



Valorisation of Organic Waste

– Effect of the Feeding Regime on Process Parameters in a Continuous Black Soldier Fly Larvae Composting System

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Master's thesis

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Title: Valorisation of Organic Waste - Effect of the Feeding Regime on Process Parameters in a Continuous Black Soldier Fly Larvae Composting System

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Abstract

Municipal solid waste- and especially organic solid waste management has become a major challenge in both developing and developed countries and is often related to the spread of diseases. At the same time, an increase in the demand for animal feed protein causes disturbances in (marine) ecosystems and nutrient recycling in soils is unsatisfactory in many places. These issues have a major negative impact on the environment. Black soldier fly composting could solve these problems; by using the larvae of the black soldier fly (*Hermetia illucens*) to process organic waste and produce two valuable products: black soldier fly larvae, which could be used as chicken and fish feed, and the residue that can function as an organic fertiliser.

The aim of this study was to identify specific process parameters leading to optimised performance of a continuous black soldier fly composting system. This was accomplished by measuring specific parameters (pH, compost maturity, total solids, organic content) on various points along two plug flow reactors operated with different larval feeding regimes (40 mg dry food/larva/day and 60 mg dry food/larva/day) and identifying differences between the two regimes.

The pH, compost maturity, total solids, organic content, prepupal weight, material reduction and biomass conversion rate did not differ significantly ($P < 0.05$) between the two feeding regimes. However, survival rate of the larvae was significantly higher (100%) when fed with 60 mg total solids food/larva/day in comparison to when following the feeding regime with 40 mg total solids food/larva/day (70%). A material reduction of 68% (85% on wet basis) and a waste-to-biomass conversion rate of 19% on a total solids basis was achieved in the system.

It was found that black soldier fly composting can process more waste than what was expected per larva and day. However, further studies are needed in order to increase the total amount of waste treated in the unit.

Keywords: Black Soldier Fly, Black Soldier Fly Larvae, Organic waste, Waste Management, Insect Assisted Waste Management

Popular science summary

Municipal solid waste, and especially organic solid waste management has become a major challenge in both developing and developed countries and is often related to the spread of diseases. At the same time, an increase in the demand for animal feed protein causes disturbances in (marine) ecosystems and nutrient recycling in soils is unsatisfactory in many places. These issues have a major negative impact on the environment. Black soldier fly composting could solve these problems by using the larvae of the black soldier fly (*Hermetia illucens*) to process organic waste and produce two valuable products: black soldier fly larvae, which could be used as chicken and fish feed, and the residue that can function as an organic fertiliser.

The results of this experiment show that a black soldier fly composting system can reduce the weight of the waste by 85%. The larvae have also shown to be very efficient in storing the organic waste as biomass in their body. Nearly 20% of the waste was converted to larval biomass. Lastly the prepupae, which is the stage where the larvae start the process of transforming into a fly, weighed around 220 mg each.

In this experiment, two feeding regimes (i.e. feeding a set amount of organic waste to a different amount of larvae in two systems) were compared. Eight parameters were measured in the two systems to identify the differences and the performance of black soldier fly composting in general. It was shown that the system where the larvae were fed with more organic waste per larva and day performed better because in the other system, more larvae died. The cause of the high mortality was probably the density of the larvae.

The results of the experiment could change the way organic waste is processed, especially in developing countries where there is currently no incentive for people to start separating their waste. If people would be able to process their organic waste with black soldier fly larvae and sell the larvae afterwards, the organic waste would become a valuable resource and a shift in the organic waste value-chain would be introduced.

Keywords: Black Soldier Fly, Black Soldier Fly Larvae, Organic waste, Waste Management, Insect Assisted Waste Management

Acknowledgement

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I want to thank my two supervisors, Cecilia Lalander and Stefan Diener, who were incredibly patient and have taught me so much. Spending the last two months writing my thesis, the daily work with them in the greenhouse seems like it took place in a another life: from unwrapping frozen faeces during our monthly “food sessions” and manually counting larvae with a pair of tweezers, from what seemed to be a never ending heap, to holding the dry residue and prepupae in your hand and seeing flies emerge from their pupae. It has been a great and unique learning experience.

I think Björn Vinnerås was right when he said that I “would have to work very hard but that I would get the best supervision possible.” I want to thank him for providing me with the opportunity to work with the team on this project. I believe that moment will have a large impact on my future.

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Table of contents

List of Tables	6
List of Figures	7
Abbreviations	8
1 Introduction	9
1.1. Problem description	10
1.2. Objectives	11
1.3. Data gathering	11
2. Theoretical background	12
2.1. The Black Soldier Fly	12
2.1.1. Generational cycle	12
2.1.2. Hygiene aspect	13
2.2. Rearing of and organic solid waste treatment by the black soldier fly	14
2.2.1. Rearing of the BSF	14
2.2.2. BSF composting system	14
2.3. Products	16
3. Materials and methods	17
3.1. Materials	18
3.2. Description of experimental set-up	19
3.3. Measurement Procedure	20
3.3.1. Sample collection	20
3.3.2. Physico-chemical analysis	20
3.4. Data analysis	22
3.4.1. Analysing the control point measurements	22
3.4.2. Calculations for the mass balance	22
4. Results	23
4.1. Parameters measured at the control points	23
4.1.1. pH	23
4.1.2. Compost maturity	23
4.1.3. TS and VS	24
4.2. Mass Balance of in- and outflows	25
5. Discussion	27

5.1.	Waste processing	27
5.2.	Product output	28
	5.2.1. Organic fertiliser	28
	5.2.2. Prepupae for animal feedstuff	29
5.3.	Outlook	29
6.	Conclusions	33
7.	Bibliography	34

List of Tables

Table 1: Input values and retention times for two continuous BSF composting systems with feeding regimes of 40mg dry food/larva/day (FR-40) and 60mg dry food/larva/day (FR-60)	20
Table 2: Combined CO ₂ and NH ₃ emissions and resulting compost maturity grades based on the Solvita [®] Respiration Test maturity index (Source: Official Solvita [®] Guideline)	21
Table 3: Compost maturity grading list and condition of compost based on the Solvita [®] Respiration Test maturity index (Source: Official Solvita [®] Guideline)	21
Table 4: Process parameter mean values and standard error (SE) for the two feeding regimes (FR-40: 40 mg/larva/day and FR-60: 60 mg/larva/day) at each Control Point (CP). Feeding regimes are based on dry food with larvae in a black soldier fly larvae plug flow system. CPs are distributed evenly over the system, CP1 being the beginning and CP5 the end of the system. Mean values followed by the same letter in the same row do not vary significantly (P<0.05). n indicates number of samples measured for each process parameter.	23
Table 6: Mean and SD from measurements (triplets) of the in- and outflow streams measured at random moments.	25
Table 7: Mean and standard deviation (SD) for parameter values of the two feeding regimes (FR-40: 40 mg/larva/day and FR-60: 60 mg/larva/day) based on the mass balances and the TS/VS measurement data.	26
Table 8: comparison of larval density as provided, the larval density with the +30% enumeration error and the larval density with survival rate correction for both feeding regimes (FR-40 and FR-60) in the continuous BSF composting systems	30

