Understanding the Life History and Feeding Behaviors of the Tobacco Hornworm (*Manduca sexta*)

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ABSTRACT

There exists a natural balance between plant life and insect herbivores, which is continuously upset by modern agriculture and pest control. Larvae of the order Lepidoptera (butterflies and moths) require a large amount of food to initiate and complete metamorphosis. These larvae are important agricultural pests, destroying large percentages of crops every year and driving up harmful pesticide use. This herbivory especially increases during the last larval instars right before pupation. Tobacco Hornworms (*Manduca sexta)* are known to have a 'wandering stage' in which they are most mobile, and also consume the most plant material. To study the impacts of this behavior, I observed six *M. sexta* larvae throughout the final two instars and recorded data regarding their consumption and growth. My observations began during the 4th instar, where all six larvae measured about 20 mm in length and weighed from 0.4–0.6 grams. The larvae all ecdysed into the 5th instar stage on Day 4 of the experiment, which was preceded by about one day of little to no consumption, movement, and growth. Between Days 5 and 6 of the experiment, all of the larvae initiated the wandering stage, and the frass production rate began to increase. Larval weight increased until reaching a threshold of 4.5–6.5 grams, which took 13 days from the start of the experiment. On Days 13 and 14, the larvae's weight dipped down after purging their guts, and they all began pupation.

KEYWORDS

entomology, Lepidoptera, plant-insect interactions, wandering stage, insect behavior

INTRODUCTION

Perhaps no insect larva is more iconic than the caterpillar, with its insatiable appetite and stunning metamorphosis. In addition to being exciting and educational pets for an elementary classroom (Goodman et al. 2001), caterpillars are also a formidable agricultural pest. These larvae of the order Lepidoptera (butterflies and moths) hatch on and eat host plants at a voracious rate, making them responsible for up to 42% of global agricultural economic losses (Sree and Varma 2015). Some of these small herbivores are specialists for specific plant families. Many plant and larvae pairs have therefore coevolved a unique relationship to maintain the balance between insect population and plant growth. For instance, plants have developed chemical defenses directly targeting their specialized pest species, or attractants to natural enemies of the pest (Baldwin 2001, Yamamoto and Fraenkel 1960). Host plants often also serve as locations for larvae to pupate into adults, upon which they become resting structures, sources of nectar, and sites of oviposition. Therefore, the quality and abundance of plant resources have a tremendous impact on herbivorous insect larval fitness factors including size and ability to metamorphosize (Davidowitz 2003). Larval behavior from egg to pupation is largely influenced by the necessity of accessing plant resources.

Caterpillar foraging behavior varies throughout the insect's life cycle. Many species of Lepidoptera larvae have been observed to go through a distinct 'wandering' stage, in which they exhibit faster, farther, and more frequent movement during their final days before pupation (Beetz et al. 2008, Dominick and Truman 1984). There are many possible reasons as to why the larvae would wander at this time. One possible cause is the induction of chemical defenses by the host plant that would deter the herbivore from the location (Gandhi et al. 2020). Plant constitutive defenses, such as physical attributes like wax or trichomes, can also impact an insect's mobility along the surface of the plant. This plant characteristic would make larger, late stage larvae more capable of movement than during smaller stages (Kariyat et.al 2017). These larger sizes are also easier targets for parasitism or predation, both common threats to Lepidopteran life. A recent study has proven that the tobacco hornworm's immune system is the weakest during the final days of the 5th instar while preparing for pupation, which makes them more susceptible for both infection and parasitism at this stage (Booth et al. 2015). Increased wandering lowers competition between larvae hatching on the same plant, allowing for more widespread herbivory. Additionally, habitat

temperature and weather events could increase the need for an insect to wander to a more protected area of the plant while feeding (Clissold and Simpson 2015).

Life stage changes may influence wandering behavior as well, such as when a larva moves to a new location safe for a lengthy metamorphosis. The Tobacco Hornworm caterpillar (*Manduca sexta*) pupates underground (Yamamoto 1968), so wandering mobility is necessary to transport the larvae off of the plant and into soil for pupation to occur. This need for relocation is a likely reason for the increased movement closer to pupation. Lowering the chances of predation or parasitism is an additional benefit of wandering. Mobile caterpillars are a more difficult target, which is especially important at this life stage when the cellular changes during each larval stage transition (Beetz et al. 2008) cause the fifth instar's immunity to lower (Booth et al. 2015). Although it is clear that consumption is crucial for the larvae's growth and survival, it is unknown why the wandering behavior is only observed in the 5th instar stadium. To further explore this plant-herbivore interaction, it is necessary to gain a deeper understanding of how the herbivore itself operates.

To study the wandering and herbivory behavior of chewing larvae such as *M. sexta*, I aimed to answer the following question: What are the feeding behaviors of *M. sexta* larvae, and how do they impact larval development? My hypothesis is that the amount of food the larva consumes will increase with the development of the larvae, culminating in the wandering stage requiring the most plant consumption. Furthermore, I hypothesized that this 'wandering' behavior has a positive effect on consumption and growth, and that higher rates of wandering would correspond to more consumption and faster development. To test this hypothesis, I investigated the following subquestions. (1) What are the growth and development patterns of the final two larval life stages of *M. sexta?* (2) How does consumption impact larval growth of *M. sexta*? And, (3) how does the *wandering stage* of *M. sexta* impact consumption of *Nicotiana benthamiana* (close relative of tobacco)? To answer these questions, I observed six *M. sexta* larvae throughout the larval stages until pupation, measuring each individual's length, weight, and frass production over time. The larvae were split into two treatment groups to determine the effects of more versus less 'wandering' behavior on these measurements.

METHODS

Study Organism

My research project focused on the "tobacco hornworm," *M. sexta*. Hornworms, including *M. sexta'*s close relative *Manduca qinquemaculata* (the tomato hornworm), are a major agricultural pest due to the voracious appetites of the larvae on important crops in the family Solanaceae (tobacco, tomato, potato) (Goodman et al. 2001). I chose to use *M. sexta* as my research organism because it is fast growing and large in size, making it ideal for a short-term experiment using relatively quick measurement techniques. This organism also has a long history of published research, resulting in much information about the species' life history, behavior, and details of rearing methods (Reynolds et al. 1985).

Manduca sexta larvae have 5 instar stages, separated by 4 larval molts. These molts are cued by the larval ability to reach specific weight thresholds by eating (Nijhout et al. 1975). The final threshold weight of a larvae is 5-7 grams, and larvae with head capsules wider than 5.1 mm will always pupate in the following molt (Davidowitz et al. 2003). Adequate consumption of the host plant is important for the larvae, as metamorphosis can be delayed if the larvae does not reach its critical weight (Davidowitz et al. 2003). *Manduca sexta* larvae consume their host plant at different rates depending on their instar stage (Nijhout et al. 1975). The final instar consists of a feeding stage, a wandering stage, and a pre-pupal stage (Beetz et al. 2008). During the pre-pupal stage, the dorsal vessel becomes more conspicuous and pulsates, as pink ommochrome pigment appears along the dorsum of the larvae (Davidowitz et al. 2003). Pupation occurs underground, so the pre-pupal larvae will burrow themselves at an average depth of 6–20 cm deep and emerge as a hawk moth approximately 22–90 days later, depending on whether or not a diapause state is induced by environmental conditions or genetic makeup of the colony (Reinecke et al. 1980).

