Sequential Post-Heading Applications for Controlling Wheat Blast: A Nine-Year Summary of Fungicide Performance in Brazil

João P. Ascari[1†], Jhonatan P. Barro[1†], Flávio M. Santana[2], José M.V. Padua[3], João L. N. Maciel[2], Douglas Lau[2], Gisele A. M. Torres[2], Cheila C. Sbalcheiro[2], Claudine D. S. Seixas[4], Augusto C. P. Goulart[5], Angelo A. B. Sussel[6], Carlos A. Schipanski[7], Débora F. Chagas[7], Maurício A. O. Coelho[8], Tatiane D. N. Montecelli[9], Daniel R. Amaral[10], Adriano A. P. Custódio[11], Lucas S. O. Moreira[11], Carlos M. Utiamada[12], Wilson S. Venâncio[13], Rita C. S. Goussain[14], Kaique S. Alves[1], Emerson M. Del Ponte[1*]

[1] Departamento de Fitopatologia, Universidade Federal de Viçosa, Viçosa, 36570-900, MG, Brazil

[2] Embrapa Trigo, Passo Fundo, 99050-970, RS, Brazil

[3] Departamento de Agricultura, Universidade Federal de Lavras, Lavras, 37200-900, MG, Brazil

[4] Embrapa Soja, Londrina, 86001-970, PR, Brazil

[5] Embrapa Agropecuária Oeste, Dourados, 79804-970, MS, Brazil

[6] Embrapa Cerrados, Planaltina, 73310-970, DF, Brazil

[7] G12 Agro Pesquisa e Consultoria Agronômica, Guarapuava, 85015-344, PR, Brazil

[8] Empresa de Pesquisa Agropecuária de Minas Gerais, Patos de Minas, 38700-970, MG, Brazil

[9] Coodetec, Cascavel, 85818-650, PR, Brazil

[10] Instituto Federal do Triângulo Mineiro Uberaba, 38064-790, MG, Brazil

[11] Área de Proteção de Plantas, Instituto Agronômico do Paraná, Londrina, 86047-902, PR, Brazil

[12] Tagro, Londrina, 86070-460, PR, Brazil

[13] Estação Experimental Agrícola Campos Gerais, Palmeira, 84130-000, PR, Brazil

[14] Instituto Federal do Mato Grosso, Campo Verde, 78840-000, MT, Brazil

[†] These authors contributed equally

*Corresponding author: delponte@ufv.br

Abstract

Wheat blast, caused by *Pyricularia oryzae Triticum* (PoT) lineage, is a major constraint to wheat production, mainly in the tropics of Brazil where severe epidemics have been more frequent. We analyzed disease and wheat yield data from 42 uniform field trials conducted during nine years (2012 to 2020) in order to assess whether the percent control and yield response were influenced by fungicide type, region (tropical or subtropical), and year. Six treatments were selected, all evaluated in at least 19 trials. Two fungicides were applied as solo active ingredients: MANCozeb, and TEBUconazole, and four were premixes: AZOXystrobin + TEBU, TriFLoXystrobin + PROThioconazole, TFLX + TEBU, and PYRAclostrobin + EPOXiconazole. Percent control, calculated from back-transforming estimates by a meta-analysis network model fitted to the log of the means, ranged from 43 to 58%, with all but PYRA + EPOX showing efficacy greater than 52% on average, not differing among them. The variation in both efficacy and yield response was explained by region and all but TEBU performed better in the subtropics than in the tropics. Yield response from using three sequential sprays was around two times greater in the subtropics (319 to 532 kg/ha) than in the tropics (149 to 241.3 kg/ha). No significant decline in fungicide efficacy or yield response was observed in nine years of study for any of the fungicides. Our results reinforce the need to improve control by adopting an integrated management approach in the tropics given the poorer performance and lower profitability, especially for the premixes, than in the subtropics.

Keywords: *Pyricularia oryzae*, chemical control, profitability, meta-analysis

Introduction

Wheat blast is caused by *Pyricularia oryzae Triticum* lineage (PoT, syn *Magnaporthe oryzae*, MoT), an ascomycete fungus first found in 1985 in the state of Paraná, Brazil (Igarashi et al. 1986). Since then, wheat blast has spread to other wheat-growing countries in South America, including Bolivia, Paraguay, Argentina, and Uruguay (Cruz and Valent 2017; Ceresini et al. 2018). About thirty years after its discovery, the global concern with wheat blast epidemics increased significantly after its report in South Asia (Malaker et al. 2016), and East Africa (Zambia) (Tembo et al. 2020).

The fungus infects both the leaves and the heads of the wheat plant, but infections that occur during the reproductive stage are more frequent and of major economic significance; complete bleaching of heads may occur, severely affecting grain filling (Cruz and Valent 2017; Ceresini et al. 2018). Foliar infections are generally mild in wheat but may increase in severity, and importance for head infections, when warm and wet weather prevails during the early season (Cruz et al. 2016; Perelló et al. 2017). An early study reported a 27% reduction in yield due to wheat blast (Goulart et al. 1990), but yield losses up to 70% may occur depending on the infection timing, weather conditions, and cultivar susceptibility (Goulart and Paiva 2000).

Although the disease is present over a wide range of latitudes, the most severe epidemics have been reported in wheat-growing regions in the tropics, during the summer/fall season sowings, such as those in Central Brazil and Bolivia (Pagani et al. 2014; Cruz and Valent 2017), and more recently, in Bangladesh (Islam et al. 2019). Because epidemics are favored by the occurrence of temperatures between 25°C and 30°C, 10 hours or more of leaf wetness, and 24 to 48 hours of relative humidity >90%, wheat blast epidemics are particularly more damaging in the tropics than in the subtropics (Uddin et al. 2003; Cardoso et al. 2008; Silva et al. 2021).

Once the wheat blast pathogen is introduced, the necrotrophic fungus is capable of infecting and surviving in other grasses nearby wheat fields which are assumed to serve as a source of inoculum for epidemics in wheat if they are present prior to the wheat season (Tosa and Chuma 2014; Urashima et al. 2017; Ceresini et al. 2019). The ability of the fungus to disperse over long distances is not entirely known but evidence has shown that *Pyricularia* spores can disperse through air currents up to 1 km distant from the source (Urashima et al. 2007), or across continents via infected seeds (Ceresini et al. 2018). Although several control methods have been explored in research, including biological control agents (Chakraborty et al. 2020a, 2020b), the combination of cultural (Coelho et al. 2016), nutritional (Xavier Filha et al. 2011; Silva et al. 2016), genetic, and chemical control have been commercially feasible and more effective in disease management (Cruz et al. 2019; Cruppe et al. 2020).

Several chemicals have been evaluated for the control of the disease since the first blast epidemics in Brazil (Goulart and Paiva 1993; Santana et al. 2013). These studies have shown that two to three sequential applications of fungicides may be required for disease control, yet with relatively modest levels of control being achieved (Santana et al. 2013). Currently, fifty-three commercial fungicides have been registered for wheat blast control in Brazil (AGROFIT 2021). These include Quinone outside Inhibitor (QoI) and Demethylation Inhibitor (DMI) marketed solely or mixed together, but also multi-site mode of action fungicides such as mancozeb.

Despite the importance of wheat blast, the number of publications on chemical control of wheat blast is relatively small and the results are inconsistent. Successful control with efficacy as high as 90% has been achieved when combining the use of QoI + DMI premix and less susceptible cultivar (Rios et al. 2016). However, levels as low as 45% efficacy have been reported in susceptible cultivars grown at very favorable environments for blast outbreaks in the Brazilian Cerrado (Pagani et al. 2014). In Bangladesh, 19-commercial fungicides had their efficacy tested on a susceptible cultivar, and the blast control levels ranged from 43 to 96% (Roy et al. 2020). A 23-environment fungicide trial conducted in Brazil and Bolivia reported various levels of efficacy for QoI, DMI, and multi-site fungicides depending on disease pressure (Cruz et al. 2019).

Meta-analytic approaches have become standard to summarize the effect of treatments for fungicide testing data, including wheat diseases (Paul et al. 2008, 2018a; Machado et al. 2017; Barro et al. 2019, 2020). Uniform fungicide trials targeting wheat blast control were established in 2011 and performed yearly following a standardized protocol. The main goal of the network is to gather information on disease control and yield loss prevention across the main growing regions. The data have been analyzed and published as technical reports (Santana *et al.*, 2014; 2016a; 2016b; 2019a; 2019b; 2020a; 2020b). Data from the first three years of the cooperative trials have been published in combination with Bolivian data (Cruz et al. 2019), but several questions remained, including those related to differences between regions, years and profitability of fungicide applications. Using data collected from an additional six-year period, totaling 26 new trials, our objectives were to: 1) obtain meta-analytic estimates of wheat head blast control efficacy and yield response; 2) evaluate whether the estimates vary over years, regions, and on different levels of disease or yield; and 3) calculate fungicide profitability based on the break-even probabilities.

Material and Methods

Data source. Data were obtained from 44 field trials conducted by researchers of the wheat blast cooperative fungicide trial network (Rede de Ensaios Cooperativos de Brusone do Trigo) during nine years (seasons) (2012 to 2020). The data have been published primarily in yearly single reports for ranking fungicide efficacy (Santana et al. 2013, 2014, 2016a, 2016b, 2016c, 2019a, 2019b, 2020a, 2020b). Data from the 2020 season has not been published.

The trials were conducted in eleven municipalities across six Brazilian States: Paraná (PR), São Paulo (SP), Distrito Federal (DF), Mato Grosso (MT), Mato Grosso do Sul (MS), and Minas Gerais (MG) (Fig. 1). The trials were grouped into two climatic regions: Tropical (19 trials [DF, MT, MS, MG]) and Subtropical (25 trials [PR, SP]).