List of Figures

- Figure 1: Simplified overview of in- and outflows for the continuous black soldier fly larvae (BSFL) composting system used in this thesis project. 10
- Figure 2: Life cycle of the black soldier fly, *Hermetia illucens* in a controlled environment at 25 °C (Sheppard et al. 1995; Tomberlin and Sheppard 2002; Holmes et al. 2012; Tomberlin et al. 2009). Each block represents a stage change and includes the amount of days that have passed since the last stage change. 12
- Figure 3: Schematic overview of the plug flow reactor and the measurement points. 17
- Figure 4: Parameter values for the two feeding regimes (40 mg/larva/day, FR-40 and 60 mg/larva/day, FR-60) at each Control Point (CP). Feeding regimes are based on dry food inoculated with larvae in a black soldier fly larvae plug flow system. CPs are distributed evenly over the system, CP1 being the beginning and CP5 the end of the system. Points resemble the mean value and error bars the SE value: a) pH; b) compost maturity, ammonia (NH₃) and carbon dioxide (CO₂); c) total solids; d) organic content. 24
- Figure 5: The mass balance of the 40 mg/larva/day feeding regime (FR-40). The data represents 20 days of in- and outflow streams in the measurement period. 25
- Figure 6: The mass balance of the 60 mg/larva/day feeding regime (FR-60). The data represents 20 days of in- and outflow streams in the measurement period. 25
- Figure 7: Schematic side view of one of the Larvero used in this experiment (top) with one unit that was filled with material (green surface) and one Larvero where the new material (green) is spread over three units instead of filling one unit (bottom). 32

Abbreviations

BSF	Black Soldier Fly
BSFL	Black Soldier Fly Larvae
Larvero (pl. Larveros)	Continuous BSF composting system
FR-40	Feeding regime with 40 mg dry food/larva/day
FR-60	Feeding regime with 60 mg dry food/larva/day
Solvita®	Compost maturity test kit
TS	Total Solids
VS	Volatile Solids
CP	Control Point
SD	Standard Deviation
SE	Standard Error: divides the SD by the square root of the number of values the SD checks
SLU	Swedish University of Agricultural Sciences

1 Introduction

Municipal solid waste management has become a challenging undertaking due to an increase in living standard and rapid urbanization, especially in low- and middle-income countries (Sharholy et al. 2007). If not collected and properly treated, the organic solid waste fraction has proven to be a catalyst in spreading diseases by sheltering and feeding various disease vectors (Sharholy et al. 2007, Ali et al. 2012, Zakir Hossain et al. 2014) and have other negative environmental impact. In contrast to the separate collection of various waste streams in developed countries, most of the waste in developing countries ends up in open dumps or other more unfavourable locations. Although a general recycle or refund system for the inorganic solid waste fraction (e.g. plastics, glass, paper and metals) is not present in most developing countries, the materials are being collected, sold and recycled. So far, there have been no financial incentives for urban dwellers in developing countries to start separating and recycling organic solid waste, which makes up more than half, and in some countries over 90%, of the total waste mass in most countries and includes food waste, as well as garden waste and other plant materials (Komakech et al. 2014, Zakir Hossain et al. 2014).

Although composting and biogas production are two commonly used technologies that can recycle organic solid waste (Zakir Hossain et al. 2014), the former does not create a high value product and is unusable for urban dwellers while the latter is material and maintenance intensive and requires a rather large initial investment (Ali et al. 2012). At the same time, prices for chicken and fish feed from fishery resources are steadily rising due to overfishing of the oceans (Alder et al. 2008, Diener, Zurbrügg, et al. 2011, Cičková et al. 2014) and agricultural crop lands are being depleted of essential nutrients needed to support plant growth in many developing countries (Nwafor and Okorie 2005). A closed nutrient cycle has been identified as a key factor in supporting the needs of future human-generations (Rockström et al. 2013). Chemical fertilisers, used on a large scale, do not only create an imbalance in the nutrient cycle but are also causing a reduction in the soil quality, eutrophication and pollution by heavy metals (Zhu, Sun, et al. 2012, Owamah et al. 2013).

Organic solid waste management with the use of larvae of the black soldier fly (BSF), *Hermetia illucens* L. (Diptera: Stratiomvidae), is an alternative which is both able to sufficiently degrade organic material and create a financial incentive for urban dwellers to separate and recycle their organic solid waste (Diener, Studt

Solano, et al. 2011). The degradation of organic solid waste by the black soldier fly larvae (BSFL) has been demonstrated to be 40-75% (Diener et al. 2009, Zheng et al. 2012, Makkar et al. 2014). The larvae have protein and fat contents around 40% and 35%, respectively (Sheppard et al. 1995, Gobbi et al. 2013, Banks et al. 2014), and can be harvested and sold as chicken, fish or pig feed. At this moment, prices for fish- and soymeal, often used as feed for chicken, fish and pigs, are around \$2,150 and \$380 per metric ton, respectively (Index Mundi, Jan 2015). The waste residue can be used as an organic fertiliser (Newton et al. 2004, Kováčik et al. 2010, Lalander et al. 2013, Wang et al. 2013, Diener et al. 2014).

1.1. Problem description

Sufficient treatment of organic solid waste and valorisation of the product – the larvae that can be used as a chicken/fish feed and the residue that can be used as an organic fertiliser – are interdependent: organic waste has to be reduced optimally while the product has to be produced as efficiently as possible. To strengthen the interdependency, an optimal larval feeding regime – supporting the desired level of waste reduction while maintaining a high product output – has to be identified. Although research has been conducted on the topic of BSF composting, there is still a lack of understanding about the optimal feeding regime of the larvae. In addition, the systems for processing organic solid waste and producing larvae were so far based on a batch process. In this study, a continuous BSF composting system (Figure 1) was studied.

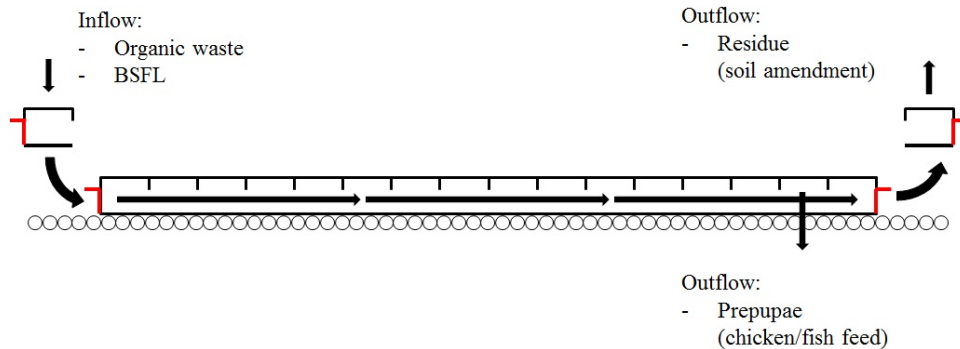


Figure 1: Simplified overview of in- and outflows for the continuous black soldier fly larvae (BSFL) composting system used in this thesis project.

