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Article

Effects of Hilling Application to Mitigate Damage from Soybean Gall Midge

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Abstract

Soybean Gall Midge is a relatively new insect species found in the midwestern United States. They are destructive to soybean crops and little research exists on how to properly deal with infestations. One proposed method is to cover the stems of soybean crops, a process known as hilling. Two studies were conducted to measure the effects of hilling on the count of soybean gall midge larvae and the resulting yield components at the end of the growing season. Results show that hilling during the early growth stages of soybean reduces the count of larvae and improves soybean growth metrics.

Keywords: soybean, hilling, soybean gall midge, infestation

1. Introduction

The first reports of a peculiar orange larvae in Nebraska came after a hailstorm in 2011, but it was not until 2018 that the soybean gall midge warranted a pest status for soybeans. It was in 2018 that entomologists received reports of larvae infestations causing damage in soybean fields, of which the species was unknown at the time. The first significant effort to determine the dispersion of the soybean gall midge was in 2019, with 63 counties reporting the insect across four states. As of 2020, the insect was identified across 114 counties in five states: Nebraska, South Dakota, Minnesota, Iowa, and Missouri (McMechan et al. 2021).

Initial reports of the gall midge larvae showed them feeding on the stems of damaged or disease-compromised soybean plants at the end of the growing season. The economic impact of the initial reports was minimal, resulting in little concern. In 2018, significant damage was reported in June due to infestation. Samples of crops were

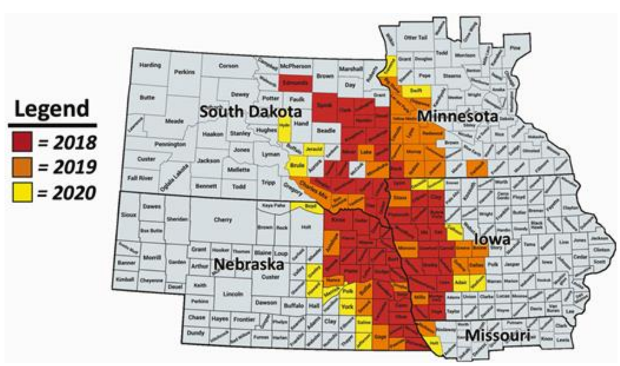


Figure 1: Locations of soybean gall midge infestations by year.

Figure 2: Photo of white and orange soybean gall midge larvae taken by Erin Hodgson.

taken and some infected soybeans had no other detectible plant diseases, raising concern that soybean gall midge could be environmentally and economically disastrous (Gagné et al. 2019).

The soybean gall midge, belonging to the Resseliella genus, starts its lifecycle when an adult lays eggs in small cracks or fissures below the cotyledonary node of soybeans. Under greenhouse conditions, white and orange larvae were observed feeding at the intersection of dead and live plant tissue, although it is unknown if the tissue was dead as a result of larval feeding. During the initial identification stage, specimens of the orange larvae were collected and were later identified to belong to the Resseliella genus. The larvae were collected on August 1, 2018 and adults emerged between 24 hours and 18 days after collection (McMechan et al. 2021).

Due to the recent immergence of this damaging pest, there are currently no recommended methods for controlling field infestations (McMechan and Hunt 2021). Given the nature of larvae feeding patterns, hilling around the stems of soybeans provides a physical barrier between the plant and egg-laying females. The studies presented here measure the effect of hilling on the count of larvae, as well as important measures of soybean growth health.

2. Methods

Two studies were conducted to measure the effect that hilling the base of soybeans has on the reduction of damage due to soybean gall midge. In the first study, referenced as the hilling study, the count of larvae was recorded during the 2022 crop cycle at approximately two week intervals. Rows of the field were blocked and partitioned into four sections. Each section received a treatment of timing for the application of hilling during the soybean growth cycle. These stages were V2, V5, and R2. These are referenced as treatments 2, 3, and 4, respectively. One section of the row was left unhilled as a control, which is refered to as treatment 1. For each treatment within a row of the field, one sample was taken from sub-rows 1 and 4.

The second study, referenced as the unhilling study, used a similar layout as the hilling study. Instead of each row of the field acting as a block, the field was partitioned into two rows and two columns for the block. This study measured the effect of removing the hilling around the stem of the soybean crops. The only difference for the application of unhilling timing was that there were seven treatments, with each treatment being an unhilling date between June 16th and August 31st. One treatment was left as the control where the crops were left unhilled at the beginning of the study. For the unhilling study, soybean gall midge larvae counts were taken approximately every two weeks, and several yield components were measured at the end of the growing season. These yield components were the number of nodes, crop height, number of seed pods, seed weight, and total seed count.

