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Manipulation of soil temperatures to influence brood emergence in the alkali bee (*Nomia melanderi*)

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Abstract – Soils in beds of commercially managed *Nomia melanderi* were manipulated with surface treatments to reduce or increase temperatures at nest depths of 20 cm. Modifying soil temperatures affected pupation rate and emergence date of *N. melanderi*. In 2010, white chalk dust delayed peak bee emergence by one week, and clear plastic mulch initiated peak emergence one week earlier, significantly increasing overall bee emergence compared to uncovered areas. In 2011, treatments included white chalk dust and plastic agricultural sheets including clear, red, black, white-on-black, blue, and brown. In 2011, clear plastic sheeting initiated early emergence while white chalk dust and white-on-black plastic plots delayed emergence. Expediting or postponing *N. melanderi* emergence could potentially allow alfalfa seed producers to extend the bees' foraging season over a greater time period, subsequently increasing pollination, seed set, and economic returns.

alfalfa / emergence / Halictidae / pollinator

1. INTRODUCTION

The alkali bee, *Nomia melanderi* Cockerell 1906 (Hymenoptera: Halictidae), is an efficient native pollinator of Eurasian alfalfa *Medicago sativa* L. (Fabaceae) in selected areas of the western USA, where the bees are actively managed by alfalfa seed producers. Alfalfa produced for seed is a cash crop in the Touchet Valley of Walla Walla County in south-central Washington, USA. *N. melanderi* is the only successfully managed native, aggregating, ground-nesting solitary bee species used for pollination in commercial agriculture in the world (Cane 2008; Pitts-Singer 2008). *N. melanderi* success as an efficient commercial pollinator for alfalfa seed production is attribut-

able to its high population density in managed nesting aggregations. Fortuitously, *N. melanderi* emergence and foraging typically coincides with alfalfa bloom (Cane 1997).

Natural populations of *N. melanderi* seek moist soils along stream and creek beds for nest sites and forage pollen from the blooms of native plants to provision brood cells. Managed *N. melanderi* bee beds provide the northwest USA with a competitive advantage in alfalfa seed production (Bohart 1970). Alfalfa seed growers that manage *N. melanderi* apply rock salt (NaCl) to the surface of their bee beds at 4.8 kgNaClm⁻² in early May prior to emergence (Johansen and Eves 1973; Johansen et al. 1982) to prevent the loss of soil moisture and control weeds (Cane 1997). Those who manage bee beds often sub-irrigate the beds with perforated polyvinyl chloride (PVC) pipe buried in trenches at depths of 61–76 cm (Johansen et al. 1982; Pitts-Singer 2008).

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N. melanderi are univoltine in Washington, and conveniently, adult emergence and peak foraging behavior tend to be synchronous with alfalfa bloom (Rust 2007; Cane 2008). To achieve the most efficient pollination and optimal seed yields, alfalfa bloom and bee flight must coincide. Spring weather varies from year to year and alfalfa bloom may not always directly match with peak *N. melanderi* emergence, though typically it blooms from early June through mid-July (Bohart 1970). In May and early June 2011, for example, rainfall reduced soil temperatures and delayed emergence of *N. melanderi*, while early blooming alfalfa varieties being produced for seed were already blooming. *N. melanderi* only forage for 6–8 weeks, so the timing of bee emergence is important to achieve ideal seed set and ensure that the bees do not emerge before bloom or too late after alfalfa bloom.

Soil temperatures have been demonstrated to have an effect on *N. melanderi* development (Stephen 1965; Rust 2007), particularly on prepupae diapausing in the soil. Stephen (1965) calculated *N. melanderi* prepupal developmental rates at controlled temperatures, documenting that development is inhibited at temperatures below 17 °C and above 35 °C and that prepupal development is fastest at 29 °C (Stephen 1965).