1.2. Objectives

The hypothesis was that a continuous system would be less labour intense and would support the microbial community, which in turn would increase the material reduction and the conversion of waste into larval biomass by increasing the material retention time.

The overall objective of this thesis was to identify specific process parameters leading to optimised performance of a continuous BSF composting system. The specific research questions were:

- How does the BSF composting system and the two feeding regimes compare to existing BSF composting systems?
- What would be the preferred feeding regime in regards to the interest for either optimum waste reduction and/or optimum product output?

The specific objectives, set to answer the research questions, were to:

- Measure the effects of two different feeding regimes on specific process parameters (pH, compost maturity, total solids, organic content, larval weight, material reduction, biomass conversion rate and survival rate);
- Identify the process parameters showing a significant difference between the two feeding regimes;
- Assess the continuous BSF composting system for its organic solid waste reduction and product output.

1.3. Data gathering

The BSF research community is rather small and therefore, most of the literature was made accessible by my supervisors. However, the focus of this thesis is not on reviewing literature but on exploring the technology by testing the hypothesis presented above on a continuous BSF composting system developed in the framework of a research and development project by the Swedish University of Agricultural Sciences (SLU) and the Swiss Federal Institute of Technology (ETH). The results of the eight measured process parameters (mentioned above) will be the main data source for answering the research questions.

2. Theoretical background

2.1. *The Black Soldier Fly*

The BSF originates from southern parts of the USA and South America and, through human activity, is now spread over the tropic, sub-tropic and warm temperate regions of the world (Sheppard et al. 1995, Makkar et al. 2014). The reason why BSF adapt so easily in other regions is because of their wide range of habitats (i.e. food sources). The larvae can feed on various manures, human excreta, rotting fruits, vegetables and carcasses (Sheppard et al. 1995, Banks et al. 2014). Another benefit of the BSFL is that they are resilient when it comes to availability of food. When food is in abundance, a larva can develop into an adult in about two weeks. However, this process can take up to four months when food is in short supply (Sheppard et al. 1995). Also the type of food or the food mixture can be of influence when looking at the adult life-history traits such as size, duration of larval stages and mortality (Gobbi et al. 2013).

2.1.1. Generational cycle

The life cycle of the BSF including the five stage changes throughout its typical 45 day life (Figure 2). As described above, the time before another stage change is heavily dependent on the availability of food and the temperature.

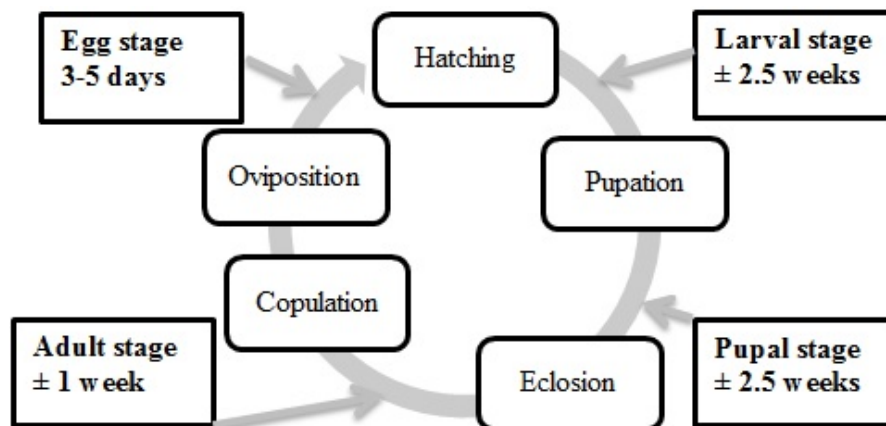


Figure 2: Life cycle of the black soldier fly, *Hermetia illucens* in a controlled environment at 25 °C (Sheppard et al. 1995; Tomberlin and Sheppard 2002; Holmes et al. 2012; Tomberlin et al. 2009). Each block represents a stage change and includes the amount of days that have passed since the last stage change.

Four days after oviposition, the event in which the female lays around 500 eggs (Sheppard et al. 1995, Holmes et al. 2012), the BSF larvae hatch. The BSFL take around 2.5 weeks to grow and crawl out of their food source as prepupae. This stage can, however, take up to four months depending on the availability of food and the temperature (Sheppard et al. 1995). The larvae have six laval stages (instars), in the sixth instar, called the prepupal stage, the colour change of the skin from white to dark-brown (May 1961). The mouth of the larvae is replaced with a hook that is used to climb out of the food source to find a suitable location for pupation (Tomberlin et al. 2009). Pupation is the process where the pupae transform into flies. The pupation location is mostly a dark cavity where the pupae will not fall prey while pupating. In many cases, the prepupae will dig itself in soil with the help of its hook (Sheppard et al. 1995). Eclosion, where the fly emerges from its pupa, occurs around 2.5 weeks after the start of the pupation (Tomberlin et al. 2009). As with the larval stage, this process can take much longer (up to five months) depending on the temperature (Furman et al. 1959, Sheppard et al. 1995). As an adult, the BSF is not a nuisance to humans: it is not an active flyer like the house fly is (Diener, Zurbrügg, et al. 2011) and it is mostly resting on vegetation. It does not seem to enter human dwellings and tries to avoid contacts with any animal at all (Čiřková et al. 2014). The flies mate and copulate about two days after eclosion. Within two days after copulation, the female finds a dark and narrow cavity near a food source to lay a new batch of eggs (Tomberlin and Sheppard 2002, Tomberlin et al. 2009, Holmes et al. 2012). Female flies die within a few hours of oviposition (Tomberlin and Sheppard 2002). Males on average live around three days longer than females (Tomberlin et al. 2009).

2.1.2. Hygiene aspect

As the BSF only comes in contact with the food source at one point, the possible pathogen transmission moment during the life of the BSF are limited. Adult black soldier flies do not feed and solely depend on their fat reserves. Therefore, the BSF is not considered a vector of diseases (Diener et al. 2009, Banks et al. 2014). Oviposition does not take place in the food source because of the relatively long hatching time of 3-5 days and the vulnerability of the eggs to fungal growth (Banks et al. 2014).