Experimental procedures. A susceptible variety, adapted for the respective region, was used in the trials (data not shown), and all agronomic practices (fertilization, weeds, and pest control) were performed according to regional recommendations (Comissão Brasileira de Pesquisa de Trigo e Triticale 2020). The trials were conducted following a completely randomized block design with four replications (a plot of 12 m^2 was a replication). All fungicides were applied three times, starting at the heading stage (60 of Zadoks growth stages) (Zadoks et al. 1974), and following 7 to 10 days apart. A backpack sprayer pressurized by CO_2 calibrated to spray 200 L/ha was used to perform the applications.

Wheat blast and grain yield assessments. Head blast incidence (INC) (proportion of diseased head) and conditional severity (SEV) (proportion of diseased spikelets in the diseased head) (Maciel et al. 2013) were visually assessed in 1-m of each of the two central rows, at wheat grain soft dough stage (85 of Zadoks growth stages) (Zadoks et al. 1974). Wheat blast index (WBI) was calculated as WBI = (INC*SEV)/100. Yield was obtained by harvesting the central rows (4 m^2) at full maturity. Grain weight and moisture were obtained for each treatment plot (fungicide + untreated). Crop yield was expressed in kg/ha at 13% moisture.

Fungicide treatment selection. To be included in the analysis, a fungicide treatment should have been tested in at least 19 trials conducted in at least four years and compared with an untreated check in the same trial. Six fungicides met the criteria, including four DMI + QoI premixes, and two single active ingredients (Table 1). After treatment selection, WBI and grain yield data were available in 42 trials each.

Network meta-analysis and inconsistency. The data were available at the plot level for all treatments for each variable of interest (WBI and grain yield). These were aggregated at the trial level, which is a typical approach used in the meta-analysis (Madden et al. 2016). We fitted an arm-based network model, also known as a two-way unconditional linear mixed model, directly to the treatment means (log-transformed or untransformed) to further obtain control efficacy and yield response (Paul et al. 2008; Machado et al. 2017). Given the statistical properties of the data (Supplemental Fig. S1), means of WBI were log-transformed, while no transformation was required to obtain the mean difference in grain yield. The arm-based model can be written as

$$\boldsymbol{Y_i} \sim \boldsymbol{N}(\boldsymbol{\mu}, \boldsymbol{\Sigma} + \boldsymbol{S_i}) \,, \qquad (1)$$

where Y_i is the vector of L (log of the means of WBI) or absolute yield (D) for the six treatments plus the untreated check for the ith study, μ is a vector representing the mean of Y_i across all studies, Σ is a 7 x 7 between-study variance-covariance matrix (for the six treatments plus the untreated check), and S_i is a within-study variance-covariance matrix for the ith study. N indicates a multivariate normal distribution.

For this approach, a measure of within-study variability is required to weight studies based on the inverse function of the sampling variance (Paul et al. 2008, 2010). Given the availability of data at the plot level, the within-study variability of L and D was calculated from the mean square error (MSE) obtained from a linear mixed model fitted to each individual trial, as described (Machado et al. 2017). Maximum likelihood estimation models were fitted to the data using the *rma.mv* function of *metafor* package (Viechtbauer 2010) of R (R Core Team 2021).

The yield response ($\overline{D}$) was calculated directly after model fitting by subtracting estimated means of fungicide treatment and untreated check (Madden et al. 2016). For percent wheat blast control ($\overline{C}$), we calculated the differences of the estimated means of the logs ($\overline{L}_{IND}$) which equals the ratio of the two means (Paul et al. 2008). The $\overline{C}$ values and their 95% confidence intervals (CIs) were obtained by back-transforming $\overline{L}_{IND}$ and the respective upper and lower limits of their 95% CIs as described in Equation 2.

$$\overline{C} = (1 - \exp(\overline{L}_{IND})) x\ 100 \quad (2)$$

Network inconsistency, or the extent to which different sources of evidence are compatible, is an important component to assess when performing a multi-arm network meta-analysis (Higgins et al. 2012). The most important source is a design-by-treatment interaction, also known as "design inconsistency", which provides a useful general framework for investigating inconsistency (Piepho 2014; Madden et al. 2016). To test for network inconsistency, we used a factorial-type ANOVA model to determine the significance of the *treatment* x *design* interaction, evaluated based on the Wald test statistic. The null hypothesis suggests that the network is consistent (Piepho 2014; Madden et al. 2016). Eight different designs (here design refers to the set of fungicide treatments in the trial) were found in the trials reporting both WBI and yield response (Supplemental Table S1).

Moderator effects. Categorical or continuous moderator variables that could explain, at least a portion of the heterogeneity of the effects across trials, were included in the network model (Equation 1) (Madden et al. 2016). The expanded model is given by

$$Y_i \sim N(\mu + \delta_i, \Sigma + S_i) \ , \quad (3)$$

where δ_i is the vector representing the moderator variable effect for the *i*th study (Paul et al. 2010). All other terms were defined previously at Equation 1.

As categorical, we created baselines for WBI and wheat grain yield based on the median of the mean values in the untreated check. For baseline disease index, the trials were divided into two groups, representing WBI_low (WBI $<$ 10%) and WBI_high (WBI $\geq$10%) disease scenarios. The baseline yield was defined as YLD_low ($<$1200 kg/ha) or YLD_high ($\geq$1200 kg/ha) based on the median yield in the untreated check plot. Finally, we created an additional categorical variable based on climatic region where trials were grouped into Tropical (DF, MT, MS, MG) and Subtropical (PR, SP) regions as mentioned previously (Fig. 1). As continuous moderators, year was included in the model to check whether there was any trend of decline in fungicide efficacy or yield response over time (Dalla Lana et al. 2018).

The moderator variables included in the model were tested using a Wald-type chi-square test to determine if their inclusion directly affected the differences in logs of WBI and the untransformed yield values (Paul et al. 2008).

Economic analysis. Monte Carlo simulations were used to produce distributions of profits that each fungicide would have in each region (Subtropical and Tropical) based on their respective yield return ($\boldsymbol{D}$, kg/ha). The profit was calculated from the difference between the income ($\boldsymbol{I}$, US$/ha) and spraying cost (US$/ha), where $\boldsymbol{I}$ is the product of $\boldsymbol{D}$ and the wheat price ($\boldsymbol{P}$, US$/kg). $\boldsymbol{D}$ was assumed to follow a truncated normal distribution between 0 and +∞, $\boldsymbol{D} \sim \boldsymbol{TN}(\boldsymbol{\mu_D},\boldsymbol{\sigma_D})$, where $\boldsymbol{\mu_D}$ is the mean $\boldsymbol{D}$ given by the estimated yield return ($\overline{\boldsymbol{D}}$), and the $\boldsymbol{\sigma_D}$ is the standard deviation which was given by standard error of ($\overline{\boldsymbol{D}}$), i.e. $\boldsymbol{SE}(\overline{\boldsymbol{D}})$. $\boldsymbol{P}$ was assumed to follow a Gamma distribution, $\boldsymbol{P} \sim \boldsymbol{Gamma}(\boldsymbol{\alpha},\boldsymbol{\beta})$, where $\boldsymbol{\alpha}$ and $\boldsymbol{\beta}$ are the shape and the rate parameters, respectively. To estimate these parameters, we gathered wheat price historical data from the AGROLINK database from January 2012 to May 2021 (AGROLINK 2021). Two methods were used to estimate the parameters: nonlinear regression using least-squares to fit the empirical cumulative distribution function to the gamma cumulative distribution function, and maximum likelihood estimation (MLE). We used a two-sample Kolmogorov-Smirnov test (KS-test) to evaluate the best estimates, in which the parameters that produce distribution with higher *P-value* in the KS-test against the empirical cumulative distribution.

The overall fungicide spraying cost (x) accounting for fungicide price of 2019/20 crop season, and operational cost of 10 U$/application for three applications are described in Table 1. The spraying cost (S_C) was assumed to follow be uniformly distributed with values varying from 5% above and below the overall fungicide spraying cost (x), therefore, $S_C \sim Unif(0.95x,1.05x)$.

We ran a total of 50,000 Monte Carlo simulations for each variable (P, D, and S_C), the distribution of I was obtained and then the distribution of profits was derived. Break-even probabilities were calculated as the relative frequency of values that the income was equal to or higher than the fungicide spraying cost ($I \geq C$).

Results

Wheat blast index (WBI) and yield data at the trial level. There was considerable variation in WBI and grain yield in the untreated plots across seasons, regions, and locations/trials (each point in Fig. 2 represents a single trial). WBI in the untreated plots of the trials ranged from 0 to 100% (median 11.06%). The median WBI was higher (12.3%) in the Tropical than in the Subtropical (8.1%) region (Fig. 2 B). Across growing seasons, the highest (89.8%) and the lowest (0.8%) WBI medians in the untreated check were recorded in the 2019 and 2017 seasons, respectively (Fig. 2 C).

Baseline yield ranged from 15.6 to 4,276.2 kg/ha (median = 1,203.5 kg/ha) across the trials. The median yield was lower in the Tropical (1,000 kg/ha) than in the Subtropical region (1,281 kg/ha) (Fig. 2 E). The highest median yield (2,173 kg/ha) was observed in 2014 and the lowest (571.6 kg/ha) in the 2019 crop season (Fig. 2 F). As expected, lower WBI and higher grain yield was observed in the fungicide treatments compared with the untreated check (Fig. 2 A to 2 D).

