

EFFICACY OF FUNGICIDES AGAINST PEANUT SMUT IN ARGENTINA

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Abstract

Peanut smut, caused by *Thecaphora frezii*, increased its incidence and prevalence in the main growing region of Argentina in the last decade becoming the main peanut disease. Despite this fact, growers continue producing peanut without any disease management strategy what is leading to a local accumulation of *T. frezii* inoculum. The goal of the present study was to assess the efficacy of 12 fungicides for controlling *T. frezii*: *in vitro* assays, pots and field experiments were performed during 2014/15 and 2015/16. *In vitro* assays classified the fungicides upon their capacity to inhibit *T. freezy* mycelium growth as: thiophanate-methyl - not fungitoxic (EC₅₀ > 100); mancozeb - moderately fungitoxic (EC₅₀ = 6.28); difenoconazole, propiconazole, cyproconazole, tebuconazole, picoxystrobin, azoxystrobin, kresoxim-methyl, pyraclostrobin, penthiopyrad and chlorothalonil - highly fungitoxic (EC₅₀ < 0.1). Azoxystrobin showed the highest control levels in pot experiments: 58.9% (2014/15) and 92% (2015/16). The greatest control efficiency in field experiments was observed for cyproconazole in 2015 and azoxystrobin in 2016, reducing peanut smut by 47.7 and 39.5% respectively. Based on our results, chemical control can be considered as a moderately efficient technique which may complement the

cultivar genetic resistance and cultural practices in an integrated approach for managing peanut smut.

Key Words: *Thecaphora frezii*; Chemical control; *in vitro* sensitivity; peanut disease; active ingredients

1. Introduction

Peanut (*Arachis hypogaea* L.) is an annual extensive crop, with its center of origin in northwestern Argentina and southeastern Bolivia (Hammons et al., 2016). Argentina produces a high-quality peanut in an area of 350,000 has, concentrating more than 90% in the province of Córdoba located at the center region of the country. More than 95% of the peanut production is exported, previously processed in situ by the industry established in the same growing region (Agüero, 2017; Calzada and Rozadilla, 2018).

Peanut smut, caused by the soil-borne fungi *Thecaphora frezii* Carranza and Lindquist, is an endemic and yield reducing disease for the argentine main growing region (Marinelli et al., 2008, 2010; Rago et al., 2014). The pathogen is a biotrophic fungus that produces sori consisting of a powdery mass of spores which colonize the seed tissue partially or totally, with potential total damage of the plant pods (Astiz Gasso et al., 2008). The infection process occurs during the crop pegging stage: when flower gynophore penetrates the soil surface, releasing an exudate that stimulate the spore germination (Marinelli et al., 2008).

Currently, no management strategies for reducing *T. frezii* infections are adopted by argentine growers, which is leading to inter-annual inoculum accumulation (Paredes et al., 2017). Granoleico peanut cultivar (known susceptibility to the smut) is planted in more than 80% of the growing region (Cazón et al., 2018; Cignetti et al., 2010; Rago et al., 2017). Breeding programs with the aim of incorporating wild resistance genes have initiated recently, but new genotypes are not expected to be adopted in the short term (Bressano et al., 2019).

Survival structure can remain viable for new infections up to four years (Cazón et al., 2016b) which implies that a short peanut crop rotation is an inefficient disease management technique in the current argentine production system. Preliminary research suggested certain levels of fungicide efficiency, applications with higher doses of DMI + QoI mixture fungicides reported to provide best control (Cazón et al., 2013; Paredes et al., 2015b). In previous studies, we observed 58% smut control by spraying a mixture of azoxystrobin and cyproconazole at a rate of 1000 cm³/ha, which is twice and half the recommended dose for peanut late leaf spot management caused by *Nothopassalora personata* (syn. *Cercosporidium personatum*) (Paredes et al., 2015a). In addition, we compare the effect of timing of the day on the fungicide smut control efficacy: fungicide applications during the night had higher control efficacy than sprayings during the day (Paredes et al., 2015c). The application timing effect can be due to the fact that peanut plants fold their leaves at night and open them in the daytime (a process known as nyctinasty), allowing for more spraying drops to reach the soil surface.

No studies of fungicide sensitiveness of local *T. frezii* populations are registered in Argentina, and this information is essential for setting up of a chemical management strategy. The lack of studies examining a wide set of fungicides to control peanut smut led us to perform this study. The objectives of this work were i) to assess the fungicides efficacy against *T. frezii* in in vitro conditions and ii) to test the field performance of fungicides against peanut smut.

