AZAHETEROCYCLES – SYNTHESIS AND THEIR ACTIVITY IN STIMULATING GROWTH AND DEVELOPMENT OF WHEAT PLANT

AZAHETEROCICLII – SINTEZĂ ŞI ACTIVITATEA LOR IN STIMULAREA CREŞTERII ŞI DEZVOLTĂRII PLANTELOR DE GRÂU

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Abstract. *One of the strategies adopted for the synthesis of the pyridazine derivatives involves nitrogen ylides, as reactive species in organic chemistry. The reaction pathway involves, in the most frequent cases, a [3+2] dipolar cycloaddition of ylides to dipolarophiles (activated alkenes and alkynes). Pyridazine compounds have an intense biological activity, being used as anticancer, antituberculosis, antihypertensive, antimicrobial agents, or antifungal. Therefore, several studies to test the biologic effect of some new pyridazine derivatives have been done using simple experiments of wheat germination/ develop of the wheat plantlets. The results showed that all the tested compounds displayed a large variety of biological activities; germination percentage, shoot and root length, fresh weights, varied as a function of structure of each investigated compound.*

Key words: pyridazine compounds, biological activity, wheat, germination, total height.

Rezumat. *O metodă accesibilă în sinteza derivaţilor piridazinici constă în folosirea cicloimoniu ilidelor, copuşi reactivi în sintezele organice. Majoritatea acestor sinteze implică cicloadiţii 3+2 dipolare ale ilidelor cu diferiţi dipolarofili (alchene şi alchine activate). Derivaţii piridazinici obţinuţi prezintă o interesantă activitate biologică, fiind utilizaţi drept agenţi cu proprietăţi anticancerigene, antituberculoase, antihipertensive, antimicrobine sau antifungice. Prin urmare, au fost efectuate diferite studii cu privire la efectul biologic pe care îl prezintă derivaţii piridazinici în germinarea seminţelor de grâu/dezvoltarea plantelor de grâu. Rezultatele obţinute au demostrat o activitate biologică variată a acestor compuşi; procentul de germinare, lungimea şi greutatea plantelor, în stare proaspătă, a variat în funcţie de structura fiecărui compus investigat.*

Cuvinte cheie: derivaţi piridazinici, activitate biologică, grâu, germinare, înălţimea totală.

INTRODUCTION

Plants are hosts to a wide range of pathogens (as fungus, bacteria, viruses), wich are responsible for various infectious diseases, causing infection of the growth and crop destruction, having significant production losses worldwide. For

this reason, plants are considered good indicators of toxic and mutagenic effects of some chemical elements and compounds, being used successfully in the detection of harmful compounds (Granata, 1999; Iqbal et al., 1998).

Also, synthesis of compounds with broad biological activity and low toxicity is an important issue in the field of biochemistry research, because pathogen agents suffer continuous mutations and even more, preparatates used in different treatments generates over time the phenomenon of resistance and/or are leading to appearance of toxic effects.

According to recent studies from the literature, the azaheterocycles derivatives are successfully used in various branches of science (analytical chemistry (Urbano et al., 1984), polymer chemistry (Mangalagiu, 2001; Surpateanu et al., 1999), physics (Kawamura, 1990), science and technology of materials (Tanaka, 1997) etc), but one of the most important applications is as intermediates in the synthesis products with pharmacological properties (Allad, 1963).

Furthermore, the dates presented in the literature testify the phytotoxic action of the pyridazine derivatives (azaheterocycles compounds) and demonstrates the possibility to use them as potential biostimulant in growth and development of horticultural plants with potential practical applications as insecticides, herbicides and pesticides) (Druta et al., 2001; Irimia et al., 2003; Mangalagiu et al., 2005; Risca et al., 2006).

In this respect, we expected that at least a part of the byproducts obtained (bioisosters with pyridazinic structure) to present possible practical applications as biologically active compounds. For this reason we tested the antibacterial and antifungal action, (Butnariu (Tucaliuc) et al., 2007; Butnariu (Tucaliuc) et al., 2009) and the effect on germination and growth of these products plantelor (Butnariu (Tucaliuc) et al., 2008).

Therefore, this paper reports the biological activity of some pyridazine derivatives on wheat germination and seedling growth.

MATERIALS AND METHODS

In synthesis of pyridazine cycloadducts, first we obtained the corresponding cycloimmonium salts, using salt method proposed by Kröhnke (Kröhnke, 1935). Therefore, we realized N-alkylation reations, by treating pyridazine with ω-bromacetophenone-p-R-substituited. After that, an accessible method for obtaining pyridazine cycloadducts is using the ylides as intermediaries. The ylides are generated *in situ*, in classical heating or under microwaves energy, in alkaline medium (TEA – in liquid phase and KF/Aliquat 336 – in interphasic catalysis / PTC) (fig. 1).

