A meta-analysis of fungicide spray schedules to minimise yield loss from powdery mildew in Australian mungbean

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8   article, it must not contain results which are not presented and substantiated in the main text and
9   should not exaggerate the main conclusions.

10   **Keywords:** keyword 1; keyword 2; keyword 3 (list three to ten pertinent keywords specific to the
11   article, yet reasonably common within the subject discipline.).

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12   Mungbean [*Vigna radiata* (L.) Wilczek] is a pulse crop primarily grown in south-east Asia for
13   human consumption as an affordable source of protein [1]. The bean pod or grain can be consumed
14   raw or added to meals after sprouting. The dried grain can also be ground into a protein enriched
15   flour for uses in noodles, biscuits and cakes [2].
16   Mungbean was first brought to Australia in the 1930s for use as a forage or green manure crop.
17   However, it was only somewhat recently, during the 1960s that mungbean was grown commercially
18   [3,4]. Prior to the 1970s the total area grown in Australia rarely exceeded 1000 hectares [3]. At the end
19   of the 1980s, between 3,000 to 10,0000 hectares were being harvested for dried beans in Australia, of
20   which mungbean is categorised by the FAO statistics data repository [5]. In the decade leading up
21   to 2018, between 35 - 86.4 thousand hectares of dried beans were planted annually. This increase is
22   attributed to high value export markets and to the improved yields in the new cultivars [6]. Currently
23   mungbean is predominantly grown in southern Queensland and northern New South Wales as a short
24   season summer legume crop. In 2019, approximately 90% of Australian mungbean was grown for
25   export with lucrative returns of up to $1300 AU per tonne [7]. Mungbean potentially yields up to three
26   tonnes per hectare [8], but due to high variability in yields between seasons and locations, the average
27   Australian farm still yields less than one tonne per hectare [4].
28   The high variability in yields can in part be attributed to a range of diseases that affect the crop [9].
29   Two major foliar diseases, tan spot or common wilt (*Curtobacterium flaccumfaciens* pv. *flaccumfaciens*)
30   and halo blight (*Pseudomonas savastanoi* pv. *phaseolicola*), are the main focus of the Australian Mungbean
31   Improvement programme's resistance breeding efforts due to lack of effective chemical controls
32   for them. While a third disease, powdery mildew, can reduce yields by up to 40% [2] in susceptible

cultivars, the damage can be mitigated through an integrated disease management strategy of fungicide treatments and cultural practices.

While the pathogens are obligate parasites, secondary hosts that allow the pathogens to over-season between mungbean crops have not yet been identified. The disease life cycle can be as short as five days for a germinating conidia to infect the host and produce new reproductive structures, conidiophores, which produce more conidia, which disseminate to new infection sites [10]. Weather conditions that favour the rapid development of the disease are cool temperatures between 22 ° and 26 °C, and a leaf surface that is not overly wet for infection [9].

Due to the requirement for cool temperatures for infection, planting date is an effective strategy for preventing the development of powdery mildew in mungbean [11]. In southern Queensland and northern New South Wales, sowing dates between late spring (November) and mid-summer (January) are advised to avoid the conducive cooler autumn temperatures, which normally commence in March. Therefore, delayed sowing dates of late January into February increase the possibility powdery mildew will infect earlier in the crop's life cycle and cause greater yield losses than in earlier sown mungbean crops.

Cultivar selection is an additional strategy to mitigate yield damage from powdery mildew. In Australia, quantitative resistance to powdery mildew has also been incorporated into few commercially available varieties. Jade-AU (pbr) has the the best disease resistance to powdery mildew (moderately susceptible), with cv. Green Diamond also containing some notable resistance [10]. However, commercially available mungbean cultivars in Australia lack sufficient resistance to be used as the primary powdery mildew management strategy. Evidence of this is shown by field trials conducted in 2016 with the moderately susceptible cultivar, Jade-AU, where yield losses of up to 32.7% were recorded [12]. Quantitative disease resistance has been observed in some breeding lines overseas [2,13], however no evidence for how they compare to Australian varietal resistance could be found. Additional strategies to cultivar choice, such as fungicide treatments, are necessary to limit yield loss when conditions for the disease are conducive.

There is almost a complete lack of peer-reviewed literature summarising fungicide efficacy on powdery mildew in Australian mungbean. Much of the experimental work has been published by funding agencies and state government extension departments as extension bulletins or other related type materials. Past field trials in eastern Australia have tested a range of fungicide active ingredients while also attempting to evaluate the best application time for the highest efficacy [9,12,14–18]. Early trials showed that for fungicide applications to protect yield, a single fungicide application at first sign of the disease with another follow-up application two weeks later, if necessary, was the most effective [10,12]. However, due to the variability in the results between seasons and experiments, few of these studies produced a clear result based on a statistical analysis. This variability presents uncertainty for the best practice of fungicide spray schedules to mitigate yield losses from powdery mildew.

The collection of experiments mentioned above and other unpublished trials present an excellent opportunity for a meta-analysis. Meta-analyses are statistical tools, which can analyse a collection of experiments, that have a similar aim, and produces a more accurate estimate of the true effect being measured. Using a meta-analysis can be useful in situations like this where several studies exist that have the same objective but may not provide a clear answer to the question due to variation in the results within the individual studies. The outcome of a meta-analysis provides a more accurate estimation of the true treatments effect because it considers the amount of variance in each study and weights the influence of each studies treatment effects according to its statistical accuracy. Typically meta-analyses consider the effect of a single treatment against a control group across multiple independent studies [19]. However meta-analyses can also consider effect differences between multiple treatments and a control group. These are called multi-variate or network meta-analyses [20]. Multi-variate meta-analyses are particularly useful when there are no direct comparisons between two treatments in any of the included studies. Indirect comparisons can be made between these two treatments if the both had direct comparisons with one or more treatments in common, assuming

no significant bias in the studies that investigated each of the treatments and which are subject to a indirect comparison [21].

