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Fosphite 53 SL (Monopotassium + dipotassium 530g/L) fungicide on the Development of Stem Rust (*Puccinia graminis* Pers f.sp *tritici*) Infection on Wheat (*Triticum aestivum* L.)

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Abstract Stem rust (*Puccinia graminis* f.sp *tritici*) of wheat (*Triticum aestivum* L.) is a major threat to wheat production. The objective of the study was to determine the effects of fungicide on the development of stem rust infection. *Kwale* cultivar was grown in the field at KALRO-Njoro in a Randomized Complete Block Design (RCBD) split-plot arrangement for main-rain (June to November, 2014) and off-rain season (February to May, 2015) by applying three treatments of Fosphite 53 SL (Monopotassium + dipotassium 530 g a.i. L^{-1}) (5L ha^{-1}, 7.5L ha^{-1}, 10L ha^{-1}), untreated plot and a standard check (Folicur 25 EW at 2.5L ha^{-1}) at three different growth stages of the wheat cultivar *Kwale*. Evaluation of disease severity was based on the modified Cobbs scale and expressed in terms of Area Under Disease Progress Curve (AUDPC). Data on yield and yield components were taken at harvest and analyzed using Statistical Analysis System (SAS) and the mean comparisons based on Least Significant Difference (LSD) at 5% probability for the separation of season, growth stage and fungicide rates. There was significant ($P \leq 0.05$) effect (season × crop stage) for AUDPC-YR and ($P \leq 0.001$) for plant height. Main-rain season, 2014 had higher values for AUDPC-SR and AUDPC-YR at 40.579% and 36.777% and lower values for plant height, tillers, spike length, spikelet per spike, TKW, biomass and yield by 17.127%, 36.874%, 19.543%, 1.27%, 3.26%,39.263%, and 40.148%, respectively compared to off-rain season, 2015. Tillering stage had lower values for AUDPC-SR and AUDPC-YR by 3.41% and 17.617% and high yield and grain weight as compared to stem elongation and heading stage. Of all the tested rates of Fosphite 53 SL (Monopotassium + dipotassium 530 g a.i. L^{-1}) only the rate of 0.5L ha^{-1} showed significant reduction in the disease pressure and positive influence in the yield and grain weight. The results suggest that spraying of Fosphite 53 SL at 0.5L ha^{-1} at tillering stage is effective in controlling stem rust hence, the adoption of effective fungicides to control stem rust pathogen and their application at the right stage of the crop can be used in reducing stem rust severity and increase yield of susceptible wheat cultivars in stem rust occurring areas.

Keywords: *cultivar, fungicide, stem rust, wheat*

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1. Introduction

Stem rust caused by *Puccinia graminis* f.sp. *tritici* (Pgt) is a destructive disease of wheat which occurs frequently in warm and humid environments such as those found in Kenya. A new and significant threat has been posed in the Eastern Africa region, due to reemergence of a new virulent race, TTKS (*Ug99*) which broke down resistance genes that had been used to control the disease [14]. In the early 1990's, wheat stem rust epidemics were experienced in Kenya following the breakdown of resistance (*Sr*36) in the popular cultivar Enkoy. A new pathotype of Pgt was detected in Eastern Africa in 1999 (*Ug99*) [9]. Since then, *Ug99* has been detected in Uganda, Kenya and Ethiopia. The pathotype has caused considerable concern because of its broad virulence spectrum that include gene *Sr*31, one of the most widely deployed stem rust resistance gene that remained effective until the detection of *Ug99*. Growing wheat in diverse agro-ecological zones in Kenya throughout the year is a major contributing factor to the pool of stem rust inoculum [2]. It is therefore difficult to prevent or reduce infection of susceptible cultivars. Regular occurrence of disease outbreaks in Kenya are due to favourable environmental conditions hence the designation of the country as a "hotspot" for stem rust [15]. Genetic resistance has provided adequate protection against stem rust disease for over 30 years [6]. However, the susceptibility of most wheat cultivars and heavy yield losses exceeding 80% [16] incurred by farmers has entailed the adoption of an integrated management strategy, hence the necessity to evaluate the available fungicides for efficacy against stem rust pathogen. Widespread growing of resistant cultivars to control the disease effectively has been adopted [4]. It is impossible to grow a profitable crop of wheat without fungicide application as all the current commercial wheat cultivars are susceptible to the new race. A stem rust severity level greater than 5% in hot-spot regions should be controlled to reduce yield losses. In the past there was no need for fungicide since there was widespread deployment and cultivation of resistant cultivars providing adequate protection [6]. Fungicides can be used as a short term management of the disease until new cultivars with genetic resistance become available. Fungicide treatments, if applied under high and moderate disease pressure at critical growth stages, may reduce large yield losses by suppressing or eliminating stem rust pathogens [7].

