Influence of Resources on *Hermetia illucens* (Diptera: Stratiomyidae) Larval Development

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ABSTRACT Arthropod development can be used to determine the time of colonization of human remains to infer a minimum postmortem interval. The black soldier fly, *Hermetia illucens* L. (Diptera: Stratiomyidae) is native to North America and is unique in that its larvae can consume a wide range of decomposing organic material, including carrion. Larvae development was observed on six resources: control poultry feed, liver, manure, kitchen waste, fruits and vegetables, and fish rendering. Larvae fed manure were shorter, weighed less, and took longer to develop. Kitchen waste produced longer and heavier larvae, whereas larvae fed fish had almost 100% mortality. Black soldier flies can colonize human remains, which in many instances can coincide with food and organic wastes. Therefore, it is necessary to understand black soldier fly development on different food resources other than carrion tissue to properly estimate their age when recovered from human remains.

KEY WORDS forensic entomology, development time, food resource, minimum postmortem interval, waste management

Forensic entomologists use arthropod evidence collected from human remains to estimate the period of insect activity and infer time of colonization (Benecke 2001). The time of colonization is a portion of the postcolonization interval, which equates to the minimum postmortem interval (min-PMI) (Tomberlin et al. 2011a,b). Knowing the min-PMI could allow investigators to identify unknown victims, narrow down the list of suspects, and validate an alibi. The min-PMI can be estimated by determining the age of the oldest immature arthropod on the decedent. However, development of an arthropod is dependent on many factors.

The nutritional value of a resource can impact the development rate of an arthropod. For example, immature flies (Diptera) commonly occur on decomposing human remains, which represent an ephemeral resource on which arthropods must compete to acquire nutrition for development before the resource disappeared. When these nutritional requirements are not met, either because of quality or quantity, development rate can be affected. Ireland and Turner (2006) showed that the blow fly, *Calliphora vomitoria* (L.) (Diptera: Calliphoridae), reared at a high density on a limited quantity of pork liver, brain, or muscle, can have a developmental difference of up to 7.5 d depending on resource. Kaneshrajah and Turner (2004) showed *Calliphora vicina* (Robineau–Desvoidy) (Diptera: Calliphoridae) larvae fed liver developed up to 2 d slower than larvae reared on other experimental tissues, such as lung, kidney, heart, and brain. Similarly, *Lucilia sericata* (Meigen) (Diptera: Calliphoridae) developed at a faster rate, and were larger, when fed pork instead of beef (Clark et al. 2006). Knowing how resources impact the development of forensically relevant arthropods can assist in the accuracy of investigations.

The black soldier fly, *Hermetia illucens* (L.) (Diptera: Stratiomyidae), has been used as evidence in previous forensic cases (Lord et al. 1991, Turchetto et al. 2001, Pujol–Luz et al. 2008). Black soldier flies have a relatively long life cycle of ≈40 d, with the larval stage lasting ≈22–24 d at 27°C (Furman et al. 1959; Sheppard et al. 1994; Tomberlin et al. 2002a,b), making them useful for estimating min-PMI for human remains that have been decomposing for weeks instead of days. In addition, unlike blow flies that feed on only tissue, soldier flies can consume a wide range of decomposing organic material, from compost and fruits to manure (James 1935). All of the feeding is done in the larval stage, with larvae acquiring a large fat body to use as energy for pupation, adult survival, and reproduction (Tomberlin et al. 2002a,b). This ability to feed on a variety of organic materials is important in relation to forensics, as human remains colonized by black soldier flies are often located in areas contaminated with other potential food substrates. In such instances, understanding if the larvae were feeding on alternate food substrates as well as the decedent would be important to know so that accurate estimates of the min-PMI can be made.

The purpose of this study was to determine if black soldier flies can successfully develop on beef liver,

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which is a common resource used in blow fly development studies, as well as five other organic materials potentially encountered at a body recovery site. We hypothesize that development of the black soldier fly will differ across organic wastes provided.

Materials and Methods

A black soldier fly colony was founded with larvae obtained from Dr. Craig Sheppard (Georgia, via RECORP Inc, in Georgetown, ON, Canada). Adult black soldier flies were maintained in a colony by using methods previously described (Sheppard et al. 2002). Adult flies were released into a 3 by 3 by 6-m cage with an artificial plant as a lekking site (Tomberlin and Sheppard 2001) in a greenhouse, which provided natural sunlight and space for mating and oviposition (Tomberlin and Sheppard 2002). Additional light was provided by using 400-W high-pressure sodium lights to maintain a photoperiod of 14:10 (L:D) h cycle throughout the year (Zhang et al. 2010). Temperature was maintained between 20 and 35°C throughout the year. Adults were provided with water by using a misting system set for 1-min intervals, twice per day, which also maintained humidity above 30% relative humidity (RH). Oviposition sites were provided by taping 8 by 4 by 0.5-cm corrugated cardboard to the side of a 50 by 25 by 20-cm container filled with poultry feed (Agribrands Purina Canada Inc., Strathroy, Ontario, Canada), that is, 17% crude protein, 2.5% crude fat, and 4.5% crude fiber. The feed was saturated with water. Cardboard flutes containing eggs were placed in a 5 by 5 by 3-cm container covered with black Weedstop Professional Landscape Fabric (Quest Home and Garden, Mississauga, ON, Canada) and housed in a growth chamber at 28°C, 60 ± 10% RH, and 24 h light cycle. Larvae were placed into a 30 by 30 by 6.5-cm container covered with black fabric mulch and fed the control poultry feed diet ad libitum (70% moisture based on preliminary experiments, 10 g food with 21 ml water). Once they had fed for 4 d, 150 larvae were selected per treatment and placed into a separate 30 by 30 by 6.5-cm container covered with black fabric mulch and placed into a growth chamber at 28°C and 60 ± 10% RH.

