

**SUSTAINABLE TILAPIA FEED DERIVED  
FROM URBAN FOOD WASTE**

A Thesis  
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The Academic Faculty

by

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## Sustainable Tilapia Feed Derived from Urban Food Waste

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*For Liam.*

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## Summary

Finding an alternative to fishmeal and fish oil in animal feeds has been a topic of increasing interest due to the pressures being put on the ocean's fisheries and the increasing world demand for animal protein. An often-overlooked source of nutrients is in the form of food waste. One third of all food produced globally ends up in landfills, wasting a huge amount of nutrients and embodied energy that could otherwise be redirected towards productive use. This study investigated the feasibility of feeding *Hermetia illucens*, the black soldier fly larvae (BSFL), grown on urban food waste, and *Lemna minor*, a species of duckweed, to tilapia in a recirculating aquaponic system as a compound feed. The study compared the growth of two groups of 58 tilapia over 44 days; one group was fed commercial pellets and the other a compound feed composed of BSFL and duckweed. The group fed the commercial pellets achieved heavier weight gain than the group fed the experimental feed but both groups resulted in steady weight gain and had similar mortality rates. Feeding the experimental feed composed of BSFL and duckweed to tilapia in an aquaponics system is an effective method of diverting food waste from the landfill and further research should be done to optimize this process.