Stem rust caused 32 to 57% of grain yield loss and 17 and 24% of thousand kernel weight reduction [15]. The use of fungicide to control wheat stem rust specifically related to stem rust race *Ug99* and its variants, has very little published information. Epidemics of leaf rust were more intense in Uruguay in 2010 and in main wheat growing areas of Argentina, but caused moderate epidemics in all countries in 2008 and 2009. In areas favorable for disease development, leaf rust can cause grain yield losses higher than 50% in severe epidemics if fungicides are not applied. Majority of cultivars are either susceptible or moderately susceptible, the fungicide application cost of controlling leaf rust in affected regions totals 50million US dollars. Farmers without resources have the option of using fungicides against *Ug99* as a last resort, however, this option is too expensive for many smallholder farmers in the developing world making them rely entirely on resistance to control wheat diseases [5].

2. Materials and Methods

2.1. Experimental Site

During the rainy season of 2014 (June-November), and dry season of 2015 (February-May), an experiment was conducted at the Kenya Agricultural and Livestock Research Organization (KALRO) Njoro, (0 20′S, 35^{o} 56′E) at 2185 m above sea level with mean minimum and maximum temperatures of 9.7°C and 23.5°C, respectively and an average annual rainfall of 900 mm. The site was chosen because environmental conditions are conducive for stem rust infection and it is a representative of wheat and barley growing areas [12].

2.2. Genotype

The cultivar *Kwale*, adapted cultivar, was used for fungicide evaluation. The cultivar shows high yield, medium maturity, but susceptibility to stem rust.

2.3. Experimental Procedure

Experimental plots were disc ploughed and harrowed twice to fine tilth suitable for wheat growing. Seeds were sown in plots of four rows of 80cm × 2.0m. Sowing was made by a seed driller at the seeding rate of 108.3kg ha^{-1}. At sowing time, diammonium phosphate (DAP) fertilizer was applied at the rate of 125kg ha^{-1} in order to supply an equivalent of 22.5kg N ha^{-1} and 25kg P ha^{-1}. A randomized Complete Block Design (RCBD) of split plot arrangement with three replicates was used. The experimental plots and rows were separated by 0.5m wide paths within and between the blocks. The fungicide was sprayed at three stages of the plant growth at three rates: Fosphite 53 SL (Monopotassium + dipotassium 530 g a.i. L^{-1}) at 5L ha^{-1}, Fosphite 53 SL Fosphite 53 SL (Monopotassium + dipotassium 530 g a.i. L^{-1}) at 7.5L ha^{-1} and Fosphite 53 SL Fosphite 53 SL (Monopotassium + dipotassium 530 g a.i. L^{-1}) at 10L ha^{-1}. One standard fungicide: Folicur 25 EW (tebuconazole 250 g a. i. L^{-1}) was applied at the rate of 2.5L ha^{-1} and non-treated control was used for comparison. At tillering stage (Zadok's growth stage 20-29) (Zadok's et al., 1974) the plots were sprayed with Buctril MC (bromoxynil ectanoate 225g ha^{-1} and MCPA Ethyl Hexyl Ester 225 g ha^{-1}) to control broad-leave weeds. The trial was top dressed with Calcium Ammonium Nitrate (CAN) at stem elongation (Zadok's GS30) [19] at the rate of 100kg ha^{-1} in order to supply additional 33 kg N ha^{-1}. Bulldock (beta-cyfluthrin) was sprayed to control pest infestation at the rate of 31 g ha^{-1} and repeated at 14-day intervals when required.