There were six types of organic waste diets tested: 1) control poultry feed, 2) pig liver, 3) pig manure, 4) kitchen waste, 5) fruits and vegetables, and 6) fish rendering. Diets were specifically chosen to give the widest range of organic waste possible, ranging from an all-meat to an all-vegetable diet. Manure was obtained from a pig farm in Ridgetown, Ontario, Canada. Kitchen waste contains animal and plant matter and was supplied by restaurants in Windsor, Ontario, Canada. Fruits and vegetables were supplied by grocery stores in Windsor, Ontario, Canada. Fish renderings were supplied from a fishery in Kingsville, Ontario, Canada. To keep the diets as consistent as possible, a vast quantity of each type of waste was individually ground to create a homogeneous diet mixture with moisture contents ranging from 71.3 to 95.6% between the diets (Table 2). The diets were packaged in sealable sandwich bags (16.5 by 15 cm) and kept frozen until needed throughout the experiment. Frozen diet was thawed 24 h before use. The effects of freezing and thawing diets were unknown.

Each of the six diet treatments started with 150 4-d-old larvae and 6 g of waste. Every day, the old waste (i.e., diet) was removed and replaced with fresh diet. When the larvae had reduced the diet by 25% in weight, the daily amount of diet provided to the larvae was increased by 5 g. Every day, the number of surviving larvae was counted (as a measure of mortality) and stage of development (larval, wandering, pupal, or adult) was recorded. These larvae were treated as "handled" larvae. Ten samples, three larvae per sample, were used to measure larvae weight. A sample of 10 larvae was used to record the length—taken as the longest measure when extended. Weight and length data collection ceased when 40% of larvae reached the wandering stage, indicated by the completely black color of the larvae and lack of feeding (Tomberlin et al. 2002a). Mortality and stage of development records were continuously recorded until all individuals either emerged as adults or died.

To correct for any potential effect of daily handling on the development of the flies, a second set of control replicates was conducted concurrently, with larvae reared on each diet exactly as previously described; however, the larvae were not measured daily, nor was diet removed. These control larvae were labeled "unhandled" larvae. Fresh diet was added at the same rate as for the handled treatments. When 40% of the larvae in the previous "handled" treatment reached the wandering stage, mortality, weight, and length were measured, as previously.

Six replicates were conducted, with each replicate originating from one generation of black soldier fly. The order of diet and larvae sampled was in a Latin square pattern. Waste samples were sent to Maxxam Analytics, Mississauga, Ontario, Canada, for nutritional analysis.

Statistical Analysis. A one-way analysis of variance (ANOVA) was used to test for differences in development time with regard to minimum, median, and maximum time to reach the wandering, pupal, and adult stages of development with a least significant distance (LSD) means separation ($P < 0.05$). Statistical analysis was performed on transformed data; however, results shown are original data. Minimum time taken to reach wandering stage and the minimum and median time to reach adult emergence were log-base-10-transformed to satisfy normality and equal variances. Similarly, minimum pupal time was transformed by $(1/\times)$. A nonparametric ANOVA (Kruskal–Wallis independent samples) was performed on development time data, except for the median time taken to reach wandering stage, because it could not be transformed.

To test the effects of handling on development, a nonparametric ANOVA (Kruskal–Wallis independent samples) test was performed to see if there was a difference between the "handled" and "unhandled" counterparts across treatments for development rate,

Table 1. Min., median, and max time taken to reach prepupal, pupal, and adult stages of development (mean days ± SE)

Diet	Wandering	Pupae	Adult
	A. Min. (mean days ± SE)		
Feed	20.17 ± 0.477C	31.67 ± 1.174B	38.83 ± 1.276D
Liver	19.17 ± 0.601C	31.83 ± 1.195B	40.17 ± 1.447CD
Manure	25 ± 0.516A	38.17 ± 1.493A	45 ± 1.211B
Kitchen waste	20.17 ± 0.543C	32.83 ± 1.078B	40 ± 1.155CD
Fruits and vegetables	21.67 ± 0.333B	33.83 ± 0.477B	42.67 ± 1.054BC
Fish	19.83 ± 0.543C	42.60 ± 2.891A	55 ± 0A
Test statistic	$N = 36$; $F_{5,30} = 17.4$; $P < 0.001$	$N = 36$; $F_{5,29} = 7.614$; $P < 0.001$	$N = 32$; $F_{5,26} = 9.582$; $P < 0.001$
	B. Median (mean days ± SE)		
Feed	23 ± 0.632BC	35.17 ± 1.276C	43.17 ± 1.302C
Liver	22.50 ± 0.719C	34.17 ± 2.072C	42.17 ± 1.887C
Manure	34 ± 1.390A	48.67 ± 2.472A	55.33 ± 2.140A
Kitchen waste	23.83 ± 0.401BC	35.33 ± 1.282C	43.33 ± 1.256C
Fruits and vegetables	28.67 ± 0.760AB	42.17 ± 1.621B	49.33 ± 1.256B
Fish	26.50 ± 0.992ABC	43.40 ± 2.857AB	55 ± 0AB
Test statistic	$N = 36$; $\chi^2 = 27.709$; $P < 0.001$	$N = 35$; $F_{5,29} = 8.969$; $P < 0.001$	$N = 32$; $F_{5,26} = 11.502$; $P < 0.001$
	C. Max (mean days ± SE)		
Feed	30 ± 0.856D	53.83 ± 3.591B	63 ± 4.082BC
Liver	32.17 ± 2.167CD	39.33 ± 1.476C	49 ± 2.066D
Manure	45.67 ± 1.358A	64.50 ± 4.169A	73.50 ± 3.128A
Kitchen waste	32.50 ± 1.875CD	45.50 ± 3.528BC	57.17 ± 4.722D
Fruits and vegetables	40.33 ± 1.542AB	54.67 ± 3.095B	68.50 ± 3.294AB
Fish	36 ± 2.745BC	46.60 ± 3.614BC	60 ± 5.00CD
Test statistic	$N = 36$; $F_{5,30} = 10.150$; $P < 0.001$	$N = 35$; $F_{5,29} = 7.088$; $P < 0.001$	$N = 32$; $F_{5,26} = 5.859$; $P < 0.001$