Considering the recent launch in November 2019 of the decision support system (DSS) PowderyMildewMBM [22], which provides a cost benefit analysis to assists growers in their decision "if" and "when" apply fungicide; a meta-analysis to evaluate and verify the best fungicide spray schedule seemed prudent. The aim of this meta-analysis was to determine, from a collection of unpublished studies, what spray management scenario provides the greatest yield protection from powdery mildew in Australian mungbean.

1. Materials and Methods

1.1. Trial criteria for inclusion and description

The data for this study were obtained through personal correspondence with colleagues and collaborating institutions. Trials undertaken in the 2013 season were conducted by the Northern Growers Alliance (NGA) [2013; 2013; 2013; 2013]. Trial data were obtained in different formats with varying levels of information. Some trials we obtained the raw data, others the reported means of each treatment, with all but one study reporting some form of variance along with the mean. Twenty six trials in total were collated.

To ensure the correct question was asked of the analysis, the data needed to conform to a strict criteria. The criteria for inclusion of trial data in the meta-analysis required, a field trial testing fungicide efficacy for powdery mildew control on mungbean in the Grains Research and Development (GRDC) northern grains regions of eastern Australia, which grows the majority of Australia's summer crops. Data from trials undertaken in this region had to include: the date when powdery mildew was first observed; disease incidence at the end of the growing season; fungicide application dates; the fungicide active ingredients; fungicide dose; and crop yield. The data were subset to only include fungicide treatments with the same mode of action; demethylase inhibitor (DMI) fungicides, tebuconazole and propiconazole were thus retained in the dataset. Subsequently the 26 trials were reduced to 17 trials (Table 1). Two more trials were also removed because grain yield variance, which was required for the meta-analysis, was not reported, reducing the number of trials down to 15.

From the trials which were included in the meta-analysis, 2 were planted in late December, 5 in January and 8 were trials planted in February (Table 1).

In the meta-analysis, to ensure sufficient replication, we made no distinction between the tebuconazole and propiconazole fungicide treatments within the trials that met the selection criteria. The design of all trials included in this analysis were randomised complete block designs, and were not previously published in peer-reviewed literature. Details of trial data are presented within a research compendium as a supplement to this publication [].

Response variable - Grain yield: Grain yield (tonne per hectare), was used as the response variable for the meta-analysis. Mean grain yields for each treatment were either obtained directly from trial reports, or calculated from the raw data when available. The sample variance was calculated from the raw data when available or the least squares statistic in the trial reports. Sample variance for the 2012 Kingaroy study was calculated from the reported least squares statistic using a T-critical value of 2.042 and the approach reported in Ngugi et al. [2011]. The T-critical value was obtained from a t-distribution table using, 30 degrees of freedom and probability of $a = 0.05$.

Similar to Paul *et al..* [2007], we fitted a multivariate random effects model $Y_i \sim N(\mu, s_i^2 + \sigma^2)$ where Y_i is the response, a vector of the grain yield measurements (tonne per hectare) for each treatment in the *i*th trial over total number (K) of trials($i = 1, \dots K$). Y_i is estimated from a normal distribution with the mean μ, sample variance of S_i^2 and a between trial variance of σ^2.

Trial Variables and moderators: While the main aim of all the identified trials were to assess the efficacy of fungicide for control of powdery mildew, some trials tested additional potential covariates in the experimental design. These additional variables included: row spacing, fungicide active ingredient,

Table 1. Description of fungicide application experiments.

Unique Trial Reference	Year	Location	Planting Date	Date First Sign	Research Organisation
mung1011/01	2011	Hermitage	2011-01-24	2011-03-28	DAF[a]
mung1011/02	2011	Kingaroy	2011-02-02	2011-03-22	DAF[a]
mung1112/01*	2012	Gatton	2012-02-20	2012-04-02	DAF[a]
mung1112/02	2012	Kingaroy	2012-02-03	2012-03-12	DAF[a]
AM1303	2013	Premer	2012-12-28	2013-02-28	NGA[b]
AM1304	2013	Marys Mount	2012-12-24	2013-03-16	NGA[b]
AM1305	2013	Goolhi	2013-01-23	2013-03-25	NGA[b]
BB1305	2013	Millmerran	2013-01-12	2013-03-13	NGA[b]
mung1415/01	2015	Hermitage	2015-01-19	2015-03-16	DAF[a]
mung1516/01	2016	Hermitage	2016-02-03	2016-03-08	DAF[a]
mung1516/02	2016	Kingaroy	2016-02-11	2016-03-09	DAF[a]
mung1516/03*	2016	Emerald	2016-02-12	2016-03-17	DAF[a]
mung1617/01	2017	Hermitage	2017-02-13	2017-03-24	USQ[c]
mung1617/02	2017	Missen Flats	2017-01-27	2017-03-07	USQ[c]
mung1718/01	2018	Wellcamp	2018-02-13	2018-03-21	USQ[c]
mung1819/01	2019	Hermitage	2018-02-04	2018-04-12	USQ[c]
mung1819/02	2019	Hermitage	2018-02-18	2018-04-12	USQ[c]

[a] Queensland Department of Agriculture, Fisheries and Forestry
[b] Northern Growers Alliance
[c] University of Southern Queensland

fungicide dose, planting date, number of fungicide applications and cultivar. To account for the variation attributed to some of these variables between trials, and within trials, a single *trial* factor was created. The *trial* random variable made distinctions between the following variables: unique trial reference code, trial location, trial season, host genotype, row spacing, and fungicide dose.