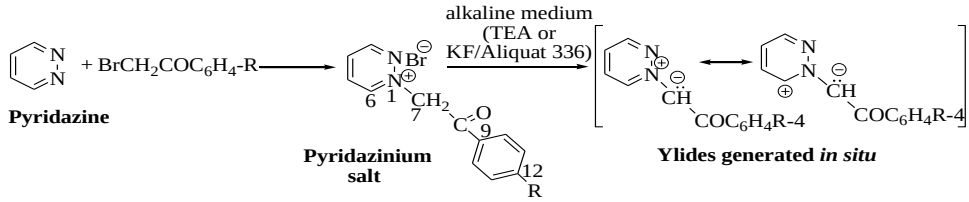

Fig. 1. Cycloimmonium salts used as precursors in the cycloaddition reaction.

By treating pyridazinium ylides, obtained *in situ*, with different activated dipolarophiles symmetrical and non- symmetrical substituted (ex. N-Phenylmaleimide, dimethyl maleate and dimethyl fumarate, acrylonitrile, dimethyl acetylendicarboxylate and ethyl propiolate) reactions occur as 3+2 dipolar cycloaddition. We obtain azabicycles derivatives (**3-8**), wich have a rapid systemic effect on plants and are active at very low concentration (fig.2).

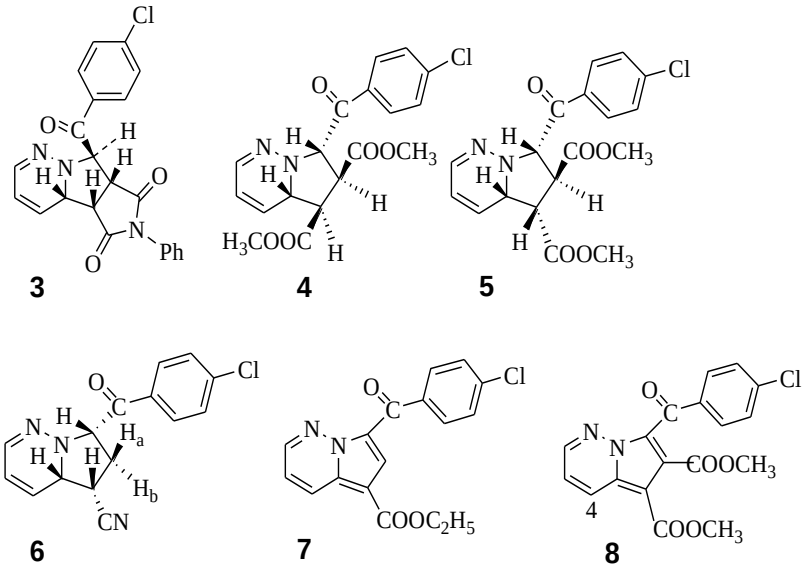

Fig. 2. Pyridazine derivatives

Germination tests were performed in a growth chamber Conviron MP4030 model G30 with programmed temperature, humidity and light. We used seed samples of wheat (*Triticum aestivum*), with specific weight 37.2 g/1000 seeds.

Thus, for this experiment, 50 seed samples of wheat were treated with 5 mL of each $5 \cdot 10^{-3}$ M solutions of pyridazine derivatives obtained in parallel with a redistilled water blank (**B**).

All the determinations were performed in triplicate or duplicate. Initially, the seed with analyzed solutions, were shaken in the tubes, at short time interval, for one hour.

Then, the seeds with their treatment solutions, were taken out and put into Petri dishes on double filter paper together. The seeds were periodically watered and the percent of germinated seeds were reported 3 days later (energy of germination, **EG**); respectively, 7 days later we reported the germination rate (**GR**). A seed with visible coleorhizae was considered germinated. After, the harvest of young wheat plants, from their seeds, was measured height (**H**, expressed as cm) and weight (**W**, expressed as grams).

RESULTS AND DISCUSSIONS

The data were validated using the Tukey test (Snedecor, 1994), with a probability of 95 %.

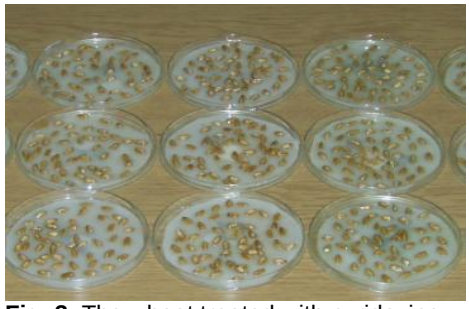

Fig. 3. The wheat treated with pyridazine derivatives in the first day of treatment.

