



## Fungicidal Activity of Seed Disinfectants Against Root Rot of Wheat in Various Types of Soils

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### ABSTRACT

We studied the efficacy of seed treatment with a variety of fungicides for control of root rot of wheat in different soil types. The effectiveness of various doses of the preparation on the basis of thiram (TMTD and TMTD-plus) and triazole containing Dividend Star, SC has been compared under the conditions of the Southern Urals in typical soils of the region (dark chestnut, southern chernozem, common chernozem). It is revealed that the effectiveness of thiram containing preparations in moderate doses (2.0–2.5 l/t for TMTD-plus and 3.0 l/t for TMTD) is higher on the dark-chestnut soils than on Chernozem. The biological efficiency of the Dividend Star does not depend on the type of soil. We compared the five pesticides with active substances from different chemical classes: Celest Top (thiamethoxam + difenoconazole + fludioxonil), Scenic Combi (clothianidin + fluoxastrobin + prothioconazole + tebuconazole), Maxim (fludioxonil), TMTD-plus (thiram), Kinto Duo (triticonazole + prochloraz) and Dividend Star (difenoconazole + cyproconazole) at the different tillage technologies under the non black soil region conditions. The effectiveness of all preparations decreases significantly at the no-till technology compared to the plowing. Most effective in plowing were Kinto Duo, Celest Top and TMTD-plus, and in zero-till – Celest Top and Scenic Combi.

**Keywords:** Wheat Root Rot, Seed Treatment, Fungicides, Soil Type, Tillage.

**HOW TO CITE THIS ARTICLE:** A. P. Glinushkin, T.A. Akimov, O.O. Beloshapkina, J. Molnár, I.I. Sycheva, S.K. Temirbekova and et al., Fungicidal activity of seed disinfectants against root rot of wheat in various types of soils, Entomol Appl Sci Lett, 2018, 5 (2):101-107

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**Received:** 02/01/2018

**Accepted:** 24/04/2018

### INTRODUCTION

The concept of the integrated plant protection demands the use of the most efficient ways to reduce the damage caused by pests. Presowing treatments of seeds have been shown to be effective in protection of both seedlings and adult plants from root rot. Root rot diseases are prevalent in many wheat-cultivating regions [1]. They cause infringements of vital processes throughout the growing season, and thus, a significant reduction of plant productivity [2]. The most common agents of this type of diseases are fungi of the genera *Fusarium* and *Bipolaris*. In addition to these, there are also fungal

pathogens from *Cercospora*, *Ophiobolus*, *Pythium* genera.

According to the Rosselkhozsentr, root rot occurred at 766.0 thousand hectares only on the spring crops in Russia in 2014 [3]. Root rot could cause substantial (up to 25%) yield losses [4]. Moreover, the composition and harmfulness of the species complex that causes root rot can vary considerably from regions to regions [5] belonging to different climatic and soil zones. The use of protective measures, including farming practices, crop rotation, and most importantly, the use of quality seeds and seed treatments practices are required to prevent root rot. There is a wide range of active compounds containing one or more active compounds and preparations containing one or more active compounds, for seed dressing.

The application of disinfectants on the soil greatly affects the effectiveness of the fungicidal seed treatment, the duration of the protective action, and toxicity to the plants. Soils vary greatly in terms of microstructure, chemical and mechanical composition, the pH of the soil solution, content of various forms of humus, microbiological activity, soil agricultural and physical properties and other parameters at the territory of the Russian Federation [6, 7]. The parameters that determine soil quality, also influence the microbial community in the rhizosphere of plants [8-10]. The pH of the soil solution significantly influences the soil's organic matter, determining the microbiota composition, on which the survival of the pathogen propagules depends [11], while the maintenance of the mobile forms of phosphorus that is available to plants affects the development of diseases of underground organs [2].

A plant protection system that takes into account the soil properties and technology of its processing should allow the optimal choice of a particular compound or preparations of certain active substances for the seeds treatment, affecting both the effectiveness of the substances against pathogens and the phytosanitary status of seeds and crop plants.

The purpose of this work was to compare the effectiveness of the fungicidal seed disinfectants belonging to a number of chemical classes against the root rot of wheat, depending on the soil types and methods of tillage in different climatic zones.

## MATERIALS AND METHODS

The standard techniques of phytopathological laboratory and field studies (root inspection, selection of soil and plant samples, isolation of the microorganisms in pure culture, microscopy, etc.) have been used. The evaluation of the seed germination was carried out in the late phase of germination (10–13 phases on the Zadoks' scale).