Rearing and Growing Conditions

I obtained one 50-count egg cup from Great Lakes Hornworm Supply, which shipped the live specimens directly to me. The hornworm cup was shipped containing an artificial diet which I immediately supplemented with *N. benthamiana* leaves. This plant, colloquially known as "Benthi", is a close relative to commercial tobacco. It is the most widely used plant material for

studies in virology, plant disease, and agricultural research regarding pathogens of Solanaceous crops (Goodin et al. 2008). *N. benthamiana* is also a host plant of *M. sexta* larvae. The plants and hornworms were kept in the Oxford Insectary Greenhouse, with temperatures set to 70℉ in the nights and 75℉ during the day. Each larvae was removed from the cup to feed solely on *N. benthamiana* upon growing to about 20 millimeters in length, which is when I also transferred six individuals to cages for my experiment. The lighting program consisted of lights turning on from 6 am to 10 pm every day of the week, with the photocell off when the outside light levels were above 850 umol/m²/sec. For the plant material, I obtained all *N. benthamania* starts from the Joint BioEnergy Institute's Shih Lab, as well as extra seeds to grow in the greenhouse throughout the experiment. Upon signs that the larvae were ready for pupation (pulsating dorsal vessel, loss of white spiracles), I provided buckets of soil for the larvae to burrow into (Davidowitz et al. 2003). After pupation was complete, I removed the pupae for weighing and sexing, and I collected the final molts to measure the width of the head capsules using a micro scale under a microscope.

Wandering Experiment: NCM Treatment

To examine the unmanipulated wandering behavior of the *M. sexta* larvae, I first observed the larvae's movements among a pair of *N. benthamiana* plants without any manipulation. In this first experiment, called the "No Controlled Movement" (NCM) Experiment, I placed a single *M. sexta* larva on a pair of 2 *N. benthamiana* plants arranged in nursery trays close enough for the leaves of each plant to be slightly touching. This arrangement was replicated 3 times. The larvae were enclosed in a mesh butterfly cage, with each plant pair spaced as far apart as possible (Figure 1a). Every day in the late afternoon, I recorded the plant location of the larva (plant A or B), the length of the caterpillar in millimeters (Figure 1b), and the number of frass pellets. I also recorded the weight of the caterpillar in grams, as well as weighing the accumulated and dried frass. When the caterpillar reached pupation, I recorded the final pupa weight and length. I also measured the head capsule width of the final molt using an ocular micrometer and a metric mini-scale.

I predicted that the caterpillars would initially stay on the original plant and begin wandering among the population upon molting into the 5th instar. If the central plant was defoliated before the 5th instar was reached, I believed that the larvae might wander at an earlier

instar stage to find more food. I expected that consumption rates would increase with instar stadium, with the highest consumption rate occurring at the 5th instar before pupation.

ACM Treatment

To determine how increased wandering impacts the larvae herbivory behavior, I carried out the NCM experiment simultaneously with 3 more replications of an "Artificially Controlled Movement" (ACM) experiment. This treatment involved moving the individual caterpillars to the other plant in the pair once a day in the late afternoon. These ACM larvae were enclosed in another mesh butterfly cage. Each day, I recorded the plant position I had artificially moved the larva to (A to B, B to A), larval length in millimeters, number of frass pellets, and weight in grams every third day. I also collected the frass pellets of each individual larva for drying and weighing. For this treatment, I predicted that the insects would get increasingly bigger, the plants would become largely defoliated, and that the caterpillars in this treatment would exhibit heavier weights overall compared to the NCM treatment.

Figure 1. Experimental conditions and measurements. a) Example of ACM and NCM treatment setup, three pairs of plants placed as far apart as possible in a mesh caterpillar cage. b) Measuring the length of the *M. Sexta* larvae in millimeters.

Data Analysis

After completing data collection until all of the caterpillars reached final pupation, I used the data to create three graphs for each individual with 'day' as the explanatory variable. These graphs included larvae length in millimeters over time, larvae weight in grams over time, and accumulated frass weight over time. Then, I combined all of the individual graphs into three figures with six data series each. I was particularly interested in seeing where the individual larvae all experienced similar changes in growth, such as notable decreases or increases in length or weight at certain times. I marked the dates of ecdysial (molting) from the fourth to the fifth instar as well as the date of pupation to determine how these life history events impacted the larval growth and consumption. Once all of the larvae completed pupation, I compared the six specimens' pupae weight and length, as well as the final headcap widths. Upon sexing the larvae after pupation, I also noted whether sex had a clear impact on instar or pupa size and weight.

RESULTS

NCM Treatment Results

The three caterpillars in the NCM treatment (labeled NCM 1, NCM 2, NCM 3) all started at a mass of 0.40 to 0.70 g, and were placed on "plant A". In the following days, I recorded their movement among the two *N. benthamiana* plants labeled A and B (Tables 1-3 in Appendix A), observing that over the course of the experiment, NCM 1 switched to plant B on day 2, and then back to plant A on day 5, before wandering around the enclosure for the rest of the experiment. NCM 2 switched to plant B on day 4, and then started wandering as well. NCM 2 moved to plant B on day 5, and then wandered until pupation. After the initiation of increased wandering on days 4 and 5, I stopped tracking plant position and frass production since the larvae were wandering considerably as well as defoliating all of the plant material, which I began replacing daily. At this stage, some larvae wandered onto the plants designated to a different individual; I noted this by writing the plant position as "NCM 2 B", meaning that the NCM 1 larvae had wandered onto "Plant B of NCM 2 individual". The growth data and frass production until wandering are shown in the accompanying graphs (Figures 2a, 2b, 3a, 3b, 4a, 4b) to visualize the larval growth and consumption over time.

