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Dietary Factors that Influences the Life History of Aedes aegypti

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A thesis submitted in partial fulfillment of the requirements for a baccalaureate degree in Biochemistry and Molecular Biology with honors in Area of Honors Biochemistry and Molecular Biology

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ABSTRACT

Aedes aegypti plays a significant role in the transmission of vector borne diseases due to its feeding behavior. While there is data that shows that this species of mosquito can feed on a variety of organisms, Ae. aegypti seem to prefer feeding on humans. Understanding factors that influence this feeding behavior will play a major role in better understanding disease-dynamics and also assist in developing artificial diets for raising mosquitoes in a laboratory. In the following report, two preservation methods – freeze drying and freeze thawing – of blood were tested to analyze the effects on life history characteristics of A. aegypti. The data showed that there were no associated risks with using freeze-thawing methods for maintenance of blood compared to using refrigerated whole blood. Freeze-drying blood had varying results between the two experiments done - one of the experiments showed a negative impact of feeding on freeze-dried blood on the survival probability and life span of the mosquitoes. Other studies went on to evaluate how certain amino acids, specifically supplementation of isoleucine and methionine, affected the average number of eggs a mosquito can lay and the average life span. When methionine was supplemented, there were negative effects on the survival probability of the mosquitoes that was both dosedependent and present only if sugar was available. The data also suggest that individual supplementation may have negative effects on average life span, while combinatorial supplementation of amino acids may decrease those effects.

TABLE OF CONTENTS

LIST OF FIGURES	iii
LIST OF TABLES	iv
ACKNOWLEDGEMENTS	V
ABSTRACT	i
Significance of This Research	1
Methods	5
Fresh, Frozen and Freeze-Dried Blood Experiments Effects of Methionine Supplementation on Aedes aegypti Life History Traits Colony Maintenance and Larval Raising Effects of Methionine and Isoleucine Supplementation Statistical Analyses	5 6 6
Results	
Effects of Blood Preservation on Life Span and Total Number of Eggs Laid	8
Supplementation of Amino Acids Results	10
The Effects that the Availability of Sugar Had on Survival Probability Supplementing with Methionine and Isoleucine	
Discussion and Conclusions	17
BIBLIOGRAPHY	22
ACADEMIC VITA	25

LIST OF FIGURES

Figure 1. The effects that various blood preservation methods have on the clutch size and survival of <i>Aedes aegypti</i> . The average total number of eggs laid (A), the average number of days alive (B), and the survival probability (C) of the mosquitoes were compared by each experimental treatment of blood preservation type provided to the mosquitoes. Bar-and-whisker plot were used. Lines extending from the plot represents the 95% confidence interval. Any dots in the data represents outliers
Figure 2. A second biological replicate of the use of various blood preservation methods. The average total number of eggs laid (A), the average life span (B), and the survival probability (C) are presented to determine the effects that feeding mosquitoes blood that has undergone one of the preservation methods listed in the methods
Figure 3 The effects of methionine supplementation to whole blood and subsequent feeding mosquitoes of a 10% w/v sucrose solution <i>ad libitum</i> . The average clutch size (A), the average number of days alive (B), and the survival probability (C) were evaluated to determine the effects such feeding would have on the mosquitoes
Figure 4. The effects of methionine supplementation to whole blood in the absence of 10% w/v sucrose availability. The average clutch size (A), the average number of days alive (B), and the survival probability (C) evaluated to determine the effects such feeding would have on the mosquitoes
Figure 5. Evaluation of survival probability and average life span of various methionine- supplemented blood fed in the presence and absence of a 10% w/v sucrose solution. In the presence and absence of sugar the average life span and survival probability of un- supplemented (A and D), low-methionine supplemented (B and E) and high methionine supplemented (C and F) are provided
Figure 6. Evaluation of various life history characteristics when offered various combinatorial amino acid supplementation and doses. The average number of eggs laid (A), the average larvae of proportion hatched (B),the survival probability (C) and the average life span (D) were used as proxies for evaluating combinatorial and individual supplementation of isoleucine and methionine

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Introduction

Significance of This Research

Mosquitoes serve as vectors for a wide range of pathogens. A major driving force behind the ability of mosquitoes to spread infections in people is due to their feeding behaviors (1,2). Some mosquitoes feed mainly on birds or mammals, while others have a wide range of hosts they can feed on. Understanding the driving characteristics behind such feeding behaviors is important to understanding disease-dynamics and transmissions.

The use of mosquito rearing facilities is integral to mosquito research and disease prevention programs. However, the cost of blood can be high in some areas, causing researchers and mass rearing facilities to become dependent on human volunteers, live animals, or slaughter house waste for blood feedings. Development of an artificial diet for mosquitoes provides more control when manipulating the diets of mosquitoes and would help ensure that the diet is uniform among their experiments. Additionally, an artificial diet that is made of ingredients that do not need to be refrigerated would allow researchers to rear mosquitoes in low resource settings, where refrigeration for blood meals may not be a viable option.