Bee bed managers already manipulate soil moisture, select sites for soil consistency, and manage weeds. Manipulation of soil temperature to influence bee emergence was a management tactic that merited further study. We have determined that by employing simple cultural methods to influence adult emergence, seed producers depending on *N. melanderi* for pollination can ensure that some bees will emerge to coincide with early- or late-blooming alfalfa varieties, potentially increasing seed set and subsequent economic returns.

2. MATERIALS AND METHODS

To manipulate soil temperature and quantify the effect on *N. melanderi* emergence in 2010, four bee

bed treatments were established: (1) white chalk dust, (2) black charcoal dust, (3) clear plastic mulch (in the form of clear plastic sheeting), and (4) untreated control. In 2010, treatments were applied prior to *N. melanderi* adult emergence on May 24. For chalk and charcoal dust treatments, just enough product was applied to cover 1 m² quadrats. For the chalk treatments, PVC frames were laid out and 1.42 kg of product was applied to the treatment area. Due to high winds in Touchet, WA, the plastic treatment was stapled to a wooden frame. The untreated control consisted of 1 m² quadrats free of additive dust treatments or plastic mulch sheets. Treatments were established in a bee bed with a history of high populations of *N. melanderi* near Touchet, WA (46° 01'02.50" N, 118°41'49.89" W). Each treatment was replicated 10 times in a completely randomized design.

Additional colors of plastic mulch sheeting and a second location were added in 2011. The treatments in 2011 consisted of (1) white chalk dust, (2) single-layer black plastic sheets, (3) red plastic sheets, (4) blue plastic, (5) clear plastic, (6) brown plastic, (7) white-on-black plastic (black side of plastic facing soil, white side of plastic facing sun), and (8) untreated control. The black charcoal dust from 2010 was not repeated in 2011. Red, blue, clear, brown, and black plastic were advertised by their vendors to warm the soil while the white-on-black plastic and white chalk were advertised as soil coolers. Plots were randomly assigned within replication for a randomized block design at each site. Site A (46°01'02.50" N, 118°41'49.89" W) and site B (46°01'55.71" N, 118°42'34.49" W) were each overlaid with five 1 m² replicate plots of each treatment. Treatments were established on May 3, 2011. The white chalk dust treatment was reapplied four times due to late spring rains totaling an accumulated 72.39 mm in May and June 2011 (Washington Agricultural Weather Network Version 2.0: AgWeatherNet, Touchet, WA station). Site A was the same site used in 2010, and site B was a naturally irrigated bee bed, not sub-irrigated by PVC pipe. Both bee beds had substantial *N. melanderi* population abundance from surveys conducted in fall 2010 (Vinchesi and Walsh, unpublished data).

In 2011, prior to the application of the plastic sheets or chalk dust, HOBO[®] soil temperature sensors were placed in two representative plots of

each treatment at each site. Sensors were placed at a 20-cm depth, where the greatest density of *N. melanderi* larvae are typically found (Bohart and Cross 1955). The HOBO® sensors were programmed to measure soil temperature every 28 min from May 3 to August 31.

In 2010, after the first detection of *N. melanderi* emergence on June 11, the clear plastic treatment was removed to allow the bees to emerge. Emergence holes were counted twice weekly beginning June 11 and ending July 15. The emergence holes were visually counted within each 1 m² plot. In 2011, all plastic was removed at each site after emergence holes were observed within every treatment on that site; emergence holes were counted three times weekly beginning June 13 and ending July 18.

Mean emergence hole counts were calculated for each date of the emergence period in 2010 and 2011. Comparisons to the control were made using Dunnett’s method. Similar comparisons were made using temperature data for each treatment. All repeated measure analysis of variance (ANOVA) and Dunnett’s comparisons were calculated using Statistix 9 program (Analytical Software, Tallahassee, FL, USA; www.statistix.com).