The random effects meta-analysis was expanded to evaluate if the residual heterogeneity could be explained by spray schedule). Spray schedule was assigned as an inside factor to the outside random factor *trial* to provide correlated random effects between the different spray schedule treatments within *trials*.

The timing of fungicide application treatments, were defined in relation to the recorded date when powdery mildew first occurred in the trial. An *Early* fungicide application treatment, referred to a fungicide spray schedule that commenced prior to the disease being observed in the trial. *Recommended* referred to a fungicide spray schedule which commenced 1 – 3 days following the first sign of disease. *Late* treatments commenced between 7 and 13 days after the first sign of disease. There were no spray schedule treatments which began between 4 and 6 days after the first sign of powdery mildew. The number of fungicide applications were binned in to two categories. A spray schedule with a *single* spray, and a spray schedule with two or three spray applications *plus*. The division of the treatments produced: 13 $Early_{single}$ treatments; 5 $Early_{plus}$; 32 $Recommended_{single}$; 46 $Recommended_{plus}$; 17 $Late_{single}$; and 20 $Late_{plus}$. $Early_{plus}$ treatments were removed from the subsequent meta-analysis, due to an insufficient sample size. What remained was the mean yield for a total of 173 treatments in the following five spray schedule categories: no spray *control*, $Early_{single}$, $Recommended_{single}$, $Recommended_{plus}$, $Late_{single}$, and $Late_{plus}$.

An ordinal 1 – 9 scale was used to describe the mean powdery mildew plot severity in most trials (Table 2). However, the 2013 NGA trials (Table 1) reported mean plot severity as the percentage of leaves covered by powdery mildew and the location of the infected leaves in the lower, middle or upper canopy. Given the ordinal scale ranked plots on the percent of foliage showing the disease from the lower canopy to the upper canopy, a conversion from percentage of the diseased canopy, to the 1 – 9 scale was straight-forward. The end of season mean plot severity, and date of first sign of disease were used to calculate the area under the disease progress curve (AUDPC) for each treatment.

Table 2. Powdery mildew severity scoring scale.

Scale	Scale.Description
1	No sign of powdery mildew
2	Small colonies in lower 1/3 of the canopy, up to 75% of the plants affected
3	Colonies in lower 1/2 of the canopy, > 75% of the plants affected
4	Colonies in lower 2/3 of the canopy, up to 75% of the plants affected
5	Colonies in lower 2/3 of the canopy, > 75% of the plants affected
6	Colonies in lower 2/3 of the canopy, 100% of the plants affected
7	Colonies in lower 2/3 of the canopy, 100% of plants, with some plants with colonies in the top 1/3 of the canopy
8	Colonies to top of the plant with > 75% of the plants affected
9	Colonies to top of the plant with > 100% of the plants affected and heavy leaf drop

159 Additional powdery mildew mean plot severity observations made in-between these two observations
160 were also included in the AUDPC calculation if they were available. AUDPC was calculated using
161 the `agricolae` package in R [25]. A categorical disease pressure factor was created by binning trials
162 into two levels based on the mean AUDPC of the no spray control plot. The two levels, "low disease
163 pressure" and "high disease pressure", were separated by the median AUDPC (153.625) of the no
164 spray control plots.

165   The mean difference in grain yield was estimated from the difference in model estimates of the
166 non-treated control and each of the spray schedule treatments using fungicide.
167 Disease pressure was tested against the basic model to determine if, when introduced as a moderator
168 variable, it could significantly explain additional heterogeneity between the spray schedule treatments.
169 A a Wald-type test was used to test the impact of the disease pressure factor.

170   Linear contrasts were used to produce the mean difference in effect sizes, and their respective
171 standard errors and 95% confidence intervals, between each spray schedule treatment.

172   Additional meta-analysis models tested variables ... for their influence on the SpraySchedule
173 variables ability to mitigate yield loss. For the variable disease pressure a Wald-type test was used to
174 test the impact of this variable on the model as a whole.

175   The data were analysed in the R statistical software environment, version 4.0.1, [26] using two
176 contributed meta-analysis packages, `metafor`, version 2-4.0, [27] and `netmeta`, version 1.2-1, [28].

2. Results

178   *control*, $Early_{single}$, $Recommended_{single}$, $Recommended_{plus}$, $Late_{single}$, and $Late_{plus}$.
179 Overall the meta-analysis indicated that, with the exception of the $Early_{single}$ treatment, all the the
180 spray schedules were effective at protecting mungbean yields relative to the no spray controls (Figure
181 1). Treatments which incorporating multiple fungicide applications, $Recommended_{plus}$ and $Late_{plus}$,
182 produced the highest mean yield protection estimates at 177.83 (SE = 41.69 kg / ha) and 147.51 kg / ha
183 (SE = 42.33 kg / ha) respectively (Table 3). Both $Recommended_{plus}$ and $Late_{plus}$ were also significantly
184 higher than single applications $Early_{single}$ (P = 0.0078 and P = 0.0025 respectively) and $Late_{single}$(P =
185 0.0476 and P = 0.0283 respectively). However, comparing $Late_{plus}$ and $Early_{single}$ should done with
186 caution given these treatments did not occur within the same trial for any of the 15 trials included
187 in the meta-analysis. Within the single fungicide application treatments `Recommended` provided the
188 highest yield protection, and was estimated at saving an average of 159.9 kg / ha (SE = 30.5 kg / ha)
189 relative to the no spray control (P < 0.0001). `Recommended` treatments were was also significantly higher
190 than `Early` treatments by 122.1 kg / ha (SE = 42.1 kg / ha)(P = 0.004). Single `Late` applications were
191 effective at significantly mitigating the effect of powdery mildew on yield by a mean of 122 kg / ha
192 (SE = 39.2 kg / ha)(P = 0.0019). However, this estimate was not significantly different to single `Early`
193 or `Recommended` treatments (P = 0.119; P = 0.278 respectively).