Current Lapses in Knowledge

Feeding behaviors of mosquitoes play a vital role in transmission of various viruses. In studies on *Culex pipiens*, it was demonstrated that these mosquitoes changed their feeding habits from favoring birds to humans as the seasons changed and birds migrated south which lead to

seasonal pattern in vector-borne diseases in humans(1,2). Not only has the behavior been demonstrated in C. pipiens, it has also been demonstrated in C. nigripalus vector for St. Louis encephalitis virus (2). Research has also shown that factors such as temperature greatly affect the life span and reproductive capacity of the mosquitoes and their ability to spread abroviruses (3). In Aedes aegypti, feeding behaviors also change with the availability of sugar, amino acids concentrations in blood, and toxic secondary metabolites (4). Understanding the factors that drive the feeding behaviors in these mosquitoes can provide a new level of understanding of disease transmission by identifying factors that influence feeding behavior. Understanding the dietary requirements of Ae. aegypti can also be used to improve feedings in mass rearing facilities on nonhuman animals. Current vector control programs have been using the bacterial endosymbiont Wolbachia pipentis as means of controlling the spread of pathogens due to the decreased replication of pathogens in Ae. aegypti infected with the endosymbiont (5,6). However, some cost to egg viability was exhibited in the Wolbachia infected mosquitoes (7). Improving blood feeding rate or supplementing blood meals with ingredients that improve egg yields could make Wolbachia-based control programs or other mass rearing efforts more efficient. Understanding of the nutrient requirements of Ae. aegypti can assist in many of these facets of vector-control programs.

A blood meal is required for many female mosquitoes to produce eggs. Proteins in the blood meal are thought to be the only requirement for eggs to develop (8-10). In mass rearing facilities and in labs conducting experiments on mosquitoes, a blood meal is used as the main dietary solution because it stimulates egg production in female mosquitoes (11). However, among bloods used in the lab, there are variations in their molecular composition, which may lead to difficulties in replicating experiments. Blood preservations methods have been tested on tse-tse flies and have shown promise in allowing those flies to maintain fecundity (13). An artificial diet or a shelf-stable version of whole blood that can be quality controlled in large batches while still generating similar fecundity to a blood meal would reduce variability and improve researchers' ability to replicate experiments. A non-blood based diet may also have other benefits such as longer shelf-life or reduced risk of blood-borne diseases.

Previous artificial diets for mosquitoes have used proteins isolated from blood such as globulins and albumin as an alternative to using whole blood (8, 10). However, artificial solutions have inconsistently resulted in similar fecundity as whole blood. For example in 1990, Kogan demonstrated that an artificial blood meal composed of only certain isolated blood proteins in a salt solution could result in similar fecundity as *Aedes aegypti* mosquitoes fed whole blood (8). This result was not repeatable for Cosgrove and Wood (1996) and the same diet did not work for other species of mosquitoes (9). This inconsistency underlines the need for more research to be conducted to better understand the nutritional requirements for *Aedes* mosquitoes. Other studies on artificial diets for mosquitoes have found that varying protein concentrations can have an effect on the fecundity of *Aedes albopictus* (10). Formulating diets from chemically defined amino acids rather than whole blood proteins and varying concentrations across a gradient will give a better understanding of what the nutritional requirements for *Aedes* mosquitoes are.

Nutritional availability at various life stages of *Aedes aegypti* affects life span and the total number of eggs that the mosquitoes were able to lay (12). In particular, previous studies demonstrated that restricted diets at the larval and adult stages of the mosquito improved the mosquitoes' average life span(4). Other experiments have shown that 9 amino acids – two of which are methionine and isoleucine – are needed for vitellogenesis in *Ae. aegypti* (14). Understanding how diet impacts traits such as the number of eggs a mosquito is able to lay and their average life

span is important due to the incubation period required for many pathogens to before the mosquito can become infectious.

Here we evaluate the effects of feeding blood preserved in different ways – freeze-dried and freeze-thawed – on the survival and egg production of *Aedes aegypti*. Life history characteristics such as egg production and survival of the mosquitoes were compared to evaluate the effect of the use of the two blood preservation methods on mosquitoes' health. Later experiments compared the aforementioned life history characteristics of the mosquitoes when blood was supplemented with methionine (typically a scarce amino acid in blood) in the presence and absence of sugar. Finally, with sugar constantly available, we evaluated the effects on egg production, survival, and hatched larval proportion of supplementing methionine and isoleucine to blood meals.