Distribution and development of the root rot were evaluated by the selection of plants in a field in the phase of tillering (21–25), flowering (61–69), and wax ripeness (75–82) using a five-point scale of infestation. Before parentheses we have specified the parameters during phase of tillering, in brackets – during the flowering

phase and wax ripeness: 0 – healthy plants, 1 – single strokes on the coleoptile, subterranean internode (the brown strokes or narrow band on the base of the stem or underground part), 2 – slight browning of the coleoptile or subterranean internode (brown stripes, covering more than half of the surface of the affected organ on the base of the stem and underground parts), 3 – strong browning of the coleoptile or subterranean internodes (solid russetting of the first stem and underground internodes), 4 – complete withering of the seedling (absence of the productive stems with the presence of 3 points symptoms).

The disease etiology was substantiated by isolating the fungal pathogens into pure cultures on potato dextrose agar in Petri plates and with microscopic examination of conidial morphology according to Khokhryakov et al. (2003) [12]. Weather information has been obtained from the local meteorological stations.

The first part of the research was carried out in 2005–2007 in the educational and experimental farm of the Orenburg State Agricultural University and other farms in the South Urals characterized by a sharply continental climate (hydrothermal coefficient 0.3–1.4) on regional varieties of spring wheat (Uchitel, Varyag, Saratovskaya 42 and others). We have studied the efficacy of different application rates of three disinfectants: Thiram (TMTD-plus, (suspension concentrate, SC) (tetramethyl-thiuram-disulphide 400 g per liter with the addition of immunostimulants (Kresacin); TMTD WSC (thiram 400 g per liter); Dividend Star SC (difenoconazole + cyproconazole 30+6.3 g per liter, respectively) on the main soil types of the region: dark chestnut (humus content 4–6%, pH 7.2–7.3), southern chernozem (humus content 3–6%, pH 7–8) and common chernozem (humus content 6–10%, pH 7–8). Seeding rates varied based on types of soils:  $3.5 \times 10^6$  on the dark chestnut;  $4.0 \times 10^6$  on the southern chernozem; ordinary chernozem –  $5.0 \times 10^6$  seeds per hectare.

The second part of the research have been conducted in temperate climate of the Non-black soil region in 2015 at the Experimental Field station of the RSAU-MAA named after K.A. Timiryazev (Moscow), where the various technologies of tillage, including plowing and no-till have been studied for several years. This al-

lowed us to assess the phytosanitary aspects of different methods of the basic tillage. Classic processing technology for the central part of the Non-black soil region is the annual plowing to a depth of 22–24 cm, while no-till technology begins to be implemented in a number of areas and farms.

The soil at the Experimental Field station is represented by sod-podzolic medium textured loam type with the following content of nutrients:  $P_2O_5$  - 150–250 mg per kg of soil,  $K_2O$  - 40–80 mg/kg, pH of the soil solution - 4.6–5.0, humus content - 2.4–2.5%. The four-year crop rotation (winter wheat + mustard for green manure after harvesting the main crop, potato, spring barley, vetch-oat mixture) has been used.

In the small plot experiment with the winter wheat line L-15 (the result of the individual selection of the Zvezda variety) we have been studied the efficacy of the following fungicide seed disinfectants: Celest Top, SC (thiamethoxam + difenoconazole + fludioxonil 262.5+25+25 g/l, respectively) - 1.2 l/t, Scenic Combi, SC (clothianidin + fluoxastrobin + prothioconazole + tebuconazole 250+37.5+37.5+5 g/l, respectively) - 1.25 l/t, Maxim, SC (fludioxonil 25 g/l) - 1.5 l/t, TMTD-plus, SC (thiram 400 g/l with the addition of kresacin) - 2.5 l/t, Kinto Duo, SC (triticonazole + prochloraz 20+60 g/l, respectively) - 2.0 l/t. The application rates of preparations were calculated according to the minimum recommended rates listed in the Directory of Pesticides and Agrochemicals (2015) [13], permitted for use on the territory of the Russian Federation (Ministry of Agriculture).

Surveys of the root rot in autumn were performed at the beginning of the three-leaf stage (10–13) at the tillering phase at autumn (21–25) and spring (25–27). Efficacy was assessed according to the development of disease relative to control variant without fungicide.

The evaluation of the chlorophyll and carotenoid content was carried out by means of the pigments extraction in 100% acetone and subsequent measurement in a spectrophotometer (SF-104) [14]. Sampling was carried out in the phase of tillering.