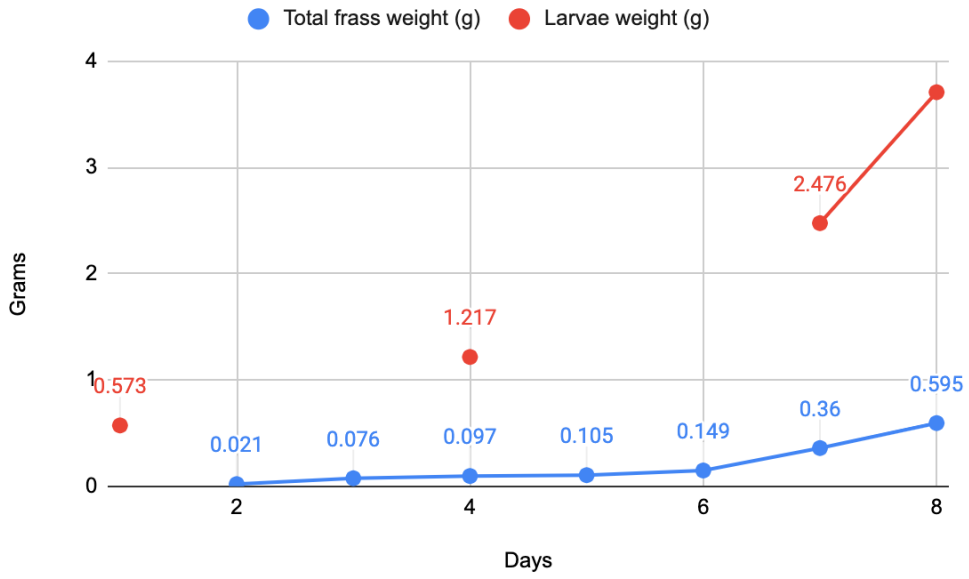

Figure 2a. NCM 1 larva weight (g) and total frass weight (g) over time.

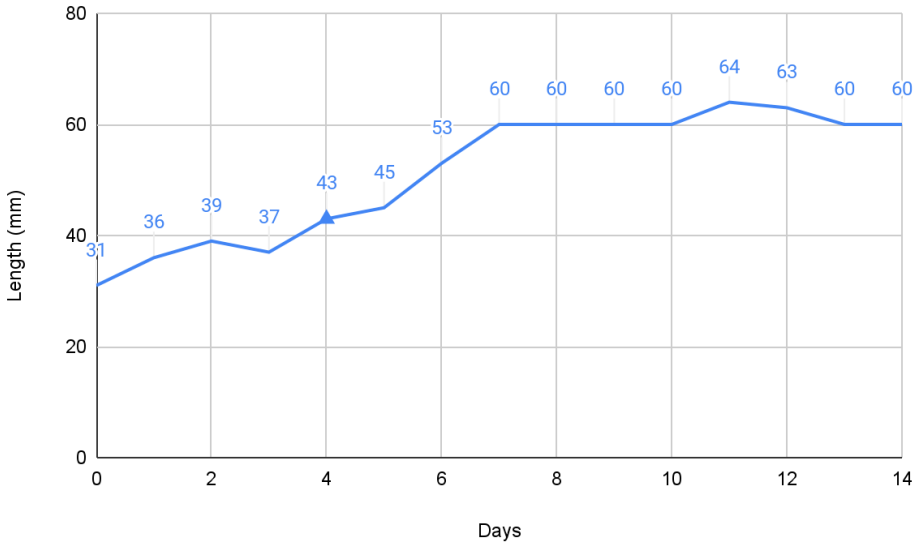

Figure 2b. NCM 1 larva length (mm) over time. Ecdysial molting noted with a triangle.

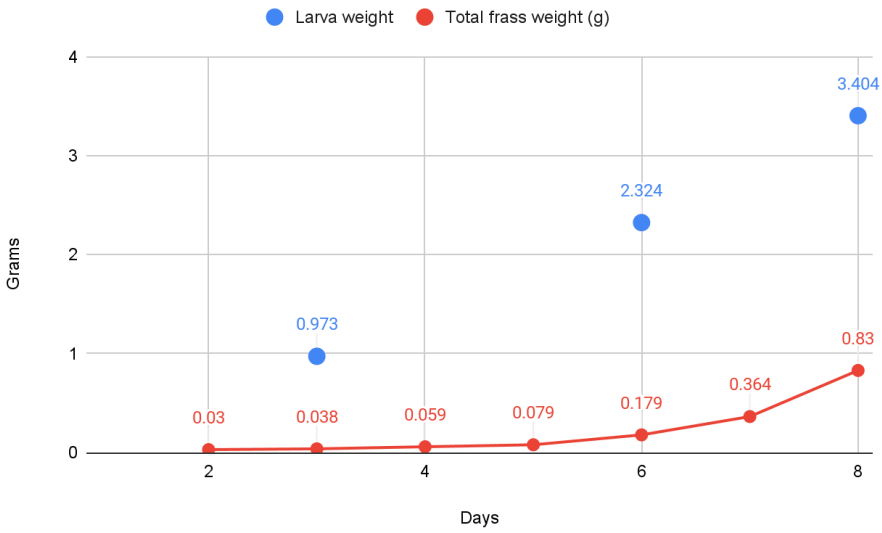

Figure 3a. NCM 2 larva weight (g) and total frass weight (g) over time.

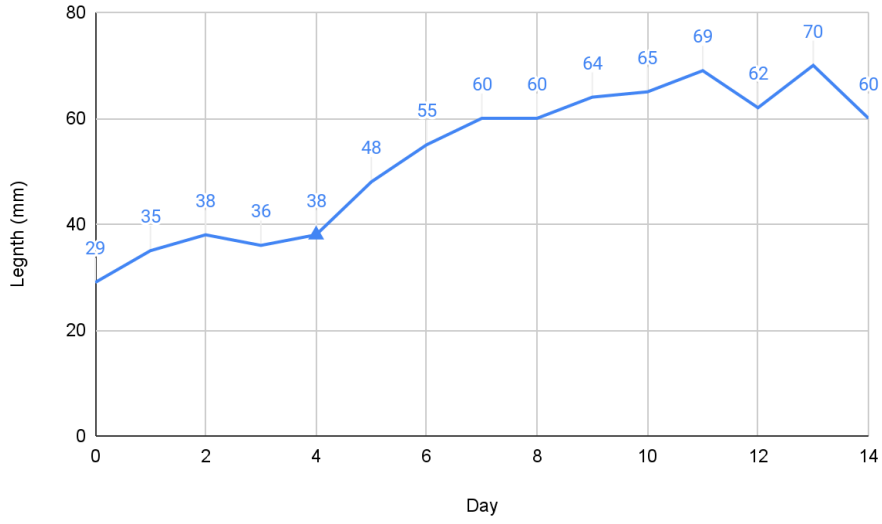

Figure 3b. NCM 2 larva length (mm) over time. Ecdysial molting noted with a triangle.