Temperature strongly influences the rates of development and growth of organisms. In poikilotherms, the rate of development through growth stages is almost completely governed by temperature (Sharpe and DeMichele 1977). Thermal time (Monteith 1977) is a convenient means for transferring biological time to clock or calendar time. Thermal time ($\Delta\tau=(T_i-T_b)\times\Delta t$) is calculated as average daily temperature (T_i) minus base temperature (T_b). Stephen (1965) determined that the base temperature was 17 °C for *N. melanderi*, and then we multiplied

the difference by the time step (Δt , in this case 1 day), with the resulting units of degree days (Campbell and Norman 1998). Daily temperatures were averaged for each of the 32 HOBO® soil sensors, thermal time was calculated, and then all mean daily temperatures were summed to calculate accumulated thermal time/degree days.

3. RESULTS

In 2010, the white chalk dust treatment on June 22 was the only treatment with a mean of less than 100 holes per m⁻²; however, by June 29, mean emergence tripled to 315.4 per m⁻² and the main peak in emergence occurred 7 days later in the foraging season of *N. melanderi* than in the untreated control plots (Table I), but it also reached 100 % emergence before the other treatments (Figure 1). The total number of emergence holes at the end of the season for the white-chalk-dust-treated plots was numerically, but not significantly ($P>0.05$), lower than the untreated control (Table I). The number of emergence holes per square meter on plots covered by the clear plastic mulch treatment was significantly greater ($P<0.01$) earlier in the season and continued to be higher throughout the season than the untreated control (Table I).

In 2011, emergence holes were observed within every treatment at site A by June 15, but first emergence holes at site B were not observed within all treatments until June 24. At site A, although no differences were observed among the treatments of total emergence hole counts over the entire season of bee flight, there

Table I. Mean emergence hole counts, 2010.

Treatment	11 Jun	17 Jun	22 Jun	29 Jun	6 Jul
Black charcoal dust	2.3±1.1	45.0±10.4	148.1±20.8	289.5±31.5	293.6±37.5
White chalk dust	0.9±0.4	9.1±2.6a	79.0±15.1a	315.4±24.3	307.4±31.9
Clear plastic	41.9±10.3a	90.7±11.3	276.4±27.5a	584.9±29.7a	603.4±39.0a
Untreated control	6.2±2.2	55.3±13.4	173.5±22.0	340.6±27.6	394.5±40.5

Mean emergence hole counts by treatment pooled by date ± standard error, 2010 ($n=10$). Emergence hole counts marked with lowercase letter “a” represent those that are significantly different from the untreated control using Dunnett’s method (June 11, 2010— $F=13.67$, $DF=3$, $P<0.001$; June 17, 2010— $F=10.73$, $DF=3$, $P<0.001$; June 22, 2010— $F=14.08$, $DF=3$, $P<0.001$; June 29, 2010— $F=23.10$, $DF=3$, $P<0.001$; July 6, 2010— $F=14.40$, $DF=3$, $P<0.001$)