Within-column and -section means followed by the same letter are not significantly different ($P > 0.05$).

with an LSD-corrected post hoc ($P < 0.05$) to test the effects of handling with regard to length and weight measurements.

The rate of larvae weight gain and rate of larvae length increase were calculated by creating a scatter plot for each type of waste treatment of each replicate and then excluding data that appeared on the upper and lower horizontal asymptotes before conducting a linear regression. For the rate of larvae weight gain per day, a nonparametric ANOVA was performed (independent-samples Kruskal–Wallis), whereas larvae length increase per day was analyzed by using an ANOVA with an LSD-corrected post hoc. The final larvae weight and length were determined by measurements taken when 40% of larvae from the handled treatment had reached the wandering stage and analyzed by using ANOVA with an LSD ($P < 0.05$)-corrected post hoc means separation.

Mortality was recorded from each waste diet at the wandering, pupal, and adult stages of development. For the wandering stage, these data were Log_{10} transformed for normality and equal variances, then analyzed by using an ANOVA with an LSD-corrected post hoc means separation ($P < 0.05$); however, results are presented as untransformed data. A nonparametric ANOVA (Kruskal–Wallis independent samples) test was used to analyze mortality at the pupal and adult stages with corrected all pairwise comparisons post hoc. All statistical analyses were performed by PASW Statistics 18.0 (SPSS, Inc., Chicago, IL).

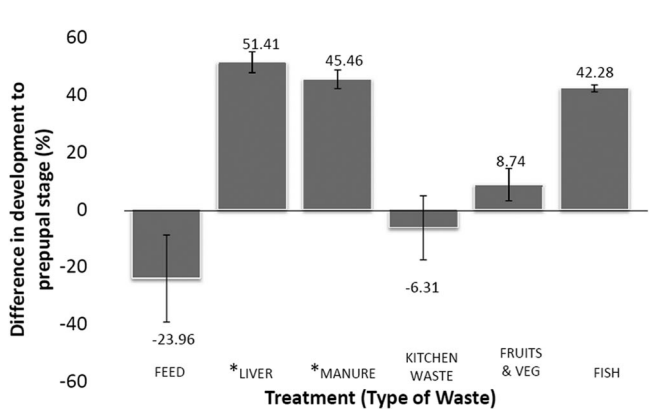

Fig. 1. Mean (±SE) difference in percentage of larvae that reached the prepupal stage at the end of the experiment between handled and unhandled larvae for six different waste treatments. The baseline zero represents unhandled larvae, whereas the bars represent handled larvae. Treatments with an asterisk represent a significant difference between handled and their unhandled counterparts ($N = 72$; $\chi^2 = 54.465$; df = 11; $P < 0.001$).

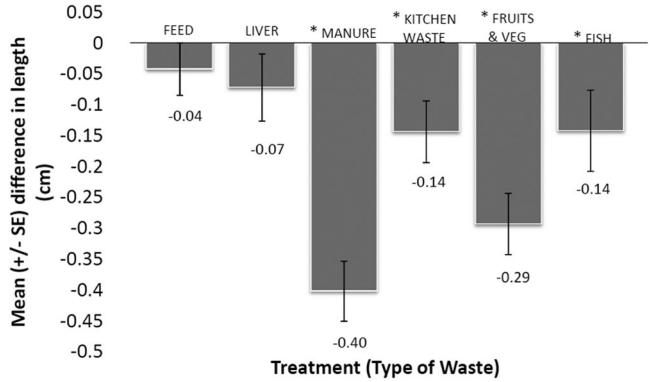

Fig. 2. Mean (±SE) difference in final larvae lengths between handled and unhandled larvae. The zero baseline represents unhandled larvae lengths, whereas bars represent handled larvae lengths; all larvae had a shorter average length when handled than when unhandled. Waste treatments with an asterisk represent a significant difference between handled larvae and their unhandled counterparts ($N = 72$; $F_{11,60} = 13.388$; $P < 0.001$).