Pathogens, disease causing microorganisms (e.g. *Salmonella* spp.), can be found in faecal matter of infected humans and animals. The BSFL have, however, shown to heavily affect the composition of organisms in the material. *Salmonella* spp., have been found to be suppressed by larval activity (Erickson and Islam 2004, Liu et al. 2008, Lalander et al. 2013), while no such inactivation have been seen for the intestinal parasite *Ascaris* spp. Furthermore, it has been found that the presence of the common house fly, *Musca domestica* L., and the lesser house fly, *Fannia canicularis* L. (Diptera: Muscidae), is reduced up to 97% when there is BSFL activity in the material (Furman et al. 1959, Sheppard 1983). A factor that can reduce organic waste associated disease transmission, as these flies are known vectors of diseases.

2.2. Rearing of and organic solid waste treatment by the black soldier fly

The BSF has shown to survive well in a controlled (e.g. temperature, light, caged *etcetera*) environment (Myers et al. 2008, Holmes et al. 2012, Gobbi et al. 2013). BSF rearing in this thesis refers to the process of the pupation, eclosion of the adults, mating by the adults, oviposition, hatching of the eggs and the hatchling nursery. The BSF composting system refers to the system in which organic waste is converted into prepupae and treatment residue.

2.2.1. Rearing of the BSF

Rearing of the BSF starts by providing a suitable climate for the adults. Especially at the point of oviposition, the temperature needs to be high. In non-tropical regions, a greenhouse is mostly used to maintain an ideal climate for BSF development (Tomberlin and Sheppard 2002, Diener, Studt Solano, et al. 2011).

The first step in the rearing process is concerned with providing a pupation medium for the prepupae that have been collected from the BSF composting system. Prepupae need a pupation medium to bury in following their instinctive need for a “safe” place for pupation. Previous studies have identified potting soil and wood shavings to facilitate the fasted pupation process (Holmes et al. 2013).

After the adults emerge, they are mostly collected in cages where water and artificial light is provided (Tomberlin and Sheppard 2002, Diener, Studt Solano, et al. 2011, Gobbi et al. 2013, Nguyen et al. 2013). In this confined space, the adults mate by in-air connection. The male has a grabbing arm that is used to “grab” the female while both are flying. The couple then finds a location to complete the copulation process (Sheppard et al. 2002).

Oviposition takes place mainly in cavities of cardboard unit provided in the fly cages and sometimes in combination with a food source (Sheppard et al. 2002, Tomberlin and Sheppard 2002, Diener, Studt Solano, et al. 2011, Gobbi et al. 2013). This part of the process is heavily influenced by temperature; Sheppard et al. (1995) has found the ideal temperature is between 27.5 °C and 37.5 °C. In addition, the oviposition site is shaded, to provide the adults with a seemingly safe place to lay their eggs (Tomberlin and Sheppard 2002).

The hatchlings crawl out of the egg and fall onto food that is provided to make them grow to a certain size. In most experimental set-ups the larvae have been kept in the food for 4-6 days before transferring them to the waste source (Tomberlin and Sheppard 2002, Diener, Studt Solano, et al. 2011).

2.2.2. BSF composting system

Designs of BSF composting systems vary and can be operated either in batches, where a batch of larvae is being fed until harvested, or in a continuous mode, where waste material and young larvae are continuously added to a system on one end while removing residue and grown prepupae on the other end. An important factor when supplying the larvae with a food source is the maximum height of the material: the material in the container must not exceed a height of 7.5-10 cm be-

cause lower layers can become anaerobic and therefore unsuitable for the BSFL (Čičková et al. 2014). In systems taking advantage of the migratory habit of the prepupae, ramps with a slope ranging from 28° to 45° were used to provide the prepupae with an “escape” route (Sheppard et al. 1995, Newton et al. 2004, St-Hilaire et al. 2007, Diener, Studt Solano, et al. 2011, Diener, Zurbrügg, et al. 2011).

The average retention time of the larvae depends on many factors such as age at inoculation, type of material and temperature of surrounding. On average, it has been found to be around ten days (Diener et al. 2009, Tomberlin et al. 2009, Yu et al. 2011, Banks et al. 2014). As they feed, some changes can be seen. First, the larvae grow as the food is converted into larval biomass; the efficiency of this process is shown by the waste-to-biomass conversion rate. In addition, tests have shown that pre-inoculation of the material with the BSFL gut bacteria improves the biomass conversion rate as well as the larval growth rate (Yu et al. 2011). Banks et al. (2014) calculated the biomass conversion rate and came to values ranging from 15-23% on wet weight basis, while Lalander et al. (2014) realised a biomass conversion rate of 11.8% on dry matter basis. Secondly, the substrate colour and texture changes. Although there has not been much research done on this topic, the natural evaporation, larval activity and introduced microbial community are the main cause for the colour and texture change (Beard and Sands 1973). The change seems to be similar for each food source and can be described as a dark, dry, odorous and granular material (Kováčik et al. 2010, Diener, Zurbrügg, et al. 2011, Zhu, Wang, et al. 2012, Wang et al. 2013). Other changes in the substrate, not directly observable, are an increase in the pH (going into a more alkaline direction) and an increase in the ammonia (NH₃) and carbon dioxide (CO₂) emissions. The emissions will gradually lower again as the substrate becomes more dry and dark (Beard and Sands 1973, Zhu, Wang, et al. 2012, Wang et al. 2013).

After the BSFL have emptied their gut and crawled out of the material as prepupae, they can be collected using the ramp with a container attached (Makkar et al. 2014). This is a good moment for harvesting because the prepupae, in their search for a dark spot to pupate, will make their way into the collection container without assistance and thus no effort is required in collecting them (Diener, Studt Solano, et al. 2011).

The rate of survival from larvae to flies in the process has been found to be 74-97% (Tomberlin et al. 2009). Yu et al. (2011) divided the larval and adult stages and reported that around 97% of the larvae crawl out as prepupae. Meyers et al. (2008) fed larvae with dairy manure and came to a survival rate of 80%.

Material reductions ranging 40-75% have been reported; the variation have been found to be depending on the type of material, temperature and larval density (Diener et al. 2009, Zheng et al. 2012, Lalander et al. 2014, Makkar et al. 2014). The treatment residue, of which the texture is described above, remains after the process.

For it to be used as an organic fertiliser, it needs post-treatment due to pathogen loads (Čičková et al. 2014). This applies however to other organic waste treatment systems as well (Alfa et al. 2014). An important factor is the water con-

tent that has to be between 50-60% to ensure aeration of the processed material (Das and Keener 1997).