Overall meta-analytic estimates of control efficacy and yield response. For all fungicides tested in the meta-analytic model, the overall WBI log response ratio ($\bar{L}_{IND}$) differed (P < 0.0001) from the untreated CHECK (Supplemental Table S2). Overall estimates of percent control efficacy ($\bar{C}$), obtained from back-transforming differences of the estimates of log of WBI ($\bar{L}_{IND}$) between the fungicide-treated and untreated plots, ranging from 43.2% and 58.0%. MANC, AZOX + TEBU, TFLX + PROT, TEBU, and TFLX + TEBU reduced WBI by at least 52% and did not differ significantly among them (P > 0.12) based on linear contrasts. PYRA + EPOX was the least effective treatment (43.2%) and did not differ from TEBU (53.5%; P = 0.07) and TFLX + TEBU (52.2%; P = 0.06) (Supplemental Table S2). The difference in percent control efficacy between the most and least effective fungicide was 14.8 percent points (Table 2). The Wald test for the treatment x design interaction showed that the network was consistent (P = 0.70).

Yield response ($\bar{D}$) was significantly higher (P < 0.0001) in all single a.i. and dual mixtures compared with the untreated CHECK (Table S3). The mean estimates of $\bar{D}$ ranged from 181.15 kg/ha to 420.10 kg/ha. The two single a.i. treatments, MANC (420.10 kg/ha) and TEBU (319.46 kg/ha) provided the greatest yield response, and linear contrasts showed no difference between them (P = 0.065). These were followed by AZOX + TEBU (301.48 kg/ha) and TFLX+PROT (299.73 kg/ha), which were not different between them (P = 0.90), but differed from TFLX+TEBU (245.34 kg/ha; P < 0.0001). All fungicides differed from PYRA + EPOX (181.15 kg/ha) with regards to yield response. The difference between the highest and lowest yield response among the treatments was 238.95 kg/ha (Table 2, Table S3). The Wald test for the treatment x design interaction showed that the network was inconsistent, meaning that results were dependent on the design (P = 0.01).

Effect of moderator variables. The categories of WBI and wheat grain yield as baselines did not affect WBI or yield (P > 0.05). Similarly, year did not affect wheat grain yield (P = 0.26) or WBI (P = 0.74), suggesting no decline in fungicide efficacy or yield response over time.

Based on the Wald test (P < 0.0001), the expanded model including the categorical moderator variable region differed statistically from the simpler model for both disease index and yield response. The control efficacy in the Subtropical region was numerically higher compared to the

Tropical region for all treatments. A difference of at least 19.1 percent points in $\overline{C}$ between regions was significant (P < 0.05) for three fungicides (PYRA + EPOX, TFLX + PROT and MANC) (Table 2). Similarly, yield response from the use of fungicides was generally higher in the Subtropical than in the Tropical region. There was a statistical difference in $\overline{D}$ between regions for all fungicides, except TEBU (P = 0.1836). Significant differences in yield responses between regions ranged from 200 to 291 kg/ha (Table 2).

In general, there was a similar pattern in the relationship between yield response and fungicide efficacy among regions. The treatment leading to the greatest mean yield response in both Tropical and Subtropical regions was MANC (Fig. 3). Similarly, MANC was the numerically most effective treatment in reducing disease index in the Subtropical region. For the Tropical region, TEBU and AZOX + TEBU exhibited a high control efficacy (~47%). Again, PYRA + EPOX provided the least yield response in both Tropical and Subtropical regions (Fig. 3).

Profits and break-even probabilities. Estimates of parameters of the Gamma distribution for the distribution of wheat prices for the 2012 to 2020 period that produced higher P-value in the KS-test were obtained using non-linear regression (P = 0.9375). The P-value in the KS-test for the parameters estimated using MLE was 0.11. The estimates of the shape ($\boldsymbol{\alpha}$) and the rate ($\boldsymbol{\beta}$) parameters were 19.4 and 0.54, respectively.

Overall, fungicides were more profitable in the subtropical region, with break-even probabilities, $P(I \geq C)$, values higher than 0.99 for all fungicides (Fig. 4). In this region, the most profitable fungicide was MANC, with a mean profit of US$269.73/ha. The least profitable fungicide in the subtropical region was PYRA+EPOX, with a mean profit of US$118.17/ha. In the tropical region, break-even probabilities of fungicides were much lower than those obtained in the subtropics. The $P(I \geq C)$ values ranged from 0.17 (PYRA+EPOX) to 0.97 (MANC) (Fig. 4). The most profitable fungicide was again MANC, with a mean profit of US$95.00/ha and the least profitable was PYRA+EPOX, with a negative mean profit of -US$17.66/ha.

Discussion

This study provides an updated summary of the effects of different fungicides applied sequentially (three sprays starting at the heading stage) for managing wheat blast. For such, we gathered data from 44 uniform field trials conducted during nine growing seasons (2012 to 2020) across several wheat-producing states in Brazil. A previous study used a portion of the data used in this work; 17 cooperative trials conducted from 2012 to 2014 in Brazil) also with the goal of comparing fungicide treatments (Cruz et al. 2019). The present study differs from the previous by evaluating a larger set of fungicide treatments, testing the effect of year and region within Brazil, and performing an economic analysis (Cruz et al. 2019). Results of our analysis generally corroborate the findings of that study for the Brazilian trials, which also reported a similar performance among the QoI+DMI premixes and a protectant fungicide. Similarly, a recent single field experiment (2019/20 season) conducted in Bangladesh reported a relatively high efficacy (65%) for MANC, which was close to our upper limit (95% confidence interval) estimate (Roy et al. 2020). The grain yield response from using MANC in the Bangladeshi study (744 kg/ha) was within the expected gain (close to the upper limit) of estimated trials conducted in the Subtropical region of Brazil. Among several differences in experimental conditions, it is worth noting the difference in the concentration of MANC that was a bit higher (800 g/kg) in that study compared with the recommended dose used in the Brazilian experiments (750 g/kg), which may explain the higher levels of efficacy and yield response.

Commercial premixes of QoI+DMI have been tested more extensively than MANC in independent research. For instance, in the study conducted by Cruz et al. (2019), the authors reported a relatively high control efficacy (58 to 68%) for picoxystrobin + cyproconazole, trifloxystrobin + tebuconazole, azoxystrobin + cyproconazole and pyraclostrobin + epoxiconazole applied twice across six field trials conducted in Bolivia from 2014 to 2015. Average yield responses obtained from applying those premixes in that study were extremely high, in the magnitude of 1,834 kg/ha (Cruz et al. 2019).

In Brazil, contrary to our findings, replicated field studies have reported higher levels of efficacy of PYRA+EPOX - ranging from 60% (Pagani et al. 2014) to 85% (Rios et al. 2016), compared with our estimates (95% CI 31 to 52%) regardless of the region. Overall, we found that this commercial premix performed the poorest among all treatments. Nevertheless, improved efficacy similar to those obtained in Bolivia, using PYRA + EPOX, can be expected especially in the southern region of Brazil, but with a high level of uncertainty (95% CI 17 to 74%).

Tebuconazole is an affordable option widely used by Brazilian growers not only for controlling wheat blast, but also for managing other important wheat diseases such as Fusarium head blight disease (Machado et al. 2017; Duffeck et al. 2020; Barro et al. 2020). The mean estimates of TEBU applied solo (54%) as well as the premix TFLX + TEBU (52%) reported in our meta-analysis were very similar to a two-year study (TEBU = 57%; TFLX + TEBU = 50%) conducted in Midwest of Brazil during 2010 and 2011 crop seasons (Pagani et al. 2014). Moreover, the yield responses from using the TEBU based fungicides reported in that study

(~400 kg/ha) (Pagani et al. 2014) were within the expected range, mainly for trials conducted in the Subtropics where yield response was greater than in the Tropical region.

In general, the levels of control were superior and, on average, 18 percent points greater in the Subtropical region (54 to 66%) compared with the Tropical region (24 to 47%). The only exception was tebuconazole fungicide for which efficacy was not influenced by region. Our findings demonstrated a similar levels of disease control reported in single studies conducted in the tropics (60%) or Subtropics (85%) (Pagani et al. 2014; Rios et al. 2016).

Results of our profitability analysis using the means and respective uncertainty of the estimates of yield return showed that, regardless of the region, the most profitable fungicide was MANC, given its lower price, and similar efficacy and yield response compared with the premixes. Although mancozeb was introduced in 1962, it is still important in the fungicide market worldwide and known to be a cost-effective fungicide (Gullino et al. 2010; Thind and Hollomon 2018). In fact, the probabilities of breaking even on costs of the commercial premixes of fungicides were generally lower, especially in the Tropical region, for all premixes (which are most costly) than MANC and TEBU, for which high break-even probabilities (> 92%) are expected.

It is worth mentioning that there are two different wheat production systems in the tropics of Brazil. The first is the irrigated and high-yielding system and where the blast is not a big concern and the non-irrigated under which the trials were established. In the latter case, sowing dates range from February to May (end of summer to begin of autumn) when the weather conditions are favorable for the disease. The combination of less susceptible cultivars and fungicide protection has been explored to improve disease management. For instance, Rios et al. (2016) reported the effect of cultivar when using PYRA + EPOX with greatest control levels using a cultivar that was considered less susceptible.

The use of premixes of single-site amended with multi-site fungicides has been tested more recently for wheat blast control. Preliminary data from the uniform fungicide trials (Santana et al. 2019b, 2019a, 2020a, 2020b) have shown grain yield benefits from adding a multi-site fungicide (mancozeb) in the mixture and more data will become available in the near future to confirm this observation. Additionally, the use of multi-site fungicides is important for managing fungicide resistance (FRAC-BR 2021). In fact, less sensitive PoT populations to QoIs and DMIs in Brazil have been reported, associated with mutations in cytochrome b gene (mainly G143A substitution) (Castroagudín et al. 2015) or *CYP51* gene (several mutation points) (Dorigan et al. 2019; Poloni et al. 2021). However, using a relatively long time series we did not find evidence of a decline in fungicide efficacy over the years for all commercial premixes amended with QoI as well as TEBU applied solo. Whether QoIs are less effective or losing efficacy over the years could not be inferred given that none of the fungicides had QoI as a sole active ingredient. If that happens to be true, and confirmed by field research, it is possible that the DMI component in the premix is responsible for the disease reduction. Further research in this area will be important to contribute to the debate around the risk associated with the use of strobilurins for wheat blast control (Castroagudín et al. 2015; Oliveira et al. 2015; Poloni et al. 2021).