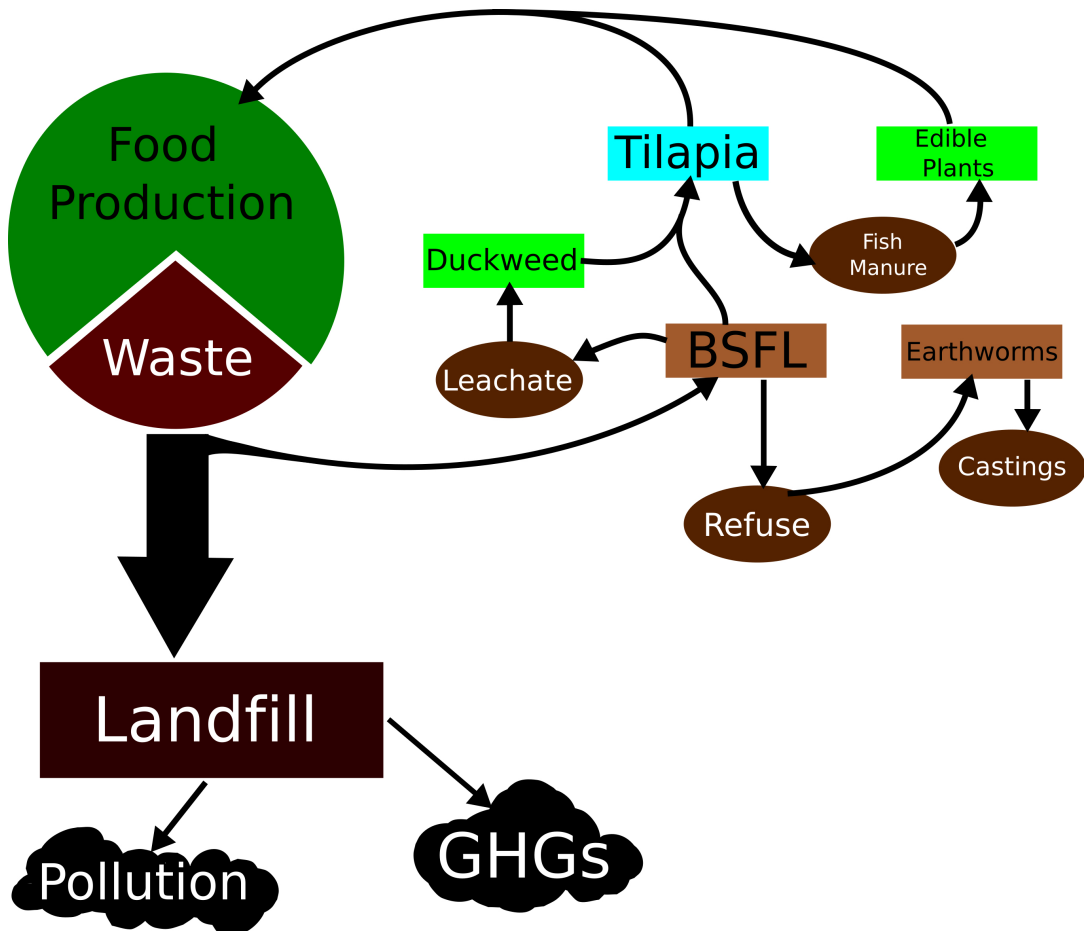
## 1. Introduction

The world's food system is facing a multitude of crises. Shortages of arable land and fresh water, climate change, biodiversity loss, ecosystem destruction, and environmental pollution loom as growing problems with potentially devastating effects if not addressed (FAO 2011b; FAO 2014a; Rockström et al. 2009). These problems are compounded by the increasing demand for animal protein created by the rapidly developing economies of large countries such as China (FAO 2003) since globally 35% of all food produced is used as animal feed (Foley et al. 2011). In addition to terrestrial animal protein, the global demand for seafood is growing. However, the ocean's fisheries have reached their harvest limits as 30% of fish stocks are fished at a biologically unsustainable level and another 60% are fished at their maximum yield (FAO 2014b). Aquaculture is expected to meet the increase in demand (Neori et al. 2007) but has traditionally been reliant on resources derived from the ocean, namely fishmeal and fish oil (Olsen and Hasan 2012; Tacon 2006; R. W. Hardy and Tacon 2002). Fishmeal has been the ideal protein source in aquaculture because of its high protein content, balanced amino acids, vitamins and minerals, essential fatty acid content, and historically cheap price (T. N. Nguyen, Davis, and Saoud 2009; Gatlin et al. 2007). Due to a limited supply and increasing demand for fishmeal (FAO 2014b; Olsen and Hasan 2012) there has been a large amount of research focused on finding alternative sources of protein in aquaculture diets (A. F. M. El-Sayed and Tacon 1997; T. N. Nguyen, Davis, and Saoud 2009; Olsen and Hasan 2012; Tacon 2006). Using plant proteins from sources such as soybean meal to replace fishmeal has yielded some positive results (El-Sayed and Tacon 1997; El-Sayed 1999; Naylor et al. 2009; Hardy 2010). However, while the use of conventional plant proteins as aquaculture feed decouples aquacultural

production from capture fisheries, it still relies on resource intensive farming and competes with other uses of commodity crops (Hardy 2010).

Utilization of waste products in aquaculture feed could help tip the balance from aquaculture being an extractive industry towards a productive one (Wohlfarth and Hulata 1987; Wu et al. 1994; Ulloa et al. 2004). An often-overlooked source of nutrients is food waste. Globally around one third of all food produced is wasted (FAO 2011a), usually ending up in a landfill where it becomes a source of organic pollution. In North America, at just the consumer level, around 100 kg of food is wasted per capita every year (FAO 2011a). The US EPA recommends food that is no longer suitable for human consumption be fed to animals (US EPA 2015). In order to utilize food waste as animal feed it must be handled and processed into a form that is economical and nutritionally viable for the animals being fed by it. Black soldier fly larvae (BSFL) can be used to digest food waste to create an animal feed that is high in protein and fat (Nguyen, Tomberlin, and Vanlaerhoven 2015) and have been successfully fed to fish (Bondari and Sheppard 1981; St-Hilaire et al. 2007; Kroeckel et al. 2012). Another organism that has attracted attention as an alternative fish feed is duckweed (Gaigher, Porath, and Granoth 1984; Hassan and Edwards 1992; Fasakin, Balogun, and Fasuru 1999; Leng, Stamboli, and Bell 1995). Duckweed is efficient at absorbing nutrients from water (Hasan and Chakrabarti 2009; Leng, Stamboli, and Bell 1995) and grows well when there is decaying organic material in the water (Skillicorn 1993). This quality might make it possible to use the refuse from BSFL culture as the nutrient source for a duckweed culture. By doing so, two types of alternative fish feed could be generated from food waste diverted from the landfill. These feeds could be then fed to tilapia in an aquaponic system. Aquaponics is a type of recirculating aquaculture that uses edible plants to perform biological filtration of the aquacultural water (Rakocy, Masser, and Losordo 2006). Combining all of these elements together would make it possible to create a

variety of marketable yields from diverted food waste (figure 1). More information about BSFL and duckweed is detailed in the following sections.



**Figure 1 System Overview-** Overview of system for diverting food waste from landfills towards productive yields. Food waste is fed to BSFL, which provide nutrients for duckweed culture. BSFL and duckweed are fed to fish in an aquaponic system, which also produces edible plants. The refuse from the BSFL could be used to feed vermiculture creating additional marketable yields of earthworms and earthworm castings.

The specific aim of this project was to evaluate the viability of a compound feed composed of black soldier fly larvae and duckweed by comparing its performance to conventional feed containing fishmeal in a recirculating aquaponic system. Three studies were conducted. The first study compared the feed conversion efficiencies of fish fed the duckweed and BSFL feed and the commercial feed containing fishmeal. The second study compared the feed assimilation efficiencies of the two feeds. The third study investigated the feed conversion efficiency BSFL to ascertain an approximate idea of how effective the BSFL cultures were at consuming waste and producing larvae. More broadly, this research seeks to broaden the arena of knowledge regarding closed loops systems of production in order to achieve maximum material and energetic yields from a set of resources.

### ***1.1. Black Soldier Fly Larvae***

The larvae of *Hermetia illucens*, the Black Soldier Fly (BSFL), possess many qualities that make them attractive for converting urban food waste into animal feed and other valuable products. They have been successfully fed to a variety of livestock animals including pigs (G. L. Newton et al. 1977), (Hale 1973), tilapia, and catfish (Bondari and Sheppard 1981).