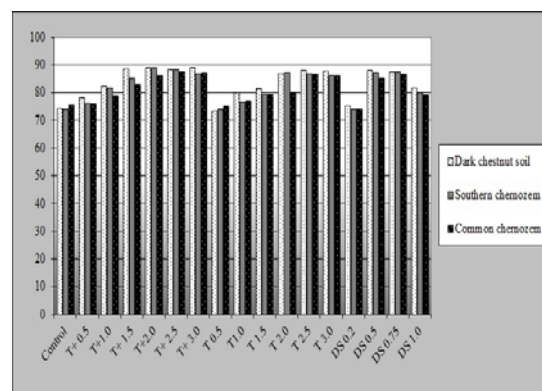
The statistical analysis method used in this work included standard deviation and confidence interval calculation.

## RESULTS AND DISCUSSION

Our data show that in the South Urals the germination rates of seeds were in almost all cases higher in the dark chestnut soils than in the southern chernozem (Figure. 1).

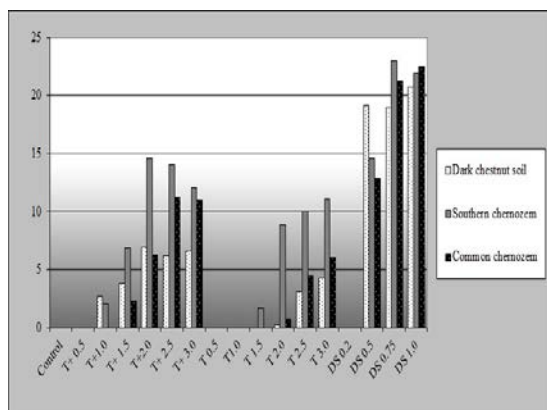
In all of the application rates of TMTD-plus we have revealed an increase in the germination of wheat seeds compared to the control: 3.8–14.8% on dark chestnut soils; 2.4–15.1% on the south chernozem; 0.3–11.6% on the common chernozem (Figure. 1). Doses above the application rate of 2 liters of TMTD-plus per tonne did not result in a further increase in seed germination. A similar trend was evident in the variant doses of TMTD.

This pesticide in the same doses was less effective on all types of soil than TMTD-plus. Also on the dark chestnut soil with the application rate of 0.5 l/t, the seed germination was not significantly higher as compared to the control. The optimal dosages were 2.0 l/t and 2.5 l/t for the TMTD-plus and TMTD, respectively (Figure. 1). The effectivity of Dividend Star, which is considered one of the best preparations against root rot, depended on its dose. The highest seed germination was observed at a rate of 0.5–0.75 l/t (recommended 0.75 l/t).



**Figure. 1.** The effect of disinfectants on seed germination of wheat (%) depending on soil zone of regional agro-ecosystems of the southern Urals, average for 2005–2007

Our data suggest that soil type has a significant influence on the effectiveness of fungicides against root rot (Figure. 2). Microbiological analysis of the affected tissues of root and stem samples showed that the causal agent of root rot in the spring wheat is the *Helminthosporium-Fusarium* spp. complex with the predominance of fungi belonging to the *Bipolaris* genus.

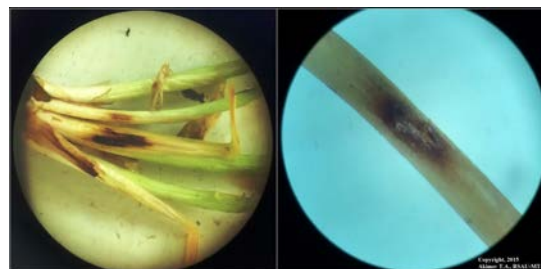


**Figure 2.** Biological efficiency (%) of the seed dressing norms in the figureht against the root rot of spring wheat (average for 2005–2007 at the South Urals)

The greatest biological efficacy of the tested pesticides was observed in the southern chernozem. This trend was most evident for TMTD and TMTD-plus in a dose of 1.5 l/t and more. The effectiveness of Dividend Star was found mainly depend on the rate of the pesticide and did not depend on the type of soil. At the rate of 0.5 l/t it was most effective in dark chestnut soil, in a medium recommended rate (0.75 l/t) – in the southern chernozem, and at the rate of 1.0 l/t – in the common chernozem. This preparation was found to be most effective against root rot in all types of soils, especially in dark chestnut soils, with the application rate of 1.0 l/t, showing a biological effectiveness of 20.7% in the average for three years. TMTD-plus with the rate of 2.0 l/t was the second most effective agent with 6.9 % efficiency. The results showed that the TMTD pesticide with the most effective rate of 3 l/t exhibited the lowest efficiency that did not exceed 5%.

