



## Article

# Sydnone Imines: A Novel Class of Plant Growth Regulators

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**Abstract:** An increase in the yield of the main cereal crops in the context of global climate changes requires additional impacts on plants. Natural and synthetic plant growth regulators (PGRs) are used to increase plant productivity and reduce the injury level caused by abiotic stressors. There is a growing need for novel highly effective plant growth stimulants to exhibit their effects at low doses and to not pose an environmental threat or injury to the crop quality. The derivatives of sydnone imine (SI), a mesoionic heterocycle possessing a 1,2,3-oxadiazole core, have been used as medicines until now but have not been used for agricultural applications. Some SI derivatives have recently been found to exhibit PGR properties. Herein, we report on the study of the PGR potential of nine SI derivatives bearing variable substituents at N(3), C(4), and N6 positions of the heterocycle designed to disclose the “molecular structure-PGR activity” relationship in this family. The SI derivatives were used in a wide concentration range ( $10^{-9}$ – $10^{-4}$  mol/L) for a pre-sowing treatment of winter wheat (*Triticum aestivum* L., two cultivars) and maize (*Zea mays* L., two hybrids) seeds in germinating experiments. All compounds were found to affect the growth of the axial organs of germinants, with the growth-stimulating or -inhibitory effect as well as its rate being considerably different for wheat and maize and, in many cases, also for roots and shoots. In addition, a pronounced concentration dependence of the effect was disclosed for many cases. The features of the molecular structure of SIs affecting their growth-regulating properties were elucidated. Compounds **4**, **6**, **7**, and **8**, which had exhibited a growth-promoting effect in germinating experiments, were used at appropriate concentrations for pot experiments on the same crops. For all compounds, the experiments showed a stimulating effect on the growth of roots (up to 80%), shoots (up to 112%), leaf area (up to 113%), fresh weights of roots (up to 83%), and aerial parts of the plants (up to 87%) or only on some of these parameters. The obtained results show a healthy outlook for the use of SI derivatives as promoting agents for improving the growth of cereal crop plants.

**Keywords:** plant growth regulators; sydnone imines; wheat; maize; seedling growth



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## 1. Introduction

The increase in the world's population, which reached eight billion people at the end of 2022 and has now (April 2023) surpassed 8.02 billion (<https://www.worldometers.info/world-population/> accessed on 26 April 2023), causes a serious need to increase food supplies. To prevent a food crisis, it is necessary to significantly increase the volume of agricultural production, especially grain crops [1,2]. By 2050, crop yields should increase by 25–70%, without significantly affecting the functioning of natural ecosystems [3]. However, since the 1960s, the rate of increase in the yield of major food crops (rice, wheat, and maize)

has slowed down [4]; moreover, the increase in yield must be achieved in a highly unstable climate because more and more extreme climatic events are expected in the future, which will have a negative impact on the growth and development of plants, ecosystems, and human living conditions [5].

Food security is threatened by global climate change [1,2]. Global processes on the Earth, both natural and anthropogenic, complicate the conditions for plant cultivation [6]. Climate change leads to increased problems for plant cultivation and population growth increases the need for crop production. Along with changing climatic conditions, it seems very difficult to provide enough food for the growing global population due to the steadily decreasing areas of arable land [1].

Climate change has increased the impact of abiotic stressors on plants, which is observed on a wide geographical scale, and poses a threat to the optimal growth and development of crops [6–8]. Therefore, plant resistance to abiotic stressors (high or low temperatures, drought, salinity, oxidative stress, etc.) is of great importance [7–9]. The effect of stress factors on vegetating plants significantly reduces the yield and quality of the crop [2]. Thus, increasing grain production is one of the main tasks of agriculture.

Cereals are sensitive to abiotic stresses; grain yields are greatly reduced if plants are exposed to unfavorable environmental factors for a long time [9]. Increasing the yield of major cereal crops under changing environmental conditions requires additional impacts on plant organisms. Various methods have been used to reduce the adverse effects of abiotic stressors. Among them, natural and synthetic plant growth regulators (PGRs) occupy a special place [10–12].

PGRs are preferred over other crop protection chemicals because they do not pose a great ecological threat or damage to crop quality [7]. It is especially important that PGRs act at low, or even ultra-low, doses, thus relieving pressure on the environment [11]. The number of publications reporting various aspects of PGR applications regarding agriculture is permanently increasing [10–16].

Both analogues of phytohormones [7,15] and compounds of other chemical natures [10,14,16] are used as PGRs. The vast majority of synthetic PGRs are either relatives of the endogenous phytohormones or the compounds acting as their antagonists that change the overall hormonal status of a plant [11]. The use of synthetic PGRs allows increasing the potential productivity and stress tolerance of crops [10].

Due to the growing problems of crop production, the need for novel highly effective crop-growth stimulants is permanently increasing. In this regard, the search for, and the screening of, more effective and ecological friendly PGRs is highly desirable.

In recent years, compounds of synthetic origins that exhibit growth-regulating and anti-stress properties have been intensively developed. Specifically, intensive studies have been initiated among sydnone imine derivatives, which are considered to be promising phytoeffectors. Sydnone imine (SI) is a sort of mesoionic heterocycle [17]; its derivatives exhibit a variety of physiological activities (vasodilatory, hypotensive, platelet anti-aggregative, etc.) and are the active substances of some drugs (Sydnophen, Sydnocarb, Molsidomine). To date, SI derivatives have not been used as agrochemicals. Recent studies have shown that many SIs exhibit plant-growth-stimulating [17–20], herbicidal [21], or herbicide antidotal properties [22,23], i.e., they behave as compounds that are promising for the rehabilitation of soils contaminated with phytotoxic pesticidal residues.

Winter wheat and maize are the worldwide leading cereal crops. They have a number of valuable food and fodder merits and are widely used in various fields. So, these crops became the species of choice for carrying out this present study. The aims of this study were to prepare the SI derivatives bearing variable substituents at the N(3), C(4), and N<sub>6</sub> positions of the heterocycle and to evaluate their PGR potential within a wide concentration range of winter wheat (*Triticum aestivum* L., cv. Mironovskaya 808 and cv. Moskovskaya 39) and maize (*Zea mays* L., ROSS 199 MV and Voronezhskij 158 SV hybrids).

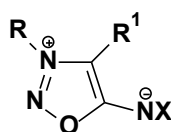
## 2. Materials and Methods

### 2.1. Plant Material

The experiments were carried out using seeds and young winter wheat (cv. Mironovskaya 808 and cv. Moskovskaya 39) and maize (hybrids ROSS 199 MV and Voronezhskij 158 SV) seedlings. The seeds were purchased from Mordovia Research Agricultural Institute (FARC North-East Branch) (Saransk, Russia).

### 2.2. Chemical Preparation

The SI derivatives 1–9 (Figure 1) were synthesized for the biological experiments. The molecular formulas of the obtained compounds were confirmed by NMR spectra. The purity of the compounds was proven by elemental analysis or by coincidence of the analytical data with those previously published in the literature for the compounds reported before.



- 1: R = CH<sub>3</sub>, R<sup>1</sup> = H, X = H · HCl
- 2: R = CH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>, R<sup>1</sup> = H, X = H · HCl
- 3: R = *N*-morpholyl, R<sup>1</sup> = CH(OH)C<sub>6</sub>H<sub>4</sub>CF<sub>3</sub>-4, X = C(O)OC(CH<sub>3</sub>)<sub>3</sub>
- 4: R = CH<sub>2</sub>CH<sub>3</sub>, R<sup>1</sup> = CH(OH)C<sub>6</sub>H<sub>4</sub>CF<sub>3</sub>-4, X = C(O)OC(CH<sub>3</sub>)<sub>3</sub>
- 5: R = CH(CH<sub>3</sub>)<sub>2</sub>, R<sup>1</sup> = CH(OH)C<sub>6</sub>H<sub>5</sub>, X = C(O)OC(CH<sub>3</sub>)<sub>3</sub>
- 6: R = CH(CH<sub>3</sub>)<sub>2</sub>, R<sup>1</sup> = CH(OH)C<sub>6</sub>H<sub>4</sub>CF<sub>3</sub>-4, X = H · HCl
- 7: R = CH(CH<sub>3</sub>)<sub>2</sub>, R<sup>1</sup> = CH(OH)C<sub>6</sub>H<sub>4</sub>CF<sub>3</sub>-4, X = C(O)OC(CH<sub>3</sub>)<sub>3</sub>
- 8: R = CH(CH<sub>3</sub>)<sub>2</sub>, R<sup>1</sup> = CH<sub>2</sub>OH, X = C(O)OC(CH<sub>3</sub>)<sub>3</sub>
- 9: R = CH(CH<sub>3</sub>)<sub>2</sub>, R<sup>1</sup> = CH<sub>2</sub>OH, X = H · HCl