2.4. Data Collection

Stem rust severity scoring begun when the plants had attained 50% susceptibility based on modified Cobbs scale where 0% = immune, and 100% = completely susceptible [8] for three readings with an interval of 10 days between heading (GS 50-69) and plant maturity (GS 70-89) [18]. Plant response to rust infection at the adult plant stage was termed "infection response". Based on the size of pustules and the associated necrosis or chlorosis, infection responses were classified into four discrete categories: R = resistant, MR = moderately resistant, MS =moderately susceptible, S = susceptible, MRMS denoted an infection response that overlaps the MR and MS categories [10]. Other agronomic traits such as date of emergence, date of flowering and date of maturity were recorded; 5 plants in the two middle rows were measured when 50% of the plants per entry had headed for parameters such as, plant height, measured from the base at ground level to the tip of plant; number of tillers were counted manually; spike length was measured from the first node where the first spikelet emerges to the spike tip; number of spikelet/spike were counted; 100 scale, while the yield was determined after drying to 12.5% HR.

2.5. Data Analysis

The data for the yield and agronomic traits, once collected and organized, was analyzed using Statistical Analysis System 9.4 portable version (SAS Institute, Inc 2012) using the following statistical model:

$$Y_{ijklm} = \mu + S_i + R_{j(i)} + F_k + SF_{ik} + V_l + SV_{il} + FV_{kl} + SFV_{ikl} + \varepsilon_{ijklm}$$

Where:

Y_{ijklm} = Observation of experimental units, μ = Overall mean, S_i = i^{th} effect due to season $R_{j(i)}$ = effect due to j^{th}

replicate, F_k = effect due to k^{th} fungicide rates, SF_{ik} = i^{th} effect due to season and k^{th}effect due to fungicide rate, V_l = effect due to l^{th} crop stage, SV_{il} = i^{th} effect due to season and l^{th} effect due to crop stage, FV_{kl} = k^{th} effect due to fungicide rate and l^{th} effect due to crop stage, SFV_{ikl} = i^{th} effect due to season, k^{th} effect due to fungicide rate and l^{th} effect due to crop stage, ε_{ijklm} = random error components.

Area under Disease Progress Curve (AUDPC) was used. It was computed using the formula by [18] and AUDPC CIMMYT programme

$$AUDPC = \sum_{i=1}^{n-1}[(t_{i+1} - t_i)(y_i + y_{i+1})/2]$$

Where;

t_i is the time in days of each reading, $y_i$is the percentage of affected part of the plant at each reading, n is the number of readings, $t\ (i+1)$ is the second assessment date of two consecutive assessment, $y\ (i+1)$ is the disease severity on assessment date $t\ (i+1)$.

3. Results

Stem rust and yellow rust infection was observed across the two seasons and was high in dry season (February to May, 2015) compared to rainy season (June to November, 2014). There was a variation in the time of disease set in in the two seasons, with the+ disease setting in earlier.

The effect of season were significant (P≤0.001) for AUDPC-SR, AUDPC-YR, plant height, tillers, spike length, biomass and yield. Significant (P≤0.01) effects (fungicide rate) were observed for AUDPC-SR and AUDPC-YR. Crop stage showed significant (P≤0.01) for AUDPC-YR. There was significant (P≤0.05) effect (season × crop stage) for AUDPC-YR and (P≤0.001) for plant height. In addition, season × fungicide rate × crop stage interaction were significant for plant height (Table 1).

Effect of fungicide rates on wheat.