2. Materials and Methods

2.1. In vitro sensitivity of fungicides based on mycelial growth inhibition test

To measure the *in vitro* sensitivity of fungicides, a mycelial growth test was carried out. A mixture of 10 *T. frezii* isolates obtained from smutted pods in 2015 in General Deheza (Córdoba province, Argentina) were used for the fungicide sensitivity test. Smut teliospores (100 mg) were suspended in aqueous solution of 0.63% NaOCl, agitated during 5 minutes, rinsed

twice with sterile distilled water and resuspended in 2 ml sterile distilled water. Disinfected teliospores were placed in potato dextrose agar made up of broth extract of healthy grains at a rate of 50 g/l (Potato Peanut Dextrose Agar, PPDA). Plates were incubated at $25 \pm 1^\circ \text{C}$ in darkness (Astiz Gasso and Wojszko, 2011). After 7-10 days, spores of *T. frezii* were germinated and colonies were transferred to a culture medium potato-dextrose agar 2% to obtain *T. frezii* pure colonies. The identities of all isolates were confirmed with specific primers for *T. frezii* (Cazón et al., 2016a).

A total of twelve fungicides products (isolated active ingredient and adjuvant) varying in their mode of action were included in the study (Table 1). Five concentrations of each active ingredient were analyzed: 0.01, 0.10, 1.00, 10.00 and 100.00 $\mu\text{g a.i./ml}$ medium. Petri plates without fungicides were used as experimental control. Each experimental unit was represented by a plate (55 mm diameter), with five repetitions per treatment.

Mycelial discs (6 mm in diameter) were extracted from actively growing culture isolates *T. frezii* (8-day-old) and transferred to the center of Petri plates containing PDA amended medium. Five replicated plates were used for each fungicide concentration incubated at 25°C in darkness.

The radial growth (colony diameter) of each isolate was measured with a digital caliper in two perpendicular directions, and it was subtracted to the original mycelial disc diameter (6 mm). The measurements were taken when the control plates mycelium reached the plate edge. The inhibition of mycelial growth relative to the treatment without fungicide was calculated:

$$GI_i = (CD_{\text{check}} - CD_i) / CD_{\text{check}} \times 100 \quad (1)$$

Where GI_i is the inhibition of i colony growth; CD is the mean colony diameter for the control without fungicide (check) or for the i fungicide. Linear model regression was fitted to the GI obtained along the logarithmic doses, and the EC_{50} was estimated by replacing components of the fitted equations.

The fungitoxicity of fungicides was classified according Tonin et al. (2015): EC₅₀<1 µg/ml is considered as highly fungitoxic; between 1 to 50 µg/ml as moderately fungitoxic; EC₅₀ > 50 µg/ml as non-toxic.

2.2. Fungicides efficacy to control peanut smut trials

Two sets of replicated experiments were conducted during growing seasons 2014/2015 (from now on “2015”) and 2015/2016 (from now on “2016”). Peanut cultivar Granoleico, was used in all experiments due to the known susceptibility to smut (Oddino et al., 2013). Twelve fungicides labeled for peanut leaf spot were tested (Table 1). A non-treated control was included in each experiment in which we sprayed water. We used one and a half fold the active ingredient dose recommended for peanut leaf spot control. One set of trials was conducted in pots and the other one in field plots.

Table 1. Fungicides included in the study: chemical group, commercial name, formulation type, doses (for peanut leaf spot and peanut smut)

Chemical group	Active ingredient	Trading name® (Company)	Formulation	Commercial rate for leaf spot (g ai/ha)	Rate used for peanut smut (g ai/ha)
QoI	Picoxystrobin	*	SC 20%	80	120
QoI	Azoxystrobin	Amistar (Syngenta)	SC 25%	90	135
QoI	Kresoxim-Methyl	*	SC 18%	125	190
QoI	Pyraclostrobin	*	SC 25%	100	150
DMI	Difenoconazole	*	SC 16%	106	160
DMI	Propiconazole	*	SC 25%	135	200
DMI	Cyproconazole	*	SC 15%	50	75
DMI	Tebuconazole	Tebuco 25 (NOVA)	SC 25%	200	300
SDHI	Penthiopyrad	*	SC 10%	80	120
EBDC	Mancozeb	Mancozeb 75 (Nufarm)	WG 75%	1100	1700
MBC	Thiophanate-Methyl	Abrigo (NOVA)	SC 50%	500	750
Chloronitriles	Chlorothalonil	Funda (Gleba)	SC 50%	1000	1500

* Experimental compounds provide by company. Rate were calculated comparatively as mixture in commercial

products

2.2.1. Field experiment

Field trials were conducted at Aceitera General Deheza (AGD) experimental station in General Deheza (province of Córdoba, Argentina - 32° 45' 20.53"S, 63° 46' 56.5"W), located in the main peanut producing region with high *T. frezii* inoculum field pressure (Paredes, 2017). The inoculum concentration in the experimental area was higher than 4500 smut teliospores/g of soil. Treatments were distributed upon a randomized complete block design with four replications. Plots consisted of four rows of peanut seedlings at 0.7-m spacing and 8 m long. Fungicides were applied at night (between 9 - 11 pm) considering the soil as spraying target, with a CO₂ pressurized backpack and handheld boom equipped with 4-nozzles (hollow cone, ALBUS ATR 80) spaced 50-cm, delivering 120 L/ha at a pressure of 310 kPa and 0.9 m/s spray velocity. The first application was done seven days after R2 stage (beginning peg) (Boote, 1982) and the second one 10 days after the first one.

