THE VALORIZATION OF A RESIDUAL PRODUCT FROM THE SYNTHESIS OF BIS (DIISOPROPYLAMINE) CHLOROPHOSPHINE AS GROWTH REGULATOR

Eugenia FAGADAR-COSMA [1], Maria LAICHICI [1], Dana VLASCICI [2], Maria MERCEA [1], Liliana OSTOPOVICI [1]

[1] Romanian Academy- Institute of Chemistry Timisoara, Bd. M. Viteazul 24, 1900-Timisoara, Romania, e-mail: efagadar@yahoo.com
[2] West University of Timisoara, Faculty of Chemistry – Biology – Geography, Inorganic Chemistry Department, Pestalozzi 16, 1900 Timisoara, Romania, e-mail: vlascici@yahoo.com

Abstract:

The purpose of the present study is to create a link between the chemical research and the agriculture, taking into account the environmental care. The paper presents the biological tests made upon wheat (Alex variety) as monocotyledonous plant and cucumber (Cornichon cultivar) as dicotyledonous plant, with diisopropylamine hydrochloride, an undesired byproduct in the synthesis of bis(diisopropylamine)chlorophosphine, which finally proved useful as growth regulating agent, functioning similar to auxine. The auxinic effect activity was established with the Tsibulskaya-Vassilev general biotest, by using concentrations between 5-100 ppm and we can conclude that the product is stimulating the risogenesis process, especially on dicotyledonous plants.

Keywords:

bis(diisopropylamine)chlorophosphine, diisopropylamine hydrochloride, Tsibulskaya-Vassilev general biotest, risogenesis process, auxinic effect

1. INTRODUCTION

This work is promoting responsible research and application of chemistry science stepping up the science / agriculture dialogue.

A chemical substance can act as a plant hormone [1,2] when it shows specific biological activity in very low concentration and also must play a fundamental role in regulating physiological phenomena in vivo, such as: cell enlargement, vascular tissue differentiation, root initiation, gravitropic and phototropic responses, and apical dominance.

This paper presents the biological tests made upon wheat (monocotyledonous) and cucumber (dicotyledonous), with a substance named *diisopropylamine hydrochloride* which was an undesired byproduct in the synthesis of

bis(diisopropylamine)chlorophosphine and which finally proved to be useful as a growth regulating agent, functioning similar to auxine. The idea of searching diisopropylamine hydrochloride as a biological active substance, started from the previous literature studies [3] which proved that compounds with secondary amino groups have local anaesthetic activity, are not toxic, and above all, their use in agriculture contribute to the reduction of environmental pollution caused by necessary packaging for removing an unnecessary product.

Auxins are responsible in promoting cell elongation [4], a process that is required before differentiation of a cell. Auxins are able to do this by promoting the intake of water, increasing the elasticity of the cell to cope with the increase of water taken in by the cell.

Some of the other effects [5-9] that auxins are known to cause are: stimulates cell elongation, stimulates differentiation of phloem and xylem, stimulates root initiation on stem cuttings and lateral root development in tissue culture, delays leaf senescence, can inhibit or promote (via ethylene stimulation) leaf and fruit abscission, can induce fruit setting and growth in some plants, involved in assimilate movement toward auxin possibly by an effect on phloem transport, delays fruit ripening , stimulates growth of flower parts. A few of these effects are to be seen as results of our presented study.

On the other hand, plants require some essential elements to function properly, mainly carbon, oxygen and hydrogen and also small quantities of nitrogen, phosphorous, potassium and magnesium, as additional elements. For example, a lack of nitrogen causes an excessive growth of roots and a red leaf base. The compound we have been tasted has benefic effect from this point of view too.

2. EXPERIMENTAL

- Reactions, reagents and all operations were carried out with protection from atmospheric moisture, using Schlenk glassware and purging inert gas.
- Reagents: phosphorus trichloride and hexane were Merck reagents for synthesis; diisopropylamine is from Sigma Aldrich Division, Germany. All chemicals used were predried and distilled from appropriate drying agents [10].
- Melting points were determined on a Bőetius apparatus.
- ^{1}H-NMR spectrum was determined in DMSO-d_6 + $CDCl_3$ solutions with a Bruker Avance DRX 400 apparatus Chemical shifts (δ) are given in ppm downfield from internal TMS.
- IR spectra were determined on a SPECORD M80 JENA.
- Elemental analysis was carried out on a CARLO ERBA 1106 analyzer.