Results

There was a significant difference in development time for the minimum, median, and maximum days it takes for larvae to reach wandering, pupal, and adult stages of development because of the different waste diets (Table 1). The time taken for larvae fed liver to reach the wandering stage was not significantly different from those fed on control feed, kitchen waste, or fish. Larvae reared on manure generally took the longest to reach the wandering stage, followed by fruits and vegetables, and then the remaining four other diets (minimum $N = 36$, $F_{5,30} = 17.4$, $P < 0.001$; median $N = 36$, $\chi^2 = 27.709$, df = 5; $P < 0.001$; maximum $N = 36$, $F_{5,30} = 10.150$, $P < 0.001$). When measuring time to develop to the pupal stage, larvae reared on liver had one of the shortest developmental times that was similar to kitchen waste. Larvae reared on manure once again generally had the longest time to reach the pupal stage, followed by fish and fruits and vegetables (Table 1: $N = 35$; $F_{5,29} = 8.969$; $P < 0.001$).

When measuring time to adult emergence, larvae reared on liver once again had the shortest maximum time needed to reach adult emergence. These results did not significantly differ from those larvae provided the control feed or kitchen waste. Larvae reared on manure and fish had the highest minimum ($N = 32$; $F_{5,26} = 9.582$; $P < 0.001$) and median ($N = 32$; $F_{5,26} = 11.502$; $P < 0.001$) times, followed by fruits and vegetables (Table 1). The longest maximum time to adult emergence was for those reared on manure, followed by fruits and vegetables ($N = 32$; $F_{5,26} = 5.859$; $P < 0.001$). Manure-fed larvae, along with fruits and vegetables as well as fish, generally had, overall, longer developmental times taken to reach wandering, pupae, and adult stages than those fed the other waste diets, whereas those fed liver, along with feed and kitchen waste, generally had the shortest developmental times.

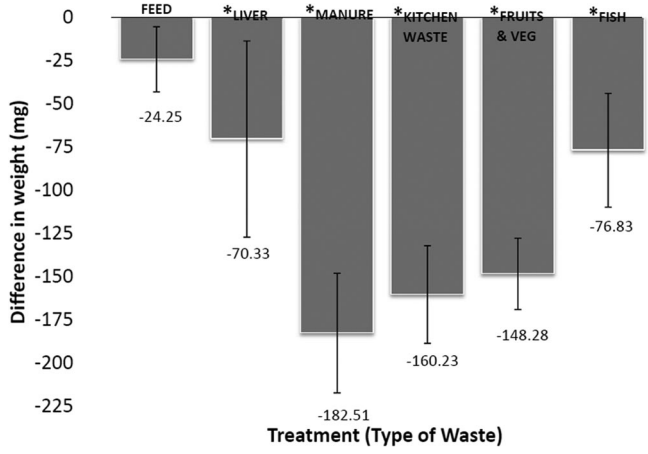

Fig. 3. Mean (±SE) difference in final larvae weights between handled and unhandled larvae. Zero baseline represents unhandled larvae weights, whereas bars represent handled larvae weights; all larvae had a lighter average weight when handled than when unhandled. Waste treatments with an asterisk represent a significant difference between handled and their unhandled counterparts ($N = 72$; $F_{11,60} = 14.261$; $P < 0.001$).

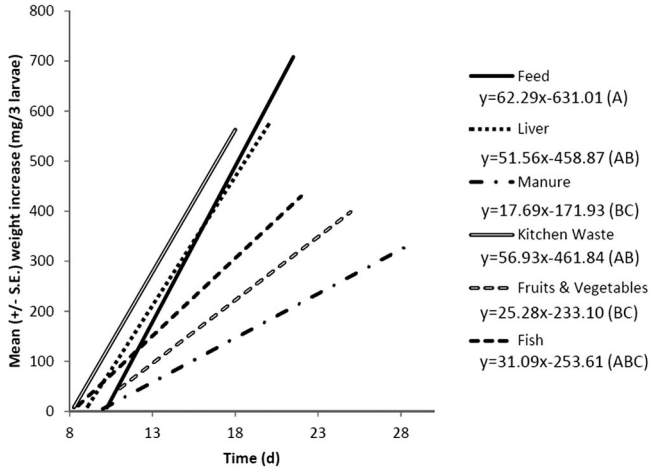

Fig. 4. Mean (±SE) rate of larvae weight gain per day for larvae reared on six different types of waste diets ($N = 36$; $\chi^2 = 28.589$; df = 5; $P < 0.001$). Means followed by the same letter are not statistically different ($P > 0.05$).

