Article

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

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1 **Simple Summary:** A simple summary goes here.

2 **Abstract:** A single paragraph of about 200 words maximum. For research articles, abstracts should
3 give a pertinent overview of the work. We strongly encourage authors to use the following style of
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6 treatments applied; 3) Results: Summarize the article's main findings; and 4) Conclusion: Indicate
7 the main conclusions or interpretations. The abstract should be an objective representation of the
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, the 1960s that mungbean was grown commercially [3,4].
18 Prior to the 1970s the total area grown in Australia rarely exceeded 1000 hectares [3]. At the end of the
19 1980s, between 3,000 to 10,0000 hectares were being harvested for dried beans in Australia, of which
20 mungbean is categorised by the FAO statistics data repository [5]. In the decade leading up to 2018
21 between 35 - 86.4 thousand hectares of dried beans were planted annually. This increase is attributed
22 high value export markets and to the improved yields in the new cultivars [6]. Currently mungbean
23 is predominantly grown in southern Queensland and northern New South Wales as a short season
24 summer legume crop. In 2019, approximately 90 % of Australian mungbean was grown for export
25 with lucrative returns of up to $1300 AU a tonne [7]. Mungbean potentially yields up to 3 tonnes per
26 hectare [8], but due to high variability in yields between seasons and locations, the average Australian
27 farm still yields less than 1 tonne per hectare [4].
28 The high variability in yields, in part, can be attributed to a range of diseases that affect the crop
29 [9]. Two major foliar diseases, tan spot or common wilt (*Curtobacterium flaccumfaciens* pv. *flaccumfaciens*)

and halo blight (*Pseudomonas savastanoi* pv. *phaseolicola*), are the main focus of the Australian Mungbean Improvement programme's resistance breeding efforts due to lack of effective chemical controls 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.

Powdery mildew is caused by two separate genera of fungi in Australia *Podosphaera xanthii* and an unnamed *Psuedoidium sp.* (Kiss and Kelly Personal Communication). 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 cooler 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, a delayed sowing date of late January into February increases the possibility powdery mildew will infect earlier in the crop development 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 the 2016 field trials 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 it's 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 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

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

128 in the *i*th trial over total number (K) of trials($i = 1, \dots K$). Y_i is estimated from a normal distribution
129 with the mean μ, sample variance of S_i^2 and a between trial variance of σ^2.

130 **Trial Variables and moderators:** While the main aim of all the identified trials were to assess the
131 efficacy of fungicide for control of powdery mildew, some trials tested additional potential covariates in
132 the experimental design. These additional variables included: row spacing, fungicide active ingredient,
133 fungicide dose, planting date, number of fungicide applications and cultivar. To account for the
134 variation attributed to some of these variables between trials, and within trials, a single *trial* factor
135 was created. The *trial* random variable made distinctions between the following variables: unique
136 trial reference code, trial location, trial season, host genotype, row spacing, and fungicide dose.

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

141 The timing of fungicide application treatments, were defined in relation to the recorded date when
142 powdery mildew first occurred in the trial. An *Early* fungicide application treatment, referred to a
143 fungicide spray schedule that commenced prior to the disease being observed in the trial. *Recommended*
144 referred to a fungicide spray schedule which commenced $1-3$ days following the first sign of disease.
145 *Late* treatments commenced between 7 and 13 days after the first sign of disease. There were no
146 spray schedule treatments which began between 4 and 6 days after the first sign of powdery mildew.
147 The number of fungicide applications were binned in to two categories. A spray schedule with a
148 *single* spray, and a spray schedule with two or three spray applications *plus*. The division of the
149 treatments produced: 13 $Early_{single}$ treatments; 5 $Early_{plus}$; 32 $Recommended_{single}$; 46 $Recommended_{plus}$';
150 17 $Late_{single}$; and 20 $Late_{plus}$. $Early_{plus}$ treatments were removed from the subsequent meta-analysis,
151 due to an insufficient sample size.
152 What remained was the mean yield for a total of 173 treatments in the following five spray schedule
153 categories: no spray *control*, $Early_{single}$, $Recommended_{single}$, $Recommended_{plus}$, $Late_{single}$, and $Late_{plus}$.
154 An ordinal $1-9$ scale was used to describe the mean powdery mildew plot severity in most trials
155 (Table 2). However, the 2013 NGA trials (Table 1) reported mean plot severity as the percentage of