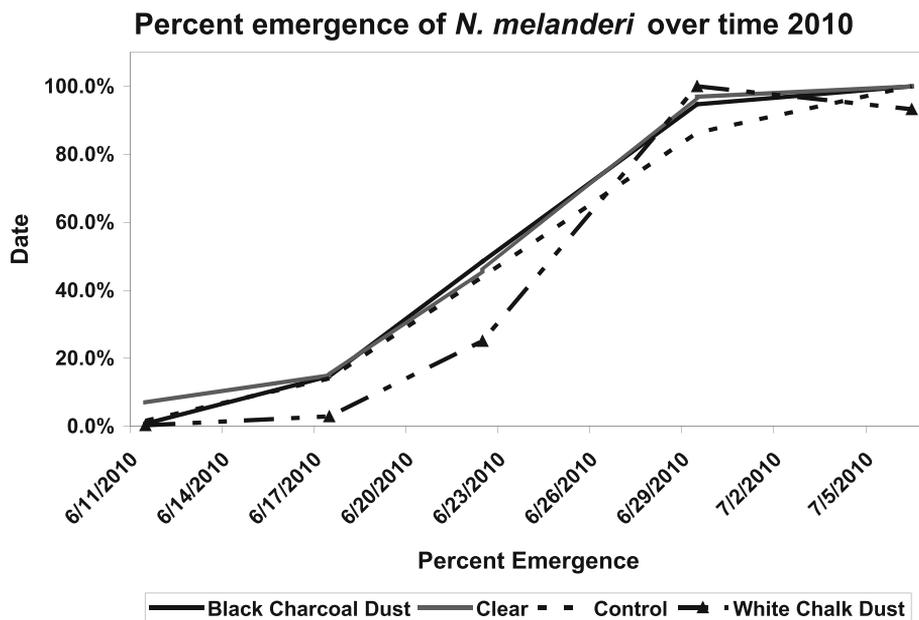


Figure 1. Percent *N. melanderi* emergence over time at site A, 2010. The white chalk dust treatment reaches 100 % emergence by June 29, while the three other treatments reach 100 % emergence by July 6.

were differences between treatments during the first week of bee emergence. An ANOVA of emergence hole counts showed that, during the first week of emergence, treatment had significant effects ($F=9.43$, $DF=7$, $P<0.001$) on mean emergence hole counts (Table II). For site A, plots treated with black, brown, white-on-black plastic, and white chalk dust had mean emergence hole counts significantly less ($P>0.05$) than the control in a Dunnett's comparison ($DF=49$, D value=2.408) of *N. melanderi* emergence on June 24 ($F=3.34$, $DF=7$, $P<0.0087$) (Table II). White-on-black plastic and white chalk dust treatments also had significantly lower means on June 27 ($F=2.57$, $DF=7$, $P<0.03$) (Table II). Also, the clear plastic treatment was significantly higher than the untreated control on the first day of observed emergence at site A ($F=4.05$, $DF=7$, $P<0.0028$). At site B, while some of the treatment plots flooded as a result of poor drainage and skewed the emergence hole counts due to bee mortality, treatment also had a significant effect ($F=8.85$, $DF=7$, $P<0.001$) on mean emergence hole counts; with the Dunnett's comparison to

control test, the clear plastic displayed significantly greater emergence hole abundance than the control throughout the first week of the nesting season ($DF=49$, D value=2.709) (Table III). Despite the later emergence dates of adult *N. melanderi* in 2011 at both sites, many of the treatments reached 100 % emergence by early July in both 2010 and 2011, though the control plots reached full emergence a week earlier in 2010 than they did in 2011 (Figures 2 and 3). The untreated control plots and clear plastic plots have similar emergence early on in 2010 and at site B in 2011, but then the clear plastic plots' emergence increases (Figures 2 and 3). The untreated controls never reach 100 % emergence at site A in 2011 (Figure 2). The site B control reaches 100 % emergence later than other treatments (Figure 3). The white-on-black plastic and white chalk dust treatments give slow emergence over the course of the study period (Figures 2 and 3).

An ANOVA was also calculated within each site to determine if treatment had any effect on accumulated degree days of the *N. melanderi*, or the time it took for *N. melanderi* to emerge as adults. Thermal time was accumulated from the

Table II. Mean emergence hole counts, 2011 site A.

Treatment	15 Jun	17 Jun	20 Jun	22 Jun	24 Jun	27 Jun	29 Jun	5 Jul
Black plastic	5.6±3.21	9.2±1.92	8±3.39	17.8±5.97	25.4±15.43a	70.2±46.24	113.6±59.28	188.4±35.21
Blue plastic	21±25.96	23.6±23.42	50±47.99	83.6±76.41	102.2±86.49	130±76.67	188.6±111.95	260.4±128.53
Brown plastic	8.2±7.98	8.8±6.76	10±7.87	18.8±12.47	37.8±31.66a	113.2±70.38	143.6±65.67	197.4±53.99
Clear plastic	65±50.6a	67.6±64.46	94±78.28	128.2±98.46	141.6±104.64	155.8±103.56	200.4±123.75	278.6±163.79
Red plastic	12.2±11.3	15.2±15.61	20.4±19.95	49.6±41.4	77.8±65.17	130.8±47.28	172.8±70.46	288.6±111.28
White-on-black plastic	8.6±6.5	12.4±10.99	13±7.68	15.2±11.51	17.4±11.63a	39.2±22.87a	71.8±29.96	150.8±45.66
White chalk dust	4.2±3.11	6±5.29	4±2.12	8.8±5.21	12.6±8.44a	49.2±27.56a	81±43.68	205.6±83.31
Untreated control	16.2±20.96	24.8±24.7	63±66.53	99±96.11	165±131.99	172.8±103.95	194.2±122.83	166.2±44.69