Methods

Fresh, Frozen and Freeze-Dried Blood Experiments

Three treatment groups were created consisting of 60 female mosquitoes placed in 18 cm³ cages. Each group was fed either fresh blood, frozen blood, or freeze dried blood through a Hemotek Machine for 40 minutes. Mosquitoes were then taken from the cage and placed into falcon tubes covered with clear mesh. Two days later, a small pool of water was added to the falcon tubes and filter paper was placed in the tube to promote and provide a surface for oviposition. After five days, the egg counts of the mosquitoes were recorded. Mosquitoes were then maintained on 10% w/v sucrose solution and monitored daily for survival to determine survival rates of each treatment group.

Effects of Methionine Supplementation on Aedes aegypti Life History Traits

A 0.65 mg/ml methionine-PBS solution was made by dissolving 6.5g L-methionine (Sigma #M9625) in 10 mL of 1x phosphate-buffered saline. Mosquitoes were then divided into three treatment groups: no-supplementation, low-methionine supplementation, and high-methionine supplementation. To the low-methionine treated group, 62.5 uL of the methionine-PBS solution was added to blood, using a micropipette to mix, to make a 3mL solution of methionine supplemented blood. To the high-methionine treated group, 125 uL of the methionine-PBS solution was added to blood to make a 3mL solution of methionine supplemented blood. To the non-supplemented group, 3 mL of human blood was offered with no additional liquid/supplement. After feeding, the mosquitoes were maintained on a 10% w/v sucrose solution. Twelve blood-fed mosquitoes were taken and placed into Eppendorf tubes. After several days, the filter papers were removed and the number of eggs that the mosquitoes laid on the filter paper and in

the water were counted. The mosquitoes were checked daily and the day each one died was recorded. The experiment was run for 45 days.

Colony Maintenance and Larval Raising

The Ae. aegypti colony (USDA Gainesville strain) used in all experiments was sourced from Benzon Research (Carlisle, PA), which had been maintained in continuous colony since 1994. In our facility, *Ae aegypti*, were kept on a 12:12 light/dark cycle, in an environmental chamber maintained at 26 deg C and 84% RH. Larvae were reared in plastic 1 L containers with ~1000 larvae per liter and fed daily on bovine liver powder (NOW Foods, Item#2450). To For hatching experiments, the larval diet was made by mixing1.88 g of liver extract in to 220mL of water. In small rearing cups, 20 mL of water was added and 1 mL of the liver slurry was added. One day after making the solutions in the small cups, the eggs were added. After 3 days, the number of larvae that hatched was counted.

Effects of Methionine and Isoleucine Supplementation

To 200 mL of 1x PBS, 260 mg of L-isoleucine (Sigma I2752) or L-methionine was added. Methionine- or isoleucine-only supplemented blood, 62.5uL of either methionine or isoleucine was added to 3 mL of blood. For low-dose treatment combination of isoleucine-methionine treatment, 62.5uL of methionine and isoleucine solutions were added to 3mL of human blood. For high-dose treatment combination of isoleucine-methionine treatment, 125 uL of methionine and isoleucine solutions were added to 3mL of human blood. For high-dose treatment combination of isoleucine-methionine treatment, 125 uL of methionine and isoleucine solutions were added to 3mL of human blood. For high-dose treatment combination of isoleucine-methionine treatment, 125 uL of methionine and isoleucine solutions were added to 3mL of human blood. After feeding, the mosquitoes were maintained on a 10% w/v sucrose solution. Twelve blood-fed mosquitoes were taken and placed into 45mL falcon tubes with a mesh cover secured by rubber bands. The following day, water and a filter paper was placed into the falcon tubes so that mosquitoes

may lay eggs on the papers in the tubes. Eggs were then collected and counted. Eggs from 5 different mosquitoes from each group were randomly selected after laying. The larvae were raised on the same solution as mentioned previously.

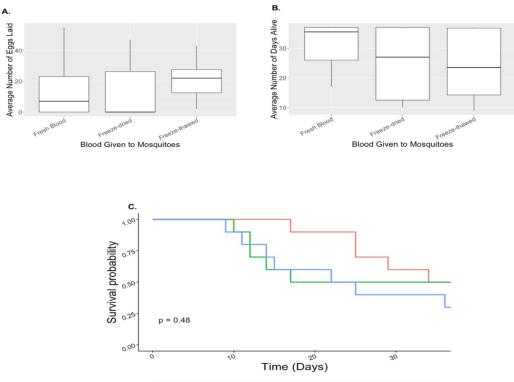
Statistical Analyses

Using the Welch Two-Sample T-Test, the total number of eggs laid, the average day of death, and the average proportion of larvae that hatched were evaluated. This T-Test was used due to the differences in sample sizes. Mosquitoes that died before three days did not have the opportunity to lay eggs. Thus, to account for such variations, Welch's T-Test was chosen. Using the Cox proportional hazard model, the proportion of mosquitoes that survived during each treatment was also evaluated.