Before describing the models fitted to the data, it is important to go through the underlying theory. The models presented are called generalized linear mixed models (GLMMs). The "generalized" part indicates that the models can be applied to various types of data. Some responses are classified as "count" data, which are values that can take on any nonnegative integers. For count data, the models are fitted with either a Poisson or negative binomial distribution. The models are fitted with a linear predictor to some component of thier corresponding distribution. This is specified by the "link function." The linear predictor is fitted directly to the data for the cases of continous data. Count data has the linear predictor fitted to the log mean of the corresponding distribution.

One particular issue that arrises with count data is that the Poisson distribution has a restriction that the mean and variance are equivalent. When this assumption is violated, there is overdispersion. The statistic used to measure overdispersion is the Pearson chi-square divided by the degrees of freedom. When this value exceeds one, there is evidence of overdispersion. A common solution to address this issue is to fit a negative binomial distribution instead, which is capable of addressing unequal means and variances, but unable to be fit to repeated measures.

The "mixed" part of GLMMs refers to random effects in model. For both studies, the field was partitioned into sections where every treatment is applied. These sections are refered to as "blocks" and allow some of the variation in responses to be attributed to them. Blocks are assumed to have an effect that normally distributed around zero with some variance.

The model fitted to the larvae counts from the hilling study was a poisson repeated measures and is shown below.

$$\eta_{ijk} = \eta + \tau_i + S_j + (\tau S)_{ij} + (B\tau)_{ik} + S(B\tau)_{ijk} \tag{1}$$

Where

- η is the intercept
- τ_i is the i^{th} hilling date
- S_j is the j^{th} sample date
- $(\tau S)_{ij}$ is the interaction between the i^{th} hilling date and the j^{th} sample date
- $(B\tau)_{ik} \sim N(0, \sigma_A^2)$ is the random interaction effect between the i^{th} treatment and the k^{th} field row
- $S(B\tau)_{ijk} \sim N(0, \sigma_B^2)$ is the random effect of the i^{th} hilling date, j^{th} sample date, and the k^{th} row

All random effects are assumed to be independent. The larvae count for the hilling study was fit with a first order ante structure, which was chosen due to having the smallest AIC during the selection process. For the GLMM specifications, $y_{ijk}|(B\tau)_{ij}, S(B\tau)_{ijk} \sim Poisson(\lambda_{ijk})$, and $\eta_{ijk} = \log(\lambda_{ijk})$.

The larvae count for the unhilling study could not converge with the conditional model, so the marginal model was used and is defined below.

$$\eta_{ijk} = \eta + \tau_i + S_j + (\tau S)_{ij} + S(B\tau)_{ijk} \tag{2}$$

The terms are defined in the same manner as the model for the hilling study. Again, the random effects are assumed to be independent. The model was fitted with a first order autogregressive structure that was chosen by having the smallest AIC. This model has the GLMM specifications $y_{ijk}|S(B\tau)_{ijk} \sim Poisson(\lambda_{ijk})$, and $\eta_{ijk} = \log(\lambda_{ijk})$.

The unhilling study yield componenets has five responses: counts of soybean nodes, pods, and seeds, and soybean height and seed weight. The full model is expressed below, but necessary adjustments made to the model can be found in Table 1.

$$\eta_{ijk} = \eta + B_i + \tau_j + (B\tau)_{ij} + \epsilon_{ijk} \tag{3}$$

Where

- η is the intercept
- $B_i \sim N(0, \sigma_B^2)$ is the effect of the i^{th} field section (block)

Response	Distribution	Link Function	Changes from full model
Count of nodes	$y\|B \sim Poisson(\lambda)$	$\eta_i = \log(\lambda_i)$	Removal of B_i and ϵ_{ijk}
Soybean height	$y\|B \sim Normal(\mu, \sigma^2)$	$\eta_i = \mu_i$	Use of CS covariance structure instead of B_i term
Count of pods	$y\|B \sim Negbin(\lambda)$	$\eta_i = \log(\lambda_i)$	Removal of B_i term and ϵ_{ijk}; KR2
Seed weight	$y\|B \sim Normal(\mu, \sigma^2)$	$\eta_i = \mu_i$	No adjustments
Count of seeds	$y\|B \sim Negbin(\lambda)$	$\eta_i = \log(\lambda_i)$	Removal of B_i term and ϵ_{ijk}; KR2

Table 1: Adjustments for the yield components data set made to the fitted model from the full model. KR2 means that the Kenward-Roger degrees of freedom approximation was used.