Procedure for synthesis of diisopropylamine hydrochloride

We obtain the diisopropylamine hydrochloride as a byproduct in our attempt to synthesize bis(diisopropylamino)chlorophosphine [11], according to equation (1):

$$PCl_3 + 4\ [(CH_3)_2CH]_2NH \rightarrow \{[(CH_3)_2CH]_2N\}_2PCl + 2\ [(CH_3)_2CH]_2NH_2^+\ Cl^- \qquad (1)$$

A solution of 10 mL (13.7g, 0.1 mol) of phosphorus trichloride in 20 mL hexane was added dropwise to a solution of 56 mL (40.4g, 0.4 mol) diisopropylamine in 180 mL hexane with continuous stirring and cooling in an ice bath. The reaction mixture

was allowed to warm to room temperature and then boiled under reflux for 30 hours. After cooling to room temperature, the reaction mixture was filtered and the precipitate, which is diisopropylamine hydrochloride, Mp: 212-214 ^oC, was washed and recrystallized from hexane. It was obtained 17g diisopropylamine hydrochloride, as byproduct. Concentration of the hexane filtrate gave 13.5 g of $(i\text{-}Pr_2N)_2PCl$, mp: 98-99^oC

Diisopropylamine hydrochloride: white crystals, m.p. 212-214 ºC; IR (KBr, cm^{-1}): 3432(ν_{NH}); 2976 (ν_{CH3}^{as}) ; 2912 (νCH); 2836 (ν_{CH3}^{s}); 1588 (δ_{NH}); 1462 (δ_{CH3}^{as}); 1376 (δ_{CH3}^{s}); 1168 (ν_{CN}). ^{1}H-NMR (DMSO-d_6 + $CDCl_3$): 3.29(m, 2 H); 1.1(dd, J=6.2 Hz, J=3.1Hz, 12 H); Elemental analysis for $C_6H_{16}NCl$ (137.5) (%): calc. C, 52.36; H, 11.63; found C, 52.61; H, 11.79.

The method for testing growth regulator activity

To establish the auxinic effect activity of diisopropylamine hydrochloride, the Tsibulskaya-Vassilev general biotest [12,13] was used.

For the determination of the biological activity of the diisopropylamine hydrochloride, as stimulator of the plant growth, laboratory tests were carried out on monocotyledonous - wheat caryopses (*Alex variety*) and on dicotyledonous - on cucumber (*Cornichon cultivar)* using the general Tsibulskaya-Vassilev biotest method comparatively with water control.

The concentrations we used were for monocotyledonous: 10 ppm, 50 ppm, and 100 ppm and for dicotyledonous 5 ppm, 10 ppm and 20 ppm. The seeds treated with bioactive compounds were held in Petri dishes on agar medium at 22^0 C during six days. After that, the biometrics measurements were carried out, watching of: the average height of plants, the average number of the roots for one plant, the average length of the roots and the dry substance on monocotyledonous and the average height of plants, the average length of principal roots, the average number of secondary roots and the dry substance for dicotyledonous.

The obtained data were calculated in percentage and compared to the water control. The results are presented in Tables and Figures 1 and 2. STATGRAPHICS PROGRAM carried out the statistical processing of the data.

3. RESULTS AND DISCUTIONS

From the data presented it could be observed that the average length of the plants increases with 3% at the concentration of 50 ppm. When the concentrations are equal with 10 and respectively 100 ppm, the registered values are below the values belonging to the water control.

By watching of the average length of the roots, we registered an increase of 15% when the concentration was 10 ppm and an increase of 8% for the concentration of 50 ppm. The concentration of 100 ppm proved to have inhibition effect (99%).

The average number of the roots on one plant increases with 12% at 10 ppm and for concentrations of 50 and 100 ppm is equal with the water control.

The dry substance reaches a significant distinct increase of 98% at the concentration of 10 ppm.