Results

Effects of Blood Preservation on Life Span and Total Number of Eggs Laid

Comparing feedings on Fresh, Freeze-thawed and Freeze-dried blood (Figure 1), there were no significant differences between the treatment groups in the first trial of the experiment conducted. The average life span for the mosquitoes among all treatment groups were not significantly different (Figure 1b) and the total number of eggs that were laid between all three groups were not significantly different (Figure 1a, p-value>0.05 between all treatments).



Treatment — Treatment=Fresh Blood — Treatment=Freeze-dried — Treatment=Freeze-thawe

Figure 1. The effects that various blood preservation methods have on the clutch size and survival of *Aedes aegypti.* The average total number of eggs laid (A), the average number of days alive (B), and the survival probability (C) of the mosquitoes were compared by each experimental treatment of blood preservation type provided to the mosquitoes. Bar-and-whisker plot were used. Lines extending from the plot represents the 95% confidence interval. Any dots in the data represents outliers.

However, when a second replicate of the experiment was performed (Figure 2), a cost to life span and clutch size was present in mosquitoes fed Freeze-dried blood. The second replicate showed that mosquitoes fed on Freeze Dried Blood laid fewer eggs than those given Fresh blood (p < 0.0005) or Freeze-thawed blood (p < 0.0005) (Figure 2A). Freeze-dried Blood fed mosquitoes also had a lower life span that was on average 5 days lower (p < 0.05) than those fed the Fresh Blood fed mosquitoes (Figure 2B). However, there was no significant difference in the average number of days alive between the mosquitoes fed Freeze-thawed or Freeze-dried Blood (Figure 2B). There was also no significant difference in the average number of days alive, survival probability or total number of eggs laid between mosquitoes fed Freeze-thawed blood and Fresh Blood (Figure 2B).

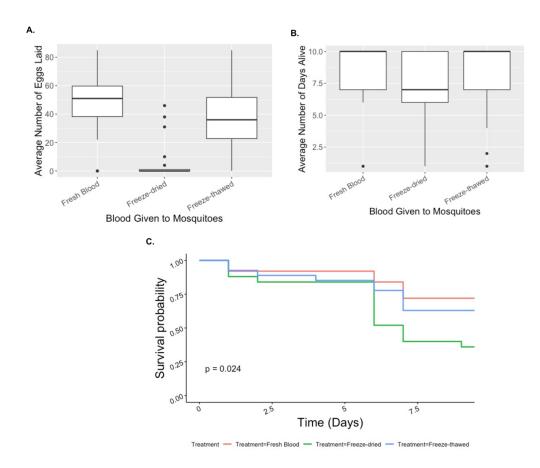


Figure 2. A second biological replicate of the use of various blood preservation methods. The average total number of eggs laid (A), the average life span (B), and the survival probability (C) are presented to determine the effects that feeding mosquitoes blood that has undergone one of the preservation methods listed in the methods.

Supplementation of Amino Acids Results

The Effects of Methionine Supplementation in the Presence of Sugar

When allowed to be fed sugar *ad libitum*, the effects of higher treatments of methionine appear to negatively impact life span of the mosquitoes with little to no effect on the clutch sizes of the mosquitoes. Figure 3A shows that when comparing Un-supplemented Blood with High Methionine and Low Methionine supplemented there were no significant differences among the average number of eggs laid. However, comparing the average number of days alive of the various supplemented blood-fed mosquitoes, high methionine supplemented blood appeared to have a negative impact of the life span of the mosquitoes compared to their un-supplemented fed counterparts (Figure 3B, p<0.05). High Methionine supplemented mosquitoes also exhibited a lower life span than their Low Methionine supplemented counter parts (Figure 3B, p = 0.0503).

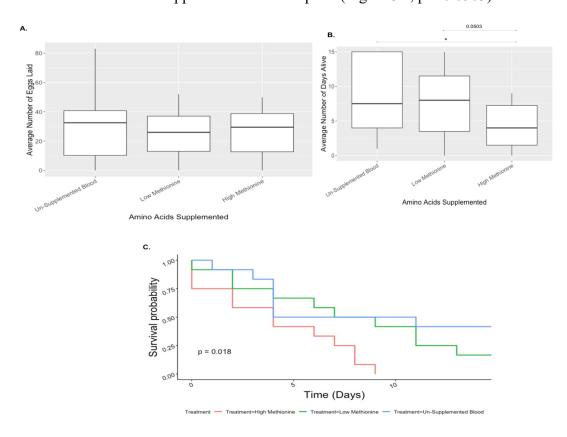


Figure 3 The effects of methionine supplementation to whole blood and subsequent feeding mosquitoes of a 10% w/v sucrose solution *ad libitum*. The average clutch size (A), the average number of days alive (B), and the survival probability (C) were evaluated to determine the effects such feeding would have on the mosquitoes.