In conclusion, the definition of the best options in fungicide programs for managing wheat blast should take into account region-specific factors that affect the performance of fungicides and also the need to extend control for other wheat diseases, a case where QoI + DMI premixes have found use not only in Brazil but elsewhere (Blandino et al. 2006; Willyerd et al. 2012; Barro et al. 2017; Paul et al. 2018b). Management tactics such as application of site-specific fungicides amended with multi-site fungicides programs; use of locally-adapted wheat cultivars carrying resistance genes against blast; and shifts in sowing time to escape conducive blast weather at heading stage, especially in the tropics, should be considered in an integrated management strategy against wheat blast. Overall, this work confirms the profitability of some site-specific and a multi-site fungicide, report their efficacy, and yield return by controlling wheat blast in both high disease-conducive region (tropical) and moderately disease-conducive region (subtropical) to blast occurrence, which may be useful for decision-making when defining fungicide programs.

Acknowledgements

The first and senior author acknowledges the financial support provided by the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq/Brazil) for scholarship and research fellowship (PQ-306857/2015-4), respectively. We also acknowledge the financial support provided by the Fundação de Amparo à Pesquisa do Estado de Minas Gerais (Fapemig/Brazil) for a research grant (APQ-03945-16). All other authors are thankful to their institutions and teams for providing the support during the conduction of field experiments and collection of the data.

Data availability statement

The data and R scripts that support the findings of this study are openly available in the Open Science Framework project at osf.io/y7sgd/. A website was generated to better visualize the scripts where all data and analyses are documented and reproducible (git.io/JGVCM).

Author's contribution

JPA and JPB contributed equally to this study. JPA, JPB and EMD conceived the idea, analyzed the data and wrote the manuscript. KSA analyzed the data and wrote the manuscript. FMS planned and coordinated the experiments; JMVP, JLNM, DL, GAMT, CCS, CDSS, ACPG, AABS, CAS, DFC, MAOC, TDNM, DRA, AAPC, LSOM, CMU, WV, and RCSG conducted the field trials and shared the data. All authors provided feedback and approved the final manuscript.

Conflict of interests

The authors declare that they have no conflict of interests.

Literature Cited

AGROFIT. 2021. Ministério da Agricultura, Pecuária e Abastecimento. Available at: http://agrofit.agricultura.gov.br/agrofit_cons/principal_agrofit_cons [Accessed March 17, 2021].

AGROLINK. 2021. O Portal do conteúdo agropecuário. Available at: https://www.agrolink.com.br/ [Accessed March 19, 2021].

Barro, J. P., Forte, C. T., Trentin, D., Scariot, M., and Milanesi, P. M. 2017. Effectiveness of different fungicide formulations and number of applications in controlling wheat leaf rust. Summa Phytopathol. 43:276–280.

Barro, J. P., Meyer, M. C., Godoy, C. V., Dias, A. R., Utiamada, C. M., Jaccoud Filho, D. de S., Wruck, D. S. M., Borges, E. P., Siqueri, F., Juliatti, F., Campos, H. D., Junior, J. N., Carneiro, L. C., Silva, L. H. C. P., Martins, M. C., Balardin, R. S., Zito, R. K., Furlan, S. H., Venancio, W. S., and Del Ponte, E. M. 2019. Performance and profitability of fungicides for managing soybean white mold: A 10-year summary of Cooperative Trials. Plant Dis. 103:2212–2220.

Barro, J., Santana, F. M., Machado, F. J., Duffeck, M. R., Lau, D., Sbalcheiro, C. C., Schipanski, C. A., Chagas, D. F., Venancio, W. S., Dallagnol, L. J., Guterres, C. W., Kuhnem, P., Feksa, H. R., and Del Ponte, E. M. 2020. Are DMI+QoI fungicide premixes during flowering worthwhile for Fusarium head blight control in wheat? a meta-analysis. Plant Dis. Available at: https://apsjournals.apsnet.org/doi/abs/10.1094/PDIS-09-20-2096-RE [Accessed April 23, 2021].

Blandino, M., Minelli, L., and Reyneri, A. 2006. Strategies for the chemical control of Fusarium head blight: Effect on yield, alveographic parameters and deoxynivalenol contamination in winter wheat grain. Eur. J. Agron. 25:193–201.

Cardoso, C. A. A., Reis, E. M., and Moreira, E. N. 2008. Development of a warning system for wheat blast caused by *Pyricularia Grisea*. Summa Phytopathol. 34:216–21.

Castroagudín, V. L., Ceresini, P. C., de Oliveira, S. C., Reges, J. T. A., Maciel, J. L. N., Bonato, A. L. V., Dorigan, A. F., and McDonald, B. A. 2015. Resistance to QoI fungicides is widespread in Brazilian populations of the wheat blast pathogen *Magnaporthe oryzae*. Phytopathology 105:284–294.

Ceresini, P. C., Castroagudín, V. L., Rodrigues, F. Á., Rios, J. A., Aucique-Pérez, C. E., Moreira, S. I., Croll, D., Alves, E., Carvalho, G., Maciel, J. L. M., and Mcdonald, B. A. 2019. Wheat blast: from its origins in South America to its emergence as a global threat: Wheat Blast. Mol. Plant Pathol. 20:155–172.

Ceresini, P. C., Castroagudín, V. L., Rodrigues, F. Á., Rios, J. A., Eduardo Aucique-Pérez, C., Moreira, S. I., Alves, E., Croll, D., and Maciel, J. L. N. 2018. Wheat Blast: Past, present, and future. Annu. Rev. Phytopathol. 56:427–456.

Chakraborty, M., Mahmud, N. U., Gupta, D. R., Tareq, F. S., Shin, H. J., and Islam, T. 2020a. Inhibitory effects of linear lipopeptides from a marine *Bacillus subtilis* on the wheat blast fungus *Magnaporthe oryzae Triticum*. Front. Microbiol. 11:1–14.

Chakraborty, M., Mahmud, N. U., Muzahid, A. N. M., Rabby, S. M. F., and Islam, T. 2020b. Oligomycins inhibit *Magnaporthe oryzae Triticum* and suppress wheat blast disease. PLOS ONE 15:1–16.

Coelho, M. A. de O., Torres, G. A. M., Cecon, P. R., and Santana, F. M. 2016. Sowing date reduces the incidence of wheat blast disease. Pesqui. Agropecu. Bras. 51:631–637.

Comissão Brasileira de Pesquisa de Trigo e Triticale. 2020. *Informações Técnicas para Trigo e Triticale*. 13th ed. Passo Fundo, RS. Brasil: Biotrigo Genética.

Cruppe, G., Cruz, C. D., Peterson, G., Pedley, K., Asif, M., Fritz, A., Calderon, L., Silva, C. L., Todd, T., Kuhnem, P., Singh, P. K., Singh, R. P., Braun, H. J., Barma N. C. D., and Valent, B. 2020. Novel sources of wheat head blast resistance in modern breeding lines and wheat wild relatives. Plant Dis. 104:35–43.

Cruz, C. D., Santana, F. M., Todd, T. C., Maciel, J. L. N., Kiyuna, J., Baldelomar, D. F., Cruz, C. P., Lau, D., Seixas, C. S., Goulart, A. C. P., Sussel, A. A., Schipanski, C. A., Chargas, D. F., Coelho, M., Montecelli, T. D. N., Utiamada, C., Custódio, A. P., Rivadeneira, M. G., Bockus, W., and Valent, B. 2019. Multi-environment assessment of fungicide performance for managing wheat head blast (WHB) in Brazil and Bolivia. Trop. Plant Pathol. 44:183–191.

Cruz, C. D., and Valent, B. 2017. Wheat blast disease: Danger on the move. Trop. Plant Pathol. 42:210–222.

Cruz, M. F. A., Rios, J. A., Araujo, L., and Ávila Rodrigues, F. 2016. Infection process of *Pyricularia oryzae* on the leaves of wheat seedlings. Trop. Plant Pathol. 41:123–127.

Dalla Lana, F., Paul, P. A., Godoy, C. V., Utiamada, C. M., da Silva, L. H. C. P., Siqueri, F. V., Forceline, C. A., Jaccound-Filho, D. S., Miguel-Wruck, D. S., Borges, E. P., Juliatti, F. C., Campos, H. D., Nunes-Junior, J., Carneiro, L. C., Canteri, M. C., Ito, M. F., Meyer, M. C., Martins, M. C., Balardin, R. S., Furlan, S. H., Carlin, V. J., and Del Ponte, E. M. 2018. Meta-analytic modeling of the decline in performance of fungicides for managing soybean rust after a decade of use in Brazil. Plant Dis. 102:807–817.

Dorigan, A. F., Carvalho, G. D., Poloni, N. M., Negrisoli, M. M., Maciel, J. L. N., and Ceresini, P. C. 2019. Resistance to triazole fungicides in *Pyricularia* species associated with invasive plants from wheat fields in Brazil. Acta Sci. Agron. 41:1–10.

Duffeck, M. R., Alves, K. S., Machado, F. J., Esker, P. D., and Del Ponte, E. M. 2020. Modeling yield losses and fungicide profitability for managing Fusarium head blight in Brazilian spring wheat. Phytopathology 110:370–378.

FRAC-BR. 2021. Comitê de Ação a Resistência a Fungicidas. Frac-Br. Available at: https://www.frac-br.org [Accessed May 31, 2021].