194   The Q_M omnibus test of moderators, shows the moderators significantly influence the model
195 ($Q_M = 31.1217$, $df = 5$, P < 0.0001) and we can reject the null hypothesis ($H_0 : \beta_1 = \beta_2 = \beta_3 = \beta_4 = 0$)
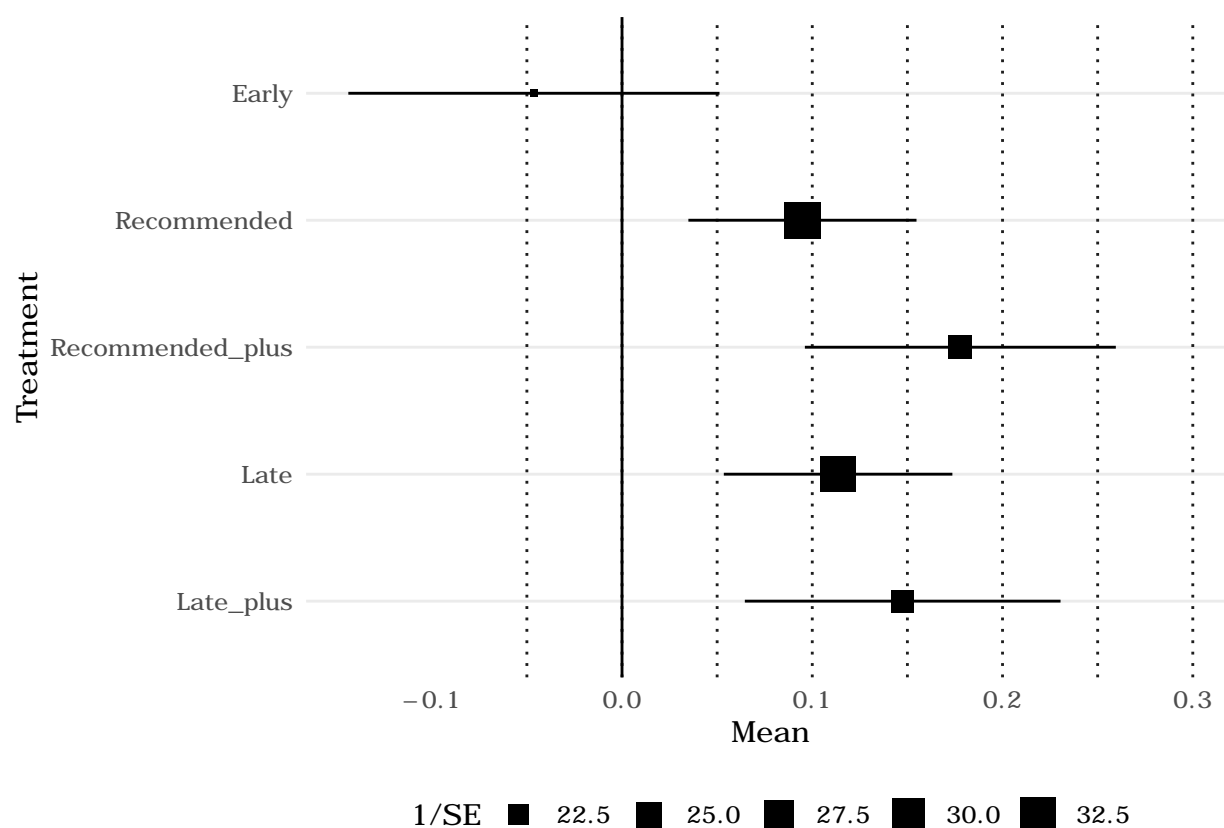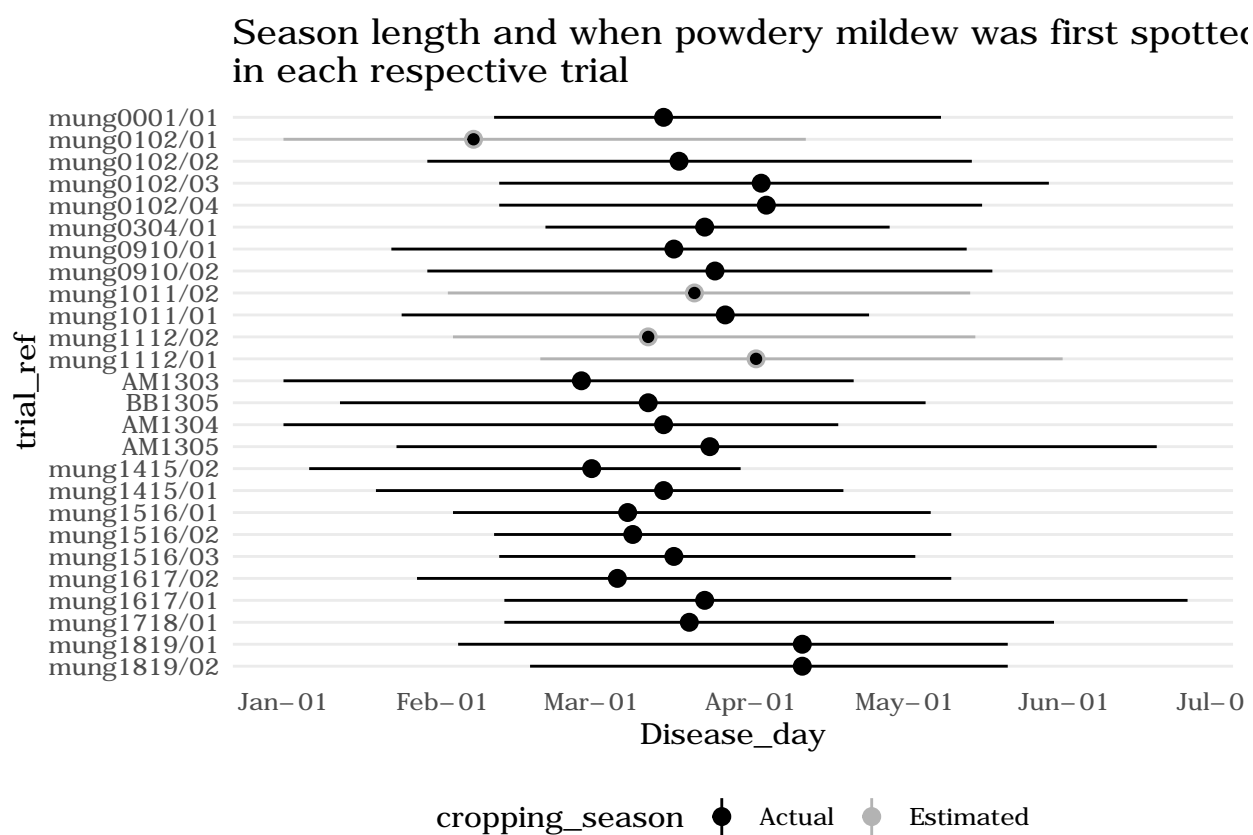
Figure 1. Yield contrasts