Mean values for the parameters observed for the different fungicide rates are given in Table 2. The effect of fungicide rate was noted on AUDPC-SR, AUDPC-YR, tillers, spike length and TKW.

Table 1. Mean squares of season, replicate, variety and variety × season on Audpc-SR, Audpc-YR, plant height, number of tillers, spike length, spikelet per spike, thousand kernel weight, biomass and yield on wheat cultivar Kwale Njoro, 2014-2015

Source of variation	df	AUPC-SR	AUDPC-YR	Plant Height (cm)	No of Tillers	Spike length (cm)	No of Spikelet/spike	Biomass (g)	Thousand Kernel weight (g)	Yield (g)
Season	1	141134.4000***	72080.100***	4536.900***	774.400***	74.547***	6.944	131915.945***	12.844	21464.071***
Rep	2	239.575	5368.125	12.678	16.678	1.579	90.844	12219.677*	5.907	1080.075
Fungicide rate	4	7110.301***	32332.774***	33.183	22.517	0.766	20.156	1356.5839	19.393	144.987
Crop stage	2	696.408	21197.033**	14.411	14.878	0.093	2.178	5076.848	6.797	871.599
Season × Crop stage	2	1015.808	11024.533*	286.233***	8.633	1.041	45.378	4178.548	7.213	812.806
Fungicide rate × Crop stage	8	158.287	2240.019	32.133	17.767	0.712	39.997	31.83.480	6.469	833.128
Season × Fungicide rate × Crop stage	12	996.311	5210.647	99.706**	11.028	0.510	21.317	3323.430	8.259	797.088
Error	58	546.917	3397.760	32.103	9.256	0.648	45.879	2887.071	6.623	522.491
Total	89									
R^2		0.855	0.644	0.784	0.694	0.716	0.261	0.595	0.423	0.591
Cv		21.986	22.699	7.474	23.442	9.580	15.659	34.282	10.937	37.175

df – degrees of freedom; Audpc – Area Under Disease Progress Curve; SR - stem rust; YR – yellow rust *, **, *** represents significance at P<0.05, P<0.01 and P<0.001, respectively.

Table 2. Effect of fungicide treatments on AUDPC-SR, AUDPC-YR, plant height, number of tillers, spike length, spikelet per spike, thousand kernel weight, biomass and yield on wheat cultivar Kwale Njoro, 2014-2015

Source of variation (Fungicide rates)	AUDPC-SR	AUDPC-YR	Plant Height (cm)	No. of Tillers	Spike length (cm)	No of Spikelet/spike	Thousand Kernel weight (g)	Biomass (g)	Yield (g)
Folicur 0.25L ha^{-1}	71.222b	182.220b	76.111a	14.167a	8.315ab	42.444a	25.333a	161.880a	64.357a
Untreated	119.861a	271.690a	72.833a	10.978ab	7.939b	40.389a	20.268b	168.720a	44.702b
Phosphite SL 0.5L ha^{-1}	111.667a	268.220a	73.833a	12.667ab	8.388ab	42.667a	22.815b	147.130a	58.832a
Phosphite SL 0.75 L ha^{-1}	115.278a	288.220a	75.111a	11.667b	8.473ab	44.111a	22.951b	150.650a	59.174a
Phosphite SL 1.0 L ha^{-1}	113.806a	273.640a	77.167a	14.111a	8.699a	44.667a	23.266b	155.300a	60.371a
General mean	106.637	256.800	75.811	12.978	8.403	43.256	23.531	156.734	61.487
$LSD_{0.05}$	15.604	38.894	3.781	2.030	0.5371	4.520	1.717	35.852	15.252

Means with the same letters are not significantly different from each other at P≤0.05.

For Fosphite 53SL application at 0.5L ha^{-1}, disease decrease was 6.836% and 1.277% for AUDPC-SR and AUDPC-YR, respectively, while mean for TKW and yield was 12.567% and 31.609% higher than the control, respectively. The rate registered a higher percentage for mean decrease in AUDPC for SR.

