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BIOLOGICAL EFFECTIVENESS OF FUNGICIDES PRIORI XTRA, FOLICUR, APPROACH PRIMA, AND MAXTROBIN XTRA FOR CONTROL OF KARNAL BUNT

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ABSTRACT

Commercial fungicides Approach Prima CS, Priori Xtra CS, Maxtrobin Xtra CS, and Folicur EW were evaluated in the field, to determine their biological effectiveness to control karnal bunt (Tilletia indica) of wheat. A completely randomized design was used with four replications. Twenty heads of cultivar Tacupeto F2001 were inoculated during the boot stage with an allantoid sporidial suspension (10,000/mL). Commercial rates indicated in the containers of each product were followed. The first application was carried out ten days after inoculation {Zadoks 56-58, (Feekes 10.4-10.5)}, and the second one ten days later. Inoculated spikes were threshed by hand and the healthy and infected kernels were counted to determine the percentage of infection. The biological effectiveness of the products evaluated were Priori Xtra 99.2, Folicur 98.9, Approach Prima 98.5, and Maxtrobin Xtra 85.3%. The untreated inoculated check had a mean of 36.9% infection. There were no statistical differences for the products evaluated for level of infection after arcsin transformation (Tukey, p = 0.05) and the coefficient of variation was 6.1%. The a thousand grain weight was 55.3, 51.1, 54.8, and 55.1 g, respectively, and 53.6 g for the untreated inoculated check. No phytotoxic effects to the wheat plants were observed.

Keywords: Karnal bunt, Tilletia indica, biological effectiveness, fungicides.

1. INTRODUCTION

Karnal bunt of wheat is caused by the fungus *Tilletia indica* Mitra, and it is the most important disease of wheat seed and grain (Fig. 1) in northwest Mexico (Fuentes-Dávila, 1997). This disease has also been reported from India (Mitra, 1931), Pakistan (Munjal, 1975), Nepal (Singh *et al*., 1989), Brazil (Da Luz *et al*., 1993), The United States of America (APHIS, 1996), Iran (Torarbi *et al*., 1996), the Republic of South Africa (Crous *et al*., 2001), and Afghanistan (CIMMYT, 2011). In Mexico, the disease was reported in 1972 by Duran. The effect on flour quality and the quarantine regulations, both, national and international, cause economic losses to farmers (SARH, 1987; Brennan *et al*., 1990; SAGARPA, 2002). Although breeding for genetic resistance has been a major goal in Mexico since the late 80´s and beginning of the 90´s (Metzger, 1986; Warham *et al*., 1986; Rajaram *et al*., 1991), an integrated management program for the disease is of primary importance. Several chemical control strategies have been evaluated since Mitra (1935, 1937) with hot water, solar energy, and seed treatments (Fuentes *et al*., 1983; Krishna and Singh, 1983; Aujla *et al*., 1986; Salazar-Huerta *et al*., 1986a,b; Smilanick *et al*., 1987; Figueroa-Lopez and Espinoza-Salazar, 1988; Warham and Prescott, 1989; Robles-Sosa and Fuentes-Dávila, 1996); although, some products inhibit teliospore germination, seed treatments do not control the disease since the life cycle of the *Tilletia indica* is different than the other smuts of wheat (Wilcoxson and Saari, 1996); similarly, the application of fungicides in soil drench does not reduce disease incidence (Valenzuela-Rodríguez, 1985). Because *Tilletia indica* causes floral infections, the application of agrochemicals during the stages of heading-flowering-anthesis of the wheat plant, is the activity by which greater control of the disease is obtained and allows a more profitable economical margin (Salazar-Huerta *et al*., 1997).

Figure 1. Symptoms of karnal bunt in the wheat grain.

Foliar application of fungicides have been evaluated for control of karnal bunt since the early 1980´s (Singh and Prasad, 1980; Singh and Singh, 1985; Smilanick *et al*., 1987; Figueroa and Valdés, 1991; Salazar-Huerta *et al*., 1997; Figueroa-López and Alvarez-Zamorano, 2000; Fuentes-Dávila *et al*., 2005, 2016, 2018; Fuentes-Dávila, 2007), and some of the commercial products evaluated include: Bavistin, Baycor, Bayfidan, Bayleton, Baytan, Bemistop, Benlate, Blitox, Ceresan, Consist, Dithane-M45, Duter, Folicur, Headline, Jewel, Kocide, Manzate, Nustar, Opus, Pointer, Sportak, Tilt, Topsin, Vangard, and Varon. Since the early experimentation with foliar application of agrochemicals, the triazol group which affects the biosynthesis of ergosterol, a primary component of the fungal cell plasma membrane (Pérez-García *et al*., 2005; Ribas e Ribas *et al*., 2016) has had the best control of the disease (Smilanick *et al*., 1987; Figueroa and Valdés, 1991; Salazar-Huerta *et al*., 1997; Figueroa-López and Alvarez-Zamorano, 2000; Fuentes-Dávila *et al*., 2005, 2016, 2018; Fuentes-Dávila, 2007), therefore, the objective of this work was to evaluate the biological effectiveness of new fungicides of the strobilurin group which affects mitochondrial respiration (Pérez-García *et al*., 2005; Feng *et al*., 2020) formulated along with a triazol component, such as Approach Prima CS DUPONT, Priori Xtra CS SYNGENTA, Maxtrobin Xtra CS HELITI, and the check triazol fungicide Folicur EW BAYER CROPSCIENCE, for control of karnal bunt in the field, under artificial inoculation.