**Figure 1.** Molecular formulas of the SI derivatives 1–9.

Chemical verification of the synthesized compounds was performed as follows. NMR spectra were recorded on a Bruker WM-400 spectrometer with an operating frequency of 400.13 MHz for <sup>1</sup>H, 100.62 MHz for <sup>13</sup>C, and 376 MHz for <sup>19</sup>F using CDCl<sub>3</sub> as the solvent. All <sup>1</sup>H and <sup>13</sup>C chemical shifts were correlated with the residual deuterated solvent signals. Trichlorofluoromethane was used as an internal standard in the <sup>19</sup>F spectra. Elemental analysis was performed on a Carlo Erba CE-1106 analyzer. Melting temperatures were determined in a quartz capillary on an Electrorthermal 1002 MEL-TEMP<sup>®</sup> and were not corrected. All reactions with organometallic compounds were performed in an atmosphere of dry argon in absolute solvents (THF was distilled over sodium benzophenone ketyl). Reactions were controlled by TLC on Sorbfil plates (Krasnodar, Russia) and Silufol UV-254 (Czech Republic). The visualization was performed by UV radiation (wavelength 254 nm) and in iodine vapor. Chromatographic separation was performed on columns filled with SiO<sub>2</sub> 40–60 μm. All commercially available reagents were purchased from ABCR (Germany) and used without further purification.

The SI derivatives 1 [24], 2 [25], 3, 4, 6, and 7 [26] were prepared by the methods described earlier. Compounds 5, 8, and 9 were synthesized as follows.

*N*<sub>6</sub>-*tert*-Butoxycarbonyl-4-( $\alpha$ -hydroxybenzyl)-3-isopropylsydnone imine (5). A 2.31 M hexane solution of *n*-BuLi (2.1 mL, 4.84 mmol) was added on stirring to a solution of *N*<sub>6</sub>-*tert*-butoxycarbonyl-3-isopropylsydnone imine (1.00 g (4.40 mmol) in 50 mL of THF at –90 °C. After stirring at –90 °C for 10 min, benzaldehyde (0.54 mL, 5.28 mmol) was added to the mixture. The cooling bath was removed and water (1 mL) was added. The solvent

was evaporated in vacuo and the products were separated by column chromatography on silica gel (CHCl<sub>3</sub>:ethyl acetate = 3:1). A crystallization from toluene/petroleum ether afforded 1.09 g of **5** (74%), m. p. 123–124 °C. <sup>1</sup>H NMR (CDCl<sub>3</sub>): 7.39 (d, *J* = 7.6 Hz, 2H) and 7.29 (d. of t., *J* = 18.8, 7.2 Hz, 3H, C<sub>6</sub>H<sub>5</sub>), 6.31 (s., 1H, CH(OH)), 4.92 (sept., *J* = 6.7 Hz, 1H, CH(CH<sub>3</sub>)<sub>2</sub>), 1.58 (d., *J* = 6.6 Hz, 3H) and 1.09 (d, *J* = 6.7 Hz, 3H, CH(CH<sub>3</sub>)<sub>2</sub>), 1.50 (s., 9H, C(CH<sub>3</sub>)<sub>3</sub>). <sup>13</sup>C NMR (CDCl<sub>3</sub>): 167.9, 159.4, 140.0, 128.6, 127.8, 125.4, 117.1, 79.2, 63.3, 56.9, 28.3, 22.1, 21.3. Calcd. for C<sub>17</sub>H<sub>23</sub>N<sub>3</sub>O<sub>4</sub> (%): C, 61.25; H, 6.95; N, 12.60. Found (%): C, 61.13; H, 6.84; N, 12.51.

*N*<sub>6</sub>-*tert*-Butoxycarbonyl-4-(hydroxymethyl)-3-isopropylsydnone imine (**8**). NaBH<sub>4</sub> (0.045 g, 1.18 mmol) was added on stirring at −20 °C to a solution of *N*<sub>6</sub>-*tert*-butoxycarbonyl-4-formyl-3-isopropylsydnone imine (0.60 g, 2.35 mmol) [19] in methanol (50 mL). The mixture was allowed to warm up to 20 °C (30 min), followed by an addition of acetic acid (0.067 mL, 1.18 mmol). The solvent was evaporated in vacuo and the products were separated by column chromatography on silica gel (CHCl<sub>3</sub>:ethyl acetate = 3:1). A crystallization from toluene/petroleum ether afforded 0.51 g of **8** (85%), m. p. 103–104 °C. <sup>1</sup>H NMR (CDCl<sub>3</sub>): 5.04 (sept., *J* = 6.6 Hz, 1H, CH(CH<sub>3</sub>)<sub>2</sub>), 4.62 (s., 2H, CH<sub>2</sub>OH), 1.71 (d., *J* = 6.7 Hz, 6H, CH(CH<sub>3</sub>)<sub>2</sub>), 1.49 (s., 9H, C(CH<sub>3</sub>)<sub>3</sub>), <sup>13</sup>C NMR (CDCl<sub>3</sub>): 170.1, 150.0, 114.3, 79.1, 56.5, 51.5, 28.2, 21.8. Calcd. for C<sub>11</sub>H<sub>19</sub>N<sub>3</sub>O<sub>4</sub> (%): C, 51.35; H, 7.44; N, 16.33. Found (%): C, 51.44; H 7.35; N, 16.39.

4-(Hydroxymethyl)-3-isopropylsydnone imine hydrochloride (**9**). To a solution of *N*<sub>6</sub>-*tert*-butoxycarbonyl-4-(hydroxymethyl)-3-isopropylsydnone imine (0.20 g, 0.78 mmol) in methanol (10 mL) was added 3.78M solution of hydrogen chloride (0.62 mL, 2.38 mmol) in dioxane. The mixture was refluxed for the disappearance of the starting material (30 min, TLC control). The solvent was evaporated in vacuo, the residue was dissolved in isopropyl alcohol (3 mL), and methyl *tert*-butyl ether (5 mL) was added. The precipitated crystals were filtered off and dried in vacuo to afford 0.136 g of **9** (83%), m. p. 128–129 °C. <sup>1</sup>H NMR (CDCl<sub>3</sub>): 9.86 (s., 2H, NH<sub>2</sub>), 6.00 (br. s., 5.31 (sept., *J* = 6.5 Hz, 1H, CH(CH<sub>3</sub>)<sub>2</sub>), 4.69 (s., 2H, CH<sub>2</sub>OH), 1.61 (d, *J* = 6.6 Hz, 6H, CH(CH<sub>3</sub>)<sub>2</sub>). <sup>13</sup>C NMR (CDCl<sub>3</sub>): 167.8, 113.7, 58.2, 49.5, 21.8. Calcd. for C<sub>6</sub>H<sub>12</sub>ClN<sub>3</sub>O<sub>2</sub> (%): C, 37.22; H, 6.25; Cl, 18.31; N, 21.70. Found (%): C, 37.32; H, 6.37; Cl, 18.51; N, 21.55.

### 2.3. Experimental Design

In the first part of this work, nine SI derivatives **1–9**, prepared as described above in Section 2.2, were used for the biological experiments as test compounds. The compounds were dissolved in distilled water or 1 mL of 75% ethanol and 10<sup>−4</sup> to 10<sup>−9</sup> mol/L solutions were exploited for an evaluation of their growth-regulating activity.

In the second part of the work, the seeds were treated with the solutions of the test compounds for 5 h, placed in vessels (50 seeds/vessel) charged with tap water, kept at room temperature for 48 h, and germinated for 7 days in a climate-controlled chamber at 21 °C at dark. Then, the germination and the length of the axial organs (root and shoot) of the seedlings were measured using standard measuring devices and the shoot to root length ratio (SRR) values were calculated. Seeds aged for the same time in tap water were used as the control. Each experiment was performed with three replications, each with 4 culture vessels with germinating seeds.

In the third part of this work, the SI derivatives **2, 4, 6, 7**, and **8** were used for seed treatment in the soil experiment. The concentrations of the compounds were chosen based on the results obtained in the previous steps: 10<sup>−6</sup> mol/L for **2**, 10<sup>−9</sup> mol/L for **4**, 10<sup>−6</sup> mol/L for **6**, 10<sup>−8</sup> mol/L for **7**, and 10<sup>−8</sup> or 10<sup>−7</sup> mol/L for **8**. SI-treated seeds, as well as water-treated seeds (control), were placed in the vessels filled with soil (1 kg of degraded chernozem) and kept under the following conditions: 22–25 °C, illumination by fluorescent lamps with a photon flux density of about 80 μmol m<sup>−2</sup> s<sup>−1</sup>, and a photoperiod of 16/8 h (day/night). The vessels were irrigated every 2 days and the soil moisture was kept at about 60 percent full humidity. After 3 weeks of growth, the plants were taken out of the soil, thoroughly washed from soil particles, and slightly dried on filter paper. The

leaves' surface area and the length of the axial organs (roots and shoots) of the seedlings, as well as the fresh and dry weight, were measured according to [27]. Each experiment was carried out in triplicate, each with 4 growth vessels with seedlings.