that there is no difference between the moderators [27]. The analysis shows there is still a significant amount of residual heterogeneity ($Q_E = 37805.5158$, $df = 149$, P < 0.0001) not captured by the spray management moderator indicating other possible moderators, which might influence grain yield.

A log-ratio test, based on a chi-squared distribution, indicated that retaining the `SpraySchedule` variable as a random interactive term provided a significantly better model (P = 0.0179). An inspection of log-likelihood profile plots showed the model, for each random interactive term, provided a reasonable estimate of τ^2, with no indication the variables were over-fit.

While the addition of disease pressure, as an interactive moderator term to `SpraySchedule`, did explain slightly more residual heterogeneity, shown by a lower $Q_E = 409.91$,. The model including disease pressure did not significantly improve the model (P = 0.4859) and did not significantly explain differences in the `SpraySchedule` term, and therefore disease pressure was not included in the final model.

3. Discussion

The current advice provided to growers for when to commence fungicide applications for control of powdery mildew, is to spray at the first sign of the disease then if necessary a follow-up spray two weeks later [10,12]. This meta-analysis confirms that advice, by showing that applying the first fungicide application, within three days of first sign will significantly mitigate yield loss from powdery mildew by between 34.89 to 154.78 kg / ha (*Recommended*, P = 0.0019). Additionally, spray schedules which commenced "late", up to 13 days after first sign of the disease were still effective at mitigating yield loss by 53.5 to 173.64 kg / ha (*Late*,P = 0.0002). Spray schedules which included follow-up fungicide applications were on average more effective at reducing yield loss due to powdery mildew by; 96.11 to 259.54 kg / ha in the *RecommendedPlus* treatments (P < 0.0001) and 64.55 to 230.47 kg / ha in the *LatePlus* treatments (P = 0.0005) (Figure 2) which again follows the advice given by Thompson et

Season length and when powdery mildew was first spotted in each respective trial

Figure 2. Season plot

Table 3. Estimated grain yield contrasts between each spray schedule.

Treatment contrasts	Estimate	Standard Error	Z-score	P-value	
Early - Recommended	0.1221	0.0421	2.8990	0.0037	**
Early - Recommended_plus	0.1587	0.0596	2.6617	0.0078	**
Early - Late	0.0843	0.0541	1.5595	0.1189	
Early - Late_plus	0.1821	0.0603	3.0203	0.0025	**
Recommended - Recommended_plus	0.0365	0.0371	0.9858	0.3242	
Recommended - Late	-0.0378	0.0349	-1.0850	0.2779	
Recommended - Late_plus	0.0600	0.0446	1.3445	0.1788	
Recommended_plus - Late	-0.0744	0.0375	-1.9812	0.0476	*
Recommended_plus - Late_plus	0.0235	0.0453	0.5174	0.6049	
Late - Late_plus	0.0978	0.0446	2.1932	0.0283	*

Note:
** - signficant at 0.005 level
* - signficant at 0.05 level

al. [12]. This meta-analysis adds certainty to the advice, which is currently given to growers, especially around any uncertainty that Early sprays might be effective at suppressing the disease. We show here Early fungicide applications, which were made prior to any sign of the disease, were not effective at mitigating yield loss.

This meta-analysis also provides a effective spray window for fungicide applications, given that spray schedules, which commenced "late" were still significantly effective at mitigating yield loss (*Late* P = 0.0002) 7 - 13 days after the first sign of disease. Growers typically do not have the time to inspect their crop regularly for the first sign of powdery mildew and therefore the disease may not be spotted within the first couple of days of it establishing. Also, weather conditions and logistical issues might prevent growers from getting machinery in the paddock to treat the disease immediately after it is spotted. These issues, and perhaps other on farm duties, might delay a timely application, permitting the disease more time to establish. Knowing a delay of up to two weeks may not lead to yield losses, can allow growers to incorporate other integrated pest management options, such as insecticide, with the fungicide spray saving on spray application costs. However attention should be made to the label requirements when first planning a spray schedule. Second applications, if they are needed, should be applied at least 10 - 14 days after the first application and not within 21 days of harvest [29]. If time permitted a second spray after the first "late" application, on average the meta-analysis shows this would be optimal, as the *LatePlus* treatment significantly limited yield loss by 97.8 kg / ha, when compared to the *Late*, single spray treatment P = 0.0283.