BSFL have been shown to effectively digest a wide range of organic waste products such as offal, kitchen waste, and fruit and vegetable waste (Nguyen, Tomberlin, and Vanlaerhoven 2015). BSFL are also useful for managing manures and when grown on chicken manure were able to reduce the amount of manure by 50% and the larvae had a resulting composition of 42% protein and 35% fat (Sheppard et al. 1994). The conversion of low value manure, or other organic waste streams, to bulk proteins and lipids creates many opportunities for developing valuable products. The ability of the

BSFL to create such high proportions of lipids has even attracted attention to how their culture might be applied to biofuel systems (Li et al. 2011).

Despite their fondness of manure, BSFL are thought of as a non-pest species (Sheppard et al. 1994) and have been shown to reduce *E. coli* levels in chicken and dairy manures (Erickson et al. 2004; Liu et al. 2008) as well as compete with house fly larvae, thereby decreasing their population (Bradley and Sheppard 1984).

The ease of harvesting the BSFL is also of immense value. The last stage of the BSFL's life cycle before pupation is a migratory stage where the larvae develops a large fat store, empties its gut, and then seeks a place away from the waste to pupate (Sheppard et al. 1994; G. L. Newton et al. 1977). This makes collecting the larvae as simple as providing a singular high point that falls into a collection bucket. Being able to harvest the larvae passively makes scaling systems much simpler and much more cost effective than more intensive systems.

After the food waste has been processed by the BSFL, the leftover biomass is nutrient rich, low-odor, humus that can be added to soils as an amendment to increase organic matter and fertility (Diener, Zurbrügg, and Tockner 2009).

One drawback of BSFL when compared to fishmeal is the presence of chitin that forms the BSFL outer membrane. Chitin is largely indigestible by fish and it should also be noted that the nitrogen content of chitin is 6.89%, which should be taken into account when formulating feed as it significantly changes the amount of digestible crude protein (Diener, Zurbrügg, and Tockner 2009).

## ***1.2. Duckweed***

Duckweed is an extremely fast growing aquatic macrophyte that floats on the surface of the water. It is distributed around the world in tropical, subtropical, and temperate climates and grows in still, fresh, or brackish water (Skillicorn 1993; Rusoff, Blakeney,

and Culley 1980). There are 37 identified species of duckweed that span five genera of the *Lemnaceae* family: *Wolffia*, *Wolffiella*, *Spirodela*, *Landolita*, and *Lemna* (Appenroth, Borisjuk, and Lam 2013). Of those species, *Lemna minor* was the only one cultured in this study. Under ideal conditions, such as growing in the presence of decaying organic matter (Skillicorn 1993), duckweed is capable of forming dense mats of colonies that grow over each other (Hillman 1961; Rusoff, Blakeney, and Culley 1980) and can double its mass in less than 48 hours (Skillicorn 1993). Duckweeds grows fastest in warm waters with a pH between 6.5-7.5 and full sun exposure but are tolerant of water temperatures between 6 and 33 °C and a large range of light intensities (Hillman 1961; Leng, Stamboli, and Bell 1995). Duckweeds are highly effective at concentrating nutrients such as phosphorus and nitrogen in their tissues and are capable of lowering the concentrations of nutrients in eutrophic waters down to trace levels (Hasan and Chakrabarti 2009; Leng, Stamboli, and Bell 1995).

Besides their profound ability of nutrient removal from water, duckweed also has an impressive nutritional profile. Efficiently managed duckweed cultures typically are composed of approximately 40% crude protein, 10% fiber, and 5% polyunsaturated fat (Hillman and Culley 1978; Leng, Stamboli, and Bell 1995). The high protein content of dried duckweed make it a good source of amino acids and protein concentrate (Rusoff, Blakeney, and Culley 1980). Additionally, the protein-rich leaves of duckweed have around one-tenth the amount of fiber as other common feed plants such as maize and soy (Leng, Stamboli, and Bell 1995; Skillicorn 1993), which further increases its potential as a highly digestible source of feed protein.

Due to its favorable nutrition, duckweed has been successfully used as a protein source for a variety of animals including fish, poultry, and even humans (Hillman and Culley 1978; Skillicorn 1993; Leng, Stamboli, and Bell 1995; Hassan and Edwards 1992). *Lemna minor*, the species cultured for this study, has been shown to be a

complete protein that contains all of the essential amino acids (Rusoff, Blakeney, and Culley 1980; Muztar, Slinger, and Burton 1978). Fresh duckweed of the *Lemna* genus performed well as feed when fed to Nile tilapia that were able to digest duckweed and efficiently assimilate its high protein content, with the ratio of duckweed offered to fish biomass gained equaling 3.4 (Hassan and Edwards 1992).

The fast growth rate of duckweed allows for large yields. It has been demonstrated that duckweeds can achieve yields of 10-20 tons dry matter/ha/year in non-laboratory settings (Leng, Stamboli, and Bell 1995). Duckweed, like many aquatic plants, is 91-95% water (Hasan and Chakrabarti 2009) so drying it out is important if the protein is to be concentrated or if there is a large transportation distance between where it grows and where it is consumed. For this reason, decentralized culture and use of duckweeds generally enhances their economic viability (Hillman and Culley 1978).