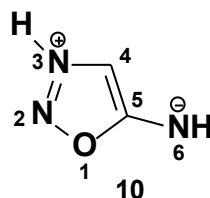
#### 2.4. Data Analysis

All measurements were performed as three independent experiments. Each parameter was assessed using 4 repetitions and at least 30 selected seedlings. For all measurements, averages and standard errors (SE) of the mean were calculated in Microsoft Excel 2007 and Statistica version 2.6. The figures and tables represent the means of all experimental data and their standard errors. The significance of differences between the water control and the SI treatments was evaluated by Student's *t*-test ( $p \leq 0.05$ ) and marked in the tables below with asterisks.

### 3. Results

#### 3.1. Design of the Sydnone Imine Derivatives 1–9

Sydnone imines are mesoionic heterocyclic compounds that are formally derived from the hypothetical molecule (10) sydnone imine itself.



It has been shown earlier that the substituents at N(3), C(4), and N(6) in SI molecules influence their growth-regulating activity. Salts 1 and 2 contain alkyl substituents at position N(3), with the substituents differing in the length of the alkyl chain and the presence of an oxygen heteroatom within the chain (compound 2). Compounds 3 and 4 differ in the type of the substituent at position N(3). Compounds 5 and 7 bear different aryl groups in the hydroxymethyl substituent at C(4). In addition, we reduced the aldehyde group in 4-formyl-3-isopropyl-N<sub>6</sub>-*tert*-butoxycarbonylsydnone imine by the action of NaBH<sub>4</sub> to afford alcohol 8. The protective *tert*-butoxycarbonyl group in the 4-hydroxymethyl compounds 7 and 8 were removed by the ordinary method [26] and hydrochlorides of the 4-hydroxymethyl substituted SIs 6 and 9 were obtained.

Thus, compounds 6 and 7 and 8 and 9 were the salts and the corresponding N<sub>6</sub>-*tert*-butoxycarbonyl derivatives. This approach allows preparations of both N<sub>6</sub>-protected and unprotected compounds, thus extending the range of the SI derivatives available for biological evaluation. SI salts are well soluble in water. The *tert*-butoxycarbonyl derivatives are more lipophilic, which may affect both the transport of these compounds in plants and their metabolism. Therefore, the derivatives representing both the salts of SIs (1, 2, 6, 9) and the *tert*-butoxycarbonyl substituted compounds (3–5, 7, and 8) were chosen for this study. The above combinations of the molecular structures could provide information regarding the structure–activity relationship in the SI series.

#### 3.2. Evaluation of the Growth-Stimulating and Growth-Inhibitory Activities of the SI Derivatives

A study of the growth-promoting and growth-inhibitory effects of the SI derivatives 1–9 on the germination of wheat seeds was carried out using five concentrations of the test compounds ( $10^{-4}$  to  $10^{-8}$  mol/L). No significant changes in the germination of wheat seeds were noted in the experiments. However, the notable effects of some compounds on the growth of axial organs of wheat seedlings (cv. Mironovskaya 808) were found (Table 1). Compounds 1, 3, and 5 showed growth-inhibitory activity, whereas compounds 4, 6, and 8 exhibited growth-promoting effects.



**Table 1.** The effect of SIs on the length of axial organs of wheat seedlings (cv. Mironovskaya 808, 7 days).

Compound	Concentration, mol/L	Root Length		Shoot Length		SRR
		mm	% of Control	mm	% of Control	
Control (H <sub>2</sub> O)	0	126.1 ± 3.1	100.0	117.5 ± 3.0	100.0	0.932
<b>1</b>	10 <sup>−4</sup>	98.9 ± 5.8 *	78.4	101.3 ± 4.2 *	86.2	1.024
	10 <sup>−5</sup>	94.9 ± 5.2 *	75.3	111.3 ± 3.6	94.7	1.173
	10 <sup>−6</sup>	94.1 ± 5.0 *	74.6	110.4 ± 3.3	94.0	1.173
	10 <sup>−7</sup>	87.2 ± 3.8 *	69.2	98.6 ± 4.8 *	83.9	1.131
	10 <sup>−8</sup>	104.7 ± 2.8 *	83.0	88.0 ± 2.9 *	74.9	0.840
<b>2</b>	10 <sup>−4</sup>	139.4 ± 4.0 *	110.5	115.5 ± 4.4	98.3	0.829
	10 <sup>−5</sup>	141.4 ± 3.2 *	112.1	100.8 ± 4.0 *	85.8	0.713
	10 <sup>−6</sup>	151.9 ± 4.4 *	120.5	115.7 ± 5.1	98.5	0.762
	10 <sup>−7</sup>	143.6 ± 4.2 *	113.9	112.5 ± 5.6	95.7	0.783
	10 <sup>−8</sup>	131.2 ± 4.7	104.0	117.1 ± 5.8	99.7	0.893
<b>3</b>	10 <sup>−4</sup>	87.0 ± 4.2 *	69.0	90.3 ± 4.4 *	76.9	1.038
	10 <sup>−5</sup>	81.5 ± 3.2 *	64.6	94.3 ± 4.4 *	80.3	1.157
	10 <sup>−6</sup>	82.4 ± 4.8 *	65.3	92.2 ± 3.0 *	78.5	1.119
	10 <sup>−7</sup>	86.2 ± 3.4 *	68.4	95.9 ± 4.8 *	81.6	1.113
	10 <sup>−8</sup>	101.6 ± 4.0 *	80.6	78.9 ± 5.4 *	67.1	0.777
<b>4</b>	10 <sup>−4</sup>	156.0 ± 3.9 *	123.7	133.9 ± 3.9 *	114.0	0.858
	10 <sup>−5</sup>	159.0 ± 4.2 *	126.1	137.0 ± 5.2 *	116.6	0.862
	10 <sup>−6</sup>	170.5 ± 4.0 *	135.2	153.1 ± 3.9 *	130.3	0.898
	10 <sup>−7</sup>	174.5 ± 6.3 *	138.4	162.2 ± 3.9 *	138.0	0.930
	10 <sup>−8</sup>	169.7 ± 5.8 *	134.6	157.7 ± 3.9 *	134.2	0.929
<b>5</b>	10 <sup>−4</sup>	103.6 ± 3.2 *	82.2	94.5 ± 6.8 *	80.4	0.912
	10 <sup>−5</sup>	103.0 ± 3.7 *	81.7	103.3 ± 6.6	87.9	1.003
	10 <sup>−6</sup>	95.0 ± 3.1 *	75.3	95.3 ± 5.7 *	81.1	1.003
	10 <sup>−7</sup>	85.0 ± 3.1 *	67.4	115.0 ± 6.0	97.8	1.353
	10 <sup>−8</sup>	92.0 ± 4.3 *	73.0	106.2 ± 6.0	90.4	1.154
<b>6</b>	10 <sup>−4</sup>	130.7 ± 4.4	103.6	149.7 ± 4.5 *	127.4	1.145
	10 <sup>−5</sup>	131.5 ± 5.4	104.3	164.5 ± 4.5 *	140.0	1.251
	10 <sup>−6</sup>	143.3 ± 6.1 *	113.6	181.3 ± 5.2 *	154.3	1.265
	10 <sup>−7</sup>	132.5 ± 4.7	105.1	140.5 ± 3.9 *	119.6	1.060
	10 <sup>−8</sup>	130.2 ± 5.2	103.3	130.2 ± 4.8 *	110.8	1.000
<b>7</b>	10 <sup>−4</sup>	131.4 ± 3.9	104.2	117.1 ± 5.2	99.7	0.891
	10 <sup>−5</sup>	136.4 ± 3.6 *	108.2	113.0 ± 5.2	96.2	0.828
	10 <sup>−6</sup>	140.8 ± 3.9 *	111.7	101.8 ± 5.2 *	86.6	0.723
	10 <sup>−7</sup>	144.2 ± 3.9 *	114.4	103.8 ± 4.5 *	88.3	0.720
	10 <sup>−8</sup>	157.5 ± 4.1 *	124.9	107.6 ± 5.2	91.6	0.683
<b>8</b>	10 <sup>−4</sup>	138.8 ± 5.0 *	110.1	118.4 ± 3.9	100.8	0.853
	10 <sup>−5</sup>	140.7 ± 3.8 *	111.6	120.7 ± 3.8	102.7	0.858
	10 <sup>−6</sup>	144.6 ± 3.8 *	114.7	128.9 ± 2.5 *	109.7	0.891
	10 <sup>−7</sup>	159.7 ± 5.4 *	126.6	137.1 ± 2.8 *	116.7	0.858
	10 <sup>−8</sup>	145.8 ± 4.0 *	115.6	132.8 ± 3.5 *	113.0	0.911
<b>9</b>	10 <sup>−4</sup>	122.1 ± 4.8	96.8	116.4 ± 3.9	99.1	0.953
	10 <sup>−5</sup>	132.0 ± 5.2	104.7	107.7 ± 4.0	91.7	0.816
	10 <sup>−6</sup>	135.0 ± 5.3	107.1	105.9 ± 2.6 *	90.1	0.784
	10 <sup>−7</sup>	130.0 ± 3.7	103.1	111.7 ± 3.1	95.1	0.859
	10 <sup>−8</sup>	123.8 ± 3.7	98.2	112.2 ± 3.6	95.5	0.906

The data highlighted in green indicate the growth-promoting effect. The data highlighted in yellow indicate growth inhibition. The bold font indicates the maximum values for the compound. \* The value significantly differs from the control at  $p = 0.05$  ( $n = 30$ ).