The Effects of Methionine Supplementation When No Sugar is Available

Mosquitoes were afforded a single blood meal with the respective methionine supplementation. After blood feeding, they were transferred to falcon tubes and maintained on water via water soaked cotton balls. In the absence of sugar, supplementation with different amounts of methionine had little effect on the average life span and total number of egg laid by the mosquitoes. In Figure 4A, Un-Supplemented Blood Fed mosquitoes had on average higher number of eggs laid, but were not significantly different from their High and Low Methionine supplements counter-parts (p > 0.05 for both comparisons). However, the Low Methionine treated groups did have the lowest average number of legs laid compared to the Un-Supplemented fed treatment group (p = 0.09). The average life span, total number of eggs laid, and survival probability of the mosquitoes were not affected by supplementation of methionine when sugar was not available (p>0.05).

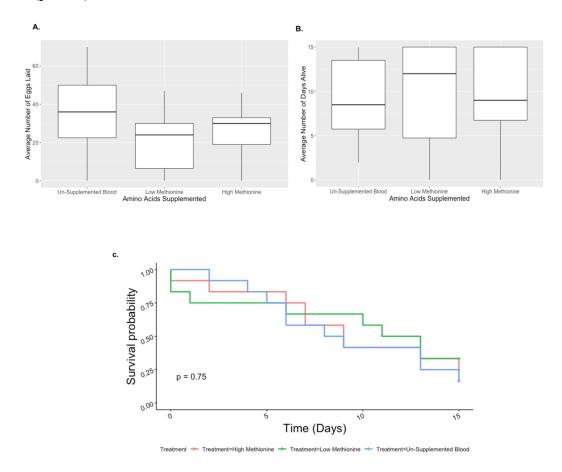


Figure 4. The effects of methionine supplementation to whole blood in the absence of 10% w/v sucrose availability. The average clutch size (A), the average number of days alive (B), and the survival probability (C) evaluated to determine the effects such feeding would have on the mosquitoes.

The Effects that the Availability of Sugar Had on Survival Probability

The greatest effects of supplementing methionine on the mosquitoes were seen when comparing mosquitoes provided sugar *ad libitum* and mosquitoes that were given water instead. Using a Cox Proportional Hazard model, Figure 5 shows that there is an increased risk of death in mosquitoes that are provided sugar at higher concentrations of methionine compared to mosquitoes given lower concentrations of methionine and sugar.

When the survival analysis of Un-supplemented Blood Fed treated mosquitoes that were sugar fed were compared to those that were not sugar fed, there was little difference in the hazard of the two groups (p = 0.78) (Figure 5A and D). Low Methionine treated groups also had no significant results differences in hazard when sugar fed and non-sugar fed when compared. However, High Methionine treated mosquitoes did have differences in their average life span and hazard ratio (HR = 1.728) (Figure 5C and 5E). Sugar fed High Methionine Blood Fed mosquitoes were more likely to die than their non-sugar fed counter parts.

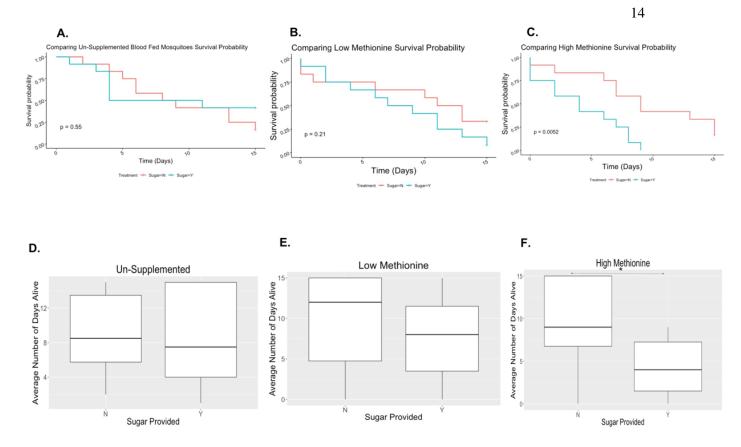


Figure 5. Evaluation of survival probability and average life span of various methionine-supplemented blood fed in the presence and absence of a 10% w/v sucrose solution. In the presence and absence of sugar the average life span and survival probability of un-supplemented (A and D), low-methionine supplemented (B and E) and high methionine supplemented (C and F) are provided.

Supplementing with Methionine and Isoleucine

Mosquitoes were provided their respective treatments and sugar *ad libitum* for the duration of the experiment. Figure 6A shows that mosquitoes fed Isoleucine-supplemented Blood had the highest average number of eggs laid. However, when T-Tests of all treatment groups were done, that there was no significant differences in the total number of eggs laid between all the treatments. When comparing High and Low Isoleucine-Methionine supplemented blood fed mosquitoes the High Isoleucine-Methionine laid more eggs during the experiment compared to the Low Isoleucine-Methionine (p = 0.072).