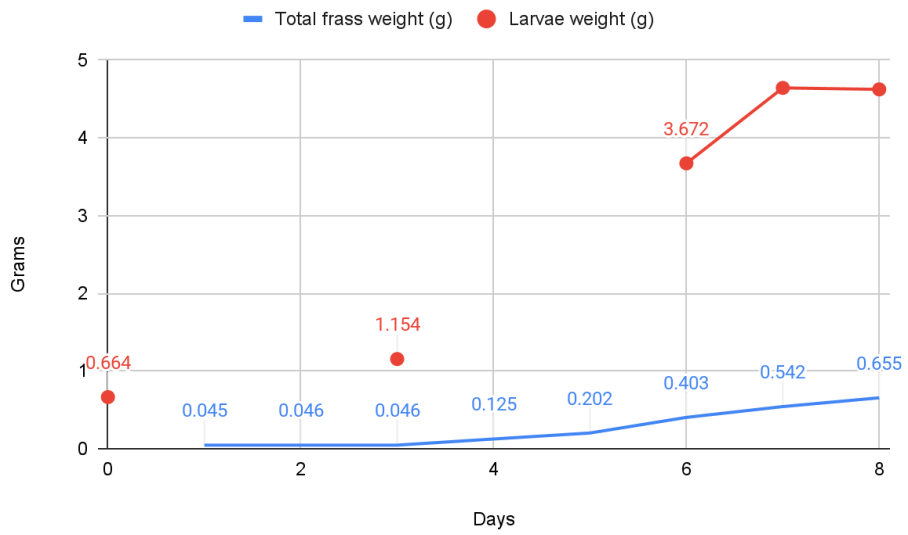

Figure 4a. NCM 3 larva weight (g) and total frass weight (g) over time.

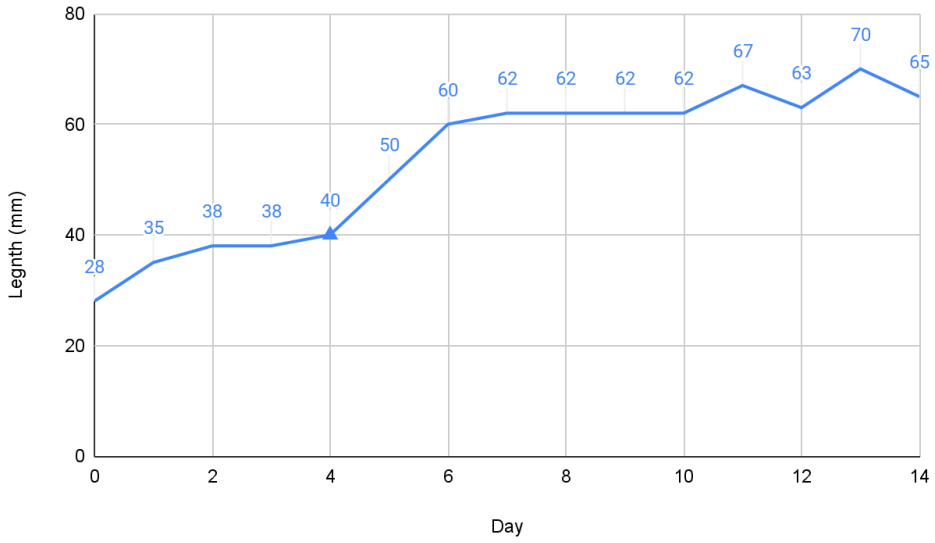

Figure 4b. NCM 3 larva length (mm) over time. Ecdysial molting noted with a triangle.

ACM Treatment Results

The three caterpillars in the ACM treatment (labeled ACM 1, ACM 2, ACM 3) all started at a mass of 0.40–0.70 g, beginning on "plant A". In the following days, they were artificially moved among two *N. benthamiana* plants (labeled A and B) once a day. On Day 9, I stopped tracking frass production, since the larvae were wandering considerably as well as defoliating all of the plant material, which I began replacing daily. The growth data and frass production until wandering are shown in accompanying graphs below (Figures 5a, 5b, 6a, 6b, 7a, 7b) to visualize the larval growth and consumption over time.

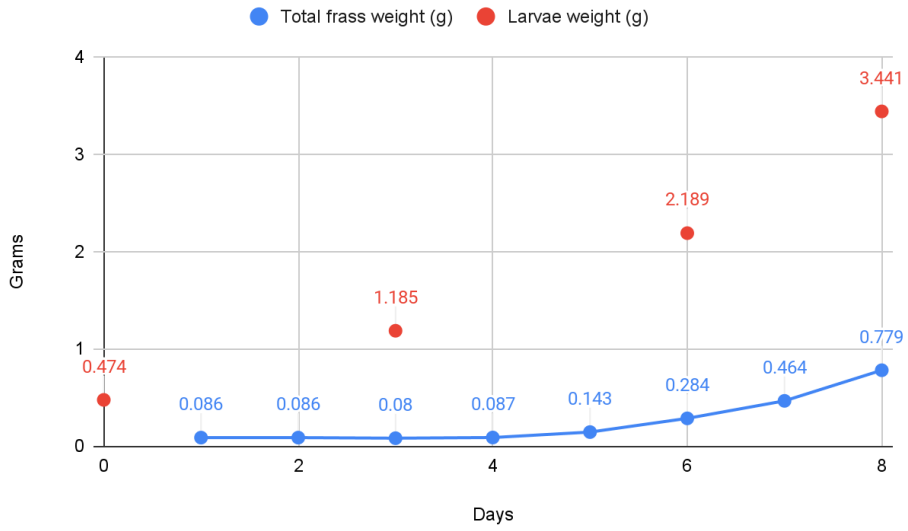

Figure 5a. ACM 1 larva weight (g) and total frass weight (g) over time.

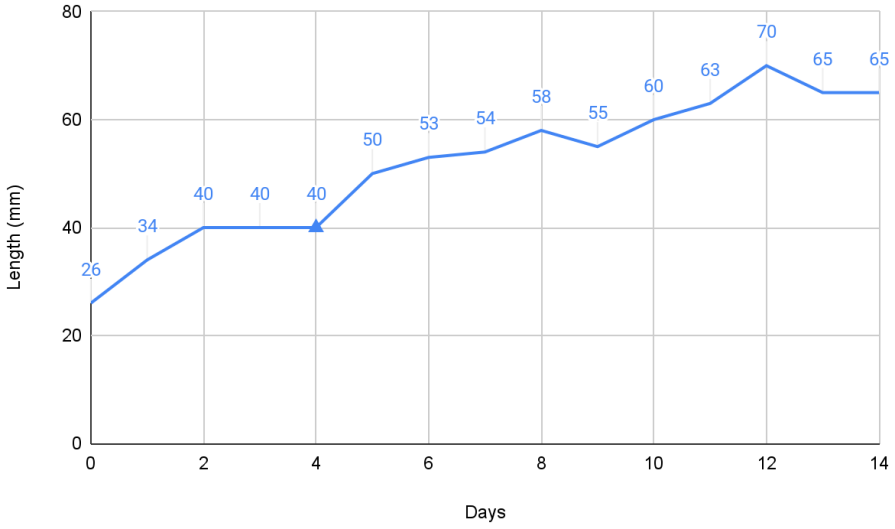

Figure 5b. ACM 1 larva length (mm) over time. Ecdysial molting noted with a triangle.