A more detailed analysis of the concentration dependence of the growth-inhibitory activity of SI derivatives showed the following regularities. Compound **1** at all concentrations decreased root growth by 17.0–30.8%. However, this compound had a weaker effect

on the shoot growth, with notable changes in growth (13.8–25.1%) being found only at concentrations of  $10^{-8}$ ,  $10^{-7}$ , and  $10^{-4}$  mol/L. Compound 3 inhibited the root and shoot growth at all concentrations, with these effects being very similar and more pronounced for roots (about 30%) than for shoots (about 20%) at all concentrations, excepting the lowest one, where the effects were found to be reversed quantitatively. Compound 5 was found to be similar to compound 1 in growth-inhibiting activity; however, the root growth was inhibited by compound 5 at all concentrations by 17.8–32.6%, while the shoot growth was inhibited only at concentrations of  $10^{-4}$  and  $10^{-6}$  mol/L by 19.6% and 18.9%, respectively.

An evaluation of the dependence of the growth-promoting effect of SIs on their concentrations showed that compound 4 at any concentration stimulated the growth of roots and shoots by 23.7% to 38.4% and 14.0 to 38.0%, respectively. Compound 6 had a stimulating effect (13.6%) on root growth only at a concentration of  $10^{-6}$  mol/L, whereas it promoted shoot growth at all concentrations by 10.8–54.3%. Compound 8 stimulated root growth (10.1–26.6%) at all concentrations, while promoted shoot growth (up to 16.7%) only at  $10^{-6}$  to  $10^{-8}$  mol/L concentrations.

Three other SI derivatives showed ambiguous effects on the growth of the axial organs of wheat. Compound 2 stimulated root growth at concentrations ranging from  $10^{-8}$  to  $10^{-5}$  mol/L, with a maximum (20.5% above the control) at  $10^{-6}$  mol/L. However, this compound slightly inhibited (14.4%) shoot growth at a concentration of  $10^{-5}$  mol/L; at lower concentrations, compound 2 showed only a growth-inhibition trend. Compound 7 behaved similarly, promoting root growth (up to 24.9%) at concentrations of  $10^{-5}$  to  $10^{-8}$  mol/L; however, it inhibited shoot growth at concentrations of  $10^{-6}$  and  $10^{-7}$  mol/L (11.7–13.4%). Finally, compound 9 showed a tendency to stimulate root growth (the differences with the control were not significant at  $p = 0.05$ ) and to inhibit shoot growth (9.9% at the concentration of  $10^{-6}$  mol/L).

It was interesting to evaluate the effect of the SI derivatives on the shoot-to-root length ratio (SRR). In most cases, the wheat seedlings (cv. Mironovskaya 808) showed an increase in SRR value under treatment with compounds 1 (except for a concentration of  $10^{-8}$  mol/L), 3 (except for a concentration of  $10^{-8}$  mol/L), 5 (except for a concentration of  $10^{-4}$  mol/L), and 6. On the contrary, a noticeable decrease in the SRR index was noted for seedlings grown from seeds treated with compounds 2, 4, 7, 8, and 9. Thus, the SI derivatives inhibiting the growth of the axial organs of wheat (cv. Mironovskaya 808) caused a notable increase in the SRR values. On the contrary, the growth-promoting compounds decreased this index.

Since the response of plants to the action of PGRs can differ not only among species but also among varieties of crop plants, we conducted similar experiments with wheat seeds cv. Moskovskaya 39. In addition, a different concentration range of the SI solutions ( $10^{-9}$ – $10^{-6}$  mol/L) was chosen for the treatment of the seeds. Compared with the results obtained with cv. Mironovskaya 808, both similarities and differences were revealed in the growth of the axial organs of wheat (Table 2). Specifically, compounds 4 and 8 were found to promote both the root and shoot growth of wheat cv. Moskovskaya 39. A promotion of the root but not shoot growth was noted for compounds 2, 7, and 9. On the contrary, compound 6 promoted shoot but not root growth. Lastly, compounds 1, 3, and 5 were disclosed to inhibit both root and shoot growth of cv. Moskovskaya 39.

**Table 2.** The effect of SIs on the length of axial organs of wheat seedlings (cv. Moskovskaya 39, 7 days).

Compound	Concentration, mol/L	Root Length		Shoot Length		SRR
		mm	% of Control	mm	% of Control	
Control (H <sub>2</sub> O)	0	100.6 ± 1.2	100.0	94.8 ± 2.7	100.0	0.942
<b>1</b>	10 <sup>−6</sup>	102.0 ± 4.2	101.4	87.6 ± 1.9	92.4	0.859
	10 <sup>−7</sup>	80.6 ± 2.3 *	80.1	69.0 ± 1.5 *	72.8	0.856
	10 <sup>−8</sup>	80.3 ± 3.4 *	79.8	53.3 ± 1.8 *	56.2	0.664
	10 <sup>−9</sup>	61.6 ± 2.1 *	61.2	64.8 ± 2.6 *	68.4	1.052
<b>2</b>	10 <sup>−6</sup>	<b>112.0 ± 2.7 *</b>	<b>111.3</b>	86.0 ± 1.7	90.7	0.768
	10 <sup>−7</sup>	98.3 ± 1.6	97.7	75.0 ± 1.2 *	79.1	0.763
	10 <sup>−8</sup>	83.0 ± 2.7 *	82.5	84.0 ± 2.4 *	88.6	1.012
	10 <sup>−9</sup>	<b>109.0 ± 1.7 *</b>	<b>108.3</b>	77.7 ± 2.7 *	82.0	0.713
<b>3</b>	10 <sup>−6</sup>	60.5 ± 1.9 *	60.1	56.3 ± 1.7 *	59.4	0.931
	10 <sup>−7</sup>	69.8 ± 2.3 *	69.4	72.5 ± 1.1 *	76.5	1.039
	10 <sup>−8</sup>	47.9 ± 1.2 *	47.6	41.3 ± 1.4 *	43.6	0.862
	10 <sup>−9</sup>	47.3 ± 1.0 *	47.0	41.8 ± 1.2 *	44.1	0.884
<b>4</b>	10 <sup>−6</sup>	97.6 ± 3.2	97.0	59.9 ± 3.6 *	63.2	0.614
	10 <sup>−7</sup>	<b>131.0 ± 1.8 *</b>	<b>130.2</b>	<b>128.8 ± 2.9 *</b>	<b>135.9</b>	0.983
	10 <sup>−8</sup>	<b>122.7 ± 1.0 *</b>	<b>122.0</b>	<b>105.6 ± 2.7 *</b>	111.4	0.861
	10 <sup>−9</sup>	<b>148.3 ± 2.9 *</b>	<b>147.4</b>	91.7 ± 1.6	96.7	0.618
<b>5</b>	10 <sup>−6</sup>	38.0 ± 1.3 *	37.8	38.8 ± 1.1 *	40.9	1.021
	10 <sup>−7</sup>	54.0 ± 2.0 *	53.7	49.1 ± 2.9 *	51.8	0.909
	10 <sup>−8</sup>	40.1 ± 2.1 *	39.9	38.0 ± 1.9 *	40.1	0.948
	10 <sup>−9</sup>	95.0 ± 1.9	94.4	76.9 ± 2.7 *	81.1	0.809
<b>6</b>	10 <sup>−6</sup>	109.5 ± 3.5	108.8	<b>149.0 ± 2.9 *</b>	<b>157.2</b>	1.361
	10 <sup>−7</sup>	77.0 ± 1.1 *	76.5	83.0 ± 3.9	87.6	1.078
	10 <sup>−8</sup>	69.5 ± 3.4 *	69.1	102.5 ± 3.0	108.1	1.475
	10 <sup>−9</sup>	103.0 ± 4.5	102.4	87.0 ± 1.7	91.8	0.845
<b>7</b>	10 <sup>−6</sup>	103.0 ± 2.0	102.4	76.0 ± 1.7 *	80.2	0.738
	10 <sup>−7</sup>	<b>114.6 ± 2.2 *</b>	<b>113.9</b>	81.8 ± 1.9 *	86.3	0.714
	10 <sup>−8</sup>	<b>121.7 ± 1.2 *</b>	<b>121.0</b>	98.3 ± 1.6	103.7	0.808
	10 <sup>−9</sup>	<b>120.7 ± 1.6 *</b>	<b>120.0</b>	<b>87.7 ± 1.1 *</b>	<b>92.5</b>	0.727
<b>8</b>	10 <sup>−6</sup>	<b>118.3 ± 2.9 *</b>	<b>117.6</b>	91.2 ± 1.2	96.2	0.771
	10 <sup>−7</sup>	<b>132.9 ± 1.6 *</b>	<b>132.1</b>	<b>104.8 ± 2.5 *</b>	<b>110.5</b>	0.789
	10 <sup>−8</sup>	107.9 ± 2.8	107.3	93.2 ± 1.8	98.3	0.864
	10 <sup>−9</sup>	<b>124.9 ± 1.1 *</b>	<b>124.2</b>	99.5 ± 2.6	105.0	0.797
<b>9</b>	10 <sup>−6</sup>	96.3 ± 1.1	95.7	84.6 ± 2.1 *	89.2	0.879
	10 <sup>−7</sup>	87.8 ± 3.4 *	87.3	66.0 ± 1.2 *	69.6	0.752
	10 <sup>−8</sup>	<b>110.0 ± 1.8 *</b>	<b>109.3</b>	96.8 ± 2.3	102.1	0.880
	10 <sup>−9</sup>	<b>114.2 ± 2.3 *</b>	<b>113.5</b>	94.8 ± 3.0	100.0	0.830