The average proportion of larvae hatched for the mosquitoes was impacted by amino acids supplemented in the blood meals provided to the mosquitoes (Figure 6B). When un-supplemented blood was given to mosquitoes, they exhibited higher proportions of larva hatched compared to both Isoleucine and High Isoleucine-Methionine supplemented blood feeds (p<0.05 and p = 0.077 respectively, Figure 6B). While the other groups did have varying proportions of larvae hatched when compared, these data were not significantly different from other treatment groups present in the data.

A survival analysis was conducted with all treatments groups to compare survival between all other treatments and the High Isoleucine-Methionine (HIM) supplemented blood. The regression coefficient for all of them compared to HIM was greater than 0, indicating that there was an increased hazard of death of each treatment compared to the HIM fed mosquitoes. However, none of the results were significantly different from one another.

Effects on the average life span of the mosquitoes for the duration of the experiment were also seen. Compared to many other treatments High Isoleucine-Methionine supplemented blood enhanced the average lifespan of the mosquitoes (Figure 6D). When compared to Isoleucine supplemented blood, more High Isoleucine-Methionine fed mosquitoes lasted for the duration of the experiment (p < 0.005, Figure 6D). When comparing High Isoleucine-Methionine (HIM) blood fed mosquitoes to Methionine supplemented blood fed mosquitoes or Low Isoleucine-Methionine (HIM) blood fed mosquitoes, the HIM mosquitoes exhibited higher life spans compared to the other two (p = 0.056 and p = 0.078 respectively, Figure 6D).

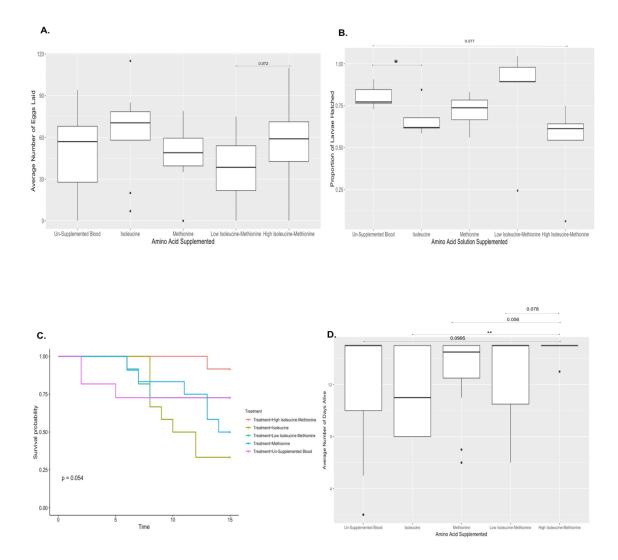


Figure 6. Evaluation of various life history characteristics when offered various combinatorial amino acid supplementation and doses. The average number of eggs laid (A), the average larvae of proportion hatched (B),the survival probability (C) and the average life span (D) were used as proxies for evaluating combinatorial and individual supplementation of isoleucine and methionine.

Discussion and Conclusions

Understanding the effects that blood preservation methods and nutrient availability have on *Aedes aegypti* is important for understanding feeding behaviors and the subsequent effects it had on disease dynamics. Another important aspect of understanding disease dynamics and disease progression in mosquitoes is the ability to use the same blood source for the duration of experiments. Thus, identifying a blood preservation method can help to extend the shelf life of the blood used – helping to reduce the amount of confounding variability that may be present from using multiple blood sources.

We show that methods such as freeze-thawing supports the maintenance of blood quality, enabling the mosquitoes to lay similar quantities of eggs as compared to their fresh-blood fed counterparts in two separate experiments. The ability of freeze-dried blood to maintain egg production and survival of mosquitoes was also evaluated. The two experiments yielded completely different results. One experiment suggested that freeze-dried blood supports the survival and egg production of the mosquitoes. However, when another experiment was performed on another group of *Ae. aegypti*, the survival was lowered in the freeze-dried treatment. Thus, freeze-dried preservation methods yield inconclusive results. We also investigate the effects that supplementing isoleucine and methionine individually and in combination would have on the life history characteristics of *Aedes aegypti*. The data demonstrate in the presence of sugar, that supplementation of methionine had a negative effect on survival with no added benefits egg production. However, these effects were not exhibited in the non-sugar fed mosquitoes, suggesting that sugar may exacerbate the effects of the protein – potentially by increasing absorption of the proteins after digestion.

