

## RESEARCH ON THE CHANGES OF SOME PHYSIOLOGICAL PARAMETERS UNDER THE ACTION OF THE TIRADIN FUNGICIDE IN PRUSSIAN CARP AND CRAYFISH

Maria Cristina Ponepal, Alina Păunescu, Octavian Drăghici, Alexandru Gabriel Marinescu  
University of Pitesti, Pitesti, Tg. din Vale nr. 1, România  
E-mail: [ponepal\\_maria@yahoo.com](mailto:ponepal_maria@yahoo.com)

### Abstract

TMTD (tetramethyl thiuram disulfide, thiram) is a general use contact fungicide with protective action, third group of toxicity. It is used to control *Botrytis* on fruit and vegetables and in seed treatment. The thiurams are toxic to algae, water fleas and fish.

The 96-hour EC<sub>50</sub> for algae growth inhibition is approximately 1 mg/l (1 ppm), the 48-hour EC<sub>50</sub> for *Daphnia* is less than 0.21 ppm and the 96-hour LC<sub>50</sub> for fish is approximately 0.1 ppm - Bluegill sunfish 0.0445 mg/l Rainbow trout 0.128 mg/l and 4 mg/l carp.

This study was carried out to analyze the effects of sublethal and lethal concentrations of Tiradin fungicide on some physiological parameters of the prussian carp (*Carassius auratus gibelio* Bloch) and crayfish (*Astacus fluviatilis*).

The Tiradin product (thiuram 70%), under the concentrations of 0.01, 0.02, 0.04, 0.08 and 0.16 ml /l water, had, overall, an inhibitory effect on oxygen consumption for the prussian carp (*Carassius auratus gibelio* Bloch) and crayfish (*Astacus fluviatilis*).

Tiradin fungicide has changed the respiratory rhythm of prussian carps in all investigated concentrations. In all the researched concentrations (0,01, 0,02, 0,04 and 0,08 0,16 ml/l water), the fungicide tiradin modified the values of breathing frequency for the prussian carp, the values recorded at the end of the experiments were, in all cases, significantly different comparing to the control values (for  $p < 0.05$ ). For the first two concentrations, the fungicide's effect is stimulating for the frequency of the breathing movements, and for the next concentrations the effect is inhibiting.

The acute test (96 hours) records mortality only in fish variants from the concentration level of 0.08 ml Tiradin / l water, but none of the variants record absolute mortality. In the variants of crayfish records mortality only after 14 days at two concentration - 0.16 and 0.31 Tiradin/l water. The Tiradin toxic effect is stronger in prussian carps in comparison to crayfish (*Astacus fluviatilis*).

Keywords: prussian carp, crayfish, fungicide, Tiradin, oxygen consumption, , breathing frequency

### 1. INTRODUCTION

Dithiocarbamates form a large group of chemicals that have numerous uses in agriculture and medicine. The thiurams are fast-curing primary accelerators for natural and synthetic rubbers and are used most in agriculture as fungicide [11]. Thiurams are biodegradable so there is little concern for ecological persistence or bioaccumulation [5]. The thiurams are toxic to algae, water fleas and fish [11], [5].

TMTD (tetramethyl thiuram disulfide, thiram) is a general use contact fungicide with protective action, third group of toxicity [6]. It is used to control *Botrytis* on fruit and vegetables and in seed treatment [6]. Thiram is irritating to the eyes, skin, and respiratory tract

[13]. In water, thiram is rapidly broken down by hydrolysis and photodegradation, especially under acidic conditions [13]. The 96-hour EC<sub>50</sub> for algae growth inhibition is approximately 1 mg/l (1 ppm) [11], the 48-hour EC<sub>50</sub> for *Daphnia* is less than 0.21 ppm [6] and the 96-hour LC<sub>50</sub> for fish is approximately 0.1 ppm - Bluegill sunfish 0.0445 mg/l Rainbow trout 0.128 mg/l [6], [12] and 4 mg/l carp [7]. This study was carried out to analyze the effects of sublethal and lethal concentrations of Tiradin fungicide on some physiological parameters of the prussian carp (*Carassius auratus gibelio* Bloch) and crayfish (*Astacus fluviatilis*).

## 2. MATERIAL AND METHOD

Determinations were performed on samples of prussian carp (*Carassius auratus gibelio Bloch*) and crayfish (*Astacus fluviatilis*) caught in the lakes and rivers bordering Pitesti. Animals were acclimatized for 2 weeks before the completion of experiments in aquariums with a capacity of 100 l and 50 l [9], under conditions of natural photoperiodism, a period in which they were fed once a day (*ad libitum*), at around 10 am. After acclimatization in the laboratory, fish and crayfish were separated into lots and placed in two experiments.