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

156 leaves covered by powdery mildew and the location of the infected leaves in the lower, middle or
157 upper canopy. Given the ordinal scale ranked plots on the percent of foliage showing the disease
158 from the lower canopy to the upper canopy, a conversion from percentage of the diseased canopy, to
159 the 1 – 9 scale was straight-forward. The end of season mean plot severity, and date of first sign of
160 disease were used to calculate the area under the disease progress curve (AUDPC) for each treatment.
161 Additional powdery mildew mean plot severity observations made in-between these two observations
162 were also included in the AUDPC calculation if they were available. AUDPC was calculated using
163 the `agricolae` package in R [25]. A categorical disease pressure factor was created by binning trials
164 into two levels based on the mean AUDPC of the no spray control plot. The two levels, 'low disease
165 pressure' and 'high disease pressure', were separated by the median AUDPC (153.625) of the no spray
166 control plots.
167 The mean difference in grain yield was estimated from the difference in model estimates of the
168 non-treated control and each of the spray schedule treatments using fungicide.
169 Disease pressure was tested against the basic model to determine if, when introduced as a moderator
170 variable, it could significantly explain additional heterogeneity between the spray schedule treatments.
171 A a Wald-type test was used to test the impact of the disease pressure factor.
172 Linear contrasts were used to produce the mean difference in effect sizes, and their respective
173 standard errors and 95 % confidence intervals, between each spray schedule treatment.
174 Additional meta-analysis models tested variables ... for their influence on the SpraySchedule
175 variables ability to mitigate yield loss. For the variable disease pressure a Wald-type test was used to
176 test the impact of this variable on the model as a whole.
177 The data were analysed in the R statistical software environment, version 4.0.1, [26] using two
178 contributed meta-analysis packages, `metafor`, version 2-4.0, [27] and `netmeta`, version 1.2-1, [28].
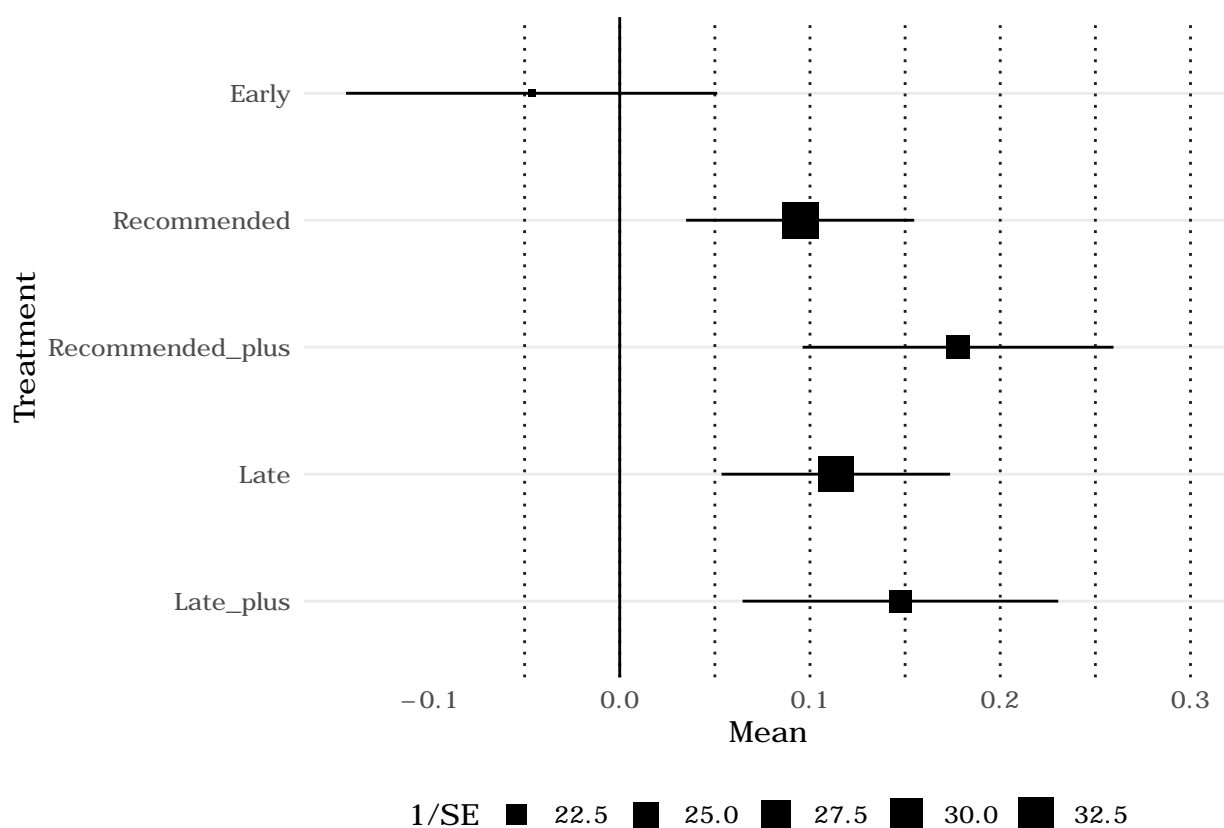
2. Results