2. MATERIALS AND METHODS

The experiment was carried out during the crop season 2018-2019 at the Norman E. Borlaug Experimental Station, located in block 910 of the Yaqui Valley at 27°22´04.64´´ latitude north and 109°55´28.26´´ longitude west, 37 masl, with climate warm [BW (h)] and extreme warm and dry [BS (h)], according to Köppen classification modified by García (1988). Sowing date was December 7, 2018 with a rate of 80 kg of seed/ha. Treatments were established in a completely randomized experimental design (Fig. 2) with four replications using bread wheat commercial cultivar Tacupeto F2001 (Camacho-Casas *et al*., 2003). The experimental plot consisted of 4 beds each with two rows 3 m long and 0.80 m between beds. The technical recommendation by INIFAP for the agronomic management was followed (Figueroa-López *et al*., 2011).

20 R4 Untreated check	19 R4 Approach Prima	18 R4 Maxtrobin Xtra	17 R4 Folicur	16 R3 Untreated check
11 R3 Maxtrobin Xtra	12 R2 Folicur	13 R2 Untreated check	14 R4 Priori Xtra	15 R3 Folicur

Figure 2. Randomized complete distribution of treatments in the field for control of karnal bunt (*Tilletia indica*), by foliar applications during the crop season fall-winter 2018-2019 in the Yaqui Valley, Sonora, Mexico.

10 R3 Approach Prima	9 R3 Priori Xtra	8 R2 Approach Prima	7 R2 Maxtrobin Xtra	6 R2 Priori Xtra
1 R1 Folicur	2 R1 Maxtrobin Xtra	3 R1 Priori Xtra	4 R1 Approach Prima	5 R1 Untreated check

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Inoculations were carried out during the boot stage (stage 49, Zadoks *et al*., 1974) by injection applying 1 mL per spike with an allantoid sporidial suspension (10,000/mL) in 20 spikes, in the central rows of each plot (Fig. 3).

Figure 3. Injection of the wheat plant during the boot stage.

Inoculum was prepared as described by Fuentes-Bueno and Fuentes-Dávila (2007). Commercial rates indicated in the containers of each product were followed (Table 1). For application of fungicides, a manual Solo backpack sprayer (15 L) was used with a single nozzle, and the volume was based on 250 L of water/ha. To avoid the carry over of the products applied, plastic barriers were used in each plot during the applications (Fig. 4). The first application was carried out ten days after inoculation {Zadoks 56-58, (Feekes 10.4-10.5)} and the second ten days later. Inoculated spikes were threshed by hand, and the percentage of infection was obtained by counting the number of infected and healthy grains from 20 inoculated spikes from each plot treated with the fungicides, and from 20 inoculated spikes from the untreated check.

Table 1. Fungicides, formulation, concentration, and rates used to control karnal bunt by foliar applications, during the crop season 2018-19, in the Yaqui Valley, Sonora, Mexico.

Treatments	Formulation and concentration^{y}	Rate^{z} L ha^{-1}
Approach Prima CS	picoxystrobin 17.9 + cyproconazole 7.1	0.40
Priori Xtra CS	azoxystrobin 18.2 + cyproconazole	0.80

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	7.27	
Maxtrobin Xtra CS	tebuconazole 22 + azoxystrobin 11	0.40
Folicur EW	Tebuconazole 25	0.50
Untreated inoculated check		

[y]Active ingredient.

[z]*Liters of commercial product.*

Figure 4. Application of fungicides in the field for control of karnal bunt.

The biological effectiveness was obtained using Abbot´s formula: effectiveness of treatments = average percentage of infection of the check – average percentage of infection of the treatment / average percentage of infection of the check x 100. The ANOVA was performed and mean comparison by Tukey´s test ($p = 0.05$) to determine statistical differences among treatments, previous arcsin transformation $\sqrt{X + 0.5}$ (Steel and Torrie, 1980). For each replication from treatments, a thousand grain weight was recorded. The phytotoxicity was evaluated ten days after each application of the fungicides, according to the EWRS scale (Table 2) (Champion, 2000).