The first experiment was carried out with prussian carps individuals with an average weight of  $10.5 \pm 1.28$  g, separated in six lots of 10 fish, subjected to Tiradin fungicide in concentrations of 0.01, 0.02, 0.04, 0.08, 0.16 ml /l water and lot of control (water).

The second experiment was carried out with crayfish individuals having an average weight of  $25.5 \pm 1.56$  g, which were subjected to tiradin fungicide concentrations of 0.01, 0.02, 0.04, 0.08, 0.16, 0.32 ml /l water and lot of control (water) (seven lots of 10 crayfish).

We mention the fact that, during all the experiments, the tiradin came from the commercial product Tiradin 70 PUS (thiram 70%). The fungicide concentrations that have been used have been established by preliminary survival test. The immersion of fish and crayfish in these solutions has been made after they have been well stirred and aired for five minutes. The water temperature has been between 18 and 20° Celsius, the immersion solution has been changed every 24 hours (semi static test) and the water has been continuously aired; the fish and crayfish have not been fed during the experiments, in order to avoid the intervention of this factor ([8], [9]).

The energetic metabolism, expressed by the oxygen consumption, was determined by using the closed respiratory chamber method (the oxygen dose in the water was established by using the Winkler chemical method) - [8], [9]. These determinations were made at intervals of 24, 48, 72, 96, 168 and respectively 336 hours.

The breathing frequency was determined at the same intervals as in the case of the energetic metabolism.

Our dates was statistically interpreted using the Anova test.

## 3. RESULTS AND DISCUSSION

In all experimental variants that have been applied on fish have only been observed three stages of the symptomatology scheme described by Schäperclaus for the intoxicated fish [8], [9]. Neurotoxic effects in rats from thiram exposure has been noticed by Lee and Peters, 1967 [1].

The first Figure shows the average frequency of the respiratory movements of prussian carps exposed to the action of Tiradin fungicide at temperatures of 18-20° C (0.01, 0.02, 0.04, 0.08, 0.16 ml /l).

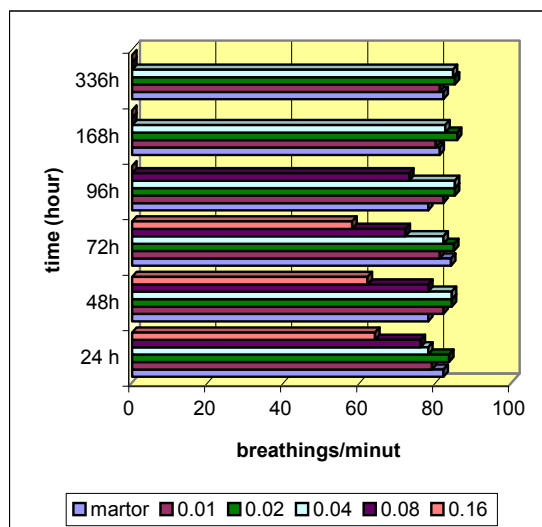


Figure 1. The influence of Tiradin fungicide upon breathing frequency on prussian carp

In all studied concentration, the Tiradin modified the values of breathing frequency; for the first two concentrations (0.01 and 0.02 ml Tiradin/l water) the fungicide effect is stimulating; at the concentration of 0.08 and 0.16 ml Tiradin/l water, effect of breathing frequency has been inhibiting. The values recorded at the end of the experiments were

significantly difference comparing to the control values.

Changes in oxygen consumption of prussian carps and crayfish exposed to the fungicide Tiradin in different concentrations are shown in Figures 2 and 3.

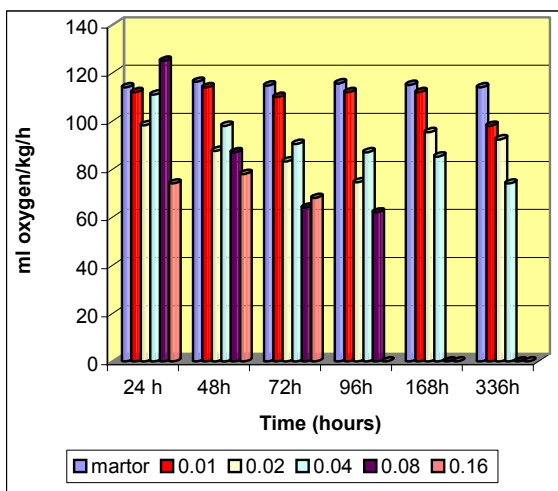


Figure 2 . The influence of the Tiradin fungicide upon oxygen consumption on prussian carp

