spring rape which is a no less significant agricultural plant, cannot fully realize their productivity potential coded in the variety genome because of their inability to sustain a vertical position. With the aim of a purposeful regulation of plant structure by using an active diethylamine compound (3-DEC) and of elucidating similarities and differences of its effect on different plant sorts, we have studied its activity with respect to barley and rape plants. Taking into account that the stem of monocotyledonous plants, graminaceous in particular, according to some anatomical signs (lasting intercalary meristem) differs from the stem of dicotyledonous plants characterized by secondary thickening, we intended to reveal the general peculiarities of stem structure in these different sorts by using the original compound 3-DEC.

As is seen from the literature on the influence of the basic compound retardant CCC on the hormones GA and IAA, as well as their changes in the plant [1, 5], it is supposed that similar processes may occur under the effect of novel compounds as well; thus, we have studied the effect of compound 3-DEC on phytohormone balance.

#### MATERIALS AND METHODS

The object of the study was the spring barley (Hordeum distichum) variety 'Auksiniai-3' and the Danish spring rape (Brassica napus L. ssp. napus) variety 'Star', which are widespread in Lithuania and adapted to our conditions. Field trials have been performed at the Lithuanian Agricultural Institute in accordance with the routine technology of barley and rape growth, adopted for the middle Lithuanian zone. The retardant activity of compound 3-DEC was evaluated by stem height, its diameter, anatomical structure in barley and rape. Employing a light microscope, the width of sclerenchyma and parenchyma was measured in barley stem anatomical slides, as well as the number of vascular bundles was calculated. In rape stems the width of the primary cortex ring and stele were measured. The number of vascular bundles was calculated in the core [6].

Phytohormone balance in rooted hypocotyls of kidney bean was determined by high pressure chromatography. This analysis was performed according to a scheme presented in [7]. Six-day-old pod hypocotyls were kept in 3-DEC derivative 1·10<sup>-4</sup> M solutions for 48 h and afterwards for 10 days in Knapp solution at 14000 lx illumination with a photoperiod of 16 h

**Cytokinin (zeatin) detection.** Conditions of highpresure chromatography (Biotronic company): ultraviolet detector, wave length 268 nm, Column col. No. (4.6 mm ×250), Nuclosil 100-5-c 18. Solvent system: acetonytril: water: acetic acid 55:44:1. Elution rate 0.9 ml/min. Zeatin retention time 3 min. Zeatin identification was carried out by comparing the retention time of synthetic zeatin (Calbiochem) and native retention duration.

ABA detection. Conditions of high-pressure chromatography (Biotronic company): ultraviolet detector, wave length 254 nm, Ultropac Column Spherisorb ODS 2.3  $\mu$  (4.6  $\times$  100 mm). Eluted with 40% methanol, elution rate 0.9 ml/min, ABA retention time 13 min. ABA was identified by comparing the retention time of synthetic (Sigma) and native ABA.

IAA detection. Conditions of high-pressure chromatography (Biotronic company): detector, fluorescing RF-530 (Shimadzu), Em = 350 nm, Ex = 280 nm. Columns: Ultropac Column. Spherisorb ODS 2.3  $\mu$  (4.6  $\times$  100 mm). Eluted with 40% methanol, elution rate 0.3–0.9 ml/min, IAA retention time 5 min. IAA identification was performed by comparing the retention time of synthetic (Sigma) and native IAA.

Gibberellin detection. The biological activity of gibberellin was determined according to Frankland and Joring (1980) by the growth of Berlin lettuce hypocotyls. Gibberellins were detected quantitatively from the calibration curve of gibberellic acid (GA<sub>3</sub>).

#### RESULTS AND DISCUSION

The main function of retardants is to stabilize the growth of axial organs; however, retardants differ in their specific effect on individual plant sorts, even varieties. For comparison, we have studied the peculiarities of the effect of compound 3-DEC on stem growth in monocotyledonous (barley) and dicotyledonous (rape) plants with respect to anatomy, morphology and physiology.

The results of our study demonstrated that compound 3-DEC [8] was an active participant in the processes of cell and tissue differentiation, causing morphological changes in barley and rape stem. It is possible that because of stretching growth inhibition of stem cells their mechanical tissues are formed more intensively, resulting in a significant increase in the diameter of the lower internodes of barley and rape storeys. Experimental data demonstrated that these processes strongly depended on the time of the treatment, i.e. on the organogenesis of plants. The optimum time of exposure to the effect of 3-DEC in barley was found to be the tillering phase, while in rape it was the phase of 4th-5th leaf development. Comparing changes in monocotyledonous and dicotyledonous plant stems one should emphasize that, despite their different anatomical structure, the effect of the novel compound on stem formation was similar: it induced mechanical tissue formation, modifying barley and rape stem growth. Under the effect of 3-DEC, the barley sclerenchyma and parenchyma ring width and the number of vascular bundles in them were found to increase by 10–15%, primary cortex ring and stele width in rape as well as vascular bundle number in stele – by 15–16% (Fig. 1).