Fosphite 53SL application at 0.75L ha^{-1}÷ showed no effect on control of stem rust and yellow rust. Also, there was a reduction in % mean increase in TKW and yield.

Fosphite 53SL application at 1.0L ha^{-1}: Disease management for stem rust was low at 5.05% over the control and -0.718% for yellow rust. The rate registered highest % in TKW at -14.792% over the control with the highest yield increase at -35.052% for all the product rates.

Folicur EW 250 application at 0.25L ha^{-1}: The mean % disease decrease for AUDPC for stem rust and yellow rust registered higher % at 40.580 and 32.930, respectively. It also had higher mean % increase in TKW and yield at -

25.960 and -43.969, respectively. The mean performance was significantly different from the Fosphite 53SL rates.

Effect of crop stage on the effectiveness of fungicide.

Table 3, shows mean values for the parameters observed for the three stages (tillering, stem elongation and heading) while the reduction was observed for yellow rust at tillering and stem elongation stage. There was reduced stem rust severity at all the growth stages: tillering, stem elongation and heading. Heading stage showed higher values for AUDPC-SR, AUDPC-YR, plant height, tillers, spike length and TKW. All the other parameters were not significantly different from each other for the three crop stages. Tillering stage had lower values for AUDPC-SR and AUDPC-YR by 3.41% and 17.617%, while it had high yield weight as compared to stem elongation and heading stage.

Table 3. Effect of three growth stages on AUDPC-SR, AUDPC-YR, plant height, number of tillers, spike length, spikelet per spike, thousand kernel weight, biomass and yield on wheat cultivar Kwale Njoro, 2014-2015

Source of variation (Growth Stage)	AUDPC-SR	AUDPC-YR	Plant Height	No. of Tillers	Spike length (cm)	No. of Spikelet/spike	Thousand Kernel weight (g)	Biomass (g)	Yield (g)
Tillering	107.033a	236.30b	75.533a	12.167a	8.385a	43.500a	23.373a	169.55a	65.898a
Stem Elongation	101.250a	247.27b	75.300a	13.333a	8.359a	42.967a	23.153a	143.54a	55.479a
Heading	110.817a	286.83a	76.600a	13.433a	8.466a	43.300a	24.065a	157.11a	63.085a
General mean	106.367	256.800	75.811	12.978	8.403	43.256	23.531	156.734	61.487
$LSD_{0.05}$	12.087	30.127	2.928	1.572	0.416	3.501	1.330	27.771	11.814

Means with the same letters are not significantly different from each other at $P \leq 0.05$.

Effect of season on the effectiveness of fungicide on wheat

Mean values for the parameters observed in two different seasons are given in Table 4. There was high disease pressure in main-rain (June to November, 2014) compared to off-rain (February to May, 2015) season by 12.472%. The effect of season was noted on AUDPC-SR, AUDPC-YR, plant height, tillers, spike length, biomass and yield. Off-rain, 2014 season had higher values for AUDPC-SR and AUDPC-YR at 40.579% and36.777% and lower values for plant height, tillers, spike length, spikelet per spike, TKW, biomass and yield by 17.127%, 36.874%, 19.543%, 1.27%, 3.26%, 39.263%, and 40.148%, respectively as compared to main-rain season. There was no significant effect of season on spikelet per spike and TKW.

Table 4. Means of main-rain, 2014 and off-rain, 2015 for AUDPC-SR, AUDPC-YR, plant height, number of tillers, spike length, spikelet per spike, thousand kernel weight, biomass and yield on wheat cultivar Kwale Njoro

Source of variation (Season)	AUDPC-SR	AUDPC-YR	Plant Height	No of Tillers	Spike length (cm)	No of Spikelet/spike	Thousand Kernel weight (g)	Biomass (g)	Yield (g)
Main-rain season, 2014	166.767b	285.10a	68.711b	10.044b	7.493b	42.978a	23.108a	118.45b	46.044b
Off-rain season, 2015	145.967a	228.50b	82.911a	15.911a	9.313a	43.533a	23.953a	195.02a	76.930a
General mean	106.367	256.800	75.811	12.978	8.403	43.256	25.531	156.734	61.487
$LSD_{0.05}$	9.869	24.598	2.391	1.284	0.340	2.858	1.086	22.675	9.646

Means with the same letters are not significantly different from each other at $P \leq 0.05$.