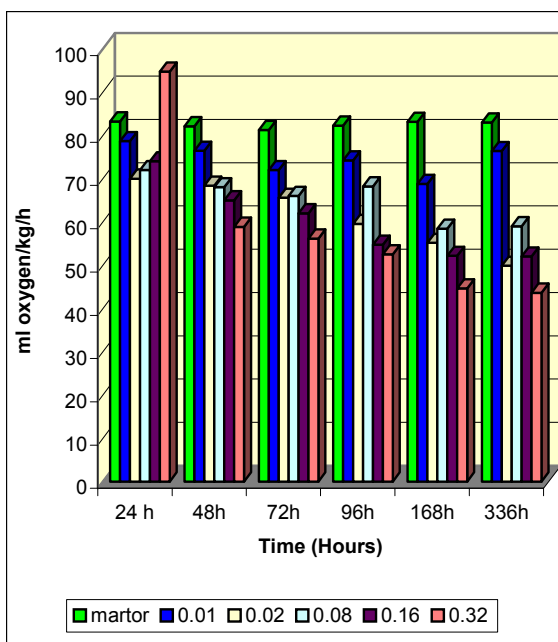


Figure 3 . The influence of the Tiradin fungicide upon oxygen consumption on crayfish (*Astacus fluviatilis*)

Tiradin had an inhibitory effect on the energy metabolism of prussian carps and crayfish –

except the first concentration of 0.01 ml Tiradin/l water.

The values of the physiological index recorded after two weeks of exposure are - between 87.5% and 49.6% of the values recorded prior to fish exposure to toxicity, significant changes for  $p < 0.05$  and between 87.49 and 48.91 of the values recorded prior to crayfish exposure to toxicity, significant changes for  $p < 0.05$ .

At the concentration of 0.01 ml Tiradin/l water, the fungicide had no significant effect to crayfish for  $p < 0.05$ .

Decreased oxygen consumption under the action of some pesticides (Dithane M 45, Reldan, Tilt,) has also been noticed by Marinescu [2] and Ponopal [3], [4], [10].

Table 1 shows the survival times on Prussian carps and *Astacus fluviatilis* during the 14 days of experiments.

TABLE 1 LETHAL EFFECT OF THE FUNGICIDE TIRADIN ON PRUSSIAN CARP AND ASTACUS FLUVIATILIS

Experiment al variants	Tirad in (ml/l water )	The number of living specimens					
		Immersion time (hours)					
		24	48	72	96	168	336
I. Prussian carp	0.01	10	10	10	10	10	10
	0.02	10	10	10	10	10	10
	0.04	10	10	10	10	8	6
	0.08	10	10	8	8	6	5
	0.16	8	7	6	4	1	1
II. Astacus fluviatili s	0.01	10	10	10	10	10	10
	0.02	10	10	10	10	10	10
	0.04	10	10	10	10	10	10
	0.08	10	10	10	10	10	10
	0.16	10	10	10	10	10	9
	0.32	10	10	10	10	10	7

The acute test (96 hours) records mortality only in fish variants from the concentration level of 0.08 ml Tiradin / l water, but none of the variants record absolute mortality. In the variants of crayfish records mortality only after 14 days at two concentration – 0.16 and 0.31 Tiradin/l water (10 and 30% after two weeks of fungicide exposure). None of the experimental

variants of crayfish recorded mortality during the first week of experience.

Analysis of Figure 2, 3 and table 1 shows that Tiradin effect is stronger on fish in comparison to crayfish. .

#### 4. CONCLUSIONS

After 7 days of exposing prussian carps to the Tiradin in concentrations of 0,01, 0,02, 0,04 and 0,08 0,16 ml/l fungicide modified the values of breathing frequency. In the two first experimental variants (0.01 and 0.02 ml/l water) Tiradin is stimulating of the breathing frequency of fish; at the concentration of 0.04, 0.08 and 0.16 ml/l water, the fungicide caused a decrease in the respiratory rhythm of prussian carps.

The fungicide Tiradin had an inhibitory effect on the energy metabolism of prussian carps and crayfish at concentrations of 0.02, 0.04, 0.08 and 0.16 ml/l water.

The Tiradin toxic effect is stronger in prussian carps in comparison to crayfish (*Astacus fluviatilis*). After two weeks of exposure to Tiradin, records mortality in crayfish only at concentrations of 0.16 and 0.32 ml Tiradin/l water.

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