Handling larvae daily had a significant effect in their development rate as well as their final length and weight. Approximately 51% more of the handled larvae fed liver reached the wandering stage than the unhandled counterpart. Similar results were recorded for those fed manure, with 45% more reaching the wandering stage than the unhandled counterpart (Fig. 1; $N = 72$; $\chi^2 = 54.465$; df = 11; $P < 0.001$). With the exception of larvae reared on liver and poultry feed, those reared on all other waste diets were shorter in length when handled than when unhandled (Fig. 2; $N = 72$, $F_{11,60} = 13.388$; $P < 0.001$). Handling resulted in decreased weight for larvae reared on all of the waste diets, except poultry feed (Fig. 3; $N = 72$; $F_{11,60} = 14.261$; $P < 0.001$). There was a difference in rate of larval weight gain per day (Fig. 4; $N = 36$; $\chi^2 = 28.589$; df = 5; $P < 0.001$) and rate of larval length increase per day (Fig. 5; $N = 36$; $F_{5,30} = 14.389$; $P < 0.001$) for larvae reared on different waste diets. Larvae generally had a higher rate of weight gain and length increase in poultry feed (62.29 mg/3 larvae/d; 0.12 cm/d), kitchen waste (56.93 mg/3 larvae/d; 0.14 cm/d), and liver (51.56 mg/3 larvae/d; 0.13 cm/d). Larvae fed manure had the slowest rate for both weight gain (17.69 mg/3 larvae/d; 0.07 cm/d) and length increase. There was also a difference in both final larval weight (Fig. 6; $N = 36$; $F_{5,30} = 14.410$; $P < 0.001$) and length (Fig. 7; $N = 36$; $F_{5,30} = 10.992$; $P < 0.001$). Similar to the per-day increases in weight and length, the final weights and lengths of larvae were greatest on poultry feed (552.96 mg/3 larvae; 2.048 cm) and kitchen waste (519.65 mg/3 larvae; 2.082 cm), followed by liver (473 mg/3 larvae; 1.95 cm). Larvae reared on manure once again had the lowest final weight (339 mg/3 larvae) and length measurements (1.76 cm).

The number of larvae successfully reaching each stage of development differed when reared on the different waste diets: wandering stage (Fig. 8; $N = 36$; $F_{5,30} = 7.322$;

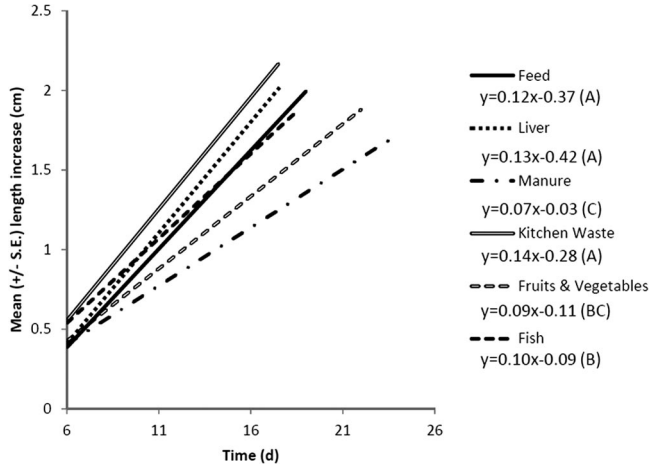

Fig. 5. Mean (±SE) rate of larvae length increase per day for larvae reared on six different waste treatments ($N = 36$; $F_{5,30} = 14.38$; $P < 0.001$). Means followed by the same letter are not statistically different ($P > 0.05$).

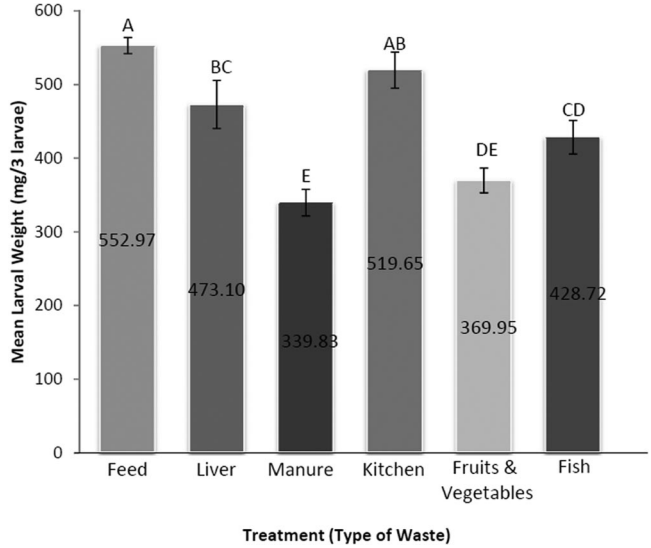

Fig. 6. Final mean (±SE) larvae weight for handled larvae reared on six different types of waste diets ($N = 36$; $F_{5,30} = 14.410$; $P < 0.001$). The mean value is given in the center of each bar. Bars topped with the same letter are not statistically different ($P > 0.05$).

$P < 0.001$), pupal stage (Fig. 9; $N = 36$; $\chi^2 = 27.974$; df = 5; $P < 0.001$), and adult stage (Fig. 10; $N = 37$; $\chi^2 = 27.524$; df = 5; $P < 0.001$). The highest overall mortality was observed from those reared on fish renderings, with 52.77% mortality for the wandering stage, 98.55% for the pupal stage, and 99.66% for the adult stage. Larvae reared on liver had the next highest mortality of 42.77, 79.11, and 84.55% at wandering, pupal, and adult stages, respectively. The third highest rates of mortality occurred on kitchen waste, at 53.33, 58.77, and 61.22%, respectively, for the three developmental stages.