### ***1.3. Feed Conversion Efficiency and Feed Assimilation***

#### ***Efficiency***

Feed conversion efficiency (FCE), also referred to as feed conversion ratio (FCR), is the amount of feed given to animal divided by the amount of animal biomass harvested as expressed below:

$$FCE = \frac{Feed\_Input}{Animal\_Biomass\_Output}$$

Different animals have very different FCEs and the exact conversion factor is dependent on a range of variable such as species, feed type, and age. According to a recent study, the FCE was around 14 for beef cattle and 2 for chickens (Peters et al. 2014). Most of the fish grown in aquaculture have a FCE between 1.5 and 2 (Olsen and Hasan 2012).

Feed assimilation efficiency refers to the amount of feed eaten that is used for metabolism or turned into additional biomass. In fish, the assimilation efficiency is typically measured by tracking the amount of feed eaten and the amount of feces produced (Belal 2005). Individual nutrients can also be tracked by knowing the amount eaten versus the amount excreted (Amirkolaie et al. 2005).

## **2. Methods and Materials**

### *2.1. Feed Conversion Efficiency Study*

#### *2.1.1. Study Setup*

The first study was designed to compare the feed conversion efficiency of the commercial feed to the experimental feed composed of BSFL and duckweed in order to assess its viability as a substitute for commercial feed made of grain and fishmeal. The fish used in the study were split into two treatment groups with one fed commercially purchased feed and the other fed the BSFL and duckweed feed. Both groups were fed known amounts and their weight was recorded at the beginning and end of the study.

The study was conducted in a 900ft<sup>2</sup> greenhouse in downtown Atlanta, Georgia. The greenhouse houses an aquaponic system that recirculates water in a closed loop. The system has one main fish tank that is a 3000-gallon raceway and is plumbed in parallel to 5 biofilter grow beds measuring 4'x24'x8". The system is continually circulated with a 3000gph pond pump and aerated with an air pump at 80L/min.

Two identical black 70-gallon stock tanks filled to approximately 50 gallons were used to house the two treatment populations. The tanks were plumbed into the existing aquaponic system so that the water leaving one bed of the plant biofilter first entered the experimental tanks and then drained into the main raceway before being recirculated back into the plant biofilters. This configuration was chosen to minimize water quality differences between the two tanks and ensure adequate nutrient removal without water exchange. Both tanks were covered with opaque corrugated PVC roofing with approximately 2" of space left on each side to allow some light to enter the tanks but prevent algae growth and keep fish from jumping out.

### 2.1.2. *Fish*

*Oreochromis aureus* (blue tilapia) were divided into two treatment populations with each initially containing 58 individuals. Juvenile fingerlings, fish that are about a finger long, were purchased from White Brook Tilapia Farm™ and grown out for 4 months in the main raceway on commercial feed before being used in the study.

### 2.1.3. *Weighing Fish*

Prior to beginning the study, the fish were caught in an improvised net cage and then individually weighed on a scale out of the water before being added to the experimental tanks. The study ran for 44 days and then fish were individually weighed again.

### 2.1.4. *Feeding Sessions*

The two populations were fed known amounts of feed until apparent satiation twice daily in 5 minute feeding sessions. A small portion of feed was added to the tank and replaced at a rate matching the feeding rate until either the allotted 5-minute session was over or the feeder observed highly reduced feeding activity for 30 seconds. Substantially decreased feeding usually coincided with the end of the 5-minute sessions. The fish consumed all of the food given as confirmed by visual observation of the feeder and the amount of feed eaten by the population was recorded at the end of each session.

### 2.1.5. *Feed Components*

The *Lemna minor* (duckweed) was obtained from the Georgia Tech Research Institute from a project cultivating duckweed for chickenfeed. The duckweed was grown

in shallow, fertilized ponds and was periodically harvested and dried in a solar dehydrator. The dried duckweed was then stored in airtight plastic bags and frozen until used in the feed preparation.

The *Hermetia illucens* (BSFL) was grown on site in specially designed Protapod™ bins purchased from an online retailer that are specifically designed for BSFL culture (ProtaPod™ 2015). The bins are designed to provide a singular high point for the wandering larvae to crawl to for collection. Each bin was fitted with a vinyl shoot leading into a sealed 5-gallon bucket to collect the mature BSFL. The bins were covered with opaque corrugated PVC roofing to prevent rain from entering the bins while allowing adult flies to enter and exit the bins, facilitating oviposition. A hole was drilled in the bottom of each bin to allow leachate to drain into 5-gallon buckets. The BSFL bins were fed fruit and vegetable waste acquired from a local smoothie shop. The BSFL were harvested and weighed immediately after the food waste additions and then frozen until use in the feed preparation. New food waste was weighed and added on a weekly basis. The amount of leachate and BSFL produced were recorded each time new food was added allowing for an approximation of the BSFL food waste to BSFL biomass conversion efficiency to be made.