Goulart, A. C. P., and Paiva, A. 1993. Evaluation of fungicides for control of wheat (*Triticum aestivum* L.) blast (*Pyricularia grisea*), 1991. In *Annual Wheat Newsletter*, Fort Collins: Department of Agronomy, Colorado State University.

Goulart, A. C. P., and Paiva, F. de A. 2000. *Avaliação de Perdas no Rendimento de Grãos de Trigo Causadas por Pyricularia grisea, no Período de 1988 a 1992, em Mato Grosso do Sul*. 7th ed. Dourados/MS: Embrapa.

Goulart, A. C. P., Paiva, F., and Mesquita, A. N. 1990. Occurrence and losses caused by wheat

(*Triticum aestivum* L.) blast (*Pyricularia oryzae* Cav.) in the state of Mato Grosso do Sul, in 1988. In *Annual Wheat Newsletter*, Fort Collins: Department of Agronomy, Colorado State University.

Gullino, M. L., Tinivella, F., Garibaldi, A., Kemmitt, G. M., Bacci, L., and Sheppard, B. 2010. Mancozeb: Past, present, and future. Plant Dis. 94:1076–1087.

Higgins, J. P. T., Jackson, D., Barrett, J. K., Lu, G., Ades, A. E., and White, I. R. 2012. Consistency and inconsistency in network meta-analysis: Concepts and models for multi-arm studies. Res. Synth. Methods 3:98–110.

Igarashi, S., Utiamada, C. M., Igarashi, L. C., Kazuma, A. H., and Lopes, R. 1986. Occurrence of *Pyricularia* sp. in wheat (*Triticum aestivum* L.) in the State of Parana, Brazil. Fitopatol. Bras. 11:351–352.

Islam, M. T., Kim, K.-H., and Choi, J. 2019. Wheat Blast in Bangladesh: The current situation and future impacts. Plant Pathol. J. 35:1–10.

Machado, F. J., Santana, F. M., Lau, D., and Del Ponte, E. M. 2017. Quantitative review of the effects of triazole and benzimidazole fungicides on Fusarium head blight and wheat yield in Brazil. Plant Dis. 101:1633–1641.

Maciel, J. L. N., Danelli, A. L. D., Boaretto, C., and Forcelini, C. A. 2013. Diagrammatic scale for the assessment of blast on wheat spikes. Summa Phytopathol. 39:162–166.

Madden, L. V., Piepho, H.-P., and Paul, P. A. 2016. Statistical models and methods for network meta-analysis. Phytopathology 106:792–806.

Malaker, P. K., Barma, N. C. D., Tiwari, T. P., Collis, W. J., Duveiller, E., Singh, P. K., Joshi, A. K., Singh, R. P., Braun, H. J., Peterson, G. L., Pedley, K. F., Farman, M. L., and Valent, B. 2016. First report of wheat blast caused by *Magnaporthe oryzae* pathotype *Triticum* in Bangladesh. Plant Dis. 100:2330.

Oliveira, S. C., Castroagudín, V. L., Maciel, J. L. N., Pereira, D. A. dos S., and Ceresini, P. C. 2015. Resistência cruzada aos fungicidas QoI azoxistrobina e piraclostrobina no patógeno da brusone do trigo *Pyricularia oryzae* no Brasil. Summa Phytopathol. 41:298–304.

Pagani, A. P. S., Dianese, A. C., and Café-Filho, A. C. 2014. Management of wheat blast with synthetic fungicides, partial resistance and silicate and phosphate minerals. Phytoparasitica 42:609–617.

Paul, P. A., Bradley, C. A., Madden, L. V., Dalla Lana, F., Bergstrom, G. C., Dill-Macky, R., Wise, K. A., Esker P. D., McMullen, M., Grybauskas, A., Kirk, W. W., Milus, E., and Ruden, K. 2018a. Effects of pre- and post anthesis applications of demethylation inhibitor fungicides on Fusarium head blight and deoxynivalenol in spring and winter wheat. Plant Dis. 102:2500–2510.

Paul, P. A., Bradley, C. A., Madden, L. V., Lana, F. D., Bergstrom, G. C., Dill-Macky, R., Esker P. D., Wise, K. A., McMullen, M., Grybauskas, A., Kirk, W. W., and Ruden, K. 2018b. Meta-analysis of the effects of QoI and DMI fungicide combinations on Fusarium head blight and deoxynivalenol in wheat. Plant Dis. 102:2602–2615.

Paul, P. A., Lipps, P. E., Hershman, D. E., McMullen, M. P., Draper, M. A., and Madden, L. V. 2008. Efficacy of triazole-based fungicides for Fusarium head blight and deoxynivalenol control in wheat: A multivariate meta-analysis. Phytopathology 98:999–1011.

Paul, P. A., McMullen, M. P., Hershman, D. E., and Madden, L. V. 2010. Meta-analysis of the effects of triazole-based fungicides on wheat yield and test weight as influenced by Fusarium head blight intensity. Phytopathology 100:160–171.

Perelló, A. E., Martinez, I., Sanabria, A., Altamirano, R., and Sibole, J. V. 2017. Pathogenicity of isolates of *Magnaporthe* spp. from wheat and grasses infecting seedlings and mature wheat plants in Argentina. Plant Pathol. 66:1149–1161.

Piepho, H. P. 2014. Network-meta analysis made easy: Detection of inconsistency using factorial analysis-of-variance models. BMC Med. Res. Methodol. 14 Available at: https://bmcmedresmethodol.biomedcentral.com/articles/10.1186/1471-2288-14-61 [Accessed March 25, 2020].

Poloni, N. M., Carvalho, G., Vicentini, S. N. C., Dorigan, A. F., Maciel, J. L. N., McDonald, B. A., Moreira, S. I., Hawkins, N., Fraaije, B. A., Kelly, D. E., Kelly, S. L., and Ceresini, P.C. 2021. Widespread distribution of resistance to triazole fungicides in Brazilian populations of the wheat blast pathogen. Plant Pathol. 70:436–448.

R Core Team. 2021. *R: A Language and Environment for Statistical Computing*. Vienna, Austria: R Foundation for Statistical Computing. Available at: https://www.R-project.org/.

Rios, J. A., Rios, V. S., Paul, P. A., Souza, M. A., Araujo, L., and Rodrigues, F. A. 2016. Fungicide and cultivar effects on the development and temporal progress of wheat blast under field conditions. Crop Prot. 89:152–160.

Roy, K. K., Anwar, M. B., Mustarin, K.-E.-, Reza, M. M. A., Muzahid-E-Rahman, M., Malaker, P. K., Barma, N. C. D., and Singh, P. K. 2020. Evaluation of different fungicides (chemical, botanical and bio-agent) in controlling wheat blast in a blast prone area in Bangladesh. Arch. Phytopathol. Plant Prot. 1–9.

Santana, F. M., Lau, D., Sbalcheiro, C. C., Aguilera, J. G., Goulart, A. C. P., Sussel, A. A. B., Schipanski, C. A., Coelho, M. A. O., Utiamada, C. M., Montecelli, T. D. N., Seixas, C. D. S., and Custódio, A. A. P. 2016a. *Eficiência de fungicidas para o controle da brusone do trigo: resultados dos ensaios cooperativos – safra 2013*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Santana, F. M., Lau, D., Sbalcheiro, C. C., Aguilera, J. G., Goulart, A. C. P., Sussel, A. A. B., Schipanski, C. A., Coelho, M. A. O., Utiamada, C. M., Montecelli, T. D. N., Seixas, C. D. S., Custódio, A. A. P., and Venâncio, W. S. 2016b. *Eficiência de fungicidas para o controle da brusone do trigo: resultados dos ensaios cooperativos – safra 2014*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Santana, F. M., Lau, D., Sbalcheiro, C. C., Goulart, A. C. P., Custódio, A. A. P., Venâncio, W. S., Goussain, R. C. S., Amaral, D. R. A., Seixas, C. D. S., and Venancio, J. F. 2016c. *Eficiência de fungicidas para controle de brusone de trigo: resultados dos ensaios cooperativos – safra 2015*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Santana, F. M., Lau, D., Sbalcheiro, C. C., Goussain, R. C. S., Venancio, W. S., Custódio, A. A. P., Moreira, L. S. O., and Sussel A. A. B. 2020a. *Eficiência de fungicidas para controle de brusone de trigo: resultados dos ensaios cooperativos, safra 2018*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Santana, F. M., Lau, D., Sbalcheiro, C. C., Sussel, A. A. B., Goussain, R. C. S., Venancio, W. S., Custódio, A. A. P., and Moreira, L. S. O. 2019a. *Eficiência de fungicidas para controle de brusone de trigo: resultados dos ensaios Cooperativos - safra 2017*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Santana, F. M., Lau, D., Sbalcheiro, C. C., Sussel, A. A. B., Seixas, C. D. S., Venancio, W. S., and Nicolau, M. 2019b. *Eficiência de fungicidas para controle da brusone de trigo: resultados dos ensaios cooperativos, safra 2016*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Santana, F. M., Lau, D., Sbalcheiro, C. C., Sussel, A. A. B., Venancio, W. S., Schipanski, C. A., and Chagas, D. F. 2020b. *Eficiência de fungicidas para controle de brusone de trigo: resultados dos ensaios cooperativos, safra 2019*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Santana, F. M., Maciel, J. N., Lau, D., Cargnin, A., Seixas, C. D. S., Bassoi, M. C., Goulart, A. C. P., Sussel, A. A. B., Schipanski, C. A., Montecelli, T. D. N., Chargas, J. H., and Guizeline, J. 2013. *Eficiência de fungicidas para o controle da brusone do trigo: resultados dos ensaios cooperativos - safra 2011*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Santana, F. M., Maciel, J. N., Lau, D., Torres, G. A. M., Cargnin, A., Seixas, C. D. S., Goulart, A. C. P., Sussel, A. A. B., Schipanski, C. A., Montecelli, Custódio, A. A. P., and Utiamada, C. M. 2014. *Eficiência de fungicidas para o controle da brusone do trigo: resultados dos ensaios cooperativos - safra 2012*. Passo Fundo, RS. Brasil: Embrapa Trigo.