Nutritional analyses show that kitchen waste, liver, and fish generally have a higher nutritional content than the control feed (Table 2). Kitchen waste contains the highest energy, carbohydrate, and calorie content, and also has relatively high fat and protein content, compared with the other diets. Liver has the highest protein content by far and relatively high energy, calorie, fat, and carbohydrate content. Fish contains the highest fat content and relatively high energy, protein, and calorie content. Carbohydrate content in fish is undetectable. Manure and fruits and

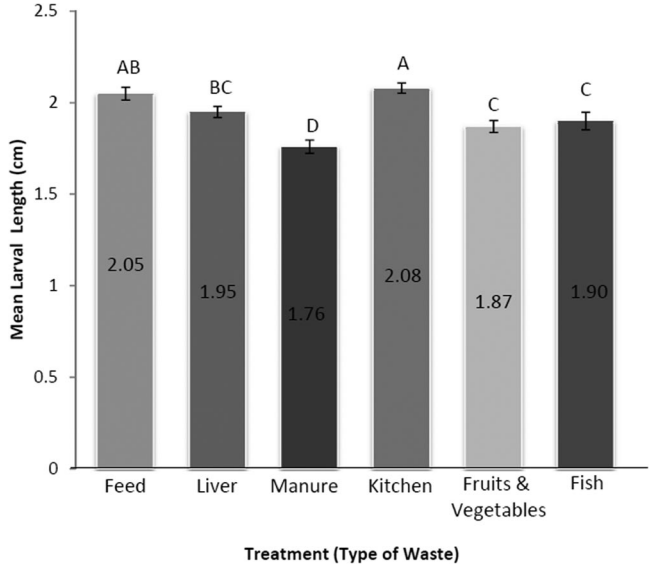

Fig. 7. Final mean (±SE) larvae length for handled larvae fed six different types of waste ($N = 36$; $F_{5,30} = 10.992$; $P < 0.001$). The mean value is given in the center of each bar. Bars topped with the same letter are not statistically different ($P > 0.05$).

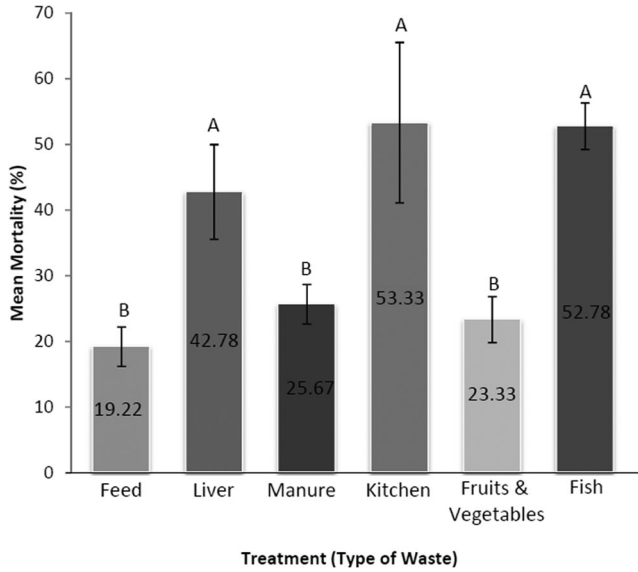

Fig. 8. Mean (±SE) percent mortality for wandering stage of development for larvae fed on six different types of waste diets ($N = 36$; $F_{5,30} = 7.322$; $P < 0.001$). The mean value is given in the center of each bar. Bars topped with the same letter are not statistically different ($P > 0.05$).

vegetable waste have lower nutritional content than control feed. Fruits and vegetables have the lowest energy, protein, calorie, and fat content, and fairly low carbohydrate content. Manure also has low energy, carbohydrate, protein, calorie, and fat content.

Discussion

As black soldier fly adults do not eat, they need to accumulate a large fat body in their larval stage to maintain adult survival (Sheppard et al. 1994). As a result, it is reasonable to conclude that larvae will take longer to acquire their fat body and take longer to complete their development if the only resources they have are diets that are low in fat. Larvae reared on kitchen waste, which had the second highest fat content after fish, developed just as fast as, or even faster in some instances than, control feed. Overall, larvae grown on manure, fruits and vegetables, and fish had longer developmental times before reaching wandering, pupal, and adult stages.

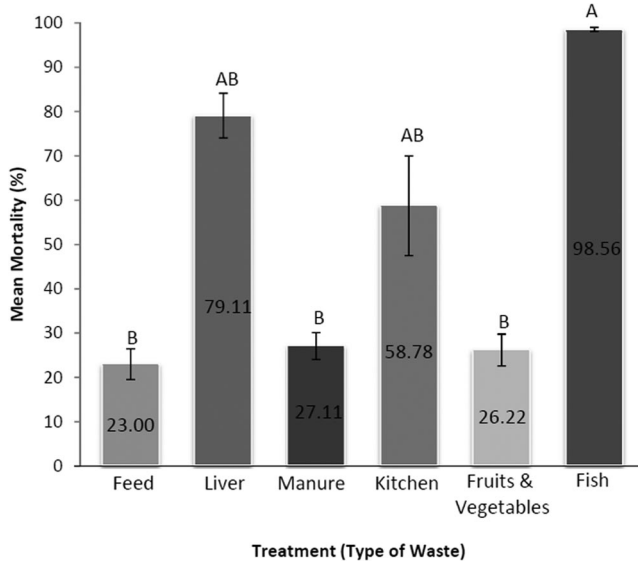

Fig. 9. Mean (±SE) percent mortality for pupal stage of development for larvae fed on six different types of waste diets ($N = 36$; $\chi^2 = 27.974$; df = 5; $P < 0.001$). The mean value is given in the center of each bar. Bars topped with the same letter are not statistically different ($P > 0.05$).