Mean emergence hole counts by treatment pooled by date±standard error, 2011, site A (*n*=5). Emergence hole counts marked with lowercase letter "a" represent those that are significantly different from the untreated control on that date using Dunnett's method (June 15, 2011—*F*=4.05, *DF*=7, *P*<0.0028; June 24, 2011—*F*=3.34, *DF*=7, *P*<0.0087; June 27, 2011—*F*=2.57, *DF*=7, *P*<0.03)

Table III. Mean emergence hole counts, 2011 site B.

Treatment	15 Jun	17 Jun	20 Jun	22 Jun	24 Jun	27 Jun	29 Jun	5 Jul
Black plastic	0±0	1±2.2	5.2±5.2	14.4±13.7	19.6±19.5	63.4±54.4	72±5.1	138±93.5
Blue plastic	2.2±3.2	4±5.5	9.4±6.7	17.6±12.4	20.8±13.1	53.2±31.5	72.6±45.2	122.2±73.3
Brown plastic	0±0	0±0	0±0	0.6±0.9	1.4±1.5	14±6.2	36.2±28.9	111.6±72.7
Clear plastic	26.4±32.0a	32.4±40.3a	38.6±37.1a	68±49.9	78.4±68.5	117.2±66.8	118.8±77.2	223.4±117.4a
Red plastic	0±0	2.2±3.9	8±8.2	20±26.9	25.4±29.6	69.6±90.3	74.4±52.3	172.4±113.5
White-on-black plastic	0±0	0±0	0.4±0.9	0.6±0.9	1.2±1.6	12.6±9.3	23±17.8	87.4±59.3
White chalk dust	0±0	0±0	0±0	0±0	0.2±0.4	4±3.5	9.8±7.2	48.4±32.4
Untreated control	0.2±0.4	3.8±3.9	4.8±4.8	30.8±30.2	45.6±45.3	76.2±64.9	86.6±79.9	76.4±68.3

Mean emergence hole counts by treatment pooled by date±standard error, site B, 2011 (*n*=5). Emergence hole counts marked with lowercase letter "a" represent those that are significantly different from the untreated control on that date using Dunnett's method (June 15, 2011—*F*=3.31, *DF*=7, *P*<0.009; June 17, 2011—*F*=2.87, *DF*=7, *P*<0.019; June 20, 2011—*F*=4.24, *DF*=7, *P*<0.002; July 5, 2011—*F*=2.26, *DF*=7, *P*<0.05)