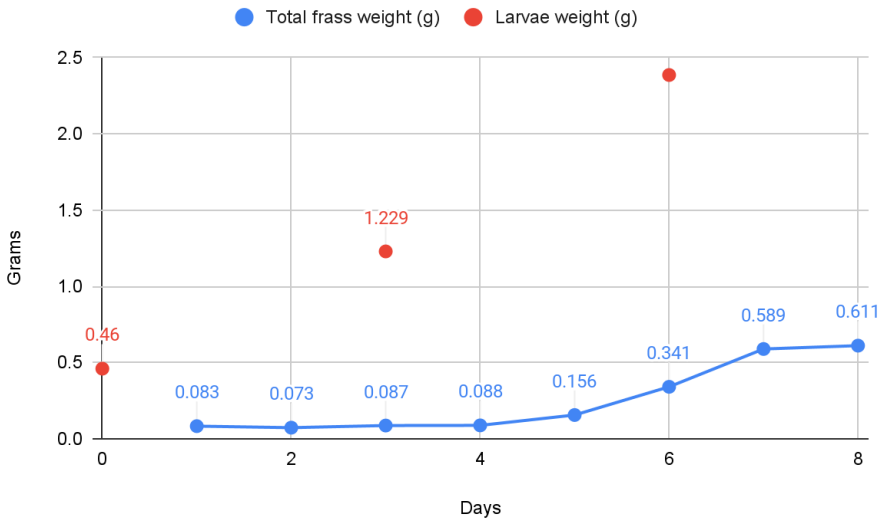

Figure 6a. ACM 2 larva weight (g) and total frass weight (g) over time.

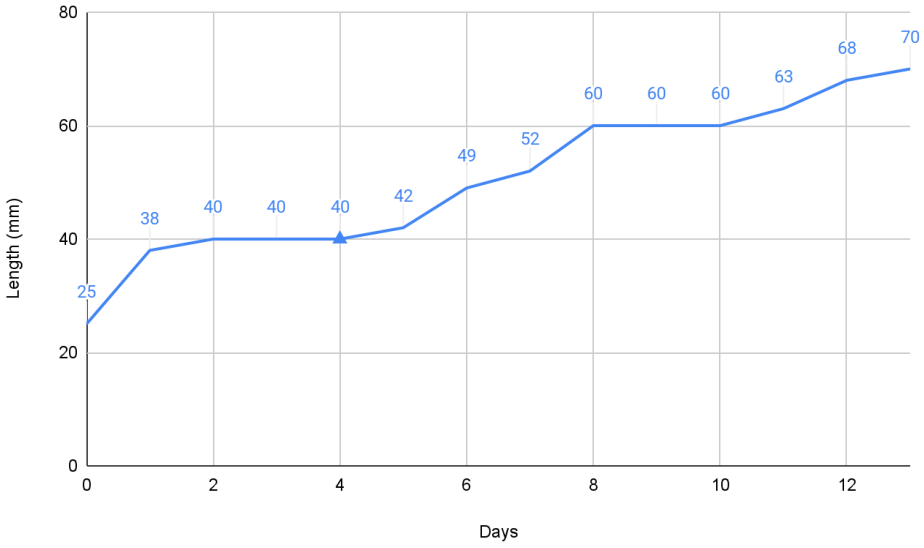

Figure 6b. ACM 2 larva length (mm) over time. Ecdysial molting noted with a triangle.

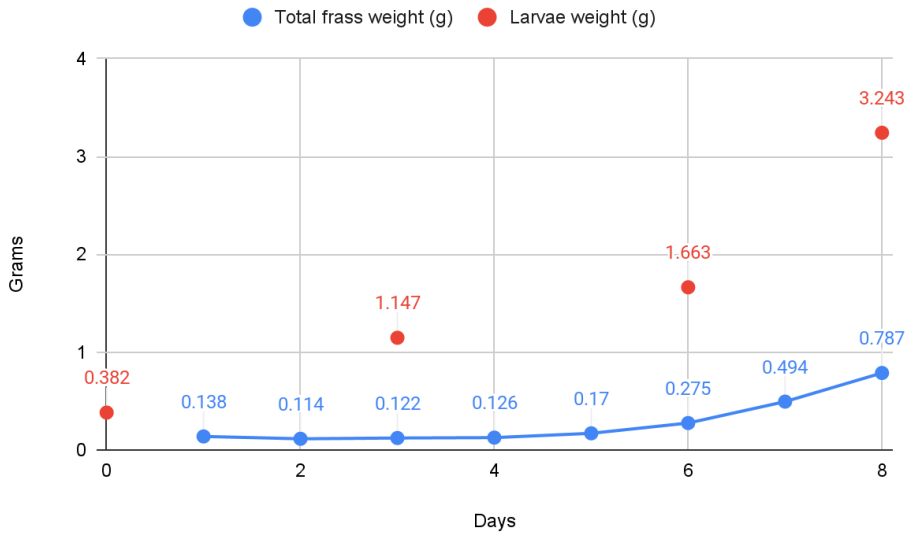

Figure 7a. ACM 3 larva weight (g) and total frass weight (g) over time.

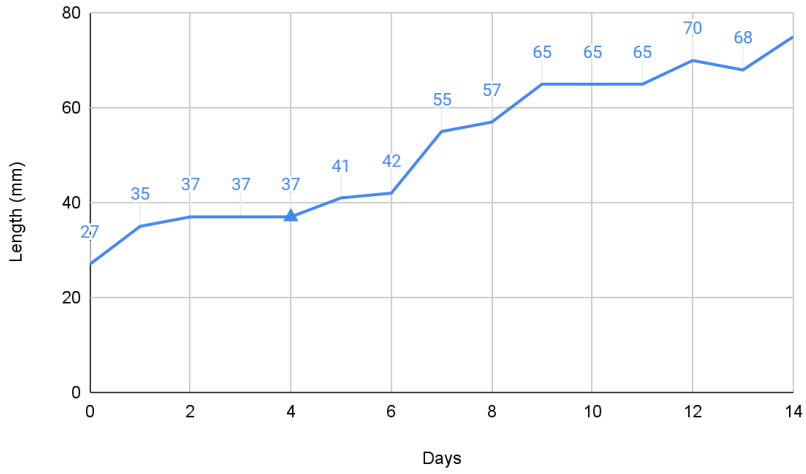

Figure 7b. ACM 3 Larva length over time. Ecdysial molting noted with a triangle.

Summary

Total frass production, which serves as a signal of consumption, is positively correlated with larval growth in the form of mass gain. A series of graphs follows, compiling data from all of the individual larvae (Figures 8a, 8b, 8c). The length, weight, and frass production over time are consistent throughout all of the larvae individuals, regardless of treatment.