#### **2.1.6. Feed Preparation**

The experimental population was fed a prepared feed of 40% dried duckweed and 60% BSFL by estimated dry mass to achieve estimated protein content of 36% matching the commercial pellets. To prepare the experimental feed, 40g of dried duckweed was combined with 225g of thawed BSFL and 503g of filtered tap water. The mixture was then blended in a high-speed blender until a homogeneous paste with uniform consistency and no visible particulates was achieved. The paste was then spread onto parchment paper on four 11"x17" baking sheets and dehydrated in the oven

at 75°C for approximately 2 hours to create dehydrated sheets. The sheets were removed from the oven, allowed to cool, and cut into approximately ¼” square pieces. The pieces were stored in an airtight container and refrigerated until use.

The control group was fed Premium Fish Food™ *Tilapia Ultimate Growout Pellets*, a commercial feed comprised of fish meal, dehulled soybean meal, ground corn, wheat middlings, brewers dried yeast, and vitamin mix, with a crude protein content of 36% (min), 6% crude fat (min), and 5% crude fiber (max). The pellets were also kept in an airtight container and refrigerated until use.

#### 2.1.7. *Nutritional Analysis*

The control and experimental feeds were sent to the University of Missouri Agricultural Experiment Station Chemical Laboratories (ESCL) for analysis. Complete amino acid profiles and crude protein, crude fat, crude fiber, ash, moisture, acid detergent fiber (ADF), neutral detergent fiber (NDF), and omega-3 content, were analyzed. Carbohydrates by difference and gross calories were calculated from the analyses.

#### 2.1.8. *Statistical Analysis and FCR*

The feed conversion ratio (FCR) of the tilapia was determined by dividing the amount of feed eaten by the weight gain of the fish over the study period. The means of the final weights of each treatment group were compared using a student’s t-test.

#### 2.1.9. *Average Daily Gain*

The average daily weight gain of the fish was calculated by subtracting the initial average weight from the final average weight and dividing by the number of days the experiment ran as expressed below:

$$\frac{\text{Mean}_{Final\_Weight} - \text{Mean}_{Initial\_Weight}}{\text{Experiment\_Length}_{(Days)}}$$

## 2.2. Fish Feed Assimilation Assay

### 2.2.1. Study Design

A feed assimilation study was conducted to assess how well the fish were able to convert the given feed into biomass. The assimilation efficiency of the fish feed into biomass was evaluated by feeding the fish a known amount of feed, collecting and weighing their feces, and determining the percentage absorbed or metabolized. Ten fish from each of the feed treatments used in the feed conversion efficiency study were individually placed into 5-gallon buckets filled with tank water filtered through a 5 $\mu$ m filter and constantly aerated. Seven fish from each treatment group received either the commercial feed or the BSFL and duckweed feed. To account for scales, mucous, or other unknown particulate shed by the fish, the other two individuals from each treatment group were controls and not fed. Following Heng et. al in 2007, all of the fish were starved for 26 hours prior to beginning the study (Heng, Ong, and Hassan 2007). The fish receiving feed in each treatment group were fed to apparent satiation and the amount of feed was recorded for each individual. The fish were monitored after feeding for gut evacuation of feces. Upon detection of feces, the fish were removed from their bucket and the feces was collected by filtering the water through a 11 $\mu$ m filter using vacuum flask. It was assumed that the fish completely evacuated their gut upon an initial observation of feces. The water from the control groups was filtered after all of the fed fish had evacuated their guts. The filter pads were placed in petri dishes and then stored in sealed plastic bags in a cooler of ice water to prevent bacterial activity during the collection period. Following the collection period, the filter pads were dried in an oven at

68°C until constant weight and weighed. The feed assimilation was calculated as the weight of the feces divided by the weight of the feed multiplied by 100.

### 2.3. *BSFL Conversion Efficiency Study*

A conversion efficiency study was done to determine how much BSFL mass could be produced from fruit and vegetable wastes from a nearby smoothie shop. The BSFL were cultured in four bins and fed all of the fruit and vegetable scraps produced from the smoothie shop every week. Additions of fruit and vegetable waste were made once per week. In order to approximate the composition of the fruit and vegetable scraps, two samples of 32-gallon trash bins were sorted and weighed on three separate occasions. The composition of the fruit and vegetable waste was broken into nine major categories and the relative contribution of each type was computed (Table 1).

<b>Type of Food Waste</b>	<b>% of Total</b>
<b>Avocado</b>	8.5
<b>Bannana</b>	20.3
<b>Citrus</b>	7.4
<b>Coffee</b>	1.2
<b>Mango &amp; Papaya</b>	17.4
<b>Melons</b>	17.7
<b>Pineapple</b>	12.1
<b>Misc</b>	10.3
<b>Juice</b>	5.1

**Table 1 Sorted Food Waste-** Sorted food waste components listed as percent of total.

To evaluate the productivity of the BSFL culture and approximate how much BSFL could be generated per culture, the conversion of food waste to BSFL was quantified. The BSFL bins were treated as “black boxes” with inputs of food waste and outputs of