The data highlighted in green indicate the growth-promoting effect. The data highlighted in yellow indicate growth inhibition. The bold font indicates the maximum values for the compound. \* The value significantly differs from the control at  $p = 0.05$  ( $n = 30$ ).

To summarize, a unidirectional effect on both cultivars of wheat was found for compounds **4** and **8** (positive) and **1**, **3**, and **5** (negative). For the rest of the SI derivatives, the effect was found to be mixed (both positive and negative). As for the SRR index, a decrease in the SRR values for cv. Moskovskaya 39 was discovered in most of the studied concentrations of SI derivatives, excepting compounds **5** and **6**. Some of the studied compounds showed an ambiguous effect on the SRR index, including an increase in the SRR values in some variants of the experiment.

The concentration range of the SI solutions of 10<sup>−9</sup>–10<sup>−6</sup> mol/L was chosen for the treatment of maize seeds. Again, the diverse effects of SI derivatives on the growth of axial organs of maize seedlings (hybrid ROSS 199 MV) were discovered (Table 3). Growth inhibition was found for compounds **3**, **6**, and **9**. Compound **3** most strongly inhibited root (by 25.6–38%) and shoot growth (by 17.9–32.1%, except for a concentration of 10<sup>−8</sup>



mol/L). Compound **6** inhibited root growth by 18.8–23.0% (except for a concentration of  $10^{-8}$  mol/L) and shoot growth by 14.1–26.0% (except for a concentration of  $10^{-9}$  mol/L).

**Table 3.** The effect of SIs on the length of maize seedlings (hybrid ROSS 199 MV, 7 days).

Compound	Concentration, mol/L	Root Length		Shoot Length		SRR
		mm	% of Control	mm	% of Control	
Control (H <sub>2</sub> O)	0	114.8 ± 2.2	100.0	78.6 ± 2.7	100.0	0.685
<b>1</b>	$10^{-6}$	110.4 ± 4.2	96.2	70.7 ± 5.7	89.9	0.640
	$10^{-7}$	113.0 ± 4.6	98.4	72.3 ± 4.5	92.0	0.640
	$10^{-8}$	127.1 ± 6.8	110.7	70.9 ± 5.4	90.2	0.556
	$10^{-9}$	111.6 ± 4.2	97.2	82.4 ± 5.2	104.8	0.739
<b>2</b>	$10^{-6}$	<b>148.1 ± 5.4 *</b>	<b>129.0</b>	84.1 ± 3.4	107.0	0.568
	$10^{-7}$	<b>141.3 ± 3.2 *</b>	123.1	<b>89.9 ± 2.4 *</b>	114.4	0.635
	$10^{-8}$	<b>130.9 ± 5.4 *</b>	114.0	88.6 ± 4.8	112.7	0.672
	$10^{-9}$	<b>126.3 ± 3.4 *</b>	110.0	<b>94.5 ± 5.4 *</b>	<b>120.2</b>	0.747
<b>3</b>	$10^{-6}$	<b>85.1 ± 3.8 *</b>	74.1	<b>53.4 ± 3.4 *</b>	67.9	0.628
	$10^{-7}$	<b>74.6 ± 4.6 *</b>	65.0	<b>64.5 ± 2.2 *</b>	82.1	0.865
	$10^{-8}$	<b>71.2 ± 2.4 *</b>	62.0	73.1 ± 2.8	93.0	1.027
	$10^{-9}$	<b>80.3 ± 3.0 *</b>	69.9	<b>59.8 ± 2.4 *</b>	76.1	0.743
<b>4</b>	$10^{-6}$	<b>132.0 ± 6.4 *</b>	115.0	78.6 ± 3.6	100.0	0.595
	$10^{-7}$	<b>136.5 ± 3.6 *</b>	118.9	<b>90.5 ± 2.9 *</b>	115.1	0.662
	$10^{-8}$	<b>146.9 ± 4.0 *</b>	128.0	<b>104.3 ± 5.1 *</b>	<b>132.7</b>	0.711
	$10^{-9}$	<b>157.4 ± 5.8 *</b>	<b>137.1</b>	84.8 ± 3.2	107.9	0.540
<b>5</b>	$10^{-6}$	<b>103.2 ± 3.3 *</b>	89.9	<b>62.9 ± 2.2 *</b>	80.0	0.684
	$10^{-7}$	119.2 ± 4.0	103.8	<b>59.8 ± 2.9 *</b>	76.1	0.626
	$10^{-8}$	111.5 ± 4.2	97.1	<b>51.4 ± 4.9 *</b>	65.4	0.633
	$10^{-9}$	<b>128.8 ± 3.8 *</b>	112.2	<b>55.9 ± 5.7 *</b>	71.1	0.770
<b>6</b>	$10^{-6}$	<b>88.4 ± 3.5 *</b>	77.0	<b>60.4 ± 2.9 *</b>	76.8	0.640
	$10^{-7}$	<b>93.2 ± 2.2 *</b>	81.2	<b>58.2 ± 3.9 *</b>	74.0	0.643
	$10^{-8}$	106.8 ± 3.4	93.0	<b>67.5 ± 3.0 *</b>	85.9	0.538
	$10^{-9}$	<b>96.0 ± 4.5 *</b>	83.6	73.9 ± 3.4	94.0	0.556
<b>7</b>	$10^{-6}$	<b>158.4 ± 4.0 *</b>	137.6	<b>101.1 ± 2.7 *</b>	128.6	0.609
	$10^{-7}$	<b>160.1 ± 4.4 *</b>	139.5	<b>103.2 ± 2.9 *</b>	<b>131.3</b>	0.500
	$10^{-8}$	<b>187.1 ± 7.4 *</b>	<b>163.0</b>	<b>100.5 ± 2.6 *</b>	127.9	0.459
	$10^{-9}$	<b>183.5 ± 6.6 *</b>	159.8	<b>102.0 ± 2.3 *</b>	129.8	0.434
<b>8</b>	$10^{-6}$	126.3 ± 5.8	110.0	87.2 ± 3.6	110.9	0.690
	$10^{-7}$	<b>168.6 ± 6.2 *</b>	146.9	<b>96.7 ± 5.0 *</b>	123.0	0.573
	$10^{-8}$	<b>200.4 ± 8.6 *</b>	<b>174.6</b>	<b>103.0 ± 1.8 *</b>	<b>131.0</b>	0.513
	$10^{-9}$	<b>138.9 ± 4.1 *</b>	121.2	84.5 ± 2.6	107.5	0.605
<b>9</b>	$10^{-6}$	<b>77.8 ± 2.2 *</b>	67.8	<b>61.4 ± 4.2 *</b>	78.1	0.797
	$10^{-7}$	<b>90.7 ± 3.4 *</b>	79.0	<b>62.9 ± 3.6 *</b>	80.0	0.693
	$10^{-8}$	<b>89.4 ± 3.6 *</b>	77.9	70.0 ± 4.6	89.1	0.782
	$10^{-9}$	<b>99.6 ± 4.6 *</b>	86.8	<b>60.8 ± 6.0 *</b>	77.4	0.606

The data highlighted in green indicate the growth-promoting effect. The data highlighted in yellow indicate growth inhibition. The bold font indicates the maximum values for the compound. \* The value significantly differs from the control at  $p = 0.05$  ( $n = 30$ ).