Silva, S. R., Custódio, A. A. P., Foloni, J. S. S., Betioli-Junior, E., and Bassoi, M. C. 2019. Nitrogen fertilization effects on wheat blast epidemics under varying field environmental conditions. Trop. Plant Pathol. 44:258–67.

Silva, B. N., Oliveira, L. M., Mochko, A. C. R., Chaves, J. A. A., Aucique-Pérez, C. E., and Rodrigues, F. A. 2021. Physiological and biochemical changes in wheat plants infected by *Pyricularia Oryzae* caused by thermal oscillations. Physiol. Mol. Plant. Path. 115:1-12.

Tembo, B., Mulenga, R. M., Sichilima, S., M'siska, K. K., Mwale, M., Chikoti, P. C., Singh, P.K., He, X., Pedley, K. F., Peterson, G. L., Singh, R. P., and Braun, H. J. 2020. Detection and characterization of fungus (*Magnaporthe oryzae* pathotype *Triticum*) causing wheat blast disease on rain-fed grown wheat (*Triticum aestivum* L.) in Zambia. PLOS ONE. 15:1-10.

Thind, T. S., and Hollomon, D. W. 2018. Thiocarbamate fungicides: reliable tools in resistance management and future outlook: Thiocarbamate fungicides. Pest Manage. Sci. 74:1547–1551.

Tosa, Y., and Chuma, I. 2014. Classification and parasitic specialization of blast fungi. J. Gen. Plant Pathol. 80:202–209.

Uddin, W., Serlemitsos, K., and Viji, G. 2003. A temperature and leaf wetness duration-based model for prediction of gray leaf spot of perennial ryegrass turf. Phytopathology 93:336–43.

Urashima, A. S., Alves, A. F., Silva, F. N., Oliveira, D., and Gazaffi, R. 2017. Host range, mating type and population structure of *Magnaporthe* sp. of a single barley field in São Paulo state, Brazil. J. Phytopathol. 165:414–424.

Urashima, A. S., Leite, S. F., and Galbieri, R. 2007. Eficiência da disseminação aérea em

Pyricularia grisea. Summa Phytopathol. 33:275–279.

Viechtbauer, W. 2010. Conducting meta-analyses in R with the metafor Package. J. Stat. Softw. 36:1–48.

Willyerd, K. T., Li, C., Madden, L. V., Bradley, C. A., Bergstrom, G. C., Sweets, L. E., McMullen, M., Ransom, J. K., Grybauskas, A., Osborne, L., Wegulo, S. N., Hershman, D. E., Bockus, W. W., Groth, D., Dill-Macky, R., Milus, E., Esker, P. D., Waxman, K. D., Adee, E. A., Ebelhar, S. A., Young, B. G., and Paul, P. A. 2012. Efficacy and stability of integrating fungicide and cultivar resistance to manage Fusarium head blight and deoxynivalenol in wheat. Plant Dis. 96:957–967.

Xavier Filha, M. S., Rodrigues, F. A., Domiciano, G. P., Oliveira, H. V., Silveira, P. R., and Moreira, W. R. 2011. Wheat resistance to leaf blast mediated by silicon. Austral. Plant Pathol. 40:28–38.

Zadoks, J. C., Chang, T. T., and Konzak, C. F. 1974. A decimal code for the growth stages of cereals. Weed Res. 14:415–421.

Table 1. Fungicide treatments evaluated for controlling wheat blast in 44 independent field trials conducted across six Brazilian states (PR, SP, DF, MT, MS, and MG) from 2012 to 2020

Fungicide a.i.[a]	**Chemical group**[b]	**Brand name**	**Nº trials**	**Dosage (mL or g/ha)**	**Cost**[d] **(US$/ha)**
AZOX+TEBU	QoI+DMI	Azimut	28	750	73
MANC	DTC	Unizeb Gold	41	3,000	50
PYRA+EPOX	QoI+DMI	Opera	28	500	45
TEBU	DMI	Tebuco Nortox	19	500	52
TFLX+PROT	QoI+DMI	Fox	44	500	82
TFLX+TEBU	QoI+DMI	Nativo[c]	44	750	75

[a] a.i. (active ingredient); AZOX + TEBU = azoxystrobin + tebuconazole; MANC = mancozeb; PYRA + EPOX = pyraclostrobin + epoxiconazole; TEBU = tebuconazole; TFLX + PROT = trifloxystrobin + prothioconazole; TFLX + TEBU = trifloxystrobin + tebuconazole.

[b] QoI = Quinone-outside inhibitors; DMI = Sterol demethylation inhibitor; DTC = Dithiocarbamate.

[c] Included in all trials as positive control to wheat blast.

[D] Overall costs (US$/ha) considering commercial prices of the 2019/20 crop season and three applications (operational cost for each application used was US$10.00/ha).

Table 2. Overall means of Wheat Blast control efficacy ($\overline{C}$) and wheat yield response ($\overline{D}$) for each fungicide treatment, relative to the untreated check, not conditioned (Overall) and conditioned (moderator analysis) to two climatic regions (Tropical [DF, MT, MS, MG] and Subtropical [PR, SP]), conducted during nine years (2012-2020) across six Brazilian states

		Control efficacy (%)					Yield return (Kg/ha)				
Fungicide[a]	**Region**	**k**[b]	$\overline{C}$	**CI$_L$**[c]	**CI$_U$**[c]	***P-value***[d]	**k**[b]	$\overline{D}$	**CI$_L$**[c]	**CI$_U$**[c]	***P-value***[d]
MANC	Overall	33	58.0	49.8	64.8		35	420.1	315.6	524.6	
	Tropical	13	42.1	26.1	54.7		14	241.3	94.7	387.9	
	Subtropical	20	66.5	40.6	81.1	0.0011	21	532.1	195.3	868.9	0.0027
TFLX + PROT	Overall	39	53.9	44.6	61.6		42	299.7	221.9	377.5	
	Tropical	15	40.8	22.9	54.5		17	149.3	44.7	254.0	
	Subtropical	24	59.9	26.4	78.2	0.0262	25	396.5	154.5	638.4	0.0004
TEBU	Overall	18	53.6	43.2	62.1		19	319.5	221.6	417.3	
	Tropical	8	47.2	28.2	61.1		9	209.0	52.7	365.3	
	Subtropical	10	56.9	13.3	78.6	0.3076	10	351.1	-14.6	716.8	0.1836
AZOX + TEBU	Overall	18	54.8	43.5	63.9		19	301.5	215.7	387.2	
	Tropical	7	47.1	26.2	62.1		8	157.0	52.7	261.3	
	Subtropical	11	58.4	10.9	80.6	0.2704	11	410.3	162.2	658.4	0.0006
TFLX + TEBU	Overall	39	52.2	43.5	59.6		42	245.3	172.9	317.8	
	Tropical	15	42.4	26.1	55.2		17	123.9	25.7	222.1	
	Subtropical	24	57.4	24.3	76.0	0.0685	25	323.4	94.6	552.2	0.0028
PYRA + EPOX	Overall	19	43.2	31.8	52.8		22	181.1	114.8	247.5	
	Tropical	8	24.2	2.81	40.9		10	29.1	-49.8	108.1	
	Subtropical	11	54.3	17.5	74.7	0.0037	12	255.0	65.4	444.6	<0.0001

[a] See Table 1 for complete information on the fungicides.

[b] number of trials that each fungicide was evaluated.

[C] upper (CI$_U$) and lower (CI$_L$) limits of the 95% confidence interval around $\overline{C}$ and $\overline{D}$.

[d] probability value (significance level) for the effect of fungicide on disease reduction and yield response (at the selected climatic regions).