BSFL. The amount of residual matter left over from the BSFL was not quantified, so rather than a true feed conversion ratio, the conversion efficiency was calculated to provide a practical metric of productivity of the BSFL culture. The following parameters were measured to determine the conversion efficiency of the BSFL cultures: the weight of food waste added to the BSFL bins, the weight of BSFL harvested from each bin, and the weight of liquid leached from the bins. The conversion efficiency was computed by dividing the weight of BSFL produced by food waste added minus the weight of leached liquids, then multiplied by 100. The equation used is shown below:

$$\frac{BSFL(lbs)}{(Feed\_Added(lbs) - Leachate(lbs))} \times 100\%$$

### 3. Results

#### 3.1. Feed Conversion Study

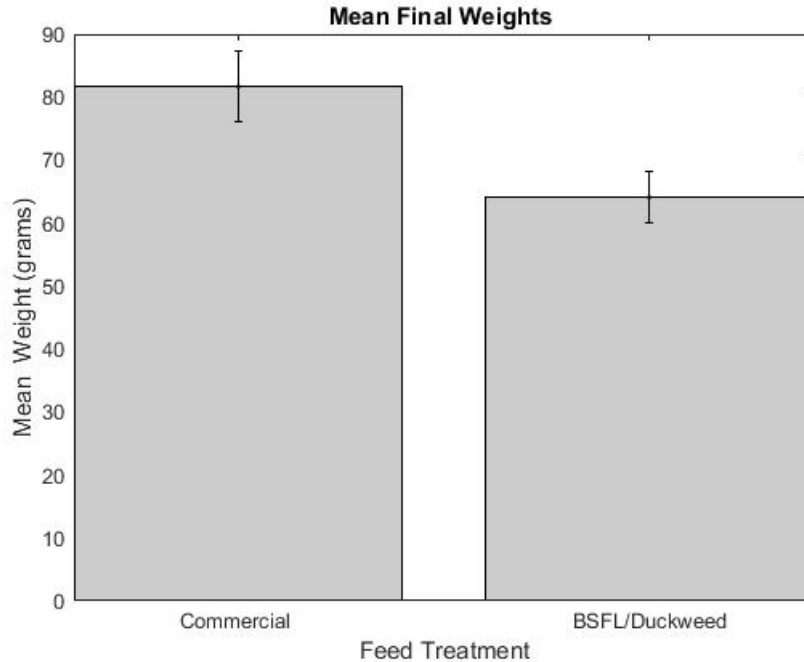
The commercial feed treatment experienced higher weight gain than the fish fed the BSFL and duckweed diet (Table 2). The two groups had similar mortality (5 in the commercial treatment and 4 in the BSFL and duckweed treatment). The FCR of the commercial feed group was 1.7 while the BSFL and duckweed group had a much less efficient FCR of 4.4. The average daily gain of the commercial treatment was about twice as much as the BSFL and duckweed treatment.

FCR Results		
Metric	Commercial Feed	BSFL/Duckweed Feed
Total Initial Population Weight (grams)	2849.2	2818.4
Total Final Population Weight (grams)	4412	3527.2
Average Initial Population Weight (grams)	48.3	47.8
Average Final Population Weight (grams)	81.7	64.1
Amount Fed (grams)	2678.6	3092.2
Initial Number of Individuals	59	59
Final Number of Individuals	54	55
Mortality	5	4
FCR	1.7	4.4
Average Daily Gain (g/day)	0.76	0.37

**Table 2 FCR Summary-** Summary of the data acquired from the feed conversion experiment.

### 3.1.1. Comparison of Means

The mean weight of the fish that were fed commercial pellets ( $N_{\text{com}} = 54$ ,  $M = 81.7\text{g}$ ,  $SD = 40.5$ ) was significantly different than the fish fed the BSFL and duckweed feed ( $N_{\text{BSFL}} = 55$ ,  $M = 64.1$ ,  $SD = 29.4$ ),  $t(107) = 2.596$ ,  $t_{\text{critical}} = 1.982$ ,  $p = 0.01076$ , using a student's two sample t-test (Figure 1). However, the mean final weight of the fish fed commercial pellets was substantially influenced by tail of the distribution (see following section). Removing the six individuals in the highest bins resulted in means that were not significantly different ( $N_{\text{com}} = 48$ ,  $M = 71.7\text{g}$ ,  $SD = 30.0$ ,  $N_{\text{BSFL}} = 55$ ,  $M = 64.1$ ,  $SD = 29.4$ ),  $t(101) = 1.287$ ,  $t_{\text{critical}} = 1.984$ ,  $p = 0.201$  using a student's two sample t-test. The fish fed the commercial pellets had a higher final weight than the fish fed the BSFL and duckweed feed, but this was mostly due to six large individuals in treatment receiving commercial feed.



**Figure 2 Final Means-** The final mean weights of the two treatment groups is shown with error bars representing the standard error of the mean.