Treatment	1	2	3	4	5	6	7
Unhilling Date	Unhilled	June 16th	July 1st	July 15th	August 1st	August 15th	August 31st

Table 2: Treatment designations for yield components from the unhilling study

- τ_j is the effect of the j^{th} unhilling date
- $(B\tau)_{ij} \sim N(0, \sigma^2_{B\tau})$ is the interaction effect of the i^{th} field section and the j^{th} unhilling date
- $\epsilon_{ijk} \sim N(0, \sigma^2_e)$ is the random error of the i^{th} field section, j^{th} unhilling date, and the k^{th} plant

The unhilling dates are expressed as numbers from one to seven in the data set and can be found in Table 2.

Each analysis used the PROC GLIMMIX procedure in SAS 9.4. For reporting differences, Tukey's honest significant differences was used to control Type I errors and issues that arrise with multiplicity.

3. Results

Hilling Study Larvae Counts

At a first glance, there appears to be no interaction in the fixed effects from Table 3 between the growth stage of hilling application and the sample date for the larvae count (p-value = 0.2314). However, the interaction plot shown in Figure 3 suggests that the count of larvae depend on the specific combinations of the hilling application and sample date. For this reason, statements about differences should include the specific levels of application date and sample date.

While several differences are considered statistically significant, the ones of interest are the differences between treatments at specific sampling dates. The only evidence of differences in larvae counts are on July 5th, 2022. These are between the control and V2 (Treatments 1 and 2, p-value = 0.0203), and between V2 and R2 (Treatments 2 and 4, p-value = 0.0351).

Effect	Num DF	Den DF	F Value	Pr > F
Trt	3	11.17	17.81	0.0001
Sample date	3	10.43	0.14	0.9352
Trt:Sample date	9	10.08	1.62	0.2314

Table 3: Type III Tests of Fixed Effects for the count of larvae from the hilling study. Figure 3 indicates some evidence of an interaction between treatment and the sample date.

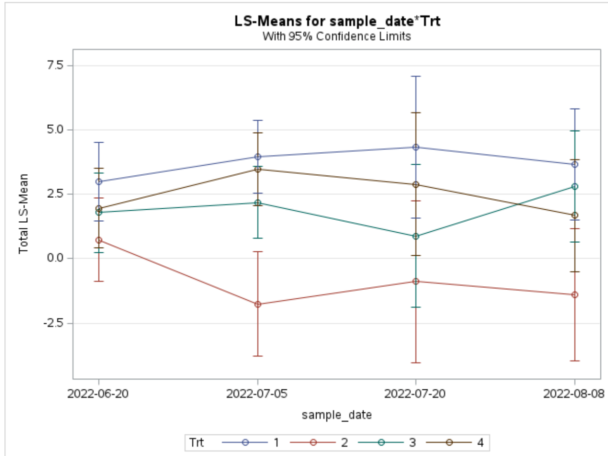


Figure 3: Least square means for the growth stage of soybean in the hilling study for the timing of hilling in the soybean growth cycle and the sample date. The responses shown are on the model scale and need to be exponentiated to show the estimates and confidence intervals for larvae counts.

Unhilling Study Larvae Counts

Similar to the hilling study, there is a significant interaction between the unhilling date and sample date (p-value $< .0001$). Those that have evidence of a difference for specific sample dates are shown in Table 4, with Figure 4 showing the least square means for the counts over time. Differences were only detected on July 1 and July 15. The results show that unhilling earlier in the growing season tended to increase the number of larvae.

Unhilling Study Yield Components

The yield components data from the unhilling study show similar results for all responses. For brevity, the results from the soybean height and total seed count will be discussed in this section, with the remaining responses located in the attached SAS output appendix. The plots for the chosen responses are located in Figure 5. For all differences discussed here, the unhilling date with the smaller response is reported first.

Soybean Height

There was evidence of a difference in unhilling dates on soybean height (p-value $< .0001$). The statistically significant differences are listed below.

- The control group (Trt 1) has evidence of a difference with all of the unhilling dates.
- Unhilling on June 16th (Trt 2) had differences in soybean heights with the latter unhilling dates, but marginal evidence of a difference with unhilling on August 31st (Trt 7) (p-value = 0.0631).
- There was also marginal evidence of a difference in soybean height when unhilling on August 31st (Trt 7) as compared to August 15th (Trt 6) (p-value = 0.547).

Soybean Seed Count

Similar to the soybean height, there is evidence of a difference in total seed counts (p-value = 0.0056)

- The control group (Trt 1) had evidence of a difference in seed counts for all unhilling dates starting on and after July 1st (Trt 3-7). The difference was marginally significant for July 1st (Trt 3, p-value = 0.0792) and August 31st (Trt 7, p-value = 0.0788).
- Unhilling on June 16th (Trt 2) had marginal evidence of a difference with August 15th (Trt 6, p-value = 0.0788).