2.2.2. Pots experiments

Two experiments with peanut plants growing in pots were conducted at INTA research facilities in Córdoba city (-31° 28' 3.8"S, -64° 8' 50.4"W). Single peanut plants were grown in 10 L pots containing a substrate composed of a 3:1 mixture of sterilized soil-vermiculite and *T. frezii* teliospores. Inoculum was added at a rate of 10,000 teliospores/g of substrate by drenching of a *T. frezii* spore suspension. *T. frezii* spores and soil used to fill the pots were obtained from the experimental area of the field trials. A completely randomized design, with five (2014/15, from now on 2015) and eight (2015/16, from now on 2016) repetitions was used for each treatment. Treatments consisted of two fungicide sprayings: at 7 to 10 days after R2 (beginning peg) and 10 days after the first one. Each fungicide application consisted of three manual sprays (approximately 2.5 ml), directed to the plant base and pegs.

2.3. Disease assessment

Smut intensity assessment was performed at physiological mature crop stage (R8, Boote, 1982) (Rago et al., 2017). Severity was visually estimated using a five-class scale (Astiz Gasso et al., 2008) which takes into account both grains disease status within the pod (Figure 1). Disease assessment in the field plots experiment was performed by collecting 1 m² from the two central grooves of each plot (and all pods produced in that area were evaluated) and in the pots experiment all the pods contained in the pots were evaluated.

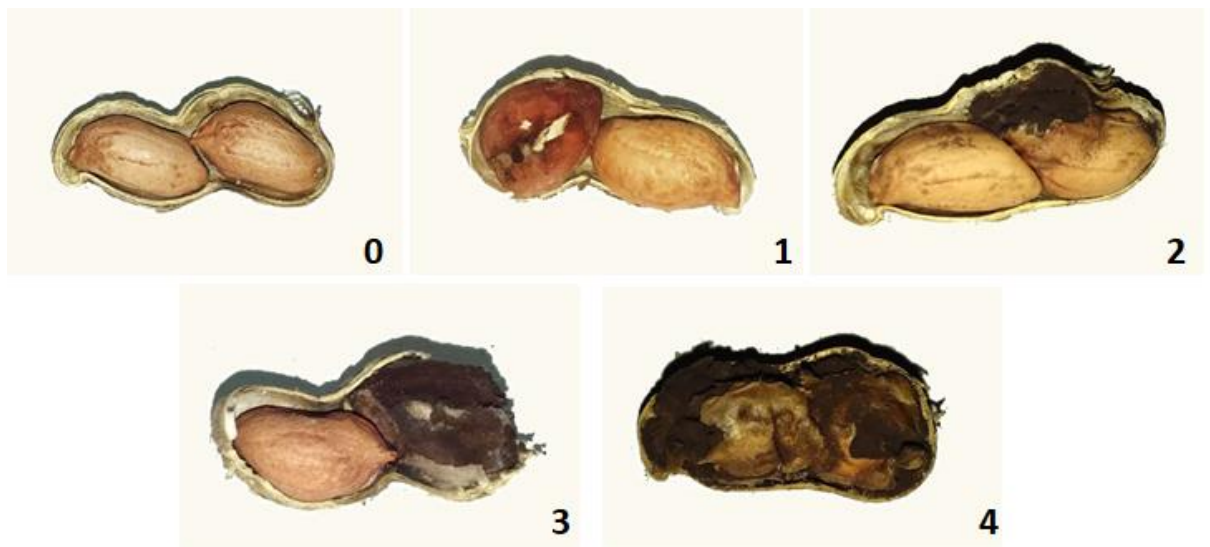


Figure 1. Five-class severity scale for peanut smut, where: 0 = both healthy kernels. 1 = presence of a small sorus in single kernel. 2 = at least one kernel with half area affected. 3 = deformed pod, with one single kernel completely smutted. 4 = deformed pod with two kernels completely smutted

For the purposes of this analysis, we used the proportion of severely damaged pods (SDP, disease class 3 and 4) as the response variable. Disease levels for these classes are considered to have impact in inoculum increase as they are lost at harvest and produces are discarded by the industry because of quality issues (Morichetti S. personal communication). SDP was calculated as:

$$SDP = (n_3 + n_4) / N \quad (2)$$

where n_3 and n_4 are the number of pods corresponding to disease severity class 3 and 4 respectively and N is the plot sample size.

Then, for graphical displaying of the results, smut control efficiency (CE) was calculated by using the following formula:

$$CE = (1 - (\text{Treatment SDP} / \text{untreated check SDP})) * 100 \quad (3)$$

2.4. Data statistical analysis

Univariate generalized linear models (GLMs) with binomial variance and logit link function were used to analyze SDP. The full model included the year - treatment interaction, which resulted significant ($P < 0.01$), then a separated model was fitted to each year. Treatment marginal means were estimated with the ‘emmeans’ function from the emmeans R package (Lenth et al., 2018) and post-hoc comparisons among them were done with Tukey test at a significance level of 5%.