4. Discussion

Fungicides can play a major role in the integrated management of the disease, until new cultivars with genetic resistance are available [6]. The use of fungicide to control stem rust disease has shown 50% higher yield in treated versus untreated plots in various wheat growing regions of Kenya [15]. Use of resistant cultivars remains the only alternative in reducing stem rust severity because of the high costs in using fungicides and the serious health problem it causes for the users and environment. Most of the fungicides tend to be effective at specific growth stages [1]. Sometimes, crops that receive three poorly timed sprays suffer as much disease as the untreated crops, suggesting that either the fungicide are applied too late, or do not effectively control the disease. There was no evident fungicide phytotoxicity in our treatments. Wheat cultivar, *Kwale*, used in the experiment did not show any visible injurious effects. There was no chlorosis of wheat in all the treated plots as well as in the plots that were not sprayed with fungicides. This showed that all the test rates of Fosphite 53SL and Folicur 250 EW were safe on *Kwale*.

The study was carried out at KALRO-Njoro which is known to have favorable weather conditions for stem rust of wheat. Plots that had the highest disease score as shown by the AUDPC, had low grain yield and TKW. This concurs with the study by [17] that reported plots with the highest AUDPC had low grain yield, thousand grain and test weights. Application of fungicide at critical growth stages under high and moderate disease pressure, may reduce large yield losses by suppressing or eliminating stem rust pathogen.

In the study, of all the tested rates of Fosphite 53 SL only the rate of 0.5L ha^{-1} showed significant reduction in the disease pressure and positive influence in the yield and grain weight. Folicur 250 EW which was the positive control was more effective than Fosphite 53 SL in reducing disease and increasing yield. There was inconsistency in the effectiveness of Fosphite 53 SL. In a study by [6] it was reported that Folicur (tebuconazole) was more effective than Triad (triadimefon) or Impact (flutriafol) in disease reduction and yield increase hence the variation in the efficacy of different fungicides in controlling stem rust. Regions with favourable weather conditions for stem rust occurrence, with stem rust severity level greater than 5% under field conditions should be controlled to reduce yield losses [6]. The three growth stages (tillering, stem elongation and heading) did not show any variation in the reduction of stem rust severity, while there was a low score for yellow rust severity at tillering stage and a high yield. This is consistent with the study made by [15,17] which showed that spraying at tillering and flowering growth stages of the crop, higher rates and more fungicide applications could achieve greater rust control and greater yield increases. Stem rust is very severe in susceptible varieties when it begins to develop in the crop before flowering. There is a clear relationship between grain yield and disease severity by demonstrating that prevention of a 1% increase in rust severity saved 2% loss in grain yield [7].

5. Conclusion

The study displayed that wheat grain yield of susceptible cultivars could be highly reduced by stem rust and yellow rust, therefore, the adoption of foliar fungicides to combat stem rust disease as a short term control strategy until resistant cultivars are developed is encouraged in Kenya. Growth stage was found to be an important factor in fungicide management of wheat rusts. Fungicidal activity of Folicur 25 EW and Fosphite 53 SL could have played a major role in their efficacy differences. The ability to suppress disease development and protect the crop canopy, which is vital for dry matter accumulation and yield significantly differ among fungicides [13]. Fosphite 53 SL foliar fungicide applied at tillering growth stage at the rate of 0.5L ha^{-1} was the most appropriate in effectively reduced/controlled the rust infection and increased grain yield. This study showed that foliar fungicide can be used to manage wheat rust as a short term control strategy. However, more research should be done to identify the right timing of application and the dosage.

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Competing Interests

The Authors have no competing interest.

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