Table 2. Values of the EWRS scale (1-9) to evaluate phytotoxicity in experimental plots, inoculated with karnal bunt and treated with Approach Prima, Priori Xtra, Maxtrobin Xtra, and Folicur in the Yaqui Valley, Sonora, Mexico, during the crop season fall-winter 2018-19.

Value (Category)	Effect on the plant
1	without effect

2	very light symptoms
3	light symptoms
4	symptoms which are not reflected on yield
5	Limit of acceptability medium damage
6	elevated damage
7	very elevated damage
8	severe damage
9	complete death

Transformation of the EWRS punctual logarithmic scale to percentage

Punctual value	Phytotoxicity (%)
1	0.0-1.0
2	1.0-3.5
3	3.5-7.0
4	7.0-12.5
5	12.5-20.0
6	20.0-30.0
7	30.0-50.0
8	50.0-99.0
9	99.0-100

3. RESULTS AND DISCUSSION

Significant statistical differences were detected between treatments and also with the untreated check, with respect to the values of percentage of infection; the coefficient of variation was 6.18% (Table 3). Mean comparison by Tukey´s test indicated that all treatments with fungicide application were effective on reducing the percentage of infection, when compared with the untreated inoculated check, which showed the highest average percentage of infection (36.95%)

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(Table 4), with a range between 34.17 and 40.41. However, Maxtrobin Xtra was statistically different than the other products. The range of the percentage of infection in plants treated with Priori Xtra was 0.09 to 0.66 and a mean of 0.26%, with Folicur the range was 0 to 0.71 and a mean of 0.37%, with Approach Prima 0.3 to 1.05 and a mean of 0.53%, and with Maxtrobin Xtra 4.66 to 6.36 and a mean of 5.42%. The biological effectiveness of the products evaluated were Priori Xtra 99.2, Folicur 98.9, Approach Prima 98.5, and Maxtrobin Xtra 85.3%. Since the early experimentation with application of fungicides during the heading-flowering-anthesis stage of the wheat plant, the triazol group which affects the biosynthesis of ergosterol, a primary component of the fungal cell plasma membrane (Pérez-García *et al*., 2005; Ribas e Ribas *et al*., 2016) has had the best control of the disease (Smilanick *et al*., 1987; Figueroa and Valdés, 1991; Salazar-Huerta *et al*., 1997; Figueroa-López and Alvarez-Zamorano, 2000; Fuentes-Dávila *et al*., 2005, 2016, 2018; Fuentes-Dávila, 2007).

Table 3. Analysis of variance of the percentage of infected grain with karnal bunt, in spikes treated with Approach Prima, Priori Xtra, Maxtrobin Xtra, and Folicur, and in spikes of an untreated check in the Yaqui Valley, Sonora, Mexico, during the crop season fall-winter 2018-19.

Source of Variation	DF	SS	MS	F value	F tab
Treatments	4	3518.48	879.62	389.08	3.06
Error	15	33.91	2.26		
Total	19				
C.V. = 6.18					

Table 4. Mean separation by Tukey´s test of the transformed percentages of infected grain with karnal bunt, in spikes treated with Approach Prima, Priori Xtra, Maxtrobin Xtra, and Folicur, in the Yaqui Valley, Sonora, Mexico, during the crop season fall-winter 2018-19.

Treatment	Infected grain		Mean separation
	Real	Transformed	
Priori Xtra	0.26	2.71	A
Folicur	0.37	2.97	A
Approach Prima	0.53	4.04	A
Maxtrobin Xtra	5.42	13.45	B
Untreated check	36.95	37.43	C