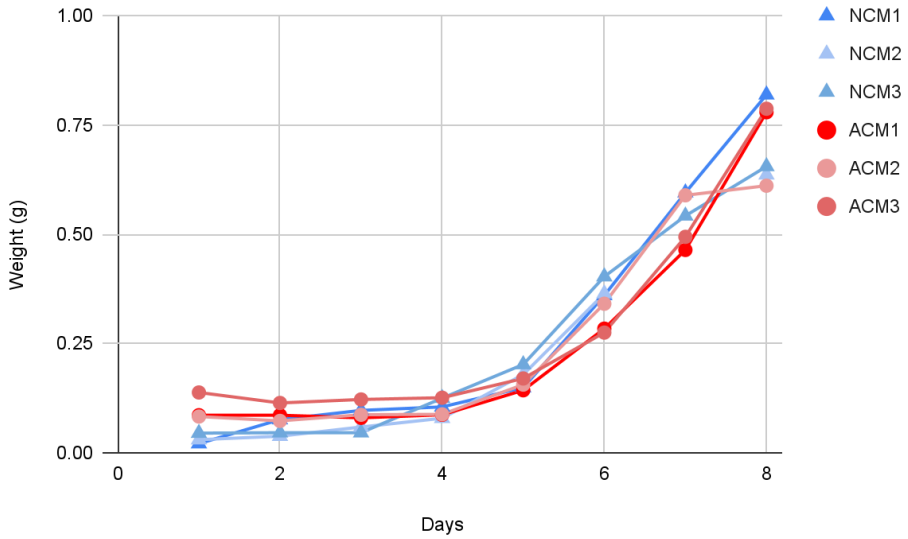

Figure 8a. All individuals total frass weight (g) over time. Frass collection stopped after Day 8 because the onset of the wandering stage made it too difficult to determine which frass pellets came from which larvae.

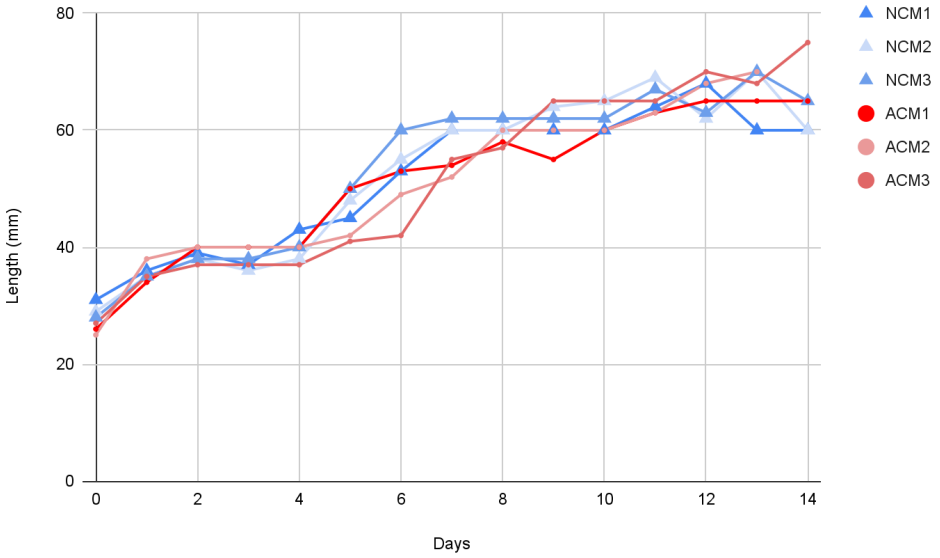

Figure 8b. All individuals larva length (mm) over time.

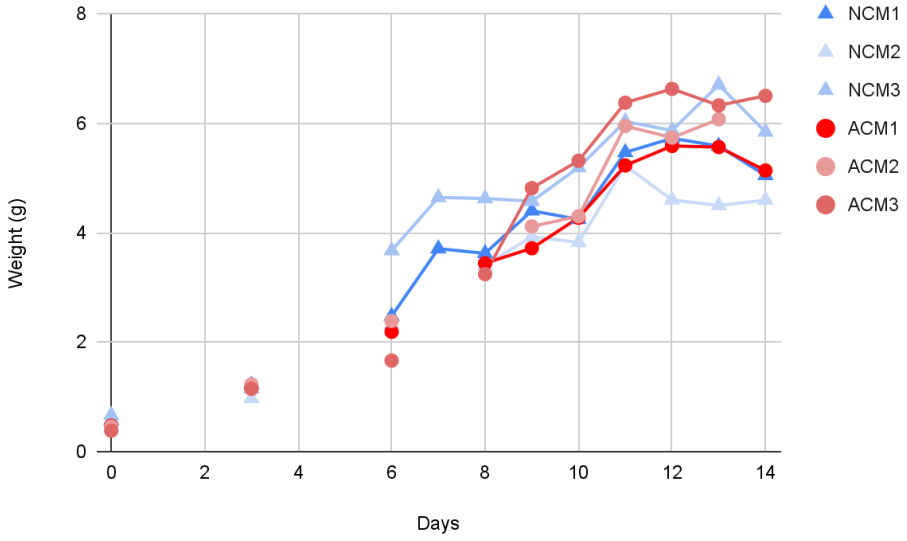

Figure 8c. All individuals' larva weight (g) over time.

Pupation Observations and Measurements

All six individuals began pupating between the 14th and 15th days. The larvae all ceased eating plant material for about 24 hours before pupation, but continued to wander around the enclosures. Once I provided them a container of soil, the larvae commenced burrowing themselves by inserting their heads and slowly digging deeper until reaching the very bottom of the container. I left the larvae to pupate and carefully dug them out after about one week in order to weigh, measure, and sex them.

The final pupa consists of a hard brown shell with a loop at the top. The lower half of the pupae wiggles or pulsates, and the casing feels fragile to the touch. Any pupae that I uncovered too soon contained gaps in the brown casing that clear liquid streamed out of, but these gaps were covered with the brown cocoon casing after a few more days in the soil. I recovered the final molt and head capsules of each of the individuals, which were split down the middle and located at the bottom of the soil container. I measured the widths of the final head capsules using an ocular micrometer and metric mini-scale (Table 7 in Appendix). My sexing results revealed that my specimens were all male except for one female, which had both the heaviest and longest pupae, with the largest headcap width. One individual, NCM 1, did not complete pupation because it failed to burrow into the soil and therefore died midway through the process.

DISCUSSION

I hypothesized that throughout the observation of the larval development, larvae would consume plant material faster over time. I also expected that the 'wandering stage' would be observable during the 5th instar, and that this period of time would include the highest rate of plant consumption. I observed six *M. sexta* larvae of the same starting age, and attempted to use two treatments of 'wandering' levels to determine the effect of increased wandering on the growth and consumption of the larvae. My observations and measurements of the larvae over time revealed key patterns in their consumption and growth with respect to life history events such as ecdysis and pupation, but my experiment failed to detect a difference in growth between wandering treatments.