Research conducted in RSAU-MTAA, was aimed at comparing the effectiveness of different preparations to protect wheat plants against root rot under conditions of non-black soil region. Microbiological analysis showed that the most common root rot pathogens were *Fusarium* (90–95%). Fungi of the genus *Bipolaris* were less common (5–10%) (Figure. 3). All treatments provided a significant increase in plant density and germination relative to the control at 7–12% (Table 1). Studied parameters were not significantly different among the various fungicide treatments. However, a treatment with Maxim was most effective on the rate of seed

germination under plowing compared to zero-till.



**Figure 3.** Root rots of winter wheat caused by a *Fusarium* sp. in RSAU-MTAA, 2015

**Table 1:** Plant density (plants/m<sup>2</sup>) and germination (%) rate of winter wheat line L-1 depending on the disinfectant and method of tillage (RSAU-MTAA 2015)

Treatment	Plowing		Zero-till	
	Plants /m <sup>2</sup>	germination, %	Plants /m <sup>2</sup>	germination, %
Control	415.7 ±19.9	75.6	380.9 ±19.0	69.3
Celest Top	485.2 ±13.3	88.2	440.0 ±18.1	80.0
Scenic Combi	479.1 ±21.2	87.1	450.1 ±19.4	81.8
Maxim	465.6 ±17.1	84.7	417.4 ±20.0	75.9
TMTD-plus	464.2 ±20.4	84.4	454.7 ±15.7	82.7
Kinto Duo	470.9 ±19.9	85.6	442.6 ±23.8	80.5

Celest Top, 1.2 l/t; Scenic Combi, 1.25 l/t; Maxim, 1.5 l/t; TMTD-plus, 2.5 l/t; Kinto Duo, 2.0 l/t

The prevalence and development of this group of diseases have been found higher in hot and dry years [2]. Summer 2015 in the Moscow region was moderately cool and wet with the hydrothermal coefficient 1.7 and unfavorable for the development of pathogens. The effectiveness of fungicides on the plots under plowing and zero-till in different ontogenesis stage is shown in (Table 2).

**Table 2:** The fungicidal effect of the disinfectants belonging to the different chemical groups on the development of winter wheat root rot under the different methods of tillage (RSAU-MTAA, 2015)

Fungicide	Plowing			Zero-till		
	Growth stage			Growth stage		
	10–15	21–25	25–27	10–15	21–25	25–27

Celest Top	53.7 ±7.1	92.7 ±6.5	60.5 ±10.8	59.2 ±7.7	46.8 ±10.1	46.4 ±11.4
Scenic Combi	62.1 ±12.4	77.2 ±7.2	81.4 ±12.9	49.2 ±12.8	38.5 ±4.5	60.1 ±8.8
Maxim	62.1 ±10.5	89.6 ±8.8	33.3 ±12.0	14.6 ±7.9	11.5 ±7.4	29.0 ±10.5
TMTD-plus	70.6 ±7.1	93.3 ±7.0	55.9 ±10.5	18.3 ±15.8	22.6 ±6.7	32.8 ±14.0
Kinto Duo	86.1 ±6.7	96.6 ±1.7	88.7 ±2.7	29.4 ±13.1	16.8 ±5.6	41.0 ±9.7

Celest Top, 1.2 l/t; Scenic Combi, 1.25 l/t; Maxim, 1.5 l/t; TMTD-plus, 2.5 l/t; Kinto Duo, 2.0 l/t

In case of plowing, all the studied pesticides were relatively effective compared to the control group at a given degree of the root rot development. In three-leaf stage the greatest biological efficiency for the development was shown by Kinto Duo and TMTD-plus– 86.1% and 70.0%, respectively. We assessed the effectiveness of the fungicides during plant development. At the onset of wintering, the efficiency of Kinto Duo increased slightly and reached 96.6%. All other fungicides also showed increased efficiency (Scenic Combi– 15%, Celest Top – 39% e.t.c.). At spring (25–27) all pesticides (mostly Maxim and TMTD-plus) exhibited lower effectiveness than at autumn.

Surveys of the root rot on the plots with direct drilling showed that tillage significantly affected the performance of the pesticides. All of them showed significantly weaker protective effect under zero tillage as compared to plowing. This is clearly evident for Maxim: at areas of plowing it was one of the most effective, however, under direct sowing conditions its biological efficacy was little. Thus, the pesticide containing only fludioxonil without other active molecules not only has not provided a protective effect but may cause additional physiological stress for seedlings. Insecticidal-fungicidal fungicides Celest Top and Scenic Combi showed the highest biological efficiency with the no-till, reducing the development of root rot by 50–60% at 10–13 stage. The greatest decrease of the efficiency was noted in Kinto Duo, Celest Top and Scenic Combi where it has decreased by up to 11–13% from the initial level. Maxim and TMTD-plus did not change its effectiveness. Spring surveys show that in plots with zero-till pesticide effectiveness were greater than at autumn. It happened because root rots development in variants without fungicides was significantly higher in zero tillage compared with plowing.