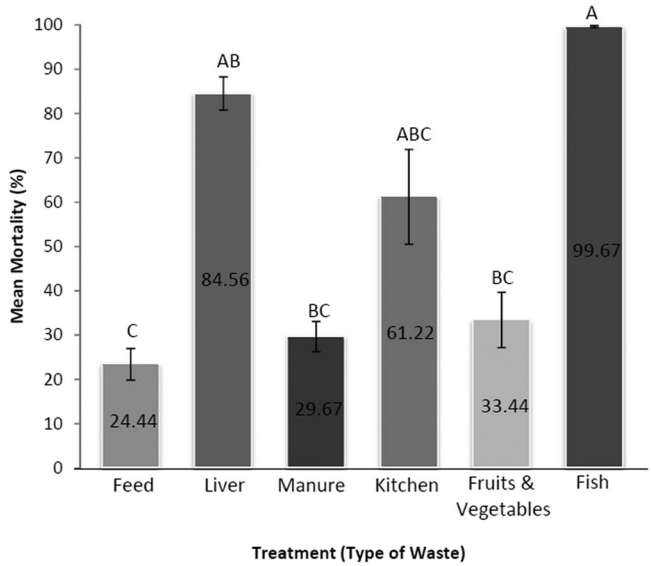

Fig. 10. Mean (±SE) percent mortality for adult stage of development for larvae fed on six different types of waste diets ($N = 36$; $\chi^2 = 27.524$; df = 5; $P < 0.001$). The mean value is given in the center of each bar. Bars topped with the same letter are not statistically different ($P > 0.05$).

Larvae reared on manure and fruits and vegetables may have performed poorly because these foods had lower energy (as measured by calories) and protein, as well as the lowest fat content per unit of dry matter. In addition to their low fat content, manure and fruits and vegetables may have caused larvae to have longer developmental times because these diets are lower in protein. Wissinger et al. (2004) showed that providing animal food resources accelerated the development of caddisfly larvae *Asynarchus nigriculus* (Banks) (Trichoptera: Limnephilidae), when compared with a detritivore's diet, by as much as 7–10 d. The availability of balanced amounts of calories, fat, and protein may also explain why larvae developing on liver had the fastest developmental times overall, followed by kitchen waste. Whereas liver is an animal product, kitchen waste is composed of protein from meat products. Kitchen waste also has the highest calorie content and the most balanced content of fat and protein.

Although it had the highest fat content, a fish diet did not cause larvae to have the fastest development time for adult emergence because fish waste may contain *too much* fat. Too much fat could be detrimental for black soldier flies; larvae may have a hard time metabolizing too much fat during the metamorphosis processes into an adult fly (Ujvari et al. 2009). Another reason why soldier fly larvae may have a difficult time developing on fish is because fish waste may contain heavy metal contamination (Gregory et al. 2005), which was not measured in this study. According to Bohadorani and Hilliker (2009), *Drosophila melanogaster* (Meigen) (Diptera: Drosophilidae) did not oviposit on mediums containing high concentrations of heavy metals because the metals would inhibit the growth of their offspring. The larvae reared on fish had almost 100% mortality at the adult stage, meaning that almost all larvae reared on fish will die before they reach the adult stage, possibly because of bioaccumulation of heavy metal contamination in the fish food web. Fish can take up the heavy metals in the water through their diet or absorption from the water (Gregory et al. 2005). Gregory et al. (2005) sampled water,

Table 2. Nutritional analysis of six different types of waste treatment diets performed by Maxxam Analytics

Quantity/100 g	Feed	Liver	Manure	Kitchen waste	Fruits and vegetables	Fish
KJ	322	468	129	583	68.5	380
Ash (g)	6.3	1.4	3	0.9	0.4	3.6
Calories	77	112	31	139	16.5	91
Proteins (g)	4.47	19.41	2.38	5.86	0.9	9.05
Fats (g)	0.626	3.25	0.148	5.62	0.065	6.55
Carbohydrates (g)	13.3	1.2	5	16.3	3.05	NA
Moisture (g)	75.2	74.7	89.5	71.3	95.6	81.9
Cis-monosaturated fatty acids (g)	0.217	0.608	0.013	2.33	0.0085	2.95
Cis-polyunsaturated fatty acids (g)	0.095	1.17	0.004	1.2	0.03	1.56
Trans-fatty acids	0.034	0.025	0.014	0.123	<0.001	0.079
Omega-3 polyunsaturated fatty acids	0.005	0.139	<0.001	0.099	0.014	1.02
Omega-6 polyunsaturated fatty acids	0.09	1.03	0.003	1.1	0.016	0.544

sediment, and fish samples from 12 lakes in Sudbury, Ontario, Canada, which is near the collection site of the fish used in this study (fisheries in Kingsville, Ontario, Canada). They discovered that some lakes had metal concentrations that violate the Ontario Provincial Water Quality Objectives by as much as an order of magnitude, and fish residing in these lakes contained high levels of cadmium, copper, nickel, and zinc. Results for larvae reared on the six different organic wastes revealed that the resource the larvae are reared on can have a significant effect on larval mortality. Diets that have the highest mortality included kitchen waste, fish, and liver.