Growth Rate Patterns

After observing the life history of *M. sexta* larvae from fourth instar to pupation, I discovered that the development and growth rates fluctuate with respect to changes in life stage such as molting or pupating. For example, each individual larva experienced a plateau in length over time, followed by periods of increasing growth until the final length of about 60–70 mm (Figure 8b). Notably, this plateau occurred on Days 2–4, and I observed ecdysis of all six larvae on Day 4. All individuals grew faster than usual between Days 5 and 6. Around Day 12, the larval weight (Figure 8c) dipped for all of the individuals, which was followed by them all pupating on Day 14 or 15. This drop in mass is likely due to the documented behavior of larvae "purging" the gut prior to pupation (Reynolds et al. 1985), as well as water loss from "body wetting" behavior used to burrow under the soil (Rienecke 1980). The final larval weights right before pupation (Figure 8b) were all consistent with the known "critical weight" of *M. sexta* larvae, 5–7 grams (Davidowitz et al. 2003). This excludes the NCM 2 individual, which pupated at a final mass of 4.595 grams, about half a gram under the typical threshold. The final headcap widths also align with a previous study stating that all larvae will pupate in the following molt after reaching a headcap width of 5.1 mm (Nijhout 1975).

Consumption Patterns

Although growth in larval weight and length corresponds with a greater accumulated amount of frass, the number of frass pellets produced drops right before molting, which occurred on Day 3 of my experiment (Figure 8a). This observation led me to erroneously conclude that the larvae were either dying or pupating during the pilot run of my study, because it also coincides with a period of very limited movement and cessation in feeding. This period of inactivity has been observed before, and termed 'molt sleep', lasting for about 14–2.5 hours before each ecdysis event (Reinecke et al. 1980, Madden and Chamberlain 1985). After ecdysis, plant consumption and resulting frass production increased immediately.

Length over time is positively correlated with frass production overall, but there is some fluctuation due to measurement error introduced by the stretchiness of the soft larval body, as well as a tendency of the larvae to shrink slightly preceding a larval molt (Figure 8b). Head capsule measurements would have been a more reliable way to track growth, but these measurements are difficult to gather from live specimens. Larval weight (Figure 8c) also increased over time until dipping right before final pupation, with most of the weight gain occurring during the wandering

stage following ecdysis into the 5th instar. Frass production (Figure 8d) also shows a dramatic increase upon entering the wandering stage. My observations coincide with the knowledge that although the consumption increases with each day of the final instar (Madden and Chamberlain 1985), the size of the gut also increases, and therefore the food stays inside the gut for longer periods of time (Reynolds 1985).

The onset of the 'wandering stage' (around Day 6) exposed a flaw in my experimental design, as I was no longer able to determine which frass pellets came from which individuals. This could have been solved by using six different enclosures. I stopped collecting frass data on Day 8 of my experiment. However, I did take note of an overall cessation in consumption right before pupating, since the larvae all stopped defoliating the plants and became immobile closely preceding pupation on Days 14 and 15.

Wandering Stage

On Day 6, after I observed the rapid growth subsequent to ecdysis, I also noted an increase in wandering activity leading to nearly complete defoliation of the plants. By Day 7, the larvae were wandering so much that I was unable to track their plant position effectively, as they were defoliating the plants faster than I could add them. Because of this "unruly behavior", my experiment failed to determine a clear difference between the impacts of imposed and natural wandering patterns. However, I was able to conclude that the wandering stage is induced between the first and second days of the 5th instar (Days 5–6 in my study), and initiates a sharp increase in plant consumption, frass production, and weight increase. This timing coincides with previous studies of *M. sexta* behavior (Dominick and Truman 1984), which investigated the effect of scotophase (light versus dark periods) on wandering. The study determined that the onset of wandering is greatly influenced by the length of the night's dark photoperiod, which initiates the release of a "prothoracicotropic hormone". The following physiological responses included many fifth-instar developmental changes, including thickened cuticle production associated with increased wandering. The interactions between environmental factors and individual growth explain why the larvae in my experiment exhibited variable onsets of their wandering stages over Days 4–6.

There are many potential reasons as to why this wandering behavior exists in Lepidopteran larvae. For example, research in plant-insect interactions has shown that chemical plant defense

mechanisms are induced by the onset of herbivory, resulting in a spike in local toxin production that would require the larvae to cease feeding (Baldwin 2001). An increase in the toxin produced by *Nicotiana*, nicotine, could be a likely deterrent of *M. sexta* from feeding on an individual plant. This hypothesis is supported by another study on *M. sexta* feeding behavior, in which host plants that release more defensive secondary compounds induced shorter larval feeding bouts as well as increased wandering around the leaf surface (Parker et al. 2023). Additionally, increased wandering during the instar stage with the weakest immune system (Booth et al. 2015) could reduce the larvae's chances of being targeted by natural predators or parasitoids, such as the *Braconidae* wasp family that lays eggs inside the larval body (McFadden, 1968).

In addition to these possibilities, 'wandering' was likely initiated as a method to find more plant material to eat. As I would often arrive to find the caterpillars wandering around their enclosures with their plants fully defoliated, it is likely that some of their increased wandering was induced by the need for more food, which would not have occurred in a natural environment. The *M. sexta* larvae also generally consumed more plant material than expected. I found myself unable to replenish their food fast enough, as they would defoliate both plants within the 24 hours between data collections. In future iterations of this experiment, I would provide more plant material for the larvae, as well as more mature plant material instead of small starts.

How does feeding relate to wandering behavior?

Manduca sexta larvae exhibit an increase of wandering behavior upon ecdysial into the 5th and final instar. Although wandering is observed both in lab conditions and in nature (Madden and Chamberlain 1985), this experiment may have induced increased wandering as a result of the larvae needing more plant material than provided. In addition to the 'molt sleep' inactivity period before ecdysis, *M. sexta* larvae also showed decreased activity and consumption in the final days before pupation. My pre-pupal observations aligned with the critical weight of 5–7 grams before pupation, as well as other pre-pupal characteristics such as a pulsating dorsal vein (Davidowitz et al. 2003).

Upon understanding the complex relationship between caterpillar growth, wandering, and consumption, scientists can aim to develop agricultural practices that balance Solanaceous crop output with sustainable pest control. For instance, detailed knowledge of when Lepidopteran pests such as *Manduca sexta* are most active or vulnerable during their life stages can inform farmers

on when to time non-chemical pest management events such as irrigation flooding or release of biological controls such as nematodes, which are proven to be most effective in controlling fifth instar larvae (Sree & Varma 2015).

Limitations and Future Directions

Although my experiment was able to measure the growth rate and consumption patterns of the *M. sexta* individuals, there are many ways the design was limited. For example, the feeding stages of *M. sexta* are easily disrupted by my observations (Reinecke et al. 1980), which could lead to a lower consumption rate than what would occur without interference. Additionally, my collection of the frass likely included inaccuracies as the pellets in the early stages were extremely small and difficult to handle. Further experimentation would also ideally involve a higher amount of replication, and should keep each individual larva in a separate cage to avoid confusion during the wandering stage. The wandering experiment should include more dramatic differences between the NCM control and the ACM treatments to effectively detect the impacts of wandering on the caterpillars.