SI derivatives **2**, **4**, **7**, and **8** exhibited growth-promoting activity. All compounds showed a pronounced activation of root growth (**2**: 10.0–29.0%; **3**: 15.0–37.1%; **7**: 37.6–67.0%; **8**: 21.2–74.6% (except for a concentration of  $10^{-6}$  mol/L)). However, their effects on shoot-growth activation were weaker and did not manifest at every concentration. Thus, compounds **2**, **4**, and **8** only significantly increased the shoot length at two concentrations, with the maximum growth enhancement being as high as 20.2, 32.7, and 31.0% of the control, respectively. Only compound **7** exhibited growth-stimulating activity at all concentrations, revealing no pronounced maximum. The remaining two compounds, **1** and **5**, either showed no growth stimulation of the axial organs or this activity was ambiguous with respect to the root and shoot growth.

As a rule, the treatment of maize seeds (ROSS 199 MB) with the SI derivatives resulted in a decrease in the SRR values. Interestingly, in the cases of an increase in the RSS index, this phenomenon took place mainly at the lowest (compounds 1–5) or, conversely, the highest concentrations of the corresponding SIs (compounds 8 and 9).

The experiments with maize hybrid Voronezhskij 158 SV showed only a poor effect of SI derivatives on plant growth (Table 4). In the majority of variants of the experiment, the difference with the water control was unreliable. A stimulating effect on the growth of axial organs was noted for the SI derivative 2 and, to a lesser extent, for compounds 5 (roots) and 4, 7, and 8 (shoots). The inhibitory effect on the growth of both roots and shoots was shown for compound 3. Compounds 1, 6, and 9 inhibited root growth, but the similar effect on shoots was poorly resolved.

**Table 4.** The effect of SIs on the length of axial organs of maize seedlings (hybrid Voronezhskij 158 SV, 7 days).

Compound	Concentration, mol/L	Root Length		Shoot Length		SRR
		mm	% of Control	mm	% of Control	
Control (H <sub>2</sub> O)	0	96.3 ± 4.3	100.0	77.2 ± 2.9	100.0	0.802
1	10 <sup>−6</sup>	63.6 ± 4.6 *	66.0	71.6 ± 5.7	92.7	1.126
	10 <sup>−7</sup>	81.6 ± 6.9	84.7	72.2 ± 4.5	93.5	0.885
	10 <sup>−8</sup>	94.3 ± 7.2	97.9	79.9 ± 5.4	103.5	0.847
	10 <sup>−9</sup>	90.6 ± 6.3	94.1	86.1 ± 7.8	111.5	0.950
2	10 <sup>−6</sup>	120.3 ± 8.1 *	124.9	90.4 ± 5.1 *	117.1	0.751
	10 <sup>−7</sup>	108.6 ± 4.8	112.8	94.8 ± 3.6 *	122.8	0.873
	10 <sup>−8</sup>	95.2 ± 8.1	98.9	104.2 ± 7.2 *	135.0	1.095
	10 <sup>−9</sup>	104.6 ± 5.1	108.6	97.5 ± 8.1 *	126.3	0.932
3	10 <sup>−6</sup>	71.1 ± 5.7 *	73.8	75.7 ± 5.1	98.1	1.065
	10 <sup>−7</sup>	71.2 ± 6.9 *	73.9	70.1 ± 3.3	90.8	0.985
	10 <sup>−8</sup>	70.6 ± 3.6	73.3	81.1 ± 4.2	105.1	1.149
	10 <sup>−9</sup>	103.0 ± 3.0	107.0	66.8 ± 3.6 *	86.5	0.649
4	10 <sup>−6</sup>	84.3 ± 5.6	87.5	84.3 ± 7.8	109.2	1.000
	10 <sup>−7</sup>	71.2 ± 5.4	73.9	108.1 ± 8.7 *	140.0	1.518
	10 <sup>−8</sup>	81.0 ± 3.0 *	84.1	89.1 ± 5.1 *	115.4	1.100
	10 <sup>−9</sup>	93.8 ± 8.7	97.4	72.8 ± 4.8	94.3	0.776
5	10 <sup>−6</sup>	102.0 ± 7.9	105.9	71.3 ± 3.3	92.4	0.699
	10 <sup>−7</sup>	100.9 ± 6	104.8	67.6 ± 4.7	87.6	0.670
	10 <sup>−8</sup>	102.9 ± 6.3	106.9	82.3 ± 5.7	106.6	0.800
	10 <sup>−9</sup>	112.5 ± 5.7 *	116.8	64.0 ± 4.1 *	82.9	0.569
6	10 <sup>−6</sup>	86.4 ± 3.9	89.7	86.0 ± 4.7	111.4	0.995
	10 <sup>−7</sup>	82.0 ± 3.3 *	85.2	67.3 ± 5.7	87.2	0.821
	10 <sup>−8</sup>	87.2 ± 6.2	90.6	73.1 ± 6	94.7	0.838
	10 <sup>−9</sup>	71.2 ± 4.5 *	73.9	77.0 ± 5.1	99.7	1.081
7	10 <sup>−6</sup>	77.9 ± 6.0 *	80.9	79.4 ± 5.1	102.8	1.019
	10 <sup>−7</sup>	86.2 ± 6.6	89.5	84.0 ± 5.7	108.8	0.974
	10 <sup>−8</sup>	102.2 ± 7.6	106.1	100.0 ± 6.8 *	129.5	0.978
	10 <sup>−9</sup>	69.3 ± 4.8 *	72.0	92.9 ± 5.3 *	120.3	1.341
8	10 <sup>−6</sup>	100.2 ± 8.7	104.0	81.9 ± 3.6	106.1	0.817
	10 <sup>−7</sup>	104.9 ± 7.8	108.9	82.9 ± 7.5	107.4	0.790
	10 <sup>−8</sup>	96.2 ± 8.4	99.9	88.2 ± 4.4 *	114.2	0.917
	10 <sup>−9</sup>	72.3 ± 3.3 *	75.1	85.3 ± 7.8	110.5	1.180
9	10 <sup>−6</sup>	99.3 ± 3.3	103.1	67.0 ± 6.3	86.8	0.675
	10 <sup>−7</sup>	82.7 ± 5.2 *	85.9	70.8 ± 3.6	91.7	0.856
	10 <sup>−8</sup>	100.9 ± 5.4	104.8	75.6 ± 6.9	97.9	0.749
	10 <sup>−9</sup>	97.9 ± 6.9	101.7	75.7 ± 4	98.1	0.773

The data highlighted in green indicate the growth-promoting effect. The data highlighted in yellow indicate growth inhibition. The bold font indicates the maximum values for the compound. \* The value significantly differs from the control at  $p = 0.05$  ( $n = 30$ ).

With maize (hybrid Voronezhskij 158 SV), an influence of the SI derivatives on the SRR index was found to be ambiguous. A notable decrease in the index value was triggered by compounds **1** (excepting a concentration of  $10^{-9}$  mol/L), **3**, **5**, and **9**. The treatment of the seeds with compound **2** (excepting a concentration of  $10^{-9}$  mol/L) resulted in an increase in the SRR value. Finally, compounds **4**, **6**, **7**, and **8** showed positive or negative effects on the SRR index depending on the concentration.

Taking into account the results described above, we selected some SI derivatives (compounds **4**, **5**, **7**, and **8**) to test their effect on plant growth for a longer time period. The seeds treated with the selected compounds were planted in pots filled with soil and the germinants were cultivated for 3 weeks.

The SI derivatives **4**, **5**, **7**, and **8** were found to differently effect the growth of wheat cv. Mironovskaya 808 (Table 5). The length of the aerial part of the plants significantly increased when treated with any of these compounds, whereas only compounds **7** and **8** promoted the growth of roots. Compound **7** appeared to be the only SI derivative to increase the leaf area compared with the control; the remaining compounds only exhibited a tendency to behave similarly. The raw weight of the plants (the sum of the roots and aerial parts) differed significantly from the control after pre-sowing treatment with all compounds, excepting for a high concentration of compound **8**. Among them, compounds **6** and **7** promoted the growth of roots and compounds **4**, **6**, and **8** promoted the same with the shoots. There were less significant differences in the dry weights compared with the control. The maximum dry weight of the roots was found for compounds **6** and **7**. The maximum dry weight of the shoots, as well as the total dry weight of the plants, was obtained after treatment with compounds **4**, **6**, and **7**. There were no significant differences in the dry weights between the variants of the experiment.