control, $Early_{single}$, $Recommended_{single}$, $Recommended_{plus}$, $Late_{single}$, and $Late_{plus}$.
Overall the meta-analysis indicated that, with the exception of the $Early_{single}$ treatment, all the the spray schedules were effective at protecting mungbean yields relative to the no spray controls (Figure @ref(fig:Figure1)). Treatments which incorporating multiple fungicide applications, $Recommended_{plus}$ and $Late_{plus}$, produced the highest mean yield protection estimates at 177.83 (SE = 41.69 kg / ha) and 147.51 kg / ha (SE = 42.33 kg / ha) respectively (Table 3). Both $Recommended_{plus}$ and $Late_{plus}$ were also significantly higher than single applications $Early_{single}$ (P = 0.0078 and P = 0.0025 respectively) and $Late_{single}$(P = 0.0476 and P = 0.0283 respectively). However, comparing $Late_{plus}$ and $Early_{single}$ should done with caution given these treatments did not occur within the same trial for any of the 15 trials included in the meta-analysis. Within the single fungicide application treatments Recommended provided the highest yield protection, and was estimated at saving an average of 159.9 kg / ha (SE = 30.5 kg / ha) relative to the no spray control (P < 0.0001). Recommended treatments were was also significantly higher than Early treatments by 122.1 kg / ha (SE = 42.1 kg / ha)(P = 0.004). Single Late applications were effective at significantly mitigating the effect of powdery mildew on yield by a mean of 122 kg / ha (SE = 39.2 kg / ha)(P = 0.0019). However, this estimate was not significantly different to single Early or Recommended treatments (P = 0.119; P = 0.278 respectively).

The Q_M omnibus test of moderators, shows the moderators significantly influence the model (Q_M = 31.1217477 , df = 5, P < 0.0001) and we can reject the null hypothesis ($H_0 : \beta_1 = \beta_2 = \beta_3 = \beta_4 = 0$) that there is no difference between the moderators [27]. The analysis shows there is still a significant amount of residual heterogeneity ($Q_E = 3.7805516 \times 10^4$, $df = 149$, 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

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

205 of log-likelihood profile plots showed the model, for each random interactive term, provided a
206 reasonable estimate of τ^2, with no indication the variables were over-fit.