Table 1. *The growth regulating activity of diisopropylamine hydrochloride on wheat*

Variant	Average length of the seedling		Average length of the roots		Average number of roots			Dry substance		
	cm	%	cm	%	No.	Difference	%	g	Difference	%
Water control	8.13	100	7.26	100	3.76	N/A	100	0.0119	N/A	100
Diisopropylamine hydrochloride										
10 ppm	8.02	99	8.37	115	4.21	+0.45	112*	0.0236	+0.0117	198***
50 ppm	8.34	103	7.87	108	3.75	–0.01	100	0.0111	–0.0008	93
100 ppm	7.74	95	7.17	99	3.75	–0.01	100	0.0108	–0.0011	91
			DL	5%	= 0.32907			= 0.00046		
				1%	= 0.73113			= 0.00103		
				0.1%	= 1.11062			= 0.00239		

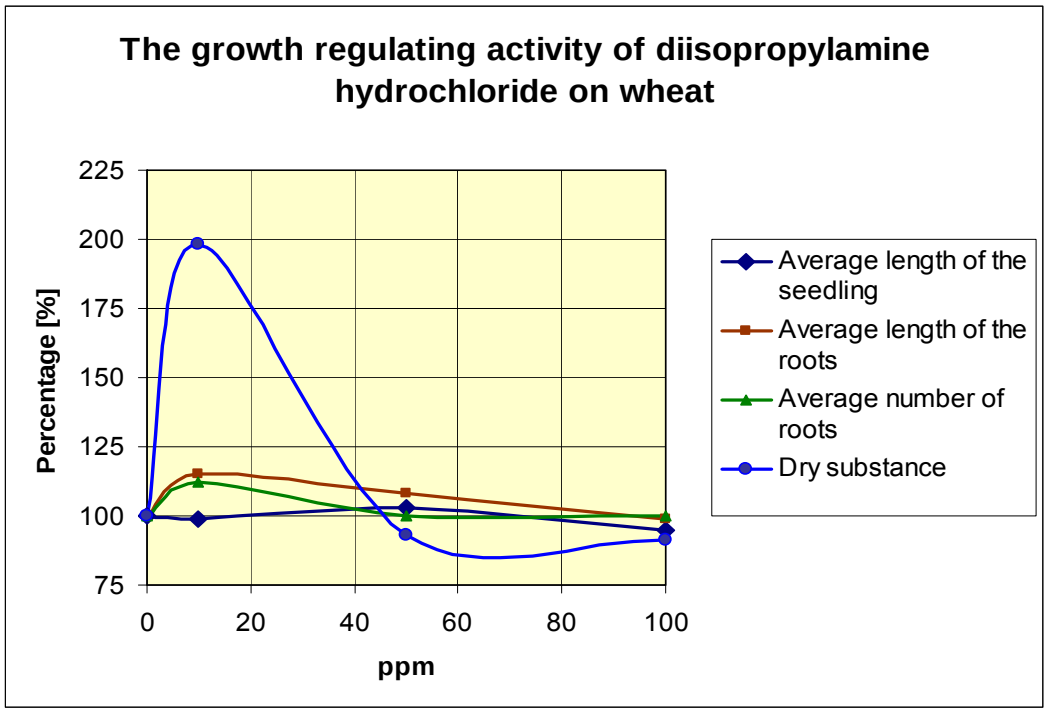

Fig. 1. *Biological activity of diisopropylamine hydrochloride on wheat*

Table 2. *The growth regulating activity of diisopropylamine hydrochloride on Cucumber*

Variant	Average length of the seedling			Average length of principal roots			Average number of secondary roots			Dry substance		
	cm	Diff.	%	cm	Diff.	%	No.	Diff.	%	g	Diff.	%
Water control	8.26	N/A	100	4.63	N/A	100	6.75	N/A	100	0.0156	N/A	100
Diisopropylamine hydrochloride												
5 ppm	9.56	+1.3	116 *	8.54	+3.91	184 ***	10.0	+3.25	148 ***	0.0256	+0.01	164 ***
10 ppm	11.35	+3.09	137 ***	5.5	+0.87	119 *	6.60	-0.15	98	0.0251	+0.0095	161 ***
20 ppm	10.13	+1.87	123 **	7.77	+3.14	168 ***	8.63	+1.88	128 **	0.0192	+0.0036	123
DL 5%	= 1.29442			= 0.64130			= 1.28157			= 0.00291		
1%	= 1.70145			= 2.03891			= 1.70034			= 0.00470		
0.1%	= 2.23141			= 2.61773			= 2.12419			= 0.00883		