2.3. Products

The prepupae, rich in protein and fat, can be used as an animal feedstuff. How rich the prepupae are in protein and fat seems not to depend too much on the food source or any other variable because the content is very similar in the various experiments done. Protein content is around 40% and fat content around 35% (Hale 1973, Sheppard et al. 1995, St-Hilaire et al. 2007, Diener et al. 2009, Gobbi et al. 2013). What seems to vary a lot is the weight of the prepupae. This ranges from a low of 48 mg dry weight per prepupa (Diener et al. 2009) to a high of 95 mg dry weight per prepupa (Yu et al. 2011). The prepupae feedstuff can be used as chicken and various fish feeds (Newton et al. 2004, Zering 2006, Diener et al. 2009, Agrawal et al. 2011, Wang et al. 2013).

The other product, the treatment residue, has a more widespread application as an organic fertiliser. The organic fertiliser can be placed in the market at a lower cost than the currently wide spread chemical fertiliser due to the wide range of (organic) waste products that could potentially be processed into organic fertiliser (Zhu, Sun, et al. 2012).

3. Materials and methods

In this study, a continuous BSF composting system was established and monitored (Figure 3,) referred to as *Larvero*. Two of these systems were compared to assess the effect of two different feeding regimes on specific parameters. Except for the amount of larvae added (resulting in different feeding regimes), the two systems were identical.

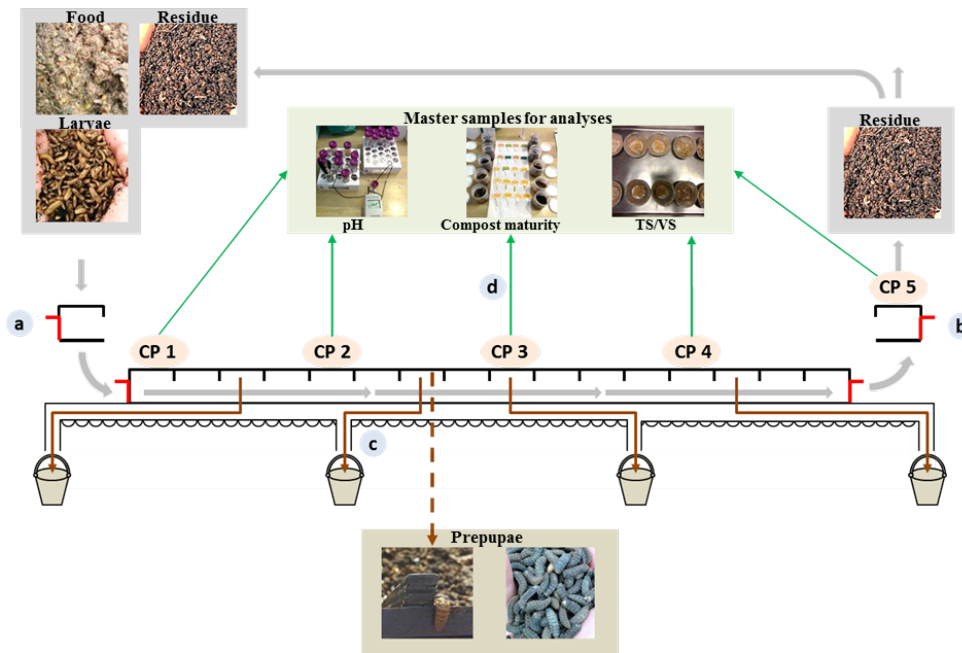


Figure 3: Schematic overview of the plug flow reactor and the measurement points.

- a: Unit with mixture of food and recycled residue is added to the *Larvero*. Larvae are inoculated.
- b: Last unit is detached and removed. Residue in the unit is sieved and partly recycled.
- c: Prepupae crawl out of the *Larvero*, in the buckets and are harvested from there.
- d: At five control points (CP) spread over the *Larvero*, master samples are taken.

The system worked as a plug-flow reactor. Organic solid waste, mixed with recycled residue, was fed into the first of a total of 17 units and larvae were added on the top. The surface area of one unit was 25x55 cm and the mixed material reached 5 cm in height giving it a volume of 6.8 L. The last unit, containing the residue, was detached and removed. The *Larvero* was then manually pushed over the conveyor rollers. As the units were pushed from one end to the other, larvae processed the material. The prepupae crawled out of the *Larvero* in search for a dark place to pupate. The prepupae eventually ended up in the buckets from where they were harvested.

In addition to the measurements taken from the in- and outflows for the mass balance, master samples were taken from five control points (CP). The points were distributed evenly over the *Larvero* and assigned to the first, last and the units with an increment of three units between the first and last unit. The samples were taken

before a new unit was added. From these master samples, subsamples for measuring the pH, compost maturity, total solids (TS) and organic content - the total volatile solids (VS), were taken.

3.1. Materials

The system was fed with a mixture of food waste (95%) and human faeces (5%). Food waste was obtained from a local restaurant on the SLU campus in Uppsala, Sweden. The restaurant serves a set variety of dishes. From observing the food waste, the main fraction was carbohydrates from potatoes and pasta. An unexpected large quantity of meat and fish was found in the food waste. The remainder consisted of vegetables, fruit, beans, sandwiches and bread. Although the restaurant has a strict waste separation system, some plastic was found in the food waste. It was mostly used to wrap the sandwiches and some packaging materials disposed of in the wrong way. The latter was removed without much effort but the sandwich wrapping material mostly stayed in the mixture. This raw mixture was minced using a heavy duty grinder. Human faeces (5% w/w) were added in order to provide indicator organism for a parallel experiment focussed on hygiene aspects of BSF composting. The faeces were collected in plastic bags and stored at -20 °C until use. The faeces were left to thaw at 27 °C for 24 hours prior to mixing, which was done in a concrete mixer for 5 min. The water content of the mixture was around 75% and caused the density to be similar to water (i.e. 1 kg = 1 L). The mixture was divided into fractions of 3.4 kg, bagged and stored at -18°C.

The larvae were produced at an onsite rearing facility. Although this facility was running at the start of the experiment, it was not able to provide enough larvae after the first week. Larvae were therefore bought from Bioflytech, a rearing facility in Spain, and from the Research Institute of Organic Agriculture FiBL in Switzerland.

Materials used to quantify flows included a two-decimal precision balance (Kern KB 2000-2NM), tools for enumerating larvae and prepupae and sieves with various diameters to screen the residue and self-harvested prepupae. The four measured parameters were pH, compost maturity, TS and the VS (also referred to as organic content). For the latter two, a drying oven and a muffle furnace were used, respectively. The pH was measured using a pre calibrated travel meter (VWR pH-110). The compost maturity was measured using the Solvita[®] Respiration Test. In addition, the temperature of the Larveros facility and material was constantly measured using temperature loggers (Tinytag plus 2 TAP-4017).