It is important to note, the studies included in this meta-analysis may not reflect a "normal" mungbean season. All the included experiments were intentionally sown late in the season, from January through to as late as early March, to ensure powdery mildew infection (Figure 2). While certain circumstances may force growers to sow mungbeans late, many may decide to fallow their fields instead of risking a powdery mildew epidemics and an early frost damaging the crop and yields. Regardless, according to the Australian Mungbean Association sowing guide [11], mungbeans should not be sown later than January in the Darling Downs, the region where the majority of trials were undertaken. If the planting guide is adhered to the mungbean would be at a lower risk of powdery mildew, reducing the requirement for fungicide applications. An example of this is if we inspect the six trials in which powdery mildew manifested 60 days or greater after planting (Trial references: AM1304, mung1819/01, mung1011/01, AM1303, AM1305 and BB1305). Of these trials, four directly compared single *Recommended* fungicide applications and *RecommendedPlus* (AM1303, AM1305, BB1305 and mung1819/01), two of which (AM1305 and mung1819/01) produced at least one treatment *RecommendedPlus* treatment with a higher grain yield the *Recommended* treatments. However, neither AM1305 or mung1819/01 neither contained *Recommended* or *RecommendedPlus* treatments with higher mean yields than the no-spray control. This indicating there is likely no benefit to applying two fungicide applications when the disease manifests later than 60 days into the growing season. Further the trials undertaken in 2013, such as Premer (AM1303), Goolhi (AM1305) Marys Mount (AM1304) and Millmerran (BB1305) reported a significant difference in powdery mildew severity between the fungicide treatments, however neither trial showed a significant difference in grain yield between treatments [16, growers alliance [15], growers alliance [17], growers alliance [14]]. Therefore, discretion is still required for if fungicide applications are necessary given the crop growth stage and forecast weather conditions. The PowderyMildewMBM considers crop growth stage and weather, to assist decision making whether to spray or not to spray [22].

PowderyMildewMBM also provides a cost-benefit analysis to growers for fungicide spray schedule scenarios. The user inputs weather, crop, and economic inputs into the DSS and the application returns the probability that one or two fungicide sprays would protect yields sufficiently to increase the net profit. The scope of the meta-analysis is limited to the timing of fungicide application and therefore we can not validate all the variables available in PowderyMildewMBM. However our results validate the decision of the app developers not to recommend prophylactic fungicide applications. If the powdery mildew is not observed in the crop the app advises the user to keep

269 monitoring for disease.

270

271 Scouting the crop for powdery mildew might not be necessary during the warmer months of the
272 season. Data from the trials included in this experiment show regardless of crop sowing date, powdery
273 mildew establishes on average in the month of March, which marks the beginning of autumn (Figure
274 2). Considering the results of the meta-analysis, weekly inspections would be ideal to ensure timely
275 fungicide applications if necessary. Scouting at intervals greater than 12 - 13 days, might risk crop
276 losses if a spray schedule does not commence within aforementioned the spray window. More work is
277 needed to understand when "late" fungicide applications are too late.

278 We estimate the costs of applying tebuconazole fungicide at $4.18 AU / ha, if fungicide is
279 purchased at $15 / L [30] and applications cost are $2 per hectare [7]. The meta-analysis estimates
280 average yield savings of between kg / ha (*Recommended*) and kg / ha (*RecommendedPlus*). These
281 average yield savings would save $ AU / ha for a single application at first sign and $ AU / ha if
282 a follow-up application is made. Lucrative returns such as these may tempt growers into applying
283 fungicide to control diseases such as powdery mildew when they are unnecessary. Excessive use
284 of fungicides are highly likely to lead ongoing problems such as fungicide resistance. *Podosphaera*
285 *xanthii* is categorised by the Australian Fungicide resistance Action Committee (FRAC) as a "High
286 risk" candidate to evolve fungicide resistance [31]. Evolution of fungicide resistance can occur
287 quickly, especially when spray schedules are not designed with potential fungicide resistance in
288 mind. Powdery mildew is one such group of fungal pathogens with an extensive track record of
289 rapidly developing fungicide resistance. In cucurbits, *P. xanthii* evolved resistance to benzimidazole
290 within three years of its registration [32] and has evolved resistance to numerous fungicide classes,
291 such as benzimidazoles, demethylation inhibitors, organophosphates, hydroxypyrimidines, quinone
292 inhibitors and quinoxalines since [33]. In cereal crops *Blumeria graminis* showed reduced sensitivity to
293 strobilurins in 1998, two years after the fungicides began being used in Europe [34].

294 According to the Fungicide Resistance Action Committee (FRAC) it is important to alternate or
295 mix fungicides with different modes of action to avoid fungicide resistance. At the time of writing, in
296 Australia one fungicide is registered with the to treat powdery mildew in mungbean, Veritas/Custodia
297 (200 g/L tebuconazole with 120 g/L azoxystrobin). This fungicide provides dual action: tebuconazole
298 disrupts C-14 demethylase, required in sterol biosynthesis for cell membranes; and azoxystrobin, a
299 Quinone outside inhibitor. Quinone is required in cell metabolism and active in the mitochondrial
300 complex III. However FRAC recommends that applications of QoIs or strobilurins, singularly or as a
301 mixture, should not be applied consecutively and alternated with a fungicide from a different group
302 [35]. For mungbean, past experiments have shown some success with sulphur applications to control
303 powdery mildew. Sulphur is a non-systemic protectant and thus provides an excellent option for
304 rotation to limit the emergence of fungicide resistance.