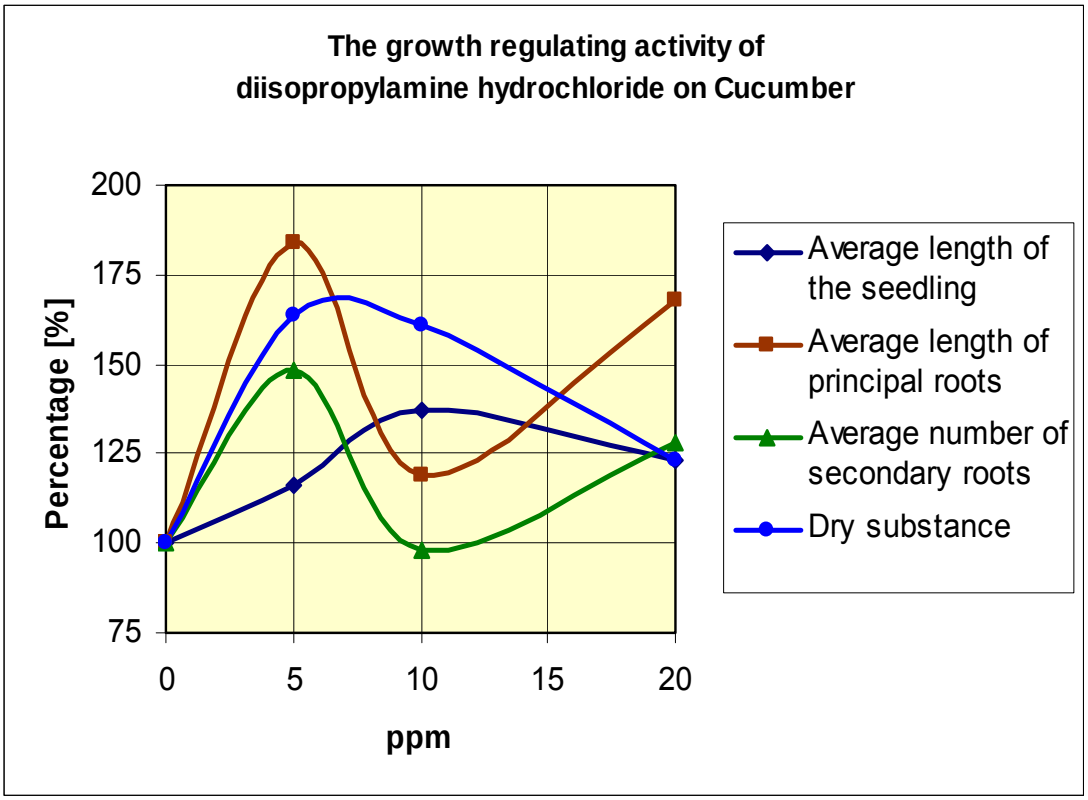

Fig. 2. *Biological activity of diisopropylamine hydrochloride on cucumber*

With respect to the dicotyledonous plant (cucumber) the average length of the seedling increases with 16% at the concentration of 5 ppm. At concentration of 10 ppm a significant distinct increase of 37% is registered, and when a concentration of 20 ppm was used the significant increase noticed, decreases to 23%.

The average length of the principal roots increase with 84% at 5 ppm, is highly with 19% at 10 ppm, and when we used a concentration of 20 ppm a distinct significant increase of 68% is noticed.

From the point of view of the average number of the secondary roots it is to be reported an increase of 48% at 5 ppm and an increase of 28% at a concentration of 20 ppm.

The statistically assured cumulative effect of protein substances is registered as significant distinct increase of 64% for 5 ppm and with an increase of 61% at 10 ppm.

4. CONCLUSIONS

By comparison of the obtained data we could remark the following:

The product we have tested is stimulating the risogenesis process of monocotyledonous plants with 12% and when referred to dicotyledonous with an increase between 28-48% with respect to the average number of roots, and an increase between 19-84% with respect to the average length of the roots.

The product has a stimulative effect on dicotyledonous seedlings when an increase of the growth of plants between 16-37% can be specified.

So we can say that the product has a significant biological activity, especially when it is applied on dicotyledonous plants.

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