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

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

212
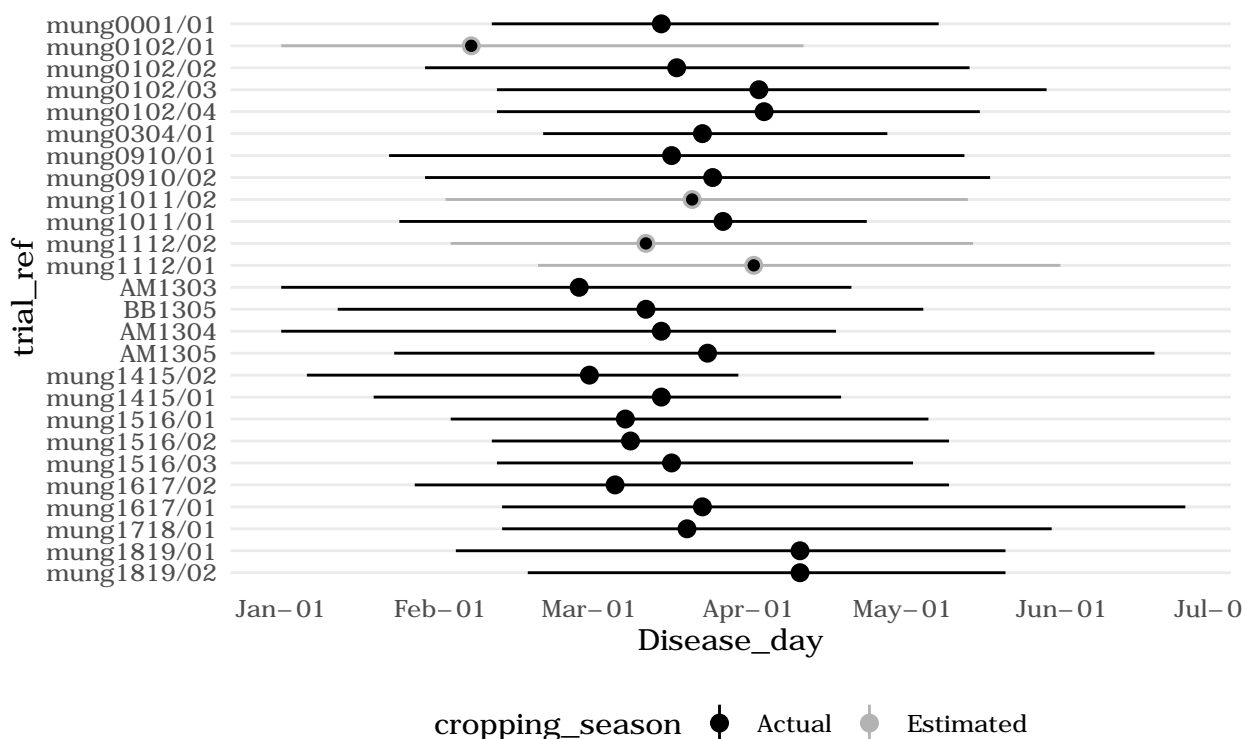
3. Discussion

214 The current advice provided to growers for when to commence fungicide applications for control
215 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). In addition, 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 @ref(fig:Figure2)) which again follows the advice given by Thompson et 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 @ref(fig:Figure2)). 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

265 stage and forecast weather conditions. The PowderyMildewMBM considers crop growth stage and
266 weather, to assist decision making whether to spray or not to spray [22].

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

275

276   Scouting the crop for powdery mildew might not be necessary during the warmer months of the
277 season. Data from the trials included in this experiment show regardless of crop sowing date, powdery
278 mildew establishes on average in the month of March, which marks the beginning of autumn (Figure
279 @ref(fig:Figure2)). Considering the results of the meta-analysis, weekly inspections would be ideal to
280 ensure timely fungicide applications if necessary. Scouting at intervals greater than 12 - 13 days, might
281 risk crop losses if a spray schedule does not commence within aforementioned the spray window.
282 More work is needed to understand when 'late' fungicide applications are too late.