### 3.1.2. *Binned Comparison of Variance*

The distribution of initial and final weights of each treatment group were compared using histograms to bin the weights in 20-gram intervals (Figure 2). The initial groups started out with similar variances (Table 4). The variance of the final weights of both treatments was substantially higher than the initial weights. The variance of the commercial feed increased much more than the variance of the BSFL and duckweed treatment group. The distributions of the final weights for each group were significantly different  $\chi^2(9, N=109)= 18.395$   $p=0.0309$ , however the tails of the distribution contributed most of the observed difference (Table 3). The histograms show that the commercial

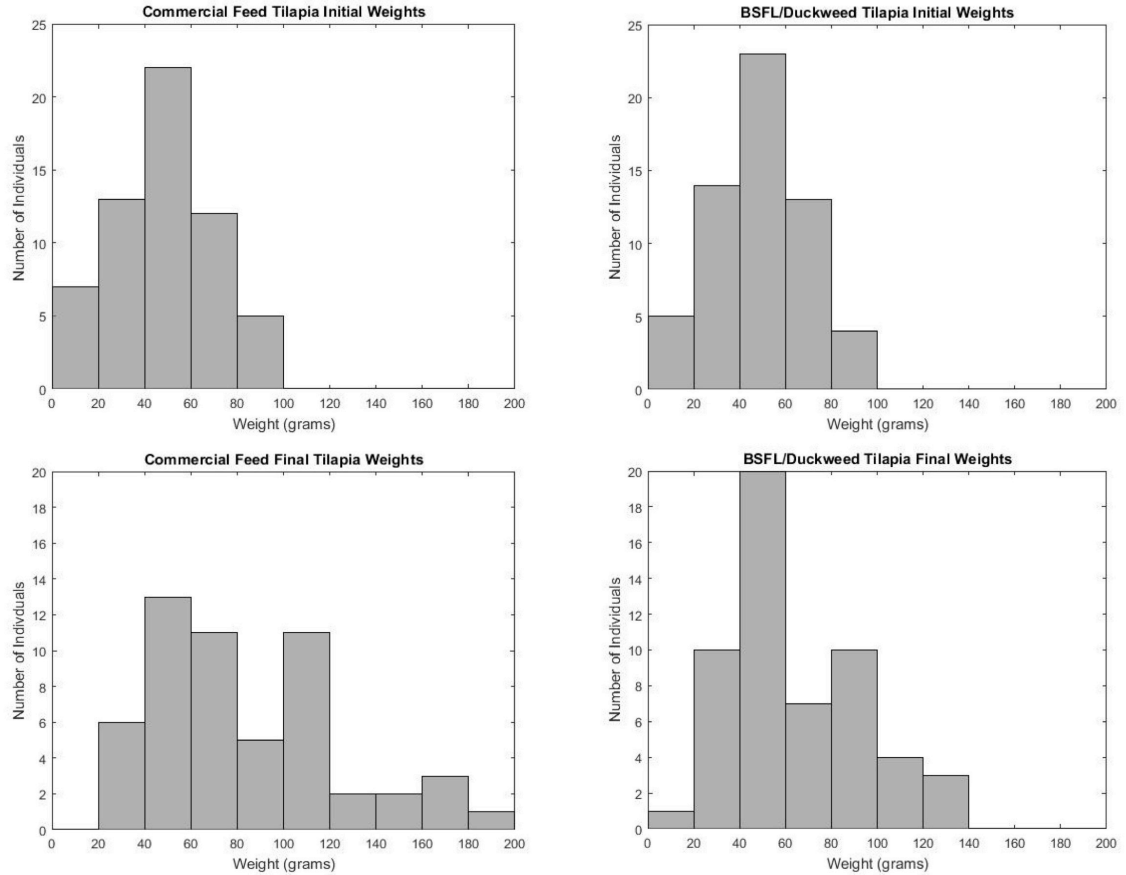
feed led to larger maximum weights of individual fish and heavier fish overall than the BSFL and duckweed feed.

Bin	Commercial Feed		BSFL Duckweed Feed	
	count	$\chi^2$	count	$\chi^2$
0-20	0	0.4954	1	0.4864
20-40	6	0.4683	10	0.4598
40-60	13	0.6859	20	0.6734
60-80	11	0.4864	7	0.4775
80-100	5	0.7954	10	0.7809
100-120	11	1.7139	4	1.6827
120-140	2	0.0919	3	0.0902
140-160	2	1.0279	0	1.0092
160-180	3	1.5418	0	1.5138
180-200	1	0.5139	0	0.5046

**Table 3 Histogram Bin Counts**-Histogram bin counts with corresponding chi-squared values.

Measure	Commercial	BSFL/Duckweed
Initial Standard Deviation	20.3	19.4
Final Standard Deviation	40.5	29.4
Initial Variance	407	370.5
Final Variance	1609.6	848.9

**Table 4 Treatment Group Standard Deviation**- Standard deviation and variance of the two treatment groups at the beginning and end of the study.



**Figure 3 Population Histograms-** Histograms of the population weights at the beginning and end of the feed conversion efficiency study.

### 3.1. Feed Assimilation Study

Several assumptions made in the design of the feed assimilation study were proven to be false and a result, no data was obtained. The first assumption was that tilapia would have completely emptied their guts after 26 hours of fasting. This was proven to be false when all 14 fish defecated into the buckets at least 33 hours after their last feeding. This also proved the assumption that the fish defecated once per feeding to be false, which was further emphasized when the fish labeled as control defecated again at 52 hours after the last feeding. An additional complication was that only three fish in

each treatment group ate the feed given, probably due to handling stress. Future attempts at this study should have a continuous fecal collection method and an adequate way of assuring the fish have emptied their guts at the beginning of the study.