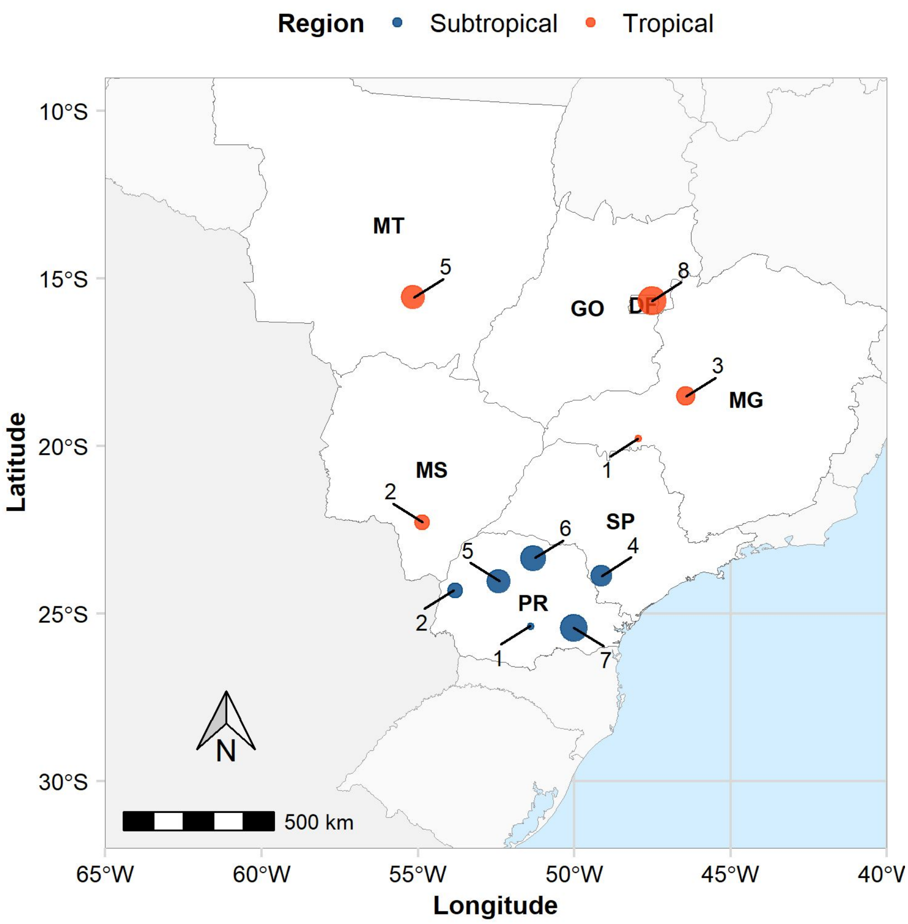

Fig. 1. Geolocation of eleven municipalities across six Brazilian states where 44 fungicide trials were conducted from 2012 to 2020 (Campo Mourão - PR, Guarapuava - PR, Campo Verde - MT, Dourados - MS, Itaberá - SP, Londrina - PR, Palmeira - PR, Palotina - PR, Patos de Minas - MG, Planaltina - DF, Uberaba - MG). Dots were colored by climatic region and the size of the circle is proportional to the number of trials conducted at each location.

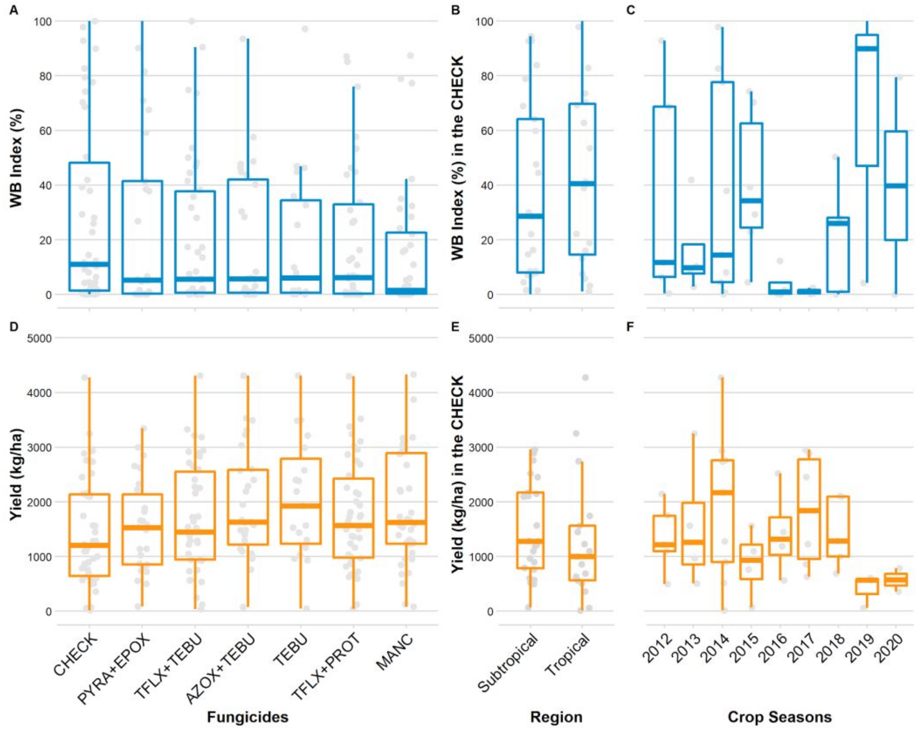

Fig. 2. Box plots depicting the means of wheat blast index (%) and wheat grain yield (kg/ha) (across years and locations) of the untreated (CHECK) and fungicide-treated plots (**A** to **D**); and the means of the same variables in the untreated plots within-region (**B** to **E**) and within-year (**C** to **F**) measured from a set of 44 field trials conducted from 2012 to 2020. Climatic regions defined in this study were: Tropical (DF, MT, MS, MG) and Subtropical (PR, SP). The thick horizontal line inside the box represents the median, the limits of the box represent the lower and upper quartiles, and the circles represent the yearly means of each treatment. See Table 1 for information on the fungicide treatments.

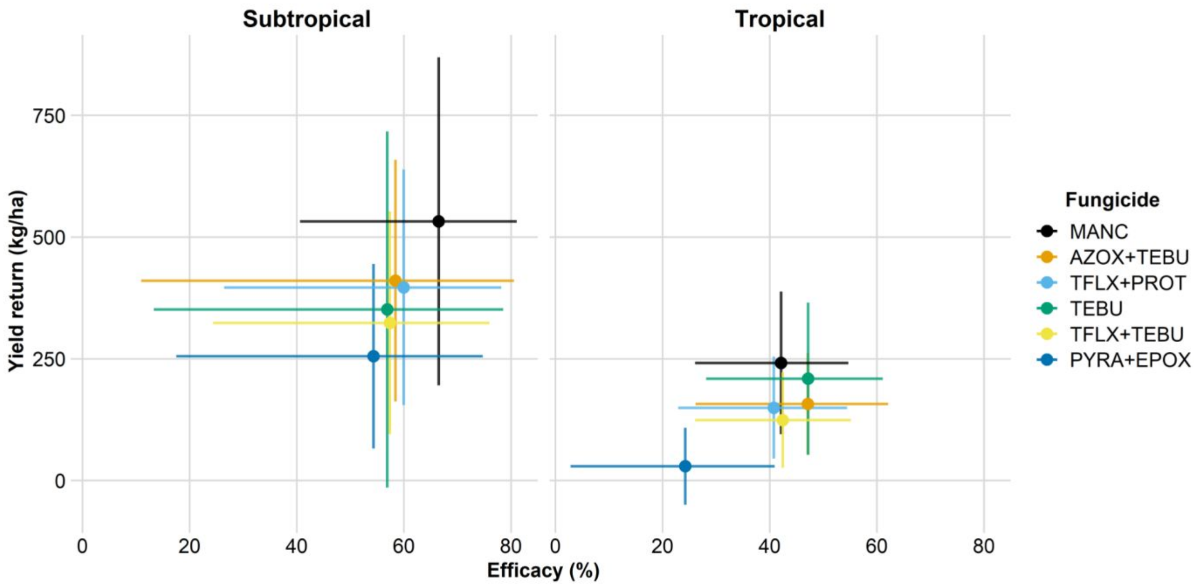

Fig. 3. Relationship between fungicide efficacy and wheat grain yield relative to the untreated check, for six selected fungicide treatments evaluated during nine years (2012 to 2020) across 44 field trials conducted across six Brazilian states (PR, SP, DF, MT, MS, and MG). Bars show the upper and lower limits of 95% confidence intervals around point estimates for both responses. See Table 1 for complete information on the evaluated fungicides.