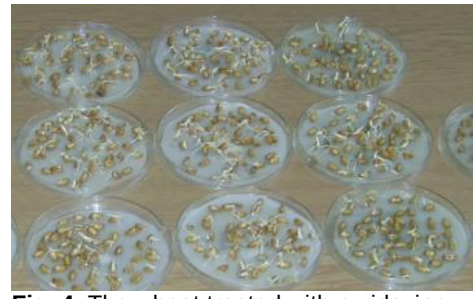

Fig. 4. The wheat treated with pyridazine derivatives in the 3rd day of treatment

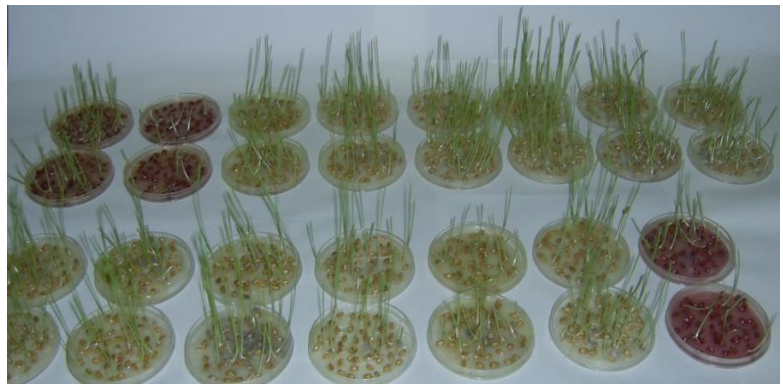

Fig. 5. The wheat treated with pyridazine derivatives in the 7rd day of treatment

The results showed that germination percentage, root length and weight of seedlings, fresh, varied as a function of the structure of each investigated compound.

Table 1

The effect of pyridazine derivatives on wheat germination (RG)

Comp.	Germination Rate (GR, %)	Number of plantlets	Comp.	Germination Rate (GR, %)	Number of plantlets
2	29 ± 4	9 ± 2	**6**	52 ± 5	23 ± 3
3	70 ± 4	25 ± 1	**7**	81 ± 3	34 ± 4
4	59 ± 6	22 ± 1	**8**	80 ± 4	34 ± 3
5	40 ± 5	20 ± 5	**B**	88 ± 5	39 ± 2

Table 2

The effect of pyridazine derivatives on wheat germination and seedling growth (the total height and the mean height of plantlets in the lot (H, Hm), (W, Wm))

Comp	H (cm)	H_{med} (cm)	W (g)	W_{med} (mg)
2	52.4 ± 13.3	4.4 ± 0.9	0.47 ± 0,41	40.03 ± 2.67
3	178.2 ± 17.2	5.9 ± 0.6	1.03 ± 0.12	34.32 ± 1.65
4	136.4 ± 3.4	5.8 ± 1.3	0.98 ± 0.08	42.32 ± 3.36
5	111.3 ± 0.3	5.5 ± 0.01	0.80 ± 0,01	39,84 ± 0,01
6	120.4 ± 4.8	6.8 ± 0.8	0.80 ± 0,13	40.35 ± 0.07
7	197.6 ± 7.2	5.3 ± 0.6	1.11 ± 0.15	28.52 ± 3.30
8	233.5 ± 25.9	6.6 ± 0.7	1.37 ± 0.14	37.05 ± 3.85
B	223.7 ± 23.2	6.5 ± 0.7	1.42 ± 0.19	41.66 ± 5.93

According to dates presented, all the pyridazine derivatives influenced dramatically the germination process of the wheat seeds and they can be divided into activators and inhibitors of plant growth of wheat.

From the class of toxic substances is considered the compound (**2**) / growth inhibitor, because the value of total plant height is only 52,4 cm, compared to the total plant height obtained for blank (223,7 cm). In terms of biochemical, it is considered that this compound disrupts plant metabolism and causes a disequilibrium in protein content and amino acids. In usually plants there is a well established proportion between the two levels, wich varies with plant age.

Compounds (7 and 8) can be considered activators of plant growth since total plant height value is very high; more, for compound (8) the total plant height exceed the value for the blank.

Good results were obtained for compounds (3) where the total plant height is 80% of the blank.

CONCLUSIONS

Experiments with wheat seed germination, showed their sensitivity to the action of substances tested and the effect of pyridazinic compounds in seed germination / growth and development of seedlings.

Pyridazine derivatives may influence germination rate and fresh plant weight, depending on the structure of each investigated compound.

Most toxic pyridazinic derivative of the series is compound (2). It presented the strongest inhibitory effect on plant growth and development. At the opposite end are compounds (**7** and **8**), good plant growth stimulators. The rest of the compounds showed a range of biological activity.

However, further research is needed to evatuate the impact of pyridazine derivatives on living organisms and especially of wheat germination.

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