### *3.2. BSFL Conversion Study*

The four BSFL bins were fed a total of 2339 pounds of fruit and vegetable scraps and together produced 57 pounds of BSFL. The average conversion ratio of fruit and vegetable waste to BSFL was 2.44% with a standard deviation of 0.33% between bins.

## 4. Discussion

The commercial feed outperformed the BSFL and duckweed feed in growth rate and weight gain but that was mostly due to a minority of the commercial population growing much larger than the rest. Gaigher et al. 1984 found that tilapia fed only fresh duckweed at a rate matching their consumption rate over the course of the day experienced an average daily gain of 1g/day (Gaigher, Porath, and Granoth 1984). Since both the commercial feed and BSFL and duckweed feed treatment groups grew well under 1g/day, (0.76g/day and 0.37g/day respectively), that would suggest the feeding rate in the feed conversion study was below the optimum rate necessary for maximum weight gain. Bondari and Sheppard 1981 found that blue tilapia fed chopped BSFL at 3% of their body weight gained an average of 0.56g/day over a 10-week period which was not significantly different from the tilapia receiving commercial pellets in that study (Bondari and Sheppard 1981). This result is closer to the weight gains observed in this study but still higher than the BSFL and duckweed feed. The difference could be due to the nutritional differences in the feed or differences in environmental conditions the fish were reared in.

The BSFL and duckweed feed probably had a higher moisture content than the commercial pellets, which could have led to the fish eating less total macronutrients despite eating a larger total mass. The chitin content of the BSFL could also lower the amount of digestible protein in the BSFL and duckweed feed which was not adjusted for in the feed formulation. To track environmental conditions, water quality was monitored twice daily at each feeding session by checking dissolved oxygen, unionized and ionized ammonia, temperature, and pH using a YSI<sup>tm</sup> pro plus multimeter. Another possible

reason for the depressed weight gain was high concentrations of ionized ammonia ( $\text{NH}_4^+$ ) that quickly rose at the beginning of the study for unknown reasons and averaged 7.1 mg/L over the course of the feed study in both treatment tanks. Unionized ammonia was undetectable or low for the entire study but prolonged exposure ionized ammonia could have negatively affected fish health.

The fish fed commercial pellets had more variable weights than the fish fed the BSFL and duckweed feed ( $\sigma^2$  commercial= 1609.6,  $\sigma^2$  BSFL/Duckweed = 848.9). However if the six largest individuals were excluded from the commercial population, the variances would be similar ( $\sigma^2$  commercial= 897.5,  $\sigma^2$  BSFL/Duckweed = 848.9). This difference could be due to differences in the physical characteristics of the feed. The commercial pellets were much harder and fish that started off small may not have been able to fit the pellets in their mouths. This could have led to the large fish getting larger and excluding the smaller fish from feeding. In contrast, the BSFL and duckweed feed was relatively soft and easy for fish to break pieces off of when they were unable to swallow them whole. Nutritional differences such as the increased fat content of the BSFL and duckweed compared to commercial pellets could also have had an effect on the weight distributions of the fish.

Despite not performing as well as the commercial pellets in overall weight gain, the BSFL and duckweed feed did produce steady growth in tilapia and resulted in a similar mortality rate as the commercial pellets. Feeding the BSFL and duckweed feed also resulted in more uniform growth than the commercial pellets, which may be desirable for aquaculture. Having all of the fish in a cohort grow at an even rate could make harvesting and planning easier and also reduce the need for grading sizes. Since the BSFL and duckweed feed did not have vitamin or mineral additives and was not as heavily researched as the commercial pellets, the performance was a satisfactory starting point for developing sustainable feed from food waste. The feed was produced

with close to no costs aside from the capital cost of the bins, collecting the food waste, and oven drying the feed. Since the tilapia were in a recirculating aquaponic system, with high value lettuces being produced as well as tilapia, the profit margins were further increased.

## 5. Conclusion

More research should be done on streamlining the production of fish food from the urban waste stream using BSFL and duckweed. The duckweed for this study was grown using inorganic fertilizer but BSFL leachate may contain enough nutrients to be a suitable alternative, which would help to completely recover wasted nutrients. The food waste processed by the BSFL could be used to feed vermiculture which would produce earthworms and valuable castings (Newton et al. 2005). Additional research on streamlining the production of the feed and using solar dehydration rather than a conventional oven to dry it would also help reduce costs and make the process more scalable. It would also be interesting to see more in depth life cycle and economic analyses of using food waste for decentralized fish feed production using BSFL and duckweed. This information would be relevant not only to those seeking to reduce costs in the aquaculture industry, but also to the subsistence farmers of the world who, more than anyone, know that wasting massive amounts of nutrients and energy is an unacceptable practice. As a society we must rethink what we label “wastes” and instead see them as resources that are to be cycled and conserved.

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