305 To extend the time that a fungicide remains effective against the target pathogen, strategic use of
306 the fungicide must be considered. Excessive fungicide sprays by a single active increase the selection
307 pressure on the pathogen to evolve resistance [35]. If any strains of the pathogen with resistance to the
308 fungicide are present in a population regular use of the fungicide will increase its proportion in the
309 population. Therefore, managing the disease with fungicides for the highest economic return is likely
310 not to require eradication of the causal pathogen. Integrated pest management practices focus on using
311 only the required number of chemical control to protect the economic return. These strategies permit
312 the presence of yield limiting diseases and pests if intervening would cost more than the economic
313 return of protection.

314 Advising use of fungicide for disease control to protect yields should also consider the harm
315 it may do to symbiotic relationships with microbiota. While some nitrogen fixing symbiont show a
316 level of tolerance to fungicides, all are detrimentally affected by increasing concentrations of fungicide
317 leading to lower nodulation and biomass accumulation [36, Shahid and Khan [37]]. Therefore if a
318 grower is relying on inoculating with rhizobia to help meet the crops nitrogen requirements, untimely

and excessive applications of a systemic fungicide for powdery mildew control may actually reduce yield.

3.1. Future work

This meta-analysis provides a ballast for the direction of future powdery mildew research in mungbean. However it does not provide enough certainty to when late fungicide applications would be too late to protect yield losses due to the disease. Future experiments should take note of physiological maturity of the crop throughout the season so "crop age" might be considered as a covariate in future analyses. In cases where mungbean physiological maturity may not have been recorded the APSIM mungbean module [38].

Early detection of powdery mildew is vital for an effective fungicidal control response. Data from the collection of 26 studies in the Northern Grain region, indicate powdery mildew is likely to manifest on crops in the first month of autumn. This data together with additional observations may permit models to predict the likely onset of the disease given weather parameters, providing an early warning system for growers to scout their crop for signs of powdery mildew. However an improved understanding of the pathogen biology is also needed to ensure models are as accurate as they can be. The population genetics, epidemiology and alternate hosts are all poorly understood in this host-pathogen relationship. The recent discovery of a second powdery mildew species, which infects mungbean highlights the paucity of knowledge on this disease.

4. Conclusion

5. Supplemental Material

Supplementary material can be found in the following research compendium.

6. Data Availability

All data and code used in the preparation of this manuscript can be found in the associated research compendium.

7. Conflict of Interest Statement

The authors of this paper have no conflicting interest in the research presented in this manuscript

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Author Contributions: For research articles with several authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used "X.X. and Y.Y. conceive and designed the experiments; X.X. performed the experiments; X.X. and Y.Y. analyzed the data; W.W. contributed reagents/materials/analysis tools; Y.Y. wrote the paper." Authorship must be limited to those who have contributed substantially to the work reported.

Conflicts of Interest: Declare conflicts of interest or state 'The authors declare no conflict of interest.' Authors must identify and declare any personal circumstances or interest that may be perceived as inappropriately influencing the representation or interpretation of reported research results. Any role of the funding sponsors in the design of the study; in the collection, analyses or interpretation of data in the writing of the manuscript, or in the decision to publish the results must be declared in this section. If there is no role, please state 'The founding sponsors

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Abbreviations

The following abbreviations are used in this manuscript:

DSS   Decision support system

References

1. Lambrides, C.J.; Godwin, I.D. Mungbean. In *Pulses, sugar and tuber crops*; Springer, 2007; pp. 69–90.
2. Chankaew, S.; Somta, P.; Isemura, T.; Tomooka, N.; Kaga, A.; Vaughan, D.A.; Srinives, P. Quantitative trait locus mapping reveals conservation of major and minor loci for powdery mildew resistance in four sources of resistance in mungbean [Vigna radiata (L.) Wilczek]. *Molecular Breeding* **2013**, *32*, 121–130. doi:10.1007/s11032-013-9856-6.
3. Lawn, R.J.; Russell, J.S. Mungbean: a grain legume for summer rainfall cropping areas of Australia. *Journal* **1978**, pp. 28–41.
4. Chauhan, Y.; Williams, R. Physiological and Agronomic Strategies to Increase Mungbean Yield in Climatically Variable Environments of Northern Australia. *Agronomy* **2018**, *8*, 83. doi:10.3390/agronomy8060083.
5. Food and Agriculture Organization of the United Nations.. FAOSTAT Statistical Database., 2020.
6. Clarry, S. The rise and rise of mungbeans, 2016.
7. Queensland Government. Mungbean (Irrigated) Darling Downs 2019, 2019.
8. Thomas.; Robertson, M.J.; Fukai, S.; Peoples, M.B. The effect of timing and severity of water deficit on growth, development, yield accumulation and nitrogen fixation of mungbean. *Field Crops Research* **2004**, *86*, 67–80. doi:10.1016/S0378-4290(03)00120-5.
9. Kelly, L.; White, J.; Sharman, M.; Brier, H.; Williams, L.; Grams, R.; Weir, D.; Mckay, A.; Sparks, A.H. Mungbean and sorghum disease update - GRDC, 2017.
10. Sparks, A.; Kelly, L. Mungbean powdery mildew management with fungicide - Field Crop Diseases | Field Crop Diseases, 2017.
11. Planting mungbeans.
12. Sue Thompson. Mungbeans vs fungus: two sprays for optimum control - Grains Research and Development Corporation. *Ground Cover* **2016**.
13. Pandey, A.K.; Burlakoti, R.R.; Kenyon, L.; Nair, R.M. Perspectives and Challenges for Sustainable Management of Fungal Diseases of Mungbean [Vigna radiata (L.) R. Wilczek var. radiata]: A Review. *Frontiers in Environmental Science* **2018**, *6*, 53. doi:10.3389/fenvs.2018.00053.
14. growers alliance, N. Powdery Mildew Mungbeans - 2013 Goolhi NSW, 2013.
15. growers alliance, N. Powdery Mildew Mungbeans - 2013 Premer NSW, 2013.
16. growers alliance, N. Powdery Mildew Mungbeans - 2013 Millmerran QLD, 2013.
17. growers alliance, N. Powdery Mildew Mungbeans - 2013 Mary's Mount NSW, 2013.
18. Thompson, S.; O'Conner, R.; Weir, D.; Conway, M.; Ainsthorpe, D.; Quinlivan, M.; Carroll, K.; Aguis, P. Fungicide management of mungbean powdery mildew, 2016.
19. Madden, L.V.; Paul, P.A. Meta-analysis for evidence synthesis in plant pathology: An overview. Phytopathology. The American Phytopathological Society, 2011, Vol. 101, pp. 16–30. doi:10.1094/PHYTO-03-10-0069.
20. Madden, L.V.; Piepho, H.P.; Paul, P.A. Statistical models and methods for network meta-analysis, 2016. doi:10.1094/PHYTO-12-15-0342-RVW.
21. Jansen, J.P.; Fleurence, R.; Devine, B.; Itzler, R.; Barrett, A.; Hawkins, N.; Lee, K.; Boersma, C.; Annemans, L.; Cappelleri, J.C. Interpreting Indirect Treatment Comparisons and Network Meta-Analysis for Health-Care