Slightly different results were obtained from the pre-sowing treatment of maize seeds with the same SI derivatives. With the ROSS 199 MB hybrid (Table 6), most of the SIs, excepting the lowest concentration of compound **8**, stimulated an elongation of the roots. However, the growth of the aerial part of the plants was significantly promoted only on the treatment of the seeds with compounds **4**, **6**, and **8** (at low concentration). Compounds **4**, **6**, and **7** enlarged the leaf area. All SI derivatives contributed to the increase in the root fresh weight, but only some compounds contributed to that of the shoots. The maximum increase in the fresh weight of plants was found for compound **7**. All SIs increased the dry weight of the roots and some performed the same with the aerial part of the plants. As a result, the dry weight of plants increased relative to the water control of the treatment with any of the studied SIs.

The treatment of maize seeds hybrid Voronezhskij 158 SV with the SI derivatives caused diverse effects on different growth parameters (Table 7). All compounds exhibited a tendency to increase the growth of both the roots and the aerial part of the plants, however the differences with the water control were not always significant. The maximal root length was obtained after treatment of the seeds with compound **8** at a concentration of  $10^{-7}$  mol/L, while the maximum value of the length of an aerial part was achieved after treatment with compound **2**. Both the leaf area and the root fresh weight were the highest in the case of treatment with compound **8** ( $10^{-8}$  mol/L), while the fresh weights of the aerial part of the plants and the whole plant were the highest after treatment with compound **4**. The maximum values of the dry weights of roots, shoots, and the whole plant were found for every SI derivative. These values significantly exceeded the water control, thus showing the high growth-stimulating effect of the SI derivatives on maize plants.

**Table 5.** The effect of SIs on the growth parameters of whet seedlings in pot experiments (cv. Mironovskaya 808, 21 days).

Compound (Concentration, mol/L)	Root Length, mm	Shoot Length, mm	S <sub>l</sub> , mm <sup>2</sup>	Fresh Weight, mg			Dry Weight				
				Root	Shoot	Σ Root + Shoot	Root, mg	Root, %	Shoot, mg	Shoot, %	Σ, mg
Control, H <sub>2</sub> O	152 ± 6.4	186 ± 7.3	1056 ± 85	47.2 ± 4.1	184.0 ± 11.5	231.2 ± 13.4	4.4 ± 0.3	9.3	17.4 ± 1.8	9.5	21.8 ± 1.5
4 (10 <sup>−9</sup> )	165 ± 7.4	286 ± 8.5 *	1285 ± 107	51.6 ± 5.9	<b>297.2 ± 24.9 *</b>	<b>348.8 ± 28.6 *</b>	5.0 ± 0.6	9.7	<b>28.8 ± 1.6 *</b>	9.7	33.8 ± 1.6 *
6 (10 <sup>−6</sup> )	168 ± 9.5	<b>394 ± 20.1 *</b>	1267 ± 72	<b>70.0 ± 9.5 *</b>	246.2 ± 26.0 *	316.2 ± 26.1 *	<b>7.2 ± 1.0 *</b>	10.3	28.6 ± 3.2 *	9.6	<b>35.8 ± 3.0 *</b>
7 (10 <sup>−8</sup> )	<b>273 ± 9.8 *</b>	320 ± 5.3 *	<b>1890 ± 144 *</b>	68.6 ± 8.1 *	228.0 ± 18.6	296.6 ± 22.4 *	6.2 ± 0.5 *	9.0	26.0 ± 2.4 *	9.2	32.2 ± 2.6 *
8 (10 <sup>−7</sup> )	175 ± 6.3 *	297 ± 12.7 *	1098 ± 76	52.4 ± 4.9	213.8 ± 22.2	266.2 ± 23.7	4.9 ± 0.5	9.4	19.4 ± 1.3	9.1	24.3 ± 1.4
8 (10 <sup>−8</sup> )	187 ± 8.3 *	323 ± 28.5 *	1391 ± 129	55.4 ± 6.2	244.2 ± 25.5 *	299.6 ± 26.8 *	5.2 ± 0.7	9.4	22.0 ± 2.9	9.0	27.2 ± 3.0

\* The value significantly differs from the control at  $p = 0.05$  ( $n = 10$ ). The bold font indicates the maximum values for the measured parameter.

**Table 6.** The effect of SIs on the growth parameters of maize seedlings in pot experiments (hybrid ROSS 199 MV, 21 days).

Compound (Concentration, mol/L)	Root Length, mm	Shoot Length, mm	S <sub>l</sub> , mm <sup>2</sup>	Fresh Weight, mg			Dry Weight				
				Root	Shoot	Σ Root + Shoot	Root, mg	Root, %	Shoot, mg	Shoot, %	Σ, mg
Control, H <sub>2</sub> O	129 ± 6.8	216 ± 9.1	1839 ± 98	145.1 ± 9.7	438.2 ± 24.5	583.3 ± 27.1	13.4 ± 0.8	9.2	33.3 ± 2.4	7.6	46.7 ± 2.2
4 (10 <sup>−9</sup> )	153 ± 7.2 *	248 ± 6.4 *	2326 ± 103 *	205.9 ± 18.6 *	557.0 ± 32.4 *	762.9 ± 38.0 *	21.4 ± 2.0 *	10.4	45.1 ± 1.6 *	8.1	66.5 ± 1.9 *
6 (10 <sup>−6</sup> )	168 ± 12.5 *	<b>394 ± 20.1 *</b>	<b>2489 ± 65 *</b>	210.0 ± 17.5 *	446.2 ± 26.0	556.2 ± 31.4	19.2 ± 1.0 *	9.1	38.4 ± 4.2	8.6	57.6 ± 3.4 *
7 (10 <sup>−8</sup> )	179 ± 17.3 *	221 ± 8.3	2122 ± 76 *	220.4 ± 15.4 *	<b>709.6 ± 20.4 *</b>	<b>930.0 ± 44.8 *</b>	21.6 ± 0.9 *	9.8	<b>58.9 ± 6.5 *</b>	8.3	<b>80.5 ± 7.4 *</b>
8 (10 <sup>−7</sup> )	<b>187 ± 13.9 *</b>	224 ± 7.2	2072 ± 138	224.0 ± 49.1 *	513.8 ± 22.2 *	737.8 ± 48.1	21.5 ± 1.5 *	9.6	46.8 ± 1.3 *	9.1	68.3 ± 2.8 *
8 (10 <sup>−8</sup> )	134 ± 8.5	320 ± 10.2 *	1944 ± 114	<b>254.2 ± 22.4 *</b>	444.2 ± 35.5	698.4 ± 36.9 *	<b>25.0 ± 1.7 *</b>	9.8	40.0 ± 3.9	9.0	65.0 ± 4.4 *

\* The value significantly differs from the control at  $p = 0.05$  ( $n = 10$ ). The bold font indicates the maximum values for the measured parameter.

**Table 7.** The effect of SIs on the growth parameters of maize seedlings in pot experiments (hybrid Voronezhskij 158 SV, 21 days).