3.2. Description of experimental set-up

The Larveros facility and the rearing facility were located in two separated rooms within a greenhouse at the campus of SLU in Uppsala, Sweden, and were kept at a constant temperature of 21 °C and 27 °C, respectively. Material was added daily on the first unit (Figure 3) and as there were 17 detachable units making up the Larvero the material stayed in the Larvero for 17 days, resulting in a *material retention* time of 17 days (Table 1). The Larveros had a self harvesting system, using a ramp as described above, where the prepupae crawled out of the larveros and drop into a gutter that was placed on both sides over the entire length of the Larveros. The average retention time of the larvae, described as the *larval retention* time, was assumed to be 10 days (The only variable that differed between the two Larveros was the amount of larvae added. In the 60 mg dry food/larva/day feeding regime (FR-60), 2750 larvae were added; while 4150 larvae were added in the 40 mg dry food/ larva/day feeding regime (FR-40) (Table 1). In both regimes, 6.8 kg food waste was added daily. As a result of the difference in larvae added, the larval densities differed (Table 1). The larval density is based on the 6.8 kg (1:1 volume to weight ratio) food ratio, not taking into account the recycled residue.

Table 1) based on literature (Diener et al. 2009, Tomberlin et al. 2009, Banks et al. 2014). On each side, the gutter had four buckets attached into which the prepupae fell, that could be detached and emptied (Figure 3). The last unit, that was removed daily, contained the residue (Figure 3). The residue was sieved and separated into three fractions. The first fraction was a dry black granular compost material that, according to the hypothesis, holds the microbial community which facilitates the processing of the material. Part of this fraction (around 10% of the food in wet weight) was added to the food in order to support the microbial activity. At the start of the measurement period, the weight of the residue out was not sufficient to supply enough residue for recycling because of the lower amount of food added in the very beginning and the long retention time. The lower amount of residue recycled had to be compensated for in the following weeks. The second fraction was the light residue, which was made up of plastic wrappings, paper and stickers, cellulose material and shed larval skins. The weight of the not-recycled fraction of the residue combined with the weight of the light residue made up the total residue outflow. The third fraction was the prepupae that did not crawl out of the system but stayed in the residue. The weight of this fraction combined with the weight of the self-harvested prepupae made up the total prepupal outflow.

The only variable that differed between the two Larveros was the amount of larvae added. In the 60 mg dry food/larva/day feeding regime (FR-60), 2750 larvae were added; while 4150 larvae were added in the 40 mg dry food/ larva/day feeding regime (FR-40) (Table 1). In both regimes, 6.8 kg food waste was added daily. As a result of the difference in larvae added, the larval densities differed (Table 1). The larval density is based on the 6.8 kg (1:1 volume to weight ratio) food ratio, not taking into account the recycled residue.

Table 1: Input values and retention times for two continuous BSF composting systems with feeding regimes of 40mg dry food/larva/day (FR-40) and 60mg dry food/larva/day (FR-60)

	Unit	FR-40	FR-60
Mass of food (wet)	kg	6.8	6.8
Mass of food (dry)	kg	1.7	1.7
Number of Larvae	ea.	4,150.0	2,750.0
Larval density	Larvae/dm ³	610.0	400.0
Larval retention time	Days	10.0	10.0
Material retention time	Days	17.0	17.0

The feeding regime (F_r), - defined as mg dry food/larva/day - was calculated as:

$$F_r = \left(\frac{F_{in}}{l} \right) / t_R \quad (1)$$

where F_{in} was the dry weight of the food (mg), l the amount of larvae and t_R the average larval retention time (days). The recycled residue was not taken into account in this calculation, as the residue was recycled within the system and thus did not add to the total amount of material in the Larveros when steady-state has been reached.

3.3. Measurement Procedure

3.3.1. Sample collection

Master samples were taken from five control points in each Larvero (Figure 3). The master samples served as the main sample from which smaller samples for the specific measurements were taken. The first samples were taken after the Larveros were in operation for 27 days. The last samples were taken 20 days later. The measurement period for the mass balance (i.e. in- and outflows of the system) was the range between the first and last sample taken: 20 days. After weighing the master samples for the mass balance, the samples were divided into five fractions. The fractions were used to measure pH, compost maturity, TS and VS. One fraction was bagged and stored in the freezer for later analysis.

3.3.2. Physico-chemical analysis

For the pH measurement, the sample was diluted with deionized water at a ratio of 1:4, shaken and left for one hour prior to analysis.

The compost maturity was measured using the Solvita® Respiration Test. It was conducted following the protocol provided by the supplier. The stage of maturity results from the combined NH₃ and CO₂ emissions of the material (

Table 2) and is expressed in eight grades (Table 3). A high grade means the compost is mature, a low grade indicated there is still a lot of respiration of carbon dioxide (CO₂) and ammonia (NH₃) in the compost and it is therefore still in an immature stage. The inflow material (CP1) was too wet to comply with the pre-conditions for the test and was therefore not included in the analysis.

Table 2: Combined CO₂ and NH₃ emissions and resulting compost maturity grades based on the Solvita[®] Respiration Test maturity index (Source: Official Solvita[®] Guideline)

		Carbon Dioxide (CO ₂) grade							
		1	2	3	4	5	6	7	8
Ammonia (NH ₃) grade	No/Very low	No	Very low	Low	Medi-um	Medi-um	Med/Hi gh	Hig h	Very high
		5	No/Very low	1	2	3	4	5	6
4	Low	1	2	3	4	5	6	7	8
3	Medium	1	1	2	3	4	5	6	7
2	High	1	1	1	2	3	4	5	6
1	Very high	1	1	1	1	1	2	3	4

Table 3: Compost maturity grading list and condition of compost based on the Solvita[®] Respiration Test maturity index (Source: Official Solvita[®] Guideline)

Grade	Compost description	Maturity group
8	Inactive, highly matured	Finished compost
7	Well matured, aged	
6	Curing	Active compost
5	Ready for curing	
4	Moderately active	
3	Active	
2	Very active	Raw compost
1	Fresh, raw	

The TS was done at 105 °C for 24 hours and the VS at 550 °C for four hours.

All the material added to and removed from the system were weighed and noted. The amount of larvae added to the system were sorted using a set of sieves with mesh sizes 2 mm, 1.0 mm and 0.6 mm and then counting the number of larvae in 10 or 15 ml (depending on the size of the larvae). The prepupae were enumerated daily by counting and weighing 200 prepupae and dividing the total weight by the average weight of one prepupa.

Temperature meters were placed in the Larveros facility and in the material of both Larveros to measure the temperature in the facility and in the material whilst being processed by the larvae.

3.4. Data analysis

3.4.1. Analysing the control point measurements

The statistical software programme R (R Core Team, 2012) was used to calculate the mean, standard deviation and standard error of the data sets. Significance was measured within a 95% confidence interval for the t-test and ANOVA ($P < 0.05$).

3.4.2. Calculations for the mass balance

The percentage biomass conversion rate (BCR) on a TS basis was calculated as:

$$BCR = \left(\frac{M_{pp} - M_l}{F_{in}} \right) * 100 \quad (2)$$

where M_l was the dry weight of the larvae inflow and M_{pp} the dry weight of the prepupae outflow. This was then divided by F_{in} , the dry weight of the inflow material (mg).