The QoI (quinone outside inhibitor) fungicides are newer products obtained from wood-rotting mushroom fungi, including *Strobilurus tenacellus* (Pers.) Singer from which the name was coined the Q_oI fungicides control an unusually wide array of fungal diseases, including diseases caused by water molds, downy mildews, powdery mildews, leaf spotting and blighting fungi, fruit rotters, and rusts. They are used on a wide variety of crops, including cereals, field crops, fruits, tree nuts, vegetables, turfgrasses, and ornamentals. (Vincelli, 2012). Strobilurins have site-specific mode of action, through inhibiting fungal respiration at the Qo site in mitochondria, causing electron transport blockage in the cytochrome bc1 complex (complex III of electron transport chain), between cytochrome b and cytochrome c1, at the Qo site by inhibiting ubiquinol-cytochrome c-oxide reductase, they reduce the respiratory process blocking the fungus cell's energy supply (ATP), leading to its death (Selim and Khalil, 2021). The strobilurin-based fungicide Headline CE (pyraclostrobin), with a wide spectrum of control of fungi (Ascomycetes, Basidiomycetes, Deuteromyces, and Oomycetes) and over 50 major plant diseases (BASF, 2022; Vademecum, 2022), showed a biological effectiveness (BE) of 86.87% in comparison with the triazol commercial fungicides Varon EC (tebuconazole), Pointer SC (flutriafol), and Opus SC (epoxyconazole) which had a BE of 97.5, 97.2, and 96.9%, respectively (Fuentes-Dávila *et al*., 2016). The combination of triazol-strobilurin fungicide Consist Max 500 SC (tebuconazole + trifloxystrobin), was evaluated at several rates for control of karnal bunt, and had a range of BE of 76 to 89%, while Opus had 93.3% (unpublished). Another combination of triazol-strobilurin fungicide under the commercial name of Juwel CS (epoxyconazole + kresoxim-methyl) was evaluated; its BE was 97.7% in comparison to the triazol fungicides Opus, Bemistop EC (propiconazole), and Folicur EW (tebuconazole) which had a BE of 98.2, 95.4, and 95.2%, respectively (Fuentes-Dávila *et al*., 2018). In this study, the three combination of triazol-strobilurin Approach Prima CS (picoxystrobin + cyproconazole), Priori Xtra CS (azoxystrobin + cyproconazole), and Maxtrobin Xtra CS (tebuconazole + azoxystrobin) showed a BE of 98.5, 99.2, and 85.3%, respectively, in comparison to the triazol fungicide Folicur with 98.9%. Although, the highest BE was shown by Priori Xtra, the rate used doubled those of the other combinations and it was higher than the rate of Folicur. The use of a single strobilurin active ingredient (pyraclostrobin) had a difference of 10.6% BE lower than the triazol fungicide (tebuconazole), while the combination trifloxystrobin with tebuconazole had 4.3% BE lower than epoxyconazole. Similarly, the combination kresoxim-methyl with epoxyconazole had 0.5% BE lower than epoxyconazole. From these results, there is no synergistic effect by the combination of fungicides from the two chemical groups, and the triazol products show better control of karnal bunt, although, statistically they might not be different. The a thousand grain weight did not have significant differences among treatments (Fig. 5); Folicur treated plots had a range of 45.3-54.4 with an average of 51.1 g, those from the untreated check 53.1-54.9 with an avg. of 53.6 g, Approach Prima 52.8-56.8 and the avg. 54.8 g, Maxtrobin Xtra 52.6-57.3 and avg. 55.1 g, and Priori Xtra 54.1-56.7 and avg. 55.3 g. The application of the different products did not cause any adverse effect on the growth and development of treated plants.

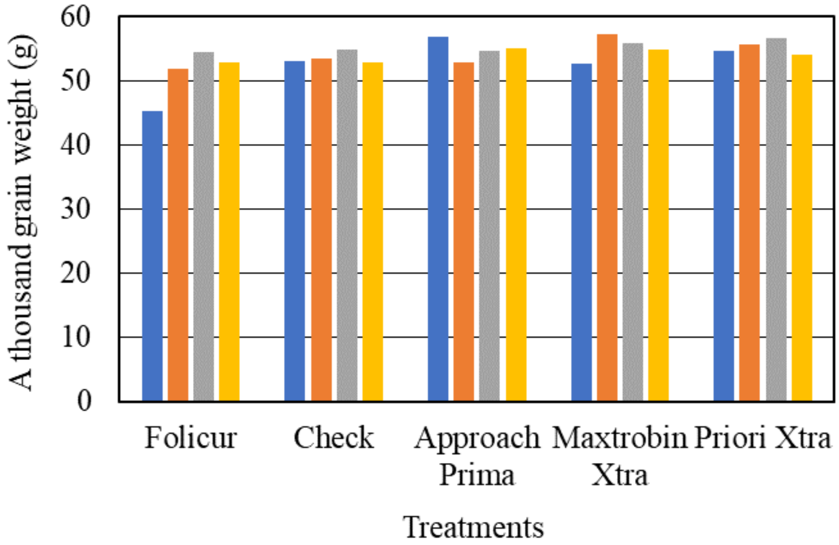

Figure 5. A thousand grain weight of bread wheat cultivar Tacupeto F2001, in four plots treated with each of four fungicides and an untreated check, during the 2018-2019 crop season, at the Norman E. Borlaug Experimental Station in the Yaqui Valley, Sonora, Mexico.

4. CONCLUSION

The biological effectiveness of Priori Xtra, Folicur, Approach Prima, and Maxtrobin Xtra for control of karnal bunt of wheat by foliar applications during heading-flowering-anthesis was 99.2, 98.9, 98.5, and 85.3%, respectively, being statistically similar. The a thousand grain weight was 55.3, 51.1, 54.8, and 55.1 g, respectively, and 53.6 g for the untreated inoculated check. According to the EWRS scale, no phytotoxicity was detected on the wheat plants treated with the four fungicides.

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