Black soldier fly larvae were able to successfully consume the six organic waste diets tested: 1) control poultry feed, 2) pig liver, 3) pig manure, 4) kitchen waste, 5) fruits and vegetables, and 6) fish rendering. Type of diet significantly affected black soldier flies' ability to develop with respect to developmental rate, size of larvae, and mortality. It is important to know the effect resource has on development of black soldier flies in forensic entomology, where larval age is used to estimate min-PMI. Knowing how black soldier flies develop on organic waste is beneficial in cases where a body may be found in landfills, dumpsters, or unkempt homes. Not knowing developmental variations that larvae undergo based on diet can lead to inaccurate measures of larval age, which in turn leads to inaccurate estimates in min-PMI.

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References Cited

Bahadorani, S., and A. Hilliker. 2009. Biological and behavioural effects of heavy metals in *Drosophila melanogaster* adults and larvae. J. Insect Behav. 22: 399–411.

Benecke, M. 2001. A brief history of forensic entomology. Forensic Sci. Int. 120: 2–14.

Clark, K., L. Evans, and R. Wall. 2006. Growth rates of the blowfly, *Lucili sericata*, on different body tissues. Forensic Sci. Int. 156: 145–149.

Furman, D. P., R. D. Young, and E. P. Catts. 1959. *Hermetia illucens* (Linnaeus) as a factor in the natural control of *Musca domestica* Linnaeus. J. Econ. Entomol. 52: 917–921.

Gregory, P. G., J. W. Rajotte, and P. Couture. 2005. Effects of industrial metals on wild fish populations along a metal contamination gradient. Ecotoxicol. Environ. Saf. 61: 287–312.

Ireland, S., and B. Turner. 2006. The effects of larval crowding and food type on the size and development of the blowfly, *Calliphora vomitoria.* Forensic Sci. Int. 15: 175–181.

James, M. T. 1935. The genus *Hermetia* in the United States (Diptera: Stratiomyidae). Bull. Brooklyn Entomol. Soc. 30: 165–170.

Kaneshrajah, G., and B. Turner. 2004. *Callliphora vicina* larvae grow at different rates on different body tissues. Int. J. Legal Med. 118: 242–244.

Lord, W. D., M. L. Goff, T. R. Adkins, and N. H. Haskell. 1991. The black soldier fly *Hermetia illucens* (Diptera: Stratiomyidae) as a potential measure of human postmortem interval: observations and case histories. J. Forensic Sci. 39: 215–222.

Pujol–Luz, J. R., P.A.C. Francez, A. Ururahy–Rodrigues, and R. Constantino. 2008. The black soldier-fly, *Hermetia illucens* (Diptera, Stratiomyidae), used to estimate the postmortem interval in a case in Amapa State, Brazil. J. Forensic Sci. 53: 476–478.

Sheppard, D. C., G. L. Newton, and S. A. Thompson. 1994. A value added manure management system using the black soldier fly. Bioresour. Technol. 50: 275–279.

Sheppard, D. C., J. K. Tomberlin, J. A. Joyce, B. C. Kiser, and S. M. Sumner. 2002. Rearing methods for the black soldier fly (Diptera: Stratiomyidae). J. Med. Entomol. 39: 695–698.

Tomberlin, J. K., and D. C. Sheppard. 2001. Lekking behavior of the black soldier fly (Diptera: Stratiomyidae). Fla. Entomol. 84: 729–730.

Tomberlin, J. K., and D. C. Sheppard. 2002. Factors influencing mating and oviposition of black soldier flies (Diptera: Stratiomyidae) in a colony. J. Entomol. Sci. 37: 345–352.

Tomberlin, J. K., D. C. Sheppard, and J. A. Joyce. 2002a. Selected life-history traits of black soldier flies (Diptera: Stratiomyidae) reared on three artificial diets. Ann. Entomol. Soc. Am. 95: 379–386.

Tomberlin, J. K., D. C. Sheppard, and J. A. Joyce. 2002b. Susceptibility of black soldier fly (Diptera: Stratiomyidae) larvae and adults to four insecticides. J. Econ. Entomol. 95: 598–602.

Tomberlin, J. K., R. Mohr, M. E. Benbow, A. M. Tarone, and S. Vanlaerhoven. 2011a. A roadmap for bridging basic and applied research in forensic entomology. Annu. Rev. Entomol. 56: 401–421.

Tomberlin, J. K., M. E. Benbow, A. M. Tarone, and R. M. Mohr. 2011b. Basic research in evolution and ecology enhances forensics. Trends Ecol. Evol. 26: 53–55.

Turchetto, M., S. Lafisca, and G. Constantini. 2001. Postmortem interval (PMI) determined by study sarcophagous biocenoses: three cases from the province of Venice (Italy). Forensic Sci. Int. 120: 28–31.

Ujvari, B., J. F. Wallman, T. Madsen, M. Whelan, and A. J. Hulbert. 2009. Experimental studies of blowfly (*Calliphora stygia*) longevity: a little dietary fat is beneficial but too much is detrimental. Comp. Biochem. Physiol. A Mol. Integr. Physiol. 154: 383–388.

Wissinger, S., J. Steinmetz, and S. Alexander. 2004. Larval cannibalism, time constraints, and adult fitness in caddisflies that inhabit temporary wetlands. Oecologia 138: 39–47.

Zhang, J., L. Huang, J. He, J. K. Tomberlin, J. Li, C. Lei, M. Sun, Z. Liu, and Z. Yu. 2010. An artificial light source influences mating and oviposition of black soldier flies, *Hermetia illucens.* J. Insect Sci. 10: 202. (insectscience.org/10.202)

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