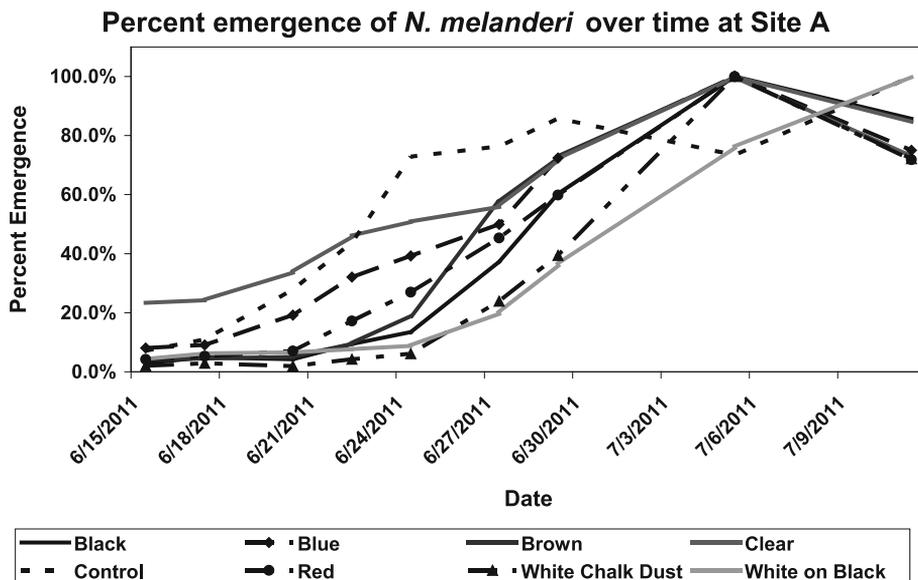


Figure 2. Percent *N. melanderi* emergence over time at site A, 2011. All treatments reach 100 % adult emergence by July 5, except for the untreated control and the white chalk dust treatments, which do not reach full emergence until July 11.

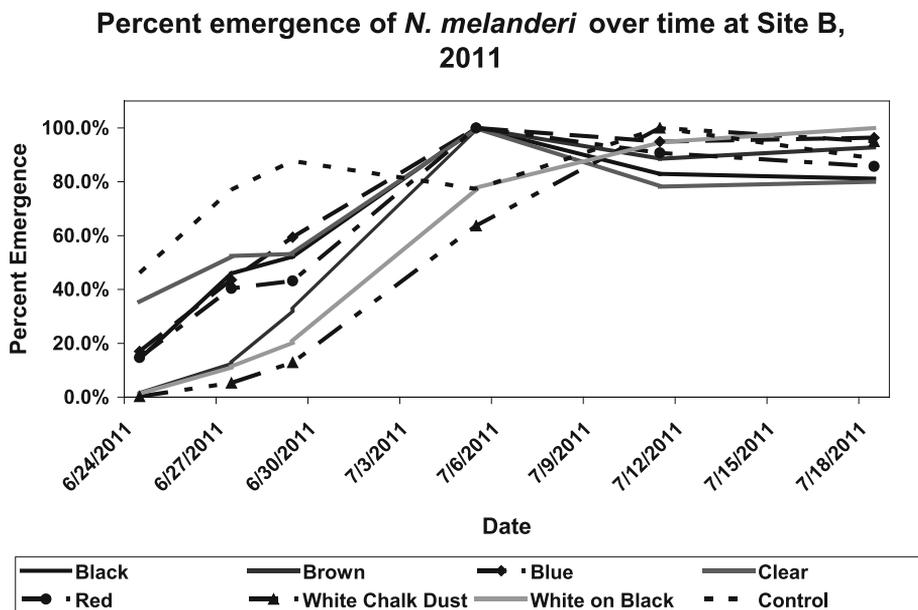


Figure 3. Percent *N. melanderi* emergence over time at site B, 2011. The control treatment and white chalk dust treatment do not reach 100 % emergence until July 11; white-on-black plastic does reach 100 % emergence until July 18; all other treatments achieve 100 % emergence by July 5.

day that treatments were placed on the bee beds until first emergence/plastic removal. Treatment had a significant effect on accumulated thermal time differences at site A ($F=20.59$, $DF=7$, $P<0.01$), with clear plastic accumulating a significantly greater number of degree days than the control and white-on-black plastic and white chalk dust accumulating less thermal time than the control (Figure 4). At site B, the same treatment effect was seen ($F=23.68$, $DF=7$, $P<0.001$), with the clear plastic treatment's accumulated thermal time significantly greater than the control and white chalk dust and white-on-black plastic treatments producing significantly lower degree-day accumulation (Figure 5).