Compound (Concentration, mol/L)	Root Length, mm	Shoot Length, mm	S <sub>l</sub> , mm <sup>2</sup>	Fresh Weight, mg			Dry Weight				
				Root	Shoot	Σ Root + Shoot	Root, mg	Root, %	Shoot, mg	Shoot, %	Σ, mg
Control, H <sub>2</sub> O	126 ± 6.1	267 ± 9.3	2182 ± 112	166.4 ± 12.3	465.0 ± 16.1	631.4 ± 17.4	15.1 ± 1.0	9.1	33.9 ± 1.6	7.3	49.0 ± 1.5
<b>2</b> (10 <sup>−6</sup> )	158 ± 14.8	<b>404 ± 16.3 *</b>	4038 ± 246 *	262.0 ± 28.0 *	607.8 ± 10.9 *	869.8 ± 27.1 *	22.6 ± 2.1 *	8.6	50.5 ± 4.9 *	8.3	73.1 ± 4.4 *
<b>4</b> (10 <sup>−9</sup> )	140 ± 11.7	375 ± 21.7 *	3691 ± 214 *	274.1 ± 26.8 *	<b>872.6 ± 67.4 *</b>	<b>1146.7 ± 72.8 *</b>	26.1 ± 0.8	9.5	70.2 ± 4.8 *	8.0	<b>96.3 ± 4.5 *</b>
<b>6</b> (10 <sup>−6</sup> )	195 ± 17.5 *	283 ± 9.1	2384 ± 126	196.5 ± 17.1	671.3 ± 8.9 *	807.8 ± 19.7 *	18.4 ± 1.4 *	9.4	61.8 ± 7.0 *	9.2	80.2 ± 6.4 *
<b>7</b> (10 <sup>−8</sup> )	151 ± 11.0	298 ± 16.8	4071 ± 317 *	162.3 ± 19.4	580.6 ± 39.9 *	742.9 ± 36.4 *	14.8 ± 1.3	9.1	48.4 ± 4.9 *	8.3	63.2 ± 5.1 *
<b>8</b> (10 <sup>−7</sup> )	<b>196 ± 19.0 *</b>	294 ± 22.7	3438 ± 143 *	226.0 ± 16.5 *	837.0 ± 15.4 *	1063.0 ± 17.6 *	19.8 ± 1.3 *	8.8	<b>72.0 ± 4.0 *</b>	8.6	91.8 ± 5.6 *
<b>8</b> (10 <sup>−8</sup> )	189 ± 18.2 *	340 ± 17.1 *	<b>4663 ± 332 *</b>	<b>304.0 ± 20.4 *</b>	669.4 ± 14.5 *	973.4 ± 22.1 *	<b>28.4 ± 3.4 *</b>	9.3	55.6 ± 5.1 *	8.3	84.0 ± 6.8 *

\* The value significantly differs from the control at  $p = 0.05$  ( $n = 10$ ). The bold font indicates the maximum values for the measured parameter.



#### 4. Discussion

It has been previously shown that some SI derivatives manifested themselves as PGRs exhibiting growth-stimulating or herbicidal activity [18,19,21], as well as herbicide antidotal properties [22,23]. Specifically, it has been disclosed that many SI derivatives used for the pre-treatment of seeds at doses of  $0.25\text{--}10\text{ g t}^{-1}$  of seeds can stimulate the growth of maize, sunflower, and winter wheat or, conversely, inhibit the growth of these crops, as well as relieve or completely eliminate the negative effect of metsulfuron-methyl (a widely used sulfonylurea herbicide) on maize growth.

In this present research, we utilized the SI derivatives 1–9 for winter wheat and maize seed treatment at significantly lower concentrations ( $10^{-8}\text{--}10^{-4}$  mol/L for winter wheat and  $10^{-9}\text{--}10^{-6}$  mol/L for maize). Compounds 4 and 8 were found to exhibit a growth-stimulating effect on both cereal species. Compound 3 solely showed a growth-inhibiting effect. The remaining six compounds exhibited either type of growth-regulating activity depending on the species. Specifically, compound 1 inhibited the growth of wheat but did not affect the maize growth. Compound 2 promoted the root growth in both wheat and maize but only the shoot growth in maize. A predominantly inhibitory effect was observed with compound 5, but one concentration ( $10^{-9}$  mol/L) caused a stimulation of the growth of maize roots. Compound 6 showed a promotion of wheat growth, more pronounced in shoots, but an inhibition of maize growth. Compound 7 showed the root growth stimulation in both crops, as well as the growth stimulation of maize shoots, but slightly inhibited the growth of wheat shoots. Lastly, compound 9 exhibited very weak growth-regulating activity on wheat and a significant growth-inhibitory effect on maize. Finally, the growth-regulating effects of SI derivatives demonstrated a significant concentration dependence and species specificity.

As for the structure–activity relationship among the SI derivatives, the results reported herein show that the substituents at every position of N(3), C(4), and N(6) may contribute to the PGR properties of the compounds. A morpholyl group at N(3) (compound 3) led to growth inhibition. On the contrary, the corresponding C(4)-unsubstituted 3-morpholyl substituted derivative promoted the growth of both roots and seedlings of maize [18]. One can see that the 3-isopropyl substituted SI derivative 7 behaves as a maize growth stimulant; meanwhile, its C(4) and N(6) unsubstituted counterparts were reported to be weak inhibitors of maize growth; an acylation of the last compound at N(6) and a substitution at C(4) contributed to the appearance of the growth-stimulating properties [18]. The presence of a longer alkyl or branched-alkyl group at N(3) provided the SI derivatives with growth-stimulating properties (compounds 4, 7, 8). The trend disclosed in this work for C(4)-substituted SI derivatives was opposite to one found earlier for the C(4)-unsubstituted derivatives [18]. In the case of C(4)-unsubstituted SIs, the lengthening of the alkyl chain at N(3), as well as its branching (an isopropyl group), contributed to a decrease in growth-stimulating properties. The presence of the N(6)-*tert*-butoxycarbonyl group on SIs (compounds 7 and 8) enhanced their growth-promoting effects compared with the corresponding hydrochloride salts (compounds 6 and 9). The 4- $\text{CF}_3$  substituent on the aryl group (compounds 4, 6, and 7) enhanced growth stimulation compared with the unsubstituted aryl derivative 5; the N(3)-morpholyl substituent (compound 3) reduced the stimulating contribution of the trifluoromethyl group (compound 7).

With wheat and maize plants, the pot experiments with the prolonged growing period showed a high growth-stimulating activity of SI derivatives selected based on the results obtained in the initial stages of this study. The different SIs were found to have different effects on various growth indicators and these effects were disclosed to have a pronounced species and variety specificity. Thus, with wheat plants, the maximum growth-stimulating activity indices were disclosed for compounds 4, 6, and 7, whereas compounds 6, 7, and 8 were the most active ones with maize hybrid ROSS 199 MV. Meanwhile, with hybrid Voronezhskij 158 SV, compounds 2, 4, and 8 behaved as the best stimulants.

At present, the conceivable mechanisms of the growth-regulating activity of SI derivatives are speculative. These compounds are known to be the exogenous donors of nitric

oxide (NO) [28], a molecule acting as a secondary messenger in both intercellular and intracellular signal transduction. In addition, superoxide anion ( $O_2^{\cdot-}$ ) is formed as a result of the metabolic transformations of SIs [28].

Both NO [28–30] and  $O_2^{\cdot-}$  [31] are involved in physiological processes in living organisms, including plants [32–35]. Specifically, NO molecule stimulates guanylate cyclase and subsequent accumulation of cycloguanosine monophosphate (cGMP) and participates in cell death and protection against pathogens [28–30].

In plants, NO acts as a homeostasis regulator [36,37]. It participates in the response to stress factors [38], including the action of heavy metals [39], salts [40], extreme temperatures [41], increased light [42], and pathogens [43]. Metalloproteins have been found to be biological targets for NO in plant organisms [30]. The advantages of sydnone imines as biologically active compounds are high resistance to hydrolysis and low toxicity [44]. In turn, superoxide anion is one of the reactive oxygen species that signals molecules in many cellular processes in plants, including their role as plant-growth regulators [31]. Thus, we believe that the physiological activity of the SI derivatives under this study is related to their ability to play the role of exogenous donors of NO and  $O_2^{\cdot-}$ . So, SIs are essentially metabolotropic compounds, whose biotransformation produces NO and  $O_2^{\cdot-}$  species, which are responsible for physiological effects. For example, the growth regulator ethephon acts in a similar way; entering the plant organism, it slowly degrades with the release of ethylene, thus influencing the hormonal ethylene status [14].

## 5. Conclusions

A study of the growth-regulating properties of nine SI derivatives revealed that all of them had an effect on the growth of the axial organs of winter wheat and maize. The trend (growth stimulation or inhibition) and rate of the effect of the SIs were found to considerably differ for wheat and maize and, in some cases, also for roots and shoots. A study of the dependence of the growth-regulating effect on the concentration of the test compounds exhibited in some cases a pronounced concentration dependence of the effect. Compounds 4 and 6–8, which had exhibited a stimulating effect in germinating experiments, were selected at appropriate concentrations for pot experiments on winter wheat and maize. In all variants of the pot experiments, the tested compounds had a stimulating effect on the growth of roots, shoots, leaf area, and fresh weight of roots and aerial parts of the plants or only on some of these parameters. There were no cases of growth inhibition. An analysis of the relationship between the molecular structures of the SI derivatives and their plant-growth regulating properties revealed that the growth-promoting effect of these compounds was influenced by the substituents at N(3), C(4), and N(6) in their molecules. The longer or branched alkyl substituents at N(3), the presence of a *tert*-butoxycarbonyl group at N(6), or a  $CF_3$  substituent at the para position of the aryl group in the substituent at C(4) contributed to the enhancement of the growth-promoting effect.

These promising compounds ought to be used in further experiments to clarify the ability of SI derivatives to antagonize the adverse effects of abiotic stressors and to elucidate the mechanisms of their actions.

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