The percentage survival rate (S_r) was calculated as:

$$S_r = \left(\frac{pp}{l} \right) * 100 \quad (3)$$

where pp was the number of prepupae out on each day, l the number of larvae added each day.

The percentage material reduction (MR) on a TS basis was calculated as:

$$MR = \left(1 - \left(\frac{M_{out}}{M_{in}} \right) \right) * 100 \quad (4)$$

where M_{out} was the total solids of the material outflows and M_{in} the total solids of the material inflow. The outflow material was made up of the residue, not taking into account the residue that is recycled in the system, and the dry weight of the master samples. The material reduction is the amount of organic that either has been transformed into volatile compounds, such as CO_2 or NH_3 , by biological processes, or is stored as biomass in the adult larvae that are removed from the system as prepupae.

4. Results

Two plug-flow continuous systems – operating under two different feeding regimes (FR40 and FR-60) – were set-up and monitored over a period of 20 days.

4.1. Parameters measured at the control points

None of the parameters measured at the CPs of the two feeding regimes resulted in significant differences between the two feeding regimes (Table 3). However, between the CPs (along the treatment line), significant differences in the values were found.

4.1.1. pH

The pH significantly increased between CP 2 and 4, where the value increased from pH 5 for both FR-40 and FR-60 to pH 8 and 7.8 for FR-40 and FR-60, respectively (and Figure 4a).

4.1.2. Compost maturity

No significant development in compost maturity occurred within the feeding regimes. Towards CP 5, there was a trend of ($P>0.05$) an increase in material maturation. In FR-60, the NH_3 and CO_2 emissions showed a significant increase (NH_3) and decrease (CO_2) between CP 3 and 4 and CP 2 and 5, respectively (and Figure 4b).

Table 4: Process parameter mean values and standard error (SE) for the two feeding regimes (FR-40: 40 mg/larva/day and FR-60: 60 mg/larva/day) at each Control Point (CP). Feeding regimes are based on dry food with larvae in a black soldier fly larvae plug flow system. CPs are distributed evenly over the system, CP1 being the beginning and CP5 the end of the system. Mean values followed by the same letter in the same row do not vary significantly ($P<0.05$). n indicates number of samples measured for each process parameter.

	Regime	n	CP 1		CP 2		CP 3		CP 4		CP 5	
			Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
pH	FR-40	7	4.6a	0.1	5.0a	0.2	6.6b	0.3	8.0c	0.1	7.9c	0.1
	FR-60	7	4.5a	0.1	5.0ab	0.3	5.9b	0.3	7.8c	0.1	7.8c	0.1
Compost maturity	FR-40	4	NA	NA	1.0	0.0	1.0	0.0	1.3	0.3	2.0	0.4
	FR-60	4	NA	NA	1.5	0.3	1.5	0.3	1.5	0.3	2.0	0.4
Ammonia (NH_3)	FR-40	4	NA	NA	5.0	0.0	4.5	0.5	3.0	0.9	3.8	0.3
	FR-60	4	NA	NA	5.0a	0.0	4.8a	0.3	2.5b	0.7	2.8b	0.3
C. dioxide (CO_2)	FR-40	4	NA	NA	1.0	0.0	1.0	0.0	2.3	0.3	2.3	0.3
	FR-60	4	NA	NA	1.3a	0.3	1.5ab	0.3	2.5ab	0.5	3.0b	0.0
TS (%)	FR-40	8	29.4a	0.7	31.4a	1	31.8a	0.7	38.1b	1.1	48.5c	2.9
	FR-60	8	31.0a	0.8	31.6a	0.9	33.2a	0.9	38.5b	1.6	52.7c	1.7
Org. con. (% TS)	FR-40	8	87.4a	0.8	85.6ab	0.5	84.7ab	0.5	81.8bc	0.8	80.7c	0.4
	FR-60	8	86.0a	0.6	85.6a	0.8	86.0a	0.5	83.1ab	0.4	81.3b	0.7

4.1.3. TS and VS

The percentage of total solids increased significantly between CP 3 and CP 5 for both regimes. The value increased from around 20% to around 50% in both regimes (and Figure 4c), while the organic content showed a small, however, significant decrease between CP 3 and CP 5. The value decreased from around 86% to 82% for both FR-40 and FR-60 (and Figure 4d).