The results presented in this paper demonstrate that the relative effects of consuming previously Freeze-thawed blood or Fresh blood are negligible – demonstrating that freeze-thawing blood can potentially be a viable option for maintaining blood supplies (Figures 1 and 2). In both experiments, the mosquitoes exhibited similar life spans and laid approximately the same number of eggs throughout the study. However, comparing Freeze-dried blood to either Fresh or Freezethawed blood showed inconclusive results. In the first experiment (Figure 1), the survival probability, average life span, and average number of eggs laid compared to the other treatment groups was not significantly different. During the second biological replicate, there was a significant reduction in the average number of eggs laid with no significant effect on life span. This may suggest that important factors required for blood feeding may have been degraded in Freeze-died blood. As noted in the literature, upon consuming a blood meal, mosquitoes on average can lay up to one hundred eggs (11). Thus, the averages presented in this experiment are quite low, making generalization of the better preservation method difficult to deduce. Nonetheless, Freeze-thawed blood does perform similarly to the Fresh Blood in both experiments. Further experiments can be done to see how long it would take before a noticeable impact on life history traits would occur if freeze-thawing blood was maintained.

As presented in previous literature, methionine and isoleucine seem to be important to egg production in mosquitoes (16). Supplementing methionine into whole blood had noticeable negative effects on the average lifespan of the mosquitoes when supplemented at both the low and high doses. However, there was no increase in egg production in either methionine groups compared to the control. The sugar fed groups did a have a significant difference in their survival probability. The High Methionine (HM) supplemented mosquitoes were more likely to die when compared to the other groups. This suggests that supplementation of methionine when sucrose is provided has a negative impact on life span with no corresponding help in the average number of eggs laid. However, when no sugar was provided, comparing the treatments showed no significant differences between the groups. One of the factors for the exhibited cost to life in the higher dose treated groups may be due to the presence of more protein compared to other nutrients that would more easily available if supplementation didn't occur. This provides evidence that a unique balance of nutrient ratios may be needed so that survival costs are not imposed on the mosquitoes. Greater cost to life may be imposed on mosquitoes who were provided sugar *ad libitum* compared to those that weren't can potentially be due to greater absorption of substances presented from the diet. Processing of purified amino acids may be more difficult for the mosquitoes than using whole proteins. If increased absorption of the amino acids occurred due to sugar availability then more energy may be exhausted on metabolizing the additional proteins, causing the exhibited reduction in life span.

When the average lifespans of mosquitoes subjected to different supplemented or unsupplemented bloods were compared based on the availability of sugar, the results of the analysis showed a decrease in survival probability for the mosquitoes that were provided sugar. This decrease in survival probability was significant when mosquitoes given sugar in the HM groups were compared to mosquitoes in the HM treatment that were not provided sugar. Thus, inclusion of sugar in methionine-supplemented blood fed mosquitoes may exacerbate costs on lifespan when mosquitoes are blood-fed without conferring any benefits to average number of eggs laid. This also suggest that methionine alone may not confer any benefits to vitellogenesis of the mosquitoes.

To evaluate any combinatorial effects of supplementing amino acids, isoleucine and methionine were supplemented to whole blood to determine the effects they would have on life history characteristics of Aedes aegypti. The results show that the presence of isoleucine and methionine at low and high doses did not have any significant effects on the average number of eggs laid by the mosquitoes. However, there was an impact on larvae proportions hatched when isoleucine-supplemented blood fed mosquitoes were compared to their wild-type counterparts. Also, when comparing High Isoleucine-Methionine (HIM) treatments to other methionine groups, on average, the mosquitoes did survive slightly longer (Figure 6D, not significant). Isoleucineonly supplementation did have a negative impact on the average lifespan of the mosquitoes when compared to HIM fed mosquitoes. Supplementing both amino acids increased survival probability and average life span at higher concentrations. The data suggests that the effects of methionine alone and isoleucine alone supplementation may not be able to improve mosquito fecundity. Instead the data demonstrates that there may be a balance in nutrients availability needed for supplementation to not have as costly an effect on the life span of the mosquitoes. Future replication of all supplementation experiments could look at using whole blood with addition of 1x-PBS as a potential control for the study – to account for any background that may result for the additional salts present in the solution.

Future studies should be geared towards understanding the effects that methionine and isoleucine have on the production of secondary metabolites within mosquitoes that may be harmful to them. Future studies can be done to see if mosquitoes raised in sterile conditions – resulting in loss of their microbiome – also exhibit similar effects from amino acid supplementation. Also, studying these effects in the context of *Wolbachia pipientis* may also be another route to go in enhancing some of the disease-prevention programs currently taking place (5,6). Furthermore,

seven other amino acids were implicated as being necessary for having effects on the gonotrophic cycle of mosquitoes (16). Conducting similar experiments as presented in this paper can help to determine what amino acids are the driving factors of the mosquito egg production versus those that may these amino acids may have a different effect on the life history characteristics in the context of disease transmission. Furthermore, research can be done to investigate how various pathogens utilize the nutrients available in mosquitoes for their own benefit. Supplementation with the amino acids presented in this work, along with looking at other blood sources food in mosquitoes in the field, can help to outline what nutrients various pathogens take advantage in the mosquitoes' diet. For such experiments survival probability, egg production, and average life span on pathogen-infected mosquitoes studies should be done to when supplementations are performed to determine if there are any impact. Thus, evaluating those effects may help to determine how pathogens hijack the nutrients present in the mosquitoes for their own benefits.