Taking into account that compound 3-DEC changes the stem structure in barley and rape, inhibiting the stem growth and stimulating mechanical tissue formation, we wanted to know if the effect of the compound manifested itself on the phytohormone level.

The function of retardants, particularly CCC, proves to be plant stem growth regulation; however, their effect on the level of plant endogenous phytohormones and inhibitors still is an object of discussion [9, 10].

We know from the literature and our previous experimental data that the inhibitory effect of retardant CCC is related to changes in GA and IAA balance in plants exposed to CCC and to the inhibition of cell elongation [1, 5, 11, 12]. There are rather strong arguments in favour of retardant CCC to be considered neither antigibberellin nor antiauxin, since its effect isn't specific with respect to one of these phytohormones. It can decrease the amount of both IAA and gibberellin-like substances [1, 5].

Our data prove that novel quaternary ammonium salt compounds such as 3-DEC, related to retardant CCC, do not exhibit any specific effect on separate

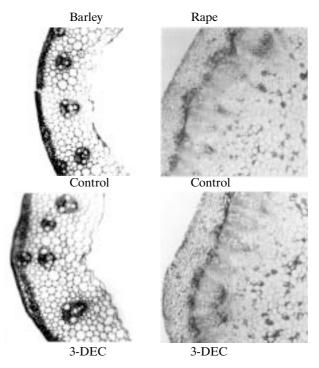


Fig. 1. The effect of compound 3-DEC on the anatomical structure of barley and rape stems: 1 - control, 2 - 3 - DEC

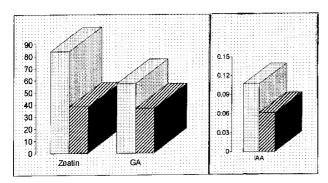


Fig. 2. The effect of compound 3-DEC on the amount of endogenous phytohormones (Zeatin, GA, IAA) in kidney been hypocotyls ( $\mu$ g/g) of dry mass: \_\_\_\_\_\_ – control, \_\_\_\_\_\_\_\_ – 3-DEC

phytohormones. The effect of 3-DEC caused changes in the metabolism of all phytohormones tested: gibberellin, auxin and cytokinins. Like in the control, only traces of ABA were detected under the effect of compound 3-DEC; the amount of IAA in the experimental variant decreased by 43%, of zeatin by 54%, of GA by 36% (Fig. 2).

We suppose that the effect of the compounds tested, inhibiting stem growth in monocotyledonous and dicotyledonous plants and forming a favourable stem structure able to protect the plants from lodging, can be related to changes in the amount of phytohormones and their ratio in barley and rape tissues.

#### References

1. Меркис АИ, Даргинавичене ЮВ, Новицкене ЛЛ. Физиология и биохимия культурных растений 1974; 69 (4): 358–64.

- 2. Муромцев ГС, Кокурин АВ, Павлова ЗН. Изв АН СССР. Сер. Биол. 1984; 5: 669–75.
- 3. Bellamine J, Penel C, Greppin H et al. Plant Growth Regulation 1998; 26: 191–4.
- 4. Bernier ZG. Ann Rev Plant Physiol 1998; 39: 175-219.
- 5. Меркис АИ, Милювене ЛИ. Физиология и биохимия культурных растений. 1975; 7 (2): 131–7.
- Kublickienė O. Histologinė technika ir praktinė histochemija. Vilnius, Mokslas, 1978: 24–36.
- 7. Miliuvienė L. Biologija 2000; 20: 280-2.
- 8. Miliuvienė L. Biologija 1999; 1: 25-7.
- 9. Судакова ЭМ. Докл РАХН 1994; 5: 14-6.
- 10. Зауралов ОА, Калмыкова ТС, Лукаткин АС. Биология 1997; 1: 80–4.
- 11. Schen HC, Zhou WJ, Xi HF et al. Acta Agric Univ Zhejinag 1991; 17: 423–26.
- 12. Zhou WJ, Xi HF. J Plant Growth Regul 1993; 12: 157–61.

#### L. Miliuvienė, V. Gavelienė

### DIETILAMINO DARINIO (3-DEC) POVEIKIS MIEŽIŲ IR RAPSŲ STIEBŲ AUGIMO MODIFIKAVIMUI

Santrauka

Straipsnyje pateikti duomenys apie ketvirtinių amonio druskų analogo, dietilamino darinio (3-DEC) fiziologinį aktyvumą, jo veikimo mechanizmą modifikuojant vienaskilčių (miežių) ir dviskilčių (rapsų) augalų stiebų augimą. Nustatyta, kad, naudojant originalų junginį 3-DEC, pasikeičiant fitohormonų kiekiui ir jų santykiui augale modifikuojamas miežių ir rapsų stiebų augimas, jų sandara, skatinamas mechaninių audinių formavimasis: padidėja miežių sklerenchimos ir parenchimos žiedų plotis bei indų plaušų kūlelių skaičius juose, taip pat pakinta rapsų pirminės žievės žiedo bei centrinio veleno plotis ir indų plaušų kūlelių skaičius centrianiame velene.