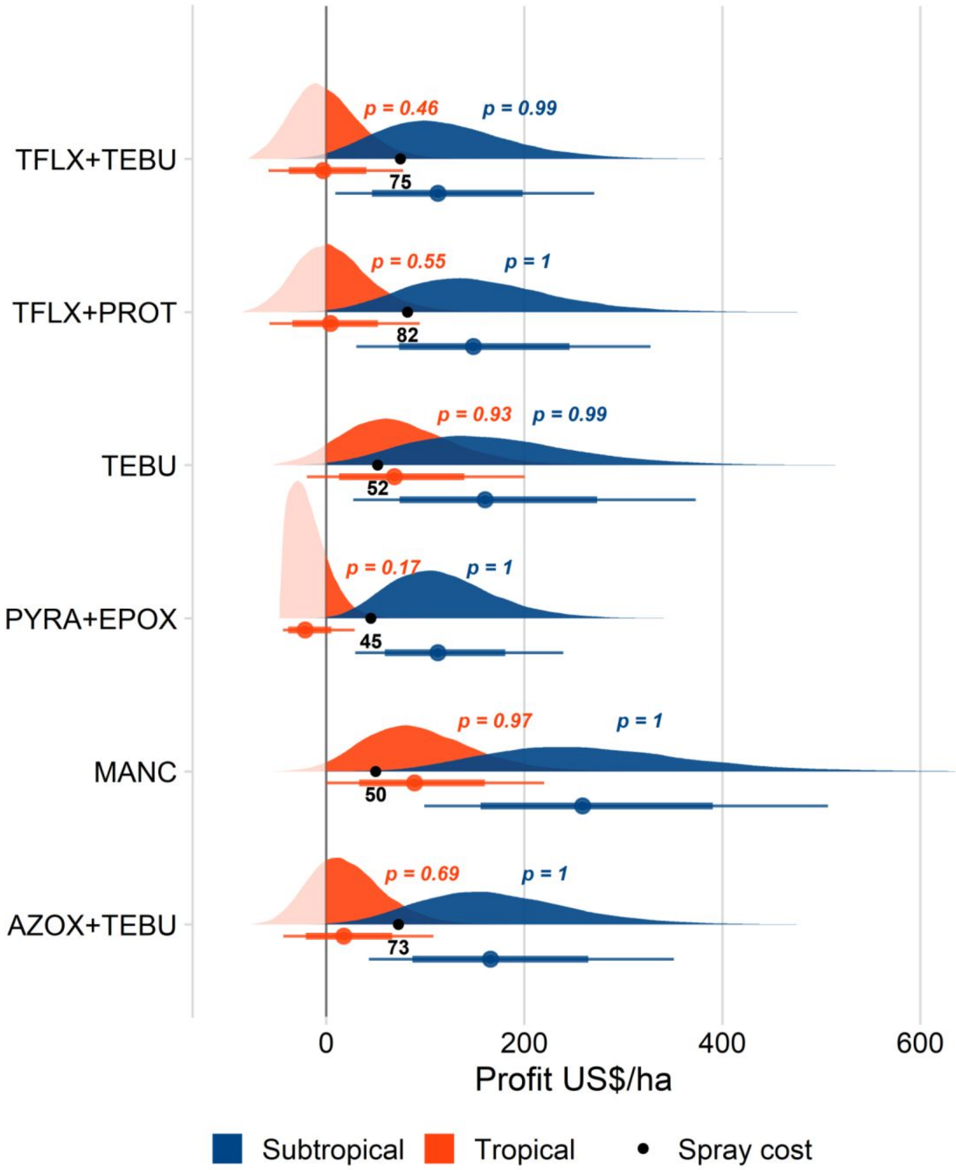

Fig. 4. Half-eye plots (density and point intervals) of profits (50,000 simulation runs) based on the meta-analytic estimate of yield return (kg/ha) for six fungicide treatments conditioned of two climatic regions (Tropical and Subtropical) evaluated during nine crop-seasons (2012 to 2020). The thinner error bars depict the 2.5 and 97.5 percentile of the distribution, while the thicker error bars represent 25 and 75 percentiles, and the solid colored point gives the median profit for each fungicide and region. *p* gives the break-even probabilities, i.e, income greater than the cost of each fungicide in each respective region. The numbered black dots represent the spraying costs in US$/ha of each fungicide.

Sequential Post-Heading Applications for Controlling Wheat Blast: A Nine-Year Summary of Fungicide Performance in Brazil

João P. Ascari et al.

Supplemental Figure and Tables

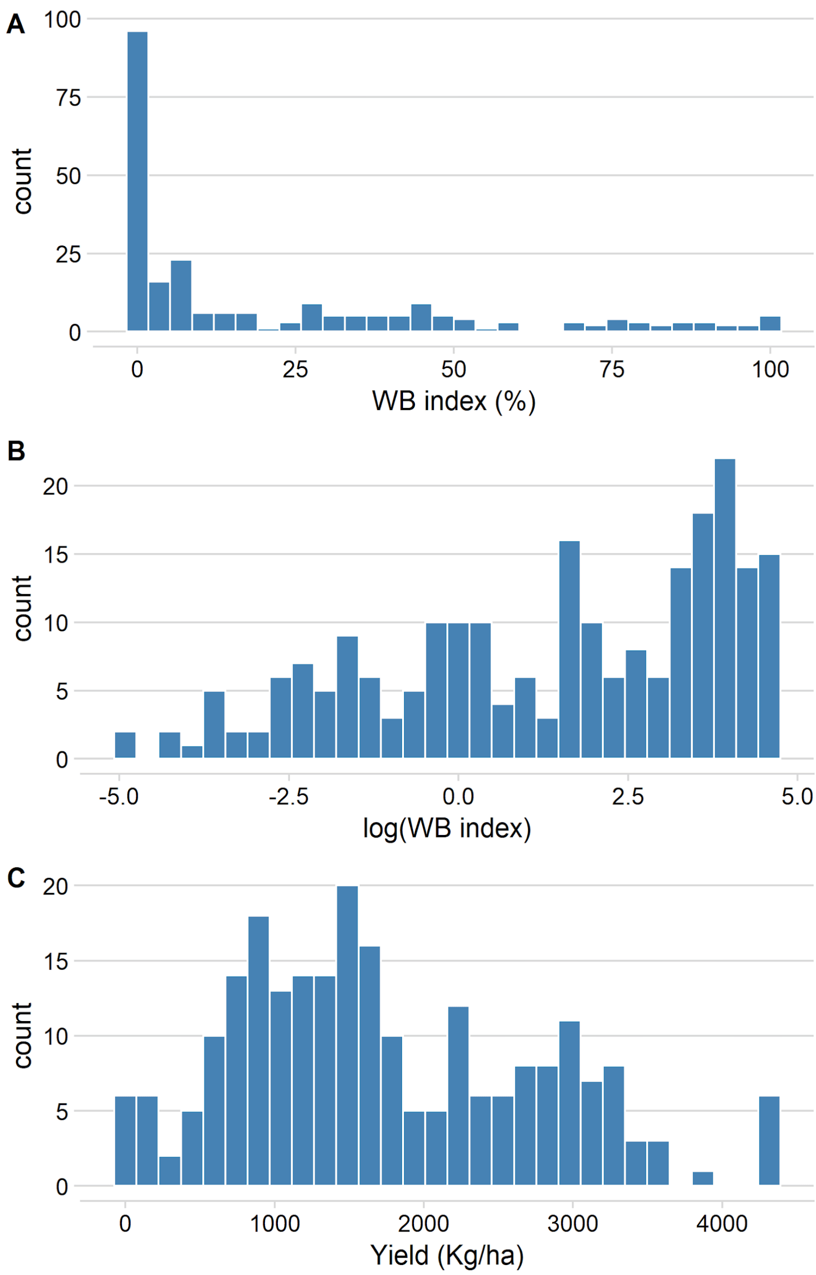

Fig. S1: Histograms for the distribution of Wheat blast index - WBI (**A**) and wheat grain yield (**C**) to check normality; **B**: log-transformed WBI data for normalizing the distribution and use in the meta-analysis models.

Table S1. Designs (set of fungicide treatments by each trial) identified in 44 independent trials conducted from 2012 to 2020 in 11 municipalities across six Brazilian states (PR, SP, DF, MT, MS and MG).

Design	Fungicides a.i. + untreated check [a]	WB index[b]	Yield[b]
1	CHECK; AZOX+TEBU; MANC; PYRA+EPOX; TEBU; TFLX+PROT; TFLX+TEBU	4	4
2	CHECK; AZOX+TEBU; PYRA+EPOX; TEBU; TFLX+PROT; TFLX+TEBU	5	5
3	CHECK; AZOX+TEBU; MANC; TEBU; TFLX+PROT; TFLX+TEBU	8	9
4	CHECK; AZOX+TEBU; MANC; TFLX+PROT; TFLX+TEBU	1	1
5	CHECK; MANC; PYRA+EPOX; TFLX+PROT; TFLX+TEBU	10	13
6	CHECK; MANC; TEBU; TFLX+PROT; TFLX+TEBU	1	1
7	CHECK; MANC;; TFLX+PROT; TFLX+TEBU	9	7
8	CHECK; TFLX+PROT; TFLX+TEBU	1	2

[a] See Table 1 for complete information about the treatments;
[b] Number of trials that each design of treatments was identified for both WB index and yield.

Table S2. Overall means and respective confidence intervals of log response ratio ($\bar{L}_{IND}$) and calculated percent control ($\bar{C}$) of Wheat Blast (WB) relative to untreated check provided by six fungicides evaluated during nine years (2012 to 2020) across 44 studies conducted in six Brazilian states (PR, SP, DF, MT, MS and MG).

		Effect Size					WB Control (%)		
Fungicide[a]	***k***[b]	$\bar{L}_{IND}$	**SE($\bar{L}$)**	***CI*$_L$**[c]	***CI*$_U$**[c]	***P value***	$\bar{C}$	***CI*$_L$**[c]	***CI*$_U$**[c]
MANC	33	-0.8677	0.0904	-1.0449	-0.6905	<0.0001	58.00	49.86	64.82
AZOX + TEBU	18	-0.7946	0.1144	-1.0188	-0.5704	<0.0001	54.82	43.46	63.89
TFLX+PROT	39	-0.7745	0.0938	-0.9583	-0.5907	<0.0001	53.90	44.60	61.64
TEBU	18	-0.7678	0.1035	-0.9706	-0.5650	<0.0001	53.59	43.16	62.11
TFLX+TEBU	39	-0.7395	0.0854	-0.9068	-0.5722	<0.0001	52.26	43.56	59.61
PYRA+EPOX	19	-0.5669	0.0939	-0.7509	-0.3829	<0.0001	43.27	31.81	52.80

[a] See Table 1 for complete information of the evaluated fungicides;
[b] number of trials that each fungicide was evaluated;
[c] upper (CI_U) and lower (CI_L) limits of the 95% confidence interval around $\bar{L}_{IND}$ and $\bar{C}$.

Table S3. Overall means and respective confidence intervals of unstandardized difference in wheat grain yield ($\overline{D}$) between fungicide-treated and untreated plots, and percent yield increase ($\overline{Y}$) for six selected fungicide treatments evaluated during nine years (2012 to 2020) across 44 studies conducted in six Brazilian states (PR, SP, DF, MT, MS and MG).

		Effect size					**Yield Return (%)**		
Fungicide[a]	***k***[b]	$\overline{D}$	**SE($\overline{D}$)**	CI_L[c]	CI_U[c]	***P* value**	$\overline{Y}$	CI_L[c]	CI_U[c]
MANC	35	420.10	53.30	315.62	524.57	<0.0001	47.55	32.25	64.62
TEBU	19	319.46	49.91	221.63	417.29	<0.0001	38.68	26.54	51.98
AZOX + TEBU	19	301.48	43.75	215.73	387.24	<0.0001	37.01	23.07	52.51
TFLX+PROT	42	299.73	39.69	221.92	377.53	<0.0001	32.42	21.84	43.93
TFLX+TEBU	42	245.34	36.98	172.85	317.83	<0.0001	29.08	19.13	39.86
PYRA+EPOX	22	181.15	33.85	114.79	247.50	<0.0001	23.76	14.62	33.64

[a] See Table 1 for complete information on the fungicides;
[b] number of trials that each fungicide was evaluated;
[c] upper (CI_U) and lower (CI_L) limits of the 95% confidence interval around $\overline{D}$ and $\overline{Y}$.