Multivariate analysis was performed to integrate all the efficacy tests (Deepak et al., 2006): principal component analysis (PCA) was conducted to the scaled variables, as: $(x - \text{mean}(x)) / \text{sd}(x)$, where x is each variable, and the scaling is done by dividing the (centered) vectors of x by their standard deviations. CE% were obtained from the field plots and peanut plants grown in pots trials (2015 and 2016). We obtained the correlation coefficients for each variable with the first and second principal components to explain the variability explained by the first and second principal component. Each fungicide (individuals, represented by points) and their performance in each trial (variables, represented by vectors) are displayed together on a biplot.

3. Results

3.1. *In vitro* sensitivity

The growth of *T. frezii* isolates was 4.76 (\pm 0.14) mm per day in non-treated control plates. A wide range of sensitivity of *T. frezii* isolates was observed to the evaluated fungicides: EC50 value ranging from 0.001 to >100. The highest mycelial growth inhibitions was observed with the DMI's fungicides, QoI's pyraclostrobin and azoxystrobin, and penthiopyrad, EC50<0.1 (Figure 2). Moderate sensitivity was observed with mancozeb (EC50 = 6.28). The lowest sensitivity of *T. frezii* was observed with thiophanate-methyl (EC50 > 100 μ g/ml). According to the EC50, only thiophanate-methyl was classified as non-toxic, and mancozeb as moderately fungitoxic, the rest of the a.i. were classified as highly fungitoxic.

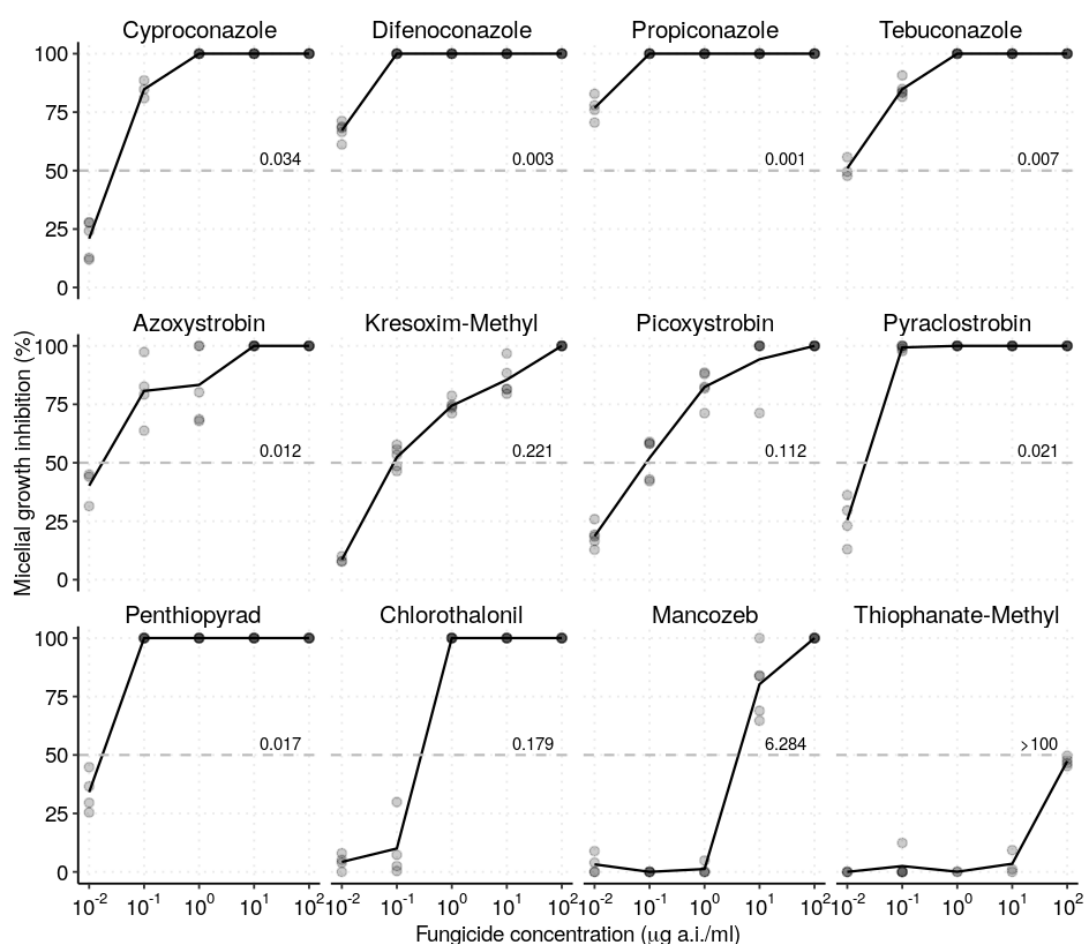


Figure 2. Mycelial growth inhibition curves according to fungicide concentration (μ g a.i./ml) for the twelve active ingredients evaluated. Numbers above the central dashed line corresponds to EC50 value.