4. DISCUSSION

In 2010, the clear plastic soil surface treatment accelerated brood emergence while the white chalk dust treatment delayed emergence of adult *N. melanderi*. In 2010, the clear plastic sheets significantly increased brood emergence through-

out the study. Unfortunately, it is impossible to directly quantify what the clear plastic did to increase emergence so significantly, due to the lack of temperature data. We can speculate that the clear plastic sheet accelerated soil warming at nest cell depth due to the significant differences in accumulated thermal time. The clear plastic may have also influenced soil moisture content or prevented drowning of the pupae following rain events. However, emergence within the clear plastic treatment in 2011 was not significantly higher than emergence within control plots as it was in 2010. With the white chalk dust, we were able to delay emergence without significantly impacting the total number of bees that emerged throughout the foraging season compared to the untreated control.

Over the course of the 2011 nesting season, there were significant differences ($P<0.05$) between treatments and the control for mean *N. melanderi* emergence hole counts. However, the reasons for these differences may be due to some of the plots being flooded in May and early June and poor drainage at site B. Site B is surrounded by a small canal and is seep

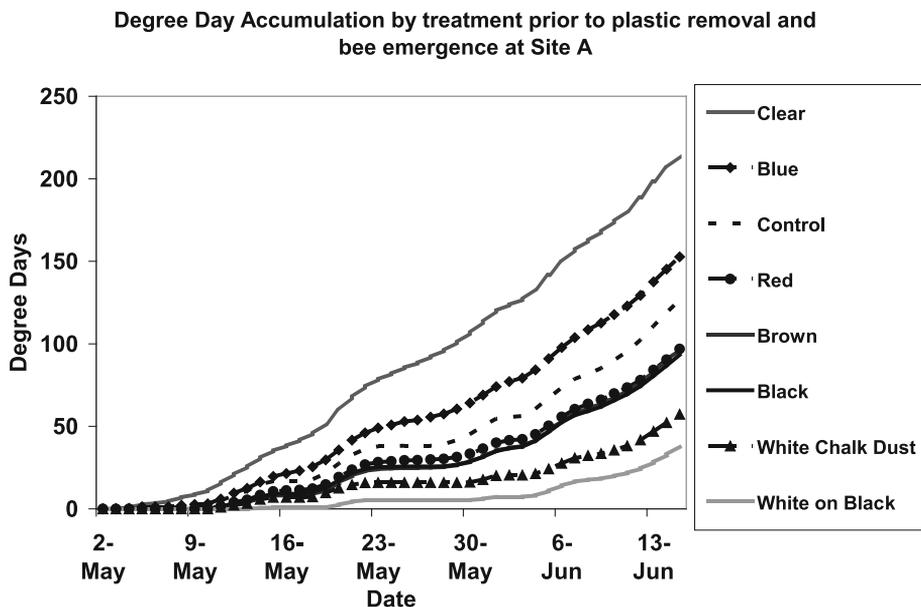


Figure 4. Degree-day accumulation by treatment prior to plastic removal and bee emergence for site A (June 15, 2011; $F=20.59$, $DF=7$, $P<0.001$).