Decision Making: Report of the ISPOR Task Force on Indirect Treatment Comparisons Good Research Practices: Part 1 Background to the task force **2011**. doi:10.1016/j.jval.2011.04.002.

22. Diggle, A. PowderyMildew MBM - Powdery Mildew management app for mungbean.

23. Ngugi, H.K.; Lehman, B.L.; Madden, L.V. Multiple Treatment Meta-Analysis of Products Evaluated for Control of Fire Blight in the Eastern United States. *Phytopathology* **2011**, *101*, 512–522. doi:10.1094/phyto-08-10-0221.

24. Paul, P.A.; Lipps, P.E.; Hershman, D.E.; Mcmullen, M.P.; Draper, M.A.; Madden, L.V. Disease Control and Pest Management A Quantitative Review of Tebuconazole Effect on Fusarium Head Blight and Deoxynivalenol Content in Wheat **2007**. *97*, 211. doi:10.1094/PHYTO-97-2-0211.

25. de Mendiburu, F. *agricolae: Statistical Procedures for Agricultural Research*, 2020. R package version 1.3-3.

26. R Core Team. *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria, 2020.

27. Viechtbauer, W. Conducting Meta-Analyses in R with the metafor Package. *Journal of Statistical Software* **2010**, *36*, 1–48. doi:10.18637/jss.v036.i03.

28. Rücker, G.; Krahn, U.; König, J.; Efthimiou, O.; Schwarzer, G. *netmeta: Network Meta-Analysis using Frequentist Methods*, 2020.

29. Permit to allow minor use of an AgVet chemical product for the control of powdery mildew in adzuji beans, mung beans and navy beans. Technical report.

30. Simpfendorfer, S.; Taylor, Z. Fungicide Management of Stripe Rust in Wheat: up-front vs in-crop options in 2011. Technical report, NSW Department of Primary Industries, 2011.

31. FRAC. Pathogen Risk List. Technical report, Fungicide Resistance Action Committee, 2019.

32. Peterson, R.A. Field resistance to benomyl in cucurbit powdery mildew. *Australian Plant Pathology Society Newsletter* **1973**, *2*, 27–28. doi:10.1071/APP9730027a.

33. Mcgrath, M.T. Fungicide Resistance in Cucurbit Powdery Mildew: Experiences and Challenges. Technical report, 2001.

34. Chin, K.M.; Chavaillaz, D.; Kaesbohrer, M.; Staub, T.; Felsenstein, F.G. Characterizing resistance risk of Erysiphe graminis f.sp. tritici to strobilurins. *Crop Protection* **2001**, *20*, 87–96. doi:10.1016/S0261-2194(00)00059-4.

35. Brent, K.J.; Hollomon, D.W. *Fungicide Resistance in Crop Pathogens: How can it be managed?*, 2nd ed.; 2007.

36. Ahemad, M.; Khan, M.S. Effect of tebuconazole-tolerant and plant growth promoting Rhizobium isolate MRP1 on pea-Rhizobium symbiosis. *Scientia Horticulturae* **2011**, *129*, 266–272. doi:10.1016/j.scienta.2011.03.036.

37. Shahid, M.; Khan, M.S. Fungicide tolerant Bradyrhizobium japonicum mitigate toxicity and enhance greengram production under hexaconazole stress. *Journal of Environmental Sciences (China)* **2019**, *78*, 92–108. doi:10.1016/j.jes.2018.07.007.

38. Robertson, M.J.; Carberry, P.S.; Huth, N.I.; Turpin, J.E.; Probert, M.E.; Poulton, P.L.; Bell, M.; Wright, G.C.; Yeates, S.J.; Brinsmead, R.B. Simulation of growth and development of diverse legume species in APSIM. *Australian Journal of Agricultural Research* **2002**, *53*, 429–446. doi:10.1071/AR01106.