3.2. Control efficacy experiments

Accumulated precipitations and mean temperature registered in General Deheza during the peanut cropping season (December to April) in 2014/15, 2015/16 are presented in Table S1. A normal crop development (without severe hydric stress) was observed in both field experiments. A total precipitation of 465 and 417 mm was recorded during the pegging phenological stage (January to March) in 2015 and 2016 respectively.

Smut pressure in pot experiments, indicated by the mean SDP at the non-treated check plants, was higher in 2015 (0.73) than 2016 (0.52). Azoxystrobin resulted the lowest SDP values in both years: 0.3 and 0.04 in 2015 and 2016, respectively. The latter values represented 59% and 92% control efficacy relative to their corresponding non-treated control. No significant differences in smut control were observed with difenoconazole (0.43 SDP, 41% control efficiency) in 2015 or from cyproconazole (0.09 SPD, 82% control efficiency) or tebuconazole (0.13 SPD, 74% control efficiency) in 2016. Intermediate smut control was observed for tebuconazole, picoxystrobin and cyproconazole in 2015, and picoxystrobin, propiconazole, mancozeb, difenoconazole and kresoxim-methyl in 2016. On the other hand, the mean SDP for chlorothalonil, penthiopyrad, thiophanate-methyl and pyraclostrobin did not differ from the nontreated check in both years (Table 2; Figure 3).

Table 2. Severely damaged pods, control efficiency, and corresponding statistics for the 12 fungicides evaluated for the control of peanut smut on pots experiments in 2015 and 2016.

Active ingredients	2015				Active ingredients	2016			
	SDP ^a	CL _L ^b	CL _U ^b	CE ^c		SDP	CL _L	CL _U	CE
Azoxystrobin	0.30	0.23	0.39	58.9	Azoxystrobin	0.04	0.02	0.09	92
Difenoconazole	0.43	0.36	0.51	41.1	Cyproconazole	0.09	0.06	0.14	82
Tebuconazole	0.51	0.44	0.58	30.1	Tebuconazole	0.13	0.08	0.19	74
Picoxystrobin	0.54	0.46	0.61	26	Picoxystrobin	0.19	0.14	0.25	62
Cyproconazole	0.56	0.48	0.63	23.3	Propiconazole	0.27	0.2	0.35	46

Kresoxim-Methyl	0.60	0.52	0.67	17.8	Mancozeb	0.28	0.21	0.36	44
Thiophanate-Methyl	0.65	0.57	0.72	11	Difenoconazole	0.29	0.22	0.36	42
Mancozeb	0.65	0.58	0.71	11	Kresoxim-Methyl	0.32	0.26	0.39	36
Pyraclostrobin	0.67	0.60	0.73	8.2	Pyraclostrobin	0.39	0.33	0.47	22
Propiconazole	0.71	0.63	0.78	2.7	Penthiopyrad	0.43	0.36	0.5	14
Penthiopyrad	0.72	0.63	0.79	1.4	Check	0.5	0.44	0.56	0
Check	0.73	0.65	0.80	0	Thiophanate-Methyl	0.51	0.43	0.58	0
Chlorothalonil	0.74	0.68	0.80	0	Chlorothalonil	0.52	0.45	0.59	0

^a Proportion of severely damaged pods (disease classes: 3 - deformed pod, with one single kernel

completely smutted and 4 - deformed pod with two kernels completely smutted).

^b Lower (CI_L) and upper (CI_U) limits of 95% confidence interval around SDP estimated mean.

^c Percentages of control efficiency of active ingredients calculated in relation to the non-treated check.