283   We estimate the costs of applying tebuconazole fungicide at
284 \$4.18 AUD / ha, if fungicide is purchased at \$15 / L [30] and
285 applications cost are $2 per hectare [@QueenslandGovernment 2019]. The meta -$
286 $analysis estimates average yield savings of between kg/ha (\text{Recommended}) and kg/ha (\text{Recommended Plus} \$).$
287 These average yield savings would save \$ AUD / ha for a single application at first sign and AUD / ha
288 if a follow-up application is made. Lucrative returns such as these may tempt growers into applying
289 fungicide to control diseases such as powdery mildew when they are unnecessary. Excessive use
290 of fungicides are highly likely to lead ongoing problems such as fungicide resistance. *Podosphaera*
291 *xanthii* is categorised by the Australian Fungicide resistance Action Committee (FRAC) as a 'High
292 risk' candidate to evolve fungicide resistance [31]. Evolution of fungicide resistance can occur
293 quickly, especially when spray schedules are not designed with potential fungicide resistance in
294 mind. Powdery mildew is one such group of fungal pathogens with an extensive track record of
295 rapidly developing fungicide resistance. In cucurbits, *P. xanthii* evolved resistance to benzimidazole
296 within three years of its registration [32] and has evolved resistance to numerous fungicide classes,
297 such as benzimidazoles, demethylation inhibitors, organophosphates, hydroxypyrimidines, quinone
298 inhibitors and quinoxalines since [33]. In cereal crops *Blumeria graminis* showed reduced sensitivity to
299 strobilurins in 1998, two years after the fungicides began being used in Europe [34].

300   According to the Fungicide Resistance Action Committee (FRAC) it is important to alternate or
301 mix fungicides with different modes of action to avoid fungicide resistance. At the time of writing, in
302 Australia one fungicide is registered with the to treat powdery mildew in mungbean, Veritas/Custodia
303 (200 g/L tebuconazole with 120 g/L azoxystrobin). This fungicide provides dual action: tebuconazole
304 disrupts C-14 demethylase, required in sterol biosynthesis for cell membranes; and azoxystrobin, a
305 Quinone outside inhibitor. Quinone is required in cell metabolism and active in the mitochondrial
306 complex III. However FRAC recommends that applications of QoIs or strobilurins, singularly or as a
307 mixture, should not be applied consecutively and alternated with a fungicide from a different group
308 [35]. For mungbean, past experiments have shown some success with sulphur applications to control
309 powdery mildew. Sulphur is a non-systemic protectant and thus provides an excellent option for
310 rotation to limit the emergence of fungicide resistance.

311   To extend the time that a fungicide remains effective against the target pathogen, strategic use of
312 the fungicide must be considered. Excessive fungicide sprays by a single active increase the selection
313 pressure on the pathogen to evolve resistance [35]. If any strains of the pathogen with resistance to the
314 fungicide are present in a population regular use of the fungicide will increase it's proportion in the

population. Therefore, managing the disease with fungicides for the highest economic return is likely not to require eradication of the causal pathogen. Integrated pest management practices focus on using only the required number of chemical control to protect the economic return. These strategies permit the presence of yield limiting diseases and pests if intervening would cost more than the economic return of protection.

Advising use of fungicide for disease control to protect yields should also consider the harm it may do to symbiotic relationships with microbiota. While some nitrogen fixing symbiont show a level of tolerance to fungicides, all are detrimentally affected by increasing concentrations of fungicide leading to lower nodulation and biomass accumulation [36, Shahid and Khan [37]]. Therefore if a 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

8. Acknowledgments

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 had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, an in the decision to publish the results'.

Abbreviations

The following abbreviations are used in this manuscript:

MDPI	Multidisciplinary Digital Publishing Institute
DOAJ	Directory of open access journals
TLA	Three letter acronym
LD	linear dichroism

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