Future directions of this project could include a variety of different Solanaceous host plants to determine how host plant variation impacts larval development and wandering. Focusing on plant impact could allow for a deeper look into the effects of nicotine content on larval growth, as some potential *M. sexta* host plants (other species in the family Solanaceae, such as *N. tabaccum*) may be less favorable for larval success than host plants with less nicotine content, such as tomatoes or potatoes (genus *Solanum*). Further investigating host plant preference of *M. sexta* will also inform which Solanaceous species are most at risk of caterpillar infestation in modern agriculture.

Broader Implications

Studying the life history and wandering behavior of *M. sexta* is necessary for developing sustainable methods of integrative pest management in agriculture. Solanaceous crops (tobacco,

tomato, potato etc.) are a lucrative part of the world agricultural economy, and many industrial farms have historically resorted to environmentally damaging pesticides to control the incredibly destructive caterpillar pests (Madden and Chamberlin 1985). My study, as well as any further research resulting from it, could help develop a method of targeting the wandering tendencies of these larvae between plants by incorporating inter-cropping or using more barriers between individual plants or rows (Sree and Varma 2015). Less chemically destructive pest management efforts could also be timed to focus on targeting the larvae at a younger stage before the wandering stage is reached, or timed to occur during the pre-ecdysis 'molt sleep' larval inactivity. Controlling *M. sexta* in agriculture is of paramount importance due to the massive abundance of Solanaceous crops that are hosts of the caterpillar larvae. Additionally, studying insect feeding behavior will be vital to our understanding of how climate change tips the balance between host plants and insect herbivores, as caterpillar consumption and wandering are believed to increase with temperature (Clissold and Simpson 2015).

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APPENDIX A: No Controlled Movement Individuals

Table 1. NCM 1 data collection. Ecdysial molting observed on Day 4, pupation on Day 14.

Day	Length (mm)	Frass #	Total frass weight (g)	Larvae weight (g)	Plant position
0	31	-	-	0.573	A
1	36	10	0.021		A
2	39	21	0.076		B
3	37	3	0.097	1.217	B
4 - ecdysis	43	2	0.105		B
5	45	12	0.149		A
6	53	16	0.36	2.476	A
7	60	25	0.595	3.708	B
8	60	35	0.819	3.622	NCM 2 B
9	60	-	-	4.4	-
10	60	-	-	4.243	-
11	64	-	-	5.466	-
12	63	-	-	5.724	-
13	60	-	-	5.583	-
14 - pupation	60	-	-	5.046	-

Table 2. NCM 2 data collection. Ecdysial molting observed on Day 4, pupation on Day 14.

Day	Length (mm)	Frass #	Total frass weight (g)	Larvae weight (g)	Plant position
0	29	-	-	0.445	A
1	35	19	0.03		A
2	38	6	0.038		A
3	36	11	0.059	0.973	A
4 - ecdysial	38	15	0.079		B
5	48	16	0.179		A
6	55	19	0.364	2.324	NCM 1 B
7	60	39	-		B
8	60	15	-	3.404	A
9	64	-	-	3.925	-
10	65	-	-	3.822	-
11	69	-	-	5.224	-
12	62	-	-	4.6	-
13	70	-	-	4.496	-
14 - pupation	60	-	-	4.595	-

Table 3. NCM 3 data collection. Ecdysial molting observed on Day 4, pupation on Day 14.

Day	Length (mm)	Frass #	Total frass weight (g)	Larvae weight (g)	Plant position
0	28	-	-	0.664	A
1	35	25	0.045		A
2	38	1	0.046		A
3	38	0	0.046	1.154	A
4 - ecdysial	40	15	0.125		A
5	50	11	0.202		B
6	60	33	0.403	3.672	NCM2 B
7	62	23	0.542	4.643	NCM2 B
8	62	20	0.655	4.624	wall
9	62	-	-	4.575	-
10	62	-	-	5.19	-
11	67	-	-	6.031	-
12	63	-	-	5.867	-
13	70	-	-	6.7	-
14 - pupation	65	-	-	5.84	-

APPENDIX B: Artificially Controlled Movement Individuals

Table 4. ACM 1 data collection. Ecdysial molting observed on Day 4, pupation on Day 14.

Day	Length (mm)	Frass #	Total frass weight (g)	Larvae weight (g)	Plant position
0	26	-	-	0.474	A
1	34	23	0.086		A to B
2	40	15	0.086		B to A
3	40	5	0.08	1.185	A to B
4 - ecdysial	40	0	0.087		B to A
5	50	11	0.143		B to A
6	53	21	0.284	2.189	extra plant to new A
7	54	29	0.464		B to new A
8	58	31	0.779	3.441	wall
9	55	19	-	3.715	B
10	60	-	-	4.272	-
11	63	-	-	5.227	-
12	70	-	-	5.584	-
13	65	-	-	5.563	-
14 - pupation	65	-	-	5.135	-

Table 5. ACM 2 data collection. Ecdysial molting observed on Day 4, pupation on Day 14.

Day	Length (mm)	Frass #	Total frass weight (g)	Larvae weight (g)	Plant position
0	25	-	-	0.46	A
1	38	26	0.083		A to B
2	40	17	0.073		B to A
3	40	3	0.087	1.229	A to B
4 - ecdysial	40	0	0.088		B to A
5	42	12	0.156		A to B
6	49	19	0.341	2.386	B to A
7	52	20	0.589		B to new A
8	60	10	0.611		B
9	60	8	-	4.113	B
10	60	-	-	4.3	-
11	63	-	-	5.948	-
12	68	-	-	5.735	-
13 - pupation	70	-	-	6.072	-

Table 6. ACM 3 data collection. Ecdysial molting observed on Day 4, pupation on Day 14.

Day	Length (mm)	Frass #	Total frass weight (g)	Larvae weight (g)	Plant position
0	27	-	-	0.382	A
1	35	35	0.138		A to B
2	37	52	0.114		B to A
3	37	0	0.122	1.147	A to B
4 - ecdysial	37	4	0.126		B to A
5	41	10	0.17		A to B
6	42	17	0.275	1.663	ACM 2 A to A new
7	55	22	0.494		B to A
8	57	33	0.787	3.243	wall
9	65	28	-	4.814	wall
10	65	-	-	5.313	-
11	65	-	-	6.374	-
12	70	-	-	6.626	-
13	68	-	-	6.325	-
14 - pupation	75	-	-	6.5	-

APPENDIX C: Final pupae measurements

Table 7. Final pupae data.

Individual	Pupa Weight (g)	Pupa length (mm)	Final headcap width (mm)	Sex
NCM 1	Died before pupation	-	5.5	-
NCM 2	2.284	40	4.92	male
NCM 3	3.814	48	6.31	female
ACM 1	3.360	45	5.2	male
ACM 2	3.390	45	5.9	male
ACM 3	3.373	45	6.03	male