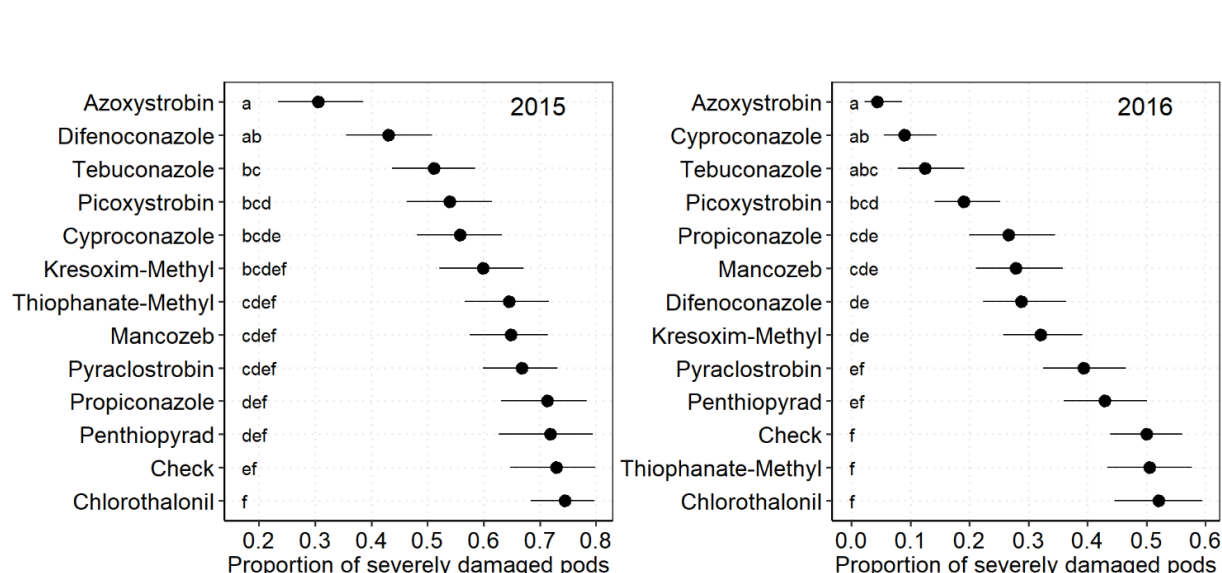


Figure 3. Proportion of severely damaged pods for different active ingredients used for the control of peanut smut on pots experiment in 2015 and 2016.

In the field experiments, mean SDP at the non-treated control was 0.39 (2015) and 0.43

(2016). Cyproconazole and azoxystrobin in 2015 presented the lowest SDP values: 0.21 and 0.25

(control efficacy of 47% and 37%, respectively) ($P < 0.05$). Azoxystrobin was the most efficient

fungicide in 2016 to control smut, with a lowest SPD: 0.26. No significant differences of SPD

with the non-treated control was observed for kresoxim-methyl, thiophanate-methyl, tebuconazole, penthiopyrad and chlorothalonil for both years (Table 3; Figure 4).

Table 3. Severely damaged pods, control efficiency, and corresponding statistics for the 12 fungicides evaluated for the control of peanut smut on field experiments in harvest 2015 and 2016.

Active ingredients	2015				Active ingredients	2016			
	SDP ^a	CL _L ^b	CL _U ^b	CE ^c		SDP	CL _L	CL _U	CE
Cyproconazole	0.21	0.19	0.23	47.7	Azoxystrobin	0.26	0.24	0.29	39.5
Azoxystrobin	0.25	0.22	0.27	37.2	Propiconazole	0.33	0.31	0.35	23.7
Mancozeb	0.29	0.27	0.31	25.8	Cyproconazole	0.34	0.32	0.37	20.9
Pyraclostrobin	0.31	0.29	0.34	20.7	Picoxystrobin	0.38	0.35	0.40	13.8
Picoxystrobin	0.32	0.29	0.35	18.9	Difenoconazole	0.38	0.35	0.40	13.3
Propiconazole	0.32	0.30	0.34	17.9	Thiophanate-Methyl	0.38	0.35	0.41	12.4
Chlorothalonil	0.33	0.30	0.35	16.8	Chlorothalonil	0.38	0.36	0.41	12.2
Kresoxim-Methyl	0.33	0.30	0.35	16.6	Mancozeb	0.38	0.35	0.41	12.0
Thiophanate-Methyl	0.35	0.32	0.37	11.7	Kresoxim-Methyl	0.39	0.36	0.43	9.9
Difenoconazole	0.36	0.34	0.38	8.2	Penthiopyrad	0.40	0.38	0.42	7.8
Tebuconazole	0.36	0.34	0.39	7.4	Pyraclostrobin	0.42	0.40	0.45	3.2
Penthiopyrad	0.37	0.35	0.40	4.8	Check	0.44	0.41	0.46	-
Check	0.39	0.36	0.42	-	Tebuconazole	0.45	0.42	0.48	0

^a Mean of the proportion of severely damaged pods (disease class 3 and 4)

^b Lower (CL_L) and upper (CL_U) limits of 95% confidence interval around SDP

^c Percentages of control efficiency of active ingredients calculated in relation to the untreated check treatment.