One of the indicators showing the physiological state of the plant is the content of pigments in leaves. The higher the content of the chlorophylls A and B, the more active the process of the photosynthesis and accumulation of sugars for the wintering. At the same time, the more the carotenoids were observed in the leaves, the weaker the process of nutrients accumulation. The results show that all fungicide preparations provided a higher chlorophyll content at the leaves of winter wheat as compared to the control (Table 3).

**Table 3:** The content of chlorophylls A and B and carotenoids (mg/g of the wet weight) in leaves of winter wheat at the tillering phase (agrobackground – plowing)

Treatment	Chlorophylls			Carotenoids
	a	b	a+b	
Control	1.29 ±0.05	0.45 ±0.02	1.74 ±0.06	0.37±0.01
Celest Top	1.47 ±0.04	0.58 ±0.05	2.05 ±0.03	0.33±0.01
Scenic Combi	1.47 ±0.13	0.57 ±0.08	2.04 ±0.21	0.33±0.03
Maxim	1.38 ±0.01	0.52 ±0.04	1.90 ±0.04	0.36±0.01
TMTD-plus	1.47 ±0.25	0.63 ±0.11	2.10 ±0.36	0.31±0.04
Kinto Duo	1.53 ±0.09	0.82 ±0.11	2.35 ±0.18	0.25±0.03

Celest Top, 1.2 l/t; Scenic Combi, 1.25 l/t; Maxim, 1.5 l/t; TMTD-plus, 2.5 l/t; Kinto Duo, 2.0 l/t

The highest content of chlorophyll and smallest content of carotenoids was found in plants treated with the Kinto Duo that differs from all other triazole antifungal preparations by the presence of the prochloraze as the second component. The second highest influence on the amount of leaf chlorophyll contents was observed for TMTD-plus having immunostimulants in its composition in addition to thiram. Insecticidal-fungicidal preparations Scenic Combi and Celest Top showed an average influence on the contents of chlorophylls and carotenoids. The highest carotenoid content in the leaves was observed after treatment with Maxim.

The results indicate that the effectiveness of disinfectants, and therefore the choice of a particular preparation in production conditions depends not only on the soil and climatic zone and the soil characteristics but also on the applied tillage technology.

## CONCLUSION

Soil type had a significant influence on the activity of the studied preparations against the root rots pathogens and on the field germination of seeds. Thiram-containing preparations have shown the highest biological effects on the southern chernozem, while the Dividend Star has not shown such a tendency. Its effectiveness depends entirely on the application rate. Seed germination was most improved in dark chestnut soils.

The most efficient preparation was Dividend Star under the soil and climatic conditions of the Southern Urals at the application rate of 0.75–1.0 l/t. TMTD-plus has been the second most efficient at the rate of 2.0–2.5 l/t (recommended 3.0–4.0 l/t). TMTD at the rate 3.0 l/t (recommended 3.0–4.0 l/t) was the least productive in terms of improving seed germination under field conditions and biological effectiveness. Thus the minimum application rates of preparations based on thiram are effective in certain cases (the presence of crop rotation, adverse for the life of the pathogen weather conditions and others).

At the sod-podzolic soil in the non-black soil region, all the studied preparations were significantly more effective when using plowing than with the direct drilling (no-till). Celest Top, TMTD-plus, Maxim and Kinto Duo have shown the highest efficiency on the plot with plowing at autumn. Moreover, the Kinto Duo and TMTD-plus provided the highest content of the chlorophylls A and B in leaves in comparison with other preparations. Only Celest Top and Scenic Combi have shown significant efficiency when using no-till technology compared to other preparations.

Thus, using either a classic non-black soil region tillage technology (plowing) or the direct drilling, the choice of the disinfectant should be carried out taking into account the spectrum of harmful organisms, the phytosanitary status of seed and soil. If a high number of phytophagous insects is predicted in the growing season, the use of insecto-fungicide preparations (Celest Top, Scenic Combi) would be optimal. If the main damage is expected from diseases transmitted with the soil or seeds, it is advisable to use TMTD-plus (for the both technologies) or Kinto Duo (only for plowing) providing a good

physiological state of plants in addition to high biological effectiveness.

*This work was partly supported by the Russian Foundation Basic Research (RFBR) under the project 15-29-05893.*

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