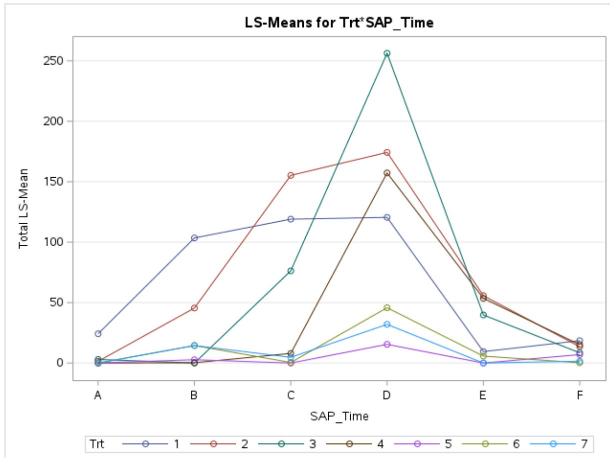
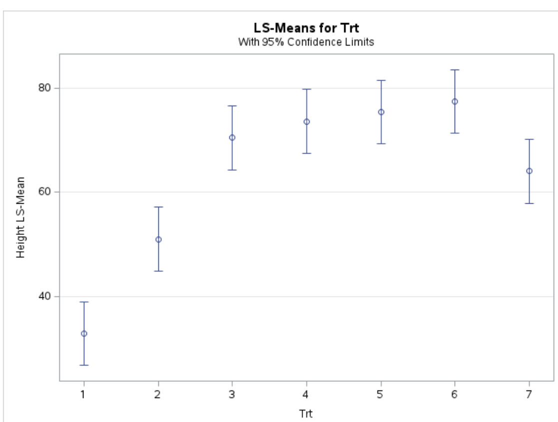
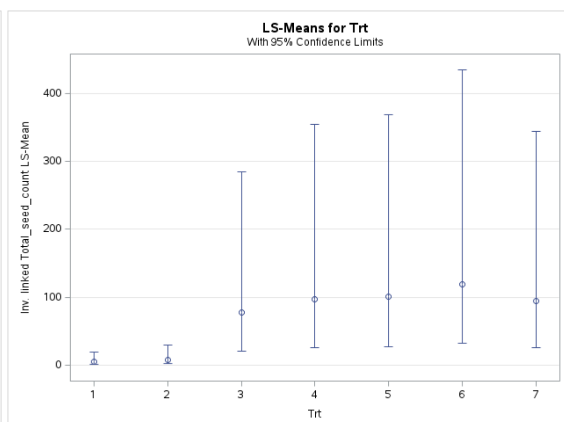


Figure 4: Least square means for larvae counts in the unhilling study. The general trend is that unhilling too early in the growing season increases the soybean gall midge larvae counts during their peak reproductice season.



(a) Soybean height.



(b) Total seed count.

Figure 5: Least square means for soybean height and total seed counts from the unhilling study and their corresponding 95% confidence intervals. Reference Table 2 for the unhilling dates.

Sample Date	Treatment differences	Estimate	P-value
July 1st	1, 4	111	0.0515*
	1, 5	119	0.0202
	1, 6	118.5	0.0215
	1, 7	114.25	0.0356
	2, 4	147.25	0.0004
	2, 5	155.25	0.0001
	2, 6	154.75	0.0001
	2, 7	150.5	0.0002
July 15th	1, 3	-135.75	0.0022
	1, 5	105	0.0973*
	2, 6	128.5	0.006
	2, 7	142.25	0.0009
	4, 5	141.75	0.0009
	4, 6	111.5	0.0487
	4, 7	125.25	0.0092

Table 4: Statistically significant differences for larvae counts in the unhilling study. The treatments can be found in Table 2, but are left as numbers to indicate the positioning of the sample dates during the growing season. The p-values were calculated using Tukey's HSD and those marked with an asterisk (*) are considered marginally significant.

4. Discussion

Soybean gall midge is a pest to soybean fields in the midwestern United States. The results of the unhilling yield component data show that unhilling on or after June 16th typically had the best results for the overall health of the crops. Unhilling too early in the growing season tended to allow for an increase in the counts of soybean gall midge larvae in July. The larvae counts in the hilling study did not have many significant differences, so it is reasonable to assume that hilling soybeans at the beginning of the growing season has the best protection against damage from the larvae.

Despite promising results, it should not be immediately concluded that hilling is definatively beneficial to reducing the damaging effects of soybean gall midge larvae. Each study was only conducted with one field located in Syracuse, Nebraska. Should the results be replicated in other locations, then generalization can occur outside of the location-specific conditions of the studies presented. Other factors that could have influenced the responses, such as weather and field conditions, were not recorded. This study is a good first step in establishing recommendations at stopping the damage caused by the soybean gall midge infestation.

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