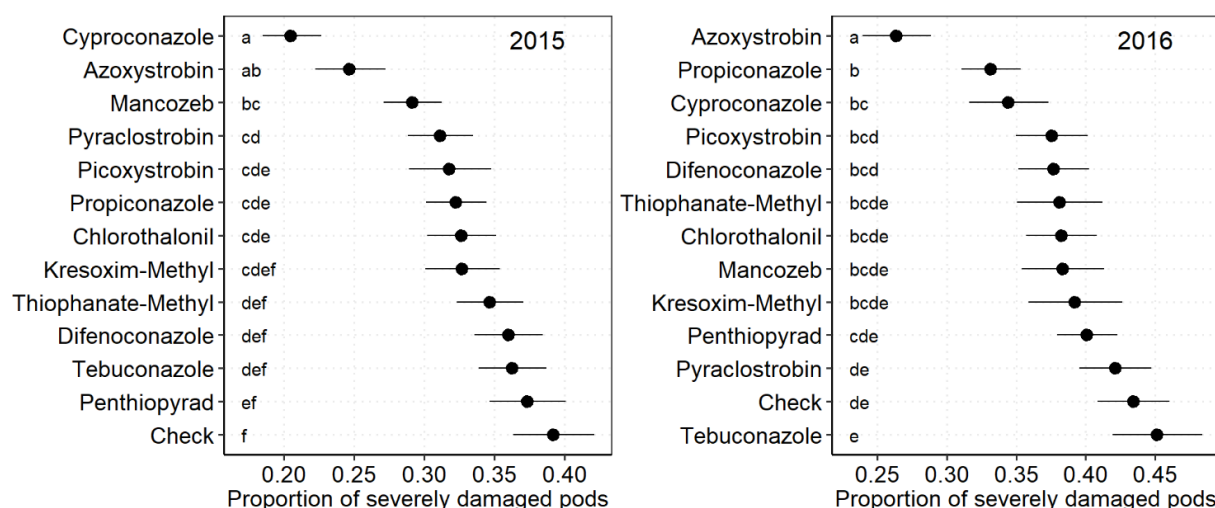


Figure 4. Proportion of severely damaged pods for different active ingredients used for the control of peanut smut on field experiments in 2015 and 2016.

3.3. Multivariate analysis

The multivariate analysis allowed us to integrate all the trials results: two independent principal components (PC) accounted for 76.5% of the total variation (56.2% and 20.3% for the PC1 and PC2 respectively). PC1 (horizontal axe) represented the control efficacy values in the field plots trials (with correlation coefficients of 0.86 and 0.77, in 2015 and 2016 respectively) and pots experiments (correlation coefficients were 0.83 and 0.73, in 2015 and 2016 respectively). The further right on the PC1-axis, the higher the efficacy control of the fungicide (Figure 5). PC2 on the y-axis represented the EC50 values: the upper in the axis, the lower the sensitivity of the smut to the fungicide. Considering this coordinate meaning, we may select as best fungicides those ones located in the right quadrants, and the lower as possible: azoxystrobin and picoxystrobin (QoI group) and cyproconazole and difenoconazole (DMI group).

We observed that control efficacy in 2015 followed a similar trend in both types of experiments (plants grown in pots and field plots), but a weak correlation among both experimental types was observed in 2016.

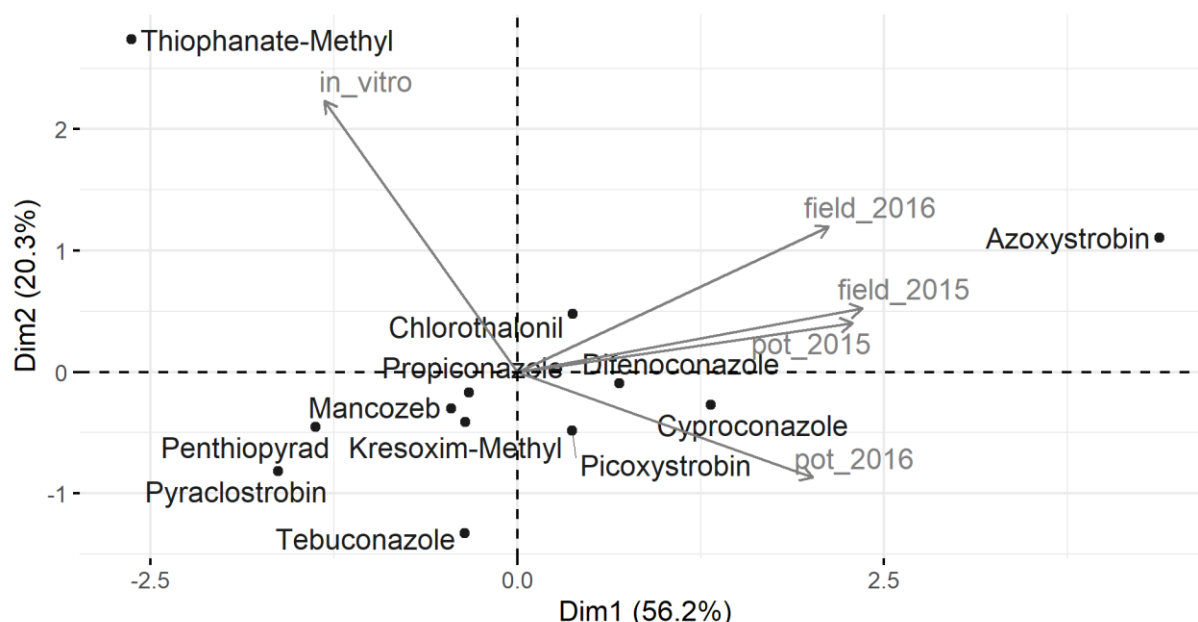


Figure 5. Biplot model showing the relationship among fungicides (individuals, represented by points) and their performance in each trial (variables, represented by vectors): in vitro experiments and pot and field experiment in 2015 and 2016.