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187 – 220

ACADEMIC VITA

Education

Pennsylvania State University, University Park, PA Schreyer's Honors College Bachelor's of Science in Biochemistry and Molecular Biology May 2019

Enrichment Programs

Millennium Scholars ProgramAugust 2015 – PresentPennsylvania State UniversityAugust 2015 – PresentSchreyer Honors CollegeAugust 2015 – PresentPennsylvania State UniversityApril 2016 – PresentPennsylvania State UniversityApril 2016 – Present

Laboratory Experiences

Andrew Read, Ph.D. Lab

The Pennsylvania State University: Department of Biology and Entomology

Evaluated housing and dietary factors that influences the life history characteristics of Aedes aegypti.

November 2015 – Present

Wrote my honors thesis.

Clare Sample, Ph.D. Lab

Penn State College of Medicine: Department of Microbiology and Immunology

Schreyer Honors College MD-PhD Summer Exposure Program

Using immunofluorescence staining and microscopy techniques I demonstrated that glycoprotein BDLF2 is a necessary envelope protein for Epstein-Barr Virus to infect epithelium.

Noah Palm, Ph.D. Lab

Yale University: Department of Immunobiology BioMed SURF Program

Using ELISAs and flow cytometry, I showed that *Akkermansia muciniphila* is an immune-stimulant commensal of the mucosal immune system and investigated the usefulness of transformed *A. muciniphilia* to stimulate the immune system for other pathogen associated antigens.

Shomyseh Sanjabi, Ph.D. Lab

University of California San Francisco: Department of Microbiology and Immunology Summer Research Training Program

Using qRT-PCR and flow cytometry, to evaluate how the absence of Sprouty 1 and Sprouty 2 affects CD8+ T Cell functionality during chronic LCMV Clone 13 infection.

Elizabeth McGraw, Ph.D. Lab

Pennsylvania State University: Department of Biology and Entomology

Using qRT-PCR and RNAi to evaluate how the presence of cadherin-87a and alphamannosidase 2a affect vector-competence of *Aedes aegypti*.

April 2018 - Present The

June 2017 - August 2017

May 2018 - August 2018

Poster Presentations

Mosquito Feeding: Do the numbers Really Matter The 2016 Undergraduate Exhibition. University Park, PA 16802 4/6/2016

The Role of BDLF2 in EBV Infection of Epithelium. Annual Biomedical Research Conference for Minority Students. Tampa, FL 11/9-12/2016

Better Blood: Can We Use Food Preservation Methods to Improve Blood Storage. The 2017 Undergraduate Exhibition. University Park, PA 4/5/2017

Akkermansia muciniphila's Ability to Stimulate the Immune System. The Annual Biomedical Research Conference for Minority Students. Phoenix, AZ 11/1-4/2017

Effects of methionine supplementation on *Aedes aegypti* **Life History Traits.** The 2018 Undergraduate Exhibition. University Park, Pa 4/18/2018

The Absence of Sprouty 1 and 2 Enhances Polyfunctional Effector T Cell Maintenance During LCMV Chronic Infection. University of California, San Francisco. 8/3/2018

Extracurricular Activities

Millennium Scholars Tutor

As a member of the Millennium Scholars Program, I was given the opportunity to tutor students in *Organic Chemistry 1 & 2*.

September 2017 – Present

The Virgin Islands Children Museum

St. Thomas, USVI

Accumulated 40.5 hours of community service mentoring students in science and arts while maintaining the displays.

<u>Awards</u>

Academic Excellence Award (\$4,500/ year)

Schreyer Honor College

Provost Award

Pennsylvania State University

Millennium Scholarship (\$15,000/4 Years)August 2015Awarded by the Pennsylvania State University to exemplary students interested in pursuing a
doctorate in STEM.

Catherine V. Beath Trustee Scholarship (\$6,000/4 years)August 2015Awarded to outstanding students in STEM by the Eberly College of Science.

Travel Award for ABRCMS

Annual Biomedical Research Conference for Minorities

May 2016

August 2015

August 2015

October 2016

Outstanding Poster Presentation

Annual Biomedical Research Conference for Minorities

FASEB MARC/IHCM Trainee Award (\$1,500)

Trainee Award to attend the Marine Biological Laboratory in Woodshole, Massachusetts Immunohistochemistry and Microscopy Module.

The Erickson Discovery Grant

Awarded for my research proposal on "Wolbachia Infection: Understanding the Requirements between Aedes aegypti and Wolbachia". Declined the offer.

November 2016

February 2017

April 2018