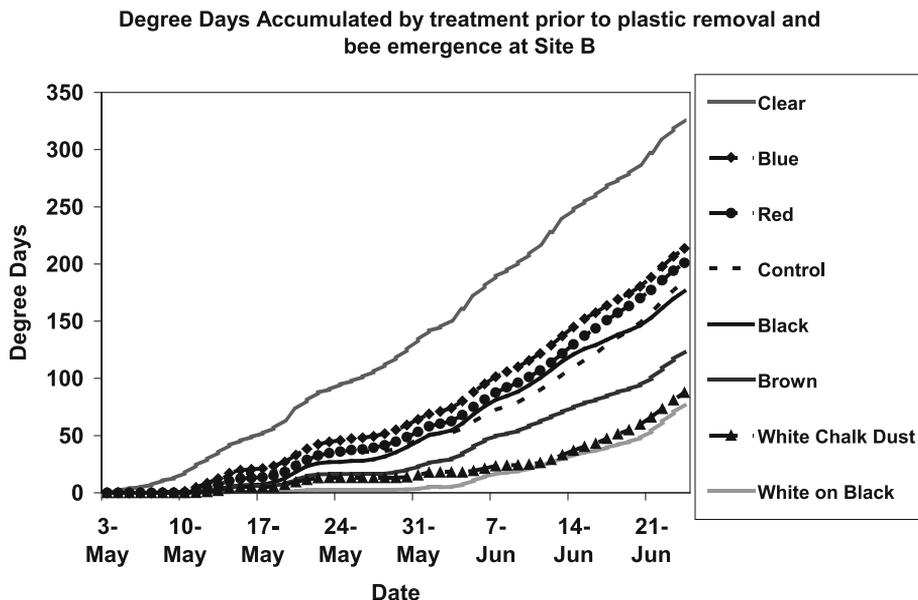


Figure 5. Degree-day accumulation by treatment prior to plastic removal and bee emergence for site B (June 24, 2011; $F=23.68$, $DF=7$, $P<0.001$).

irrigated whereas site A is sub-irrigated with PVC pipe and pumped water. Observing only the first weeks of emergence at site B, the clear plastic treatment expedited soil warming, and emergence was observed earlier in these plots than in the control plots. Brown, white-on-black plastic, and white chalk dust treatments showed both delayed and reduced emergence and cooler soil temperatures compared to the untreated control plots (Table III).

We concluded that the white treatments reflected sunlight instead of absorbing it, hence keeping the soil underneath cooler prior to emergence (Ham et al. 1993; Bonachela et al. 2012) and slowing the development rate by not accumulating sufficient thermal time for initiation of bee emergence until a later date (Figures 4 and 5). In general, *N. melanderi* emergence in 2011 was later than 2010 emergence, which can be attributed to late spring rains, less sunshine, and cooler air temperatures keeping the soil temperatures lower than some previous recorded years. Between June 1 and June 15, mean soil temperature at 20 cm was 19.4 °C in 2011, 19.7 °C in 2010, 22.1 °C in

2009, 18.8 °C in 2008, unavailable in 2007, 19.6 °C in 2006, and 20.6 °C in 2005 (AgWeatherNet).

The 2010 and 2011 results demonstrate that clear plastic accelerates bee development and subsequent emergence by letting visible light through, preventing infrared light from escaping, and suppressing evaporation, thus heating the soil (Ham et al. 1993; Bonachela et al. 2012). The 2011 soil temperature data provide evidence that the soil is warmer, and therefore, *N. melanderi* pupae accumulate more thermal time leading to earlier bee emergence under the clear plastic mulch treatment. Clear plastic mulch sheeting is a treatment that alfalfa seed growers could readily apply to accelerate *N. melanderi* emergence to pollinate the early bloom period of alfalfa. White chalk dust and white-on-black plastic can be used to slow emergence to ensure that bees will be foraging on any late-blooming alfalfa.

Alfalfa seed growers and owners of *N. melanderi* beds could apply these treatments in large strips to 5–10 % of their bee beds to

ensure emergence of *N. melanderi* throughout the entire bloom season since adults are active for only 4 to 6 weeks. Delaying development on areas of the bee bed with white-on-black plastic or white chalk dust would result in a controlled, staggered emergence of *N. melanderi*. An advantage to having *N. melanderi* emerge over time is to make certain any early or late blooming alfalfa is tripped to set the seed. If all the bees emerged at once, there is the potential that not enough alfalfa will be blooming to sustain all of the foraging bees. Environmental conditions differing between the two bee beds in this study allowed for different emergence dates and rates. Though most treatments between the two sites reached 100 % emergence at the same time (Figures 2 and 3), those at site A began their emergence earlier in 2011 than those at site B (Tables II and III).

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Effet de modifications expérimentales de la température du sol sur l'émergence du couvain de l'abeille des terres alcalines (*Nomia melanderi*)

Luzerne / émergence / Halictidae / pollinisateur / USA

Manipulationen der Bodentemperatur zur Beeinflussung des Schlüpfzeitpunkts der Alkalibiene (*Nomia melanderi*)

Alfalfa / Schlupf / Halictidae / Bestäuber

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