4. Discussion

Since its re-emergence in 2006, peanut smut has continued increasing its prevalence and incidence in the main Argentine peanut-growing region, without good perspectives for the future due to the current absence of adoption of management practices for controlling smut (Rago et al., 2017). By means of the present two-year fungicide-screening study we observed variability in the available set of commercial fungicides, including highly efficacious active ingredients to non-effective ones. To the best of our knowledge this is a first study assessing the chemical management as a technique to maintain peanut smut in low intensity levels.

In vitro mycelial growth inhibition tests showed a general good performance of the DMI's group, QoI's azoxystrobin and pyraclostrobin and SDHI's penthiopyrad. The two last chemical groups are powerful inhibitors of spore germination (Fungicide Resistance Action Committee [FRAC] group 11 and 7), a process that was not evaluated in the *in vitro* tests. This fact could

278 have a potential sub-estimation effect of the evaluated active ingredients since the smut spore
279 germination is a fundamental first step on epidemic onset after the gynophore exudates
280 stimulation (Astiz Gasso et al., 2008; Marinelli et al., 2008). In relation to this both fungi cycle
281 processes, Kosiada (2011) observed a weak correlation between mycelial growth and
282 germination of teliospores and basidiospores according to the sensitivity of some fungicides for
283 head smut of corn (*Sphacelotheca reiliana*).

284 Another weak correlation was reported between results of laboratory and field conditions
285 experiments (Hollowell et al., 2003), which led us to perform pots and field trials for assessing
286 the fungicide efficacy to control peanut smut. However, in both years of the study we observed
287 that azoxystrobin was the fungicide with highest smut control efficacy levels in pot or field
288 experiments.

289 Pavez Badilla et al. (2013), showed different levels of sensitivity *in vitro* for
290 pyraclostrobin and kresoxim-methyl (QoI) in front of the *Venturia inaequalis*, nevertheless they
291 assure that it is very difficult to extrapolate this type of investigation to the field conditions due
292 to the action mode. Differences in control levels for peanut smut were observed compared to *in*
293 *vitro* results. The DMIs a.i. showed EC50 lower than the rest of the chemical groups.
294 Cyproconazole has the highest EC50 comparatively with other DMIs, however it is one of the
295 a.i. that shows the highest control efficiency in field and pots experiments. For its part,
296 chlorothalonil and penthiopyrad (SDHI) have an EC50 like QoI, however did not demonstrate
297 disease control efficiency. Azoxystrobin, picoxystrobin, cyproconazole and tebuconazole were
298 the a.i. best responses demonstrated in the assessment; however, they are not the lowest EC50
299 within the chemical groups to which they belong.

300 The lack of consistency of the observed results between pot and field experiments can be
301 due to differences in soil properties in both experimental conditions: soil humidity content may
302 lead to different fungicide degradation rates or particles absorption. Further studies may be done

for a better understanding of the latter process and consequently a more efficient use of fungicides against *T. frezii* infections. Paredes et al. (data not published) observed in growing chambers experiments that the lower the soil humidity, the higher the incidence of smut in peanut plants.

QoI, SDHI and DMI are the most common fungicides used to control peanut diseases in crops. These fungicides groups performed less than 40-50% effective to peanut smut control in field, even when used at higher doses comparatively leaf spot (Rago et al., 2017). Mutations in genes encoding fungicide targets of pathogenic fungi are often the cause of developed resistance to QoI, SDHI and DMI fungicides (Sierotzki and Scalliet, 2013). Nevertheless, research on the molecular basis of *T. frezii* fungicide resistance has not been possible due to the lack of genetic information. Recently, Arias et al., (2019) report the complete mitogenome of *T. frezii* and provide molecular tools to study fungicide target genes and suggests potential resistance to strobilurin and carboxamide fungicides.

Individual performances of cyproconazole and azoxystrobin had the highest control efficacy in the present study. However, higher performances were observed as mixture formulations of the fungicides in previous field experiments under the same disease pressure (4400 teliospores/g of soil): azoxystrobin + cyproconazole and azoxystrobin + picoxystrobin reduced smut severity by 58 and 47% respectively relative to non-treated control (Paredes et al., 2015b, 2015a). This higher performance of the fungicides as mixtures could be due to a “potentiation” effect (Cid, 2014).

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Supplementary tables

Table S1: Data were extracted from AGD local station data: Rainfall (monthly precipitation), average monthly maximum and minimum temperatures in the experimental peanut area during 2014/15; 2015/16.

Month	Rainfall (mm)		Temperature (°C)					
	2015/16	2016/17	Mean		Average max.		Average min.	
			2015	2016	2015	2016	2015	2016
December	38	122	22	24	32	31	14	16
January	89	123	24	23	32	30	18	18
February	255	249	21	24	28	30	16	18
March	121	45	21	19	28	26	16	13
April	57	143	20	15	28	21	13	11
Acumulated	560	682						