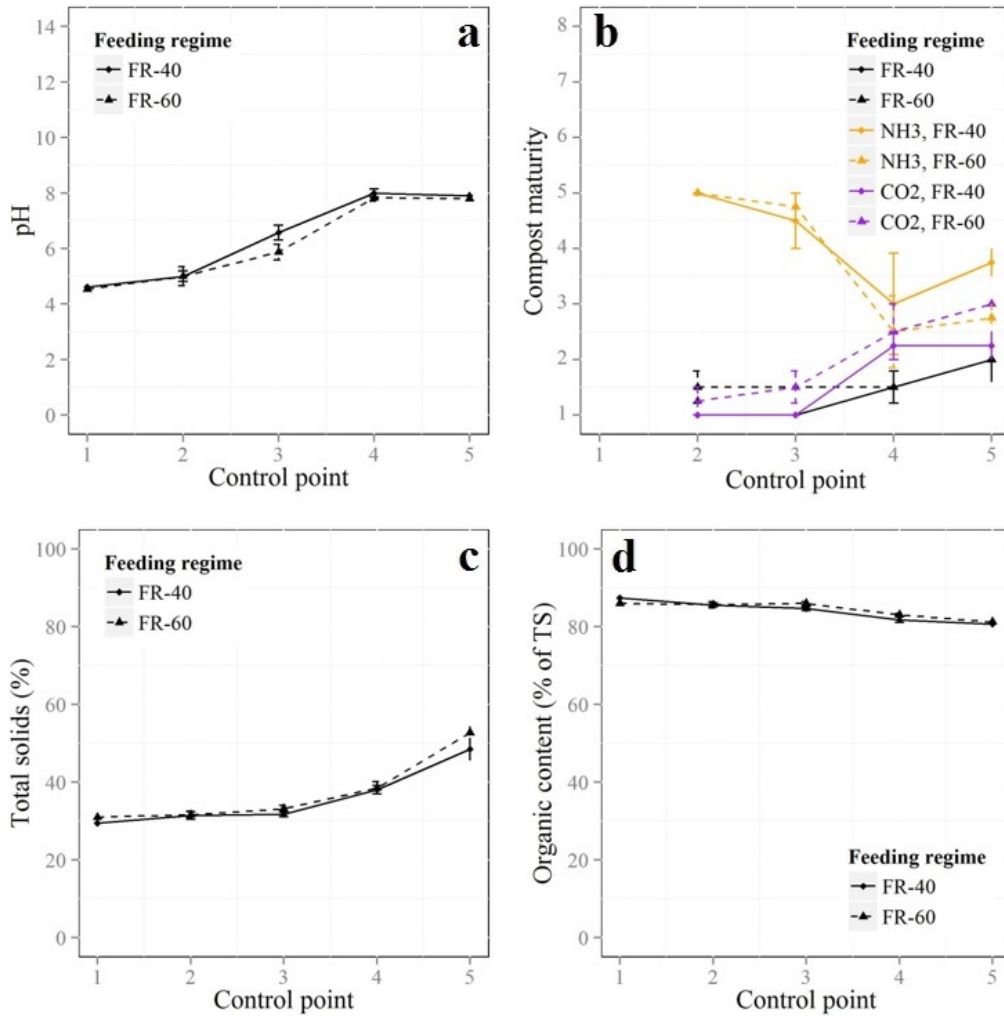


Figure 4: Parameter values for the two feeding regimes (40 mg/larva/day, FR-40 and 60 mg/larva/day, FR-60) at each Control Point (CP). Feeding regimes are based on dry food inoculated with larvae in a black soldier fly larvae plug flow system. CPs are distributed evenly over the system, CP1 being the beginning and CP5 the end of the system. Points resemble the mean value and error bars the SE value: a) pH; b) compost maturity, ammonia (NH₃) and carbon dioxide (CO₂); c) total solids; d) organic content.

4.2. Mass Balance of in- and outflows

The total solids and organic content measurements of the in- and outflows (Table 5) were used to calculate the flows of the mass balance (Figure 5-Figure 6). The results are displayed in Table 6.

Table 5: Mean and SD from measurements (triplets) of the in- and outflow streams measured at random moments.

	TS (%)		VS (% TS)	
	Mean (n=3)	SD	Mean (n=3)	SD
Food	24.2	1.8	90.4	3.4
Residue (FR-40/FR-60)	48.5/52.7	8.3/4.8	80.7/81.3	1.0/2.0
Larvae	23.9	0.6	91.6	1.7
Prepupae	41.0	0.8	81.6	4.1

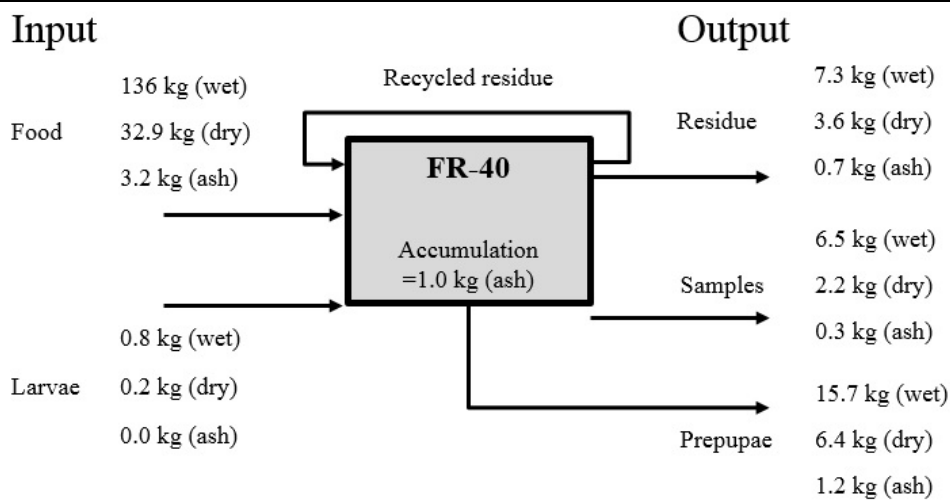


Figure 6: The mass balance of the 40 mg/larva/day feeding regime (FR-40). The data represents 20 days of in- and outflow streams in the measurement period.

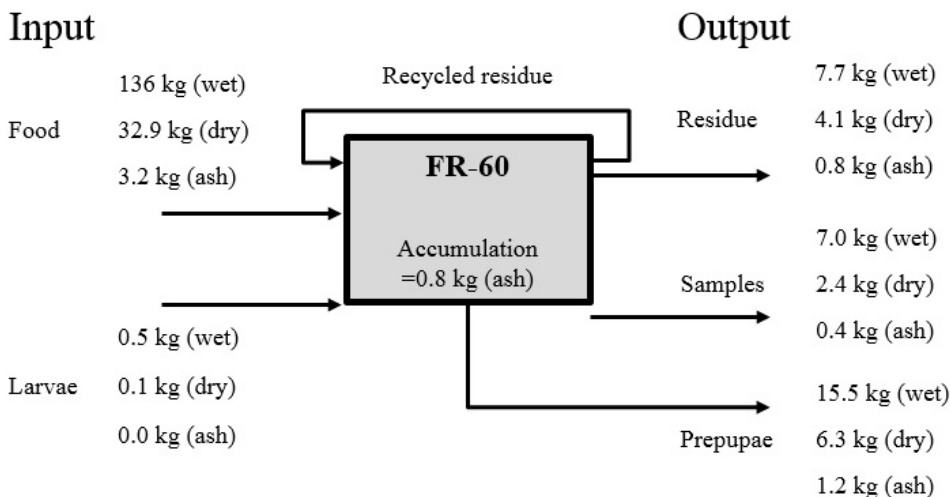


Figure 5: The mass balance of the 60 mg/larva/day feeding regime (FR-60). The data represents 20 days of in- and outflow streams in the measurement period.

An accumulation of material in both Larveros occurred (Figure 5 -Figure 6). The accumulation of FR-40 (1.0 kg ash) was somewhat higher than the accumulation in FR-60 (0.8 kg ash).

The weight of the prepupae harvested from the Larveros showed an insignificant difference between the two feeding regimes. FR-40 had an average wet and dry weight of 221 mg and 91 mg, respectively. With an average wet and dry weight of 224 mg and 92 mg respectively, FR-60 had nearly the same values (Table 6).

The survival rate of both feeding regimes shows a significant difference between the two feeding regimes (Table 6). In FR-40, 70% of the larvae added were eventually harvested as prepupae. The survival rate was higher for FR-60, where 100% of the larvae were eventually harvested as prepupae.

The material reduction shows similar results between the two feeding regimes (Table 6). In both feeding regimes, around 85% (wet weight basis), 69% (total solids basis) and 71% (organic content basis) of the food was converted into volatile compounds or built into larval biomass.

There was no significant difference in the biomass conversion rate between the two feeding regimes (Table 6). With a 19% conversion of the (dry) food into larval biomass, FR-40 and FR-60 gave the same results.

Table 6: Mean and standard deviation (SD) for parameter values of the two feeding regimes (FR-40: 40 mg/larva/day and FR-60: 60 mg/larva/day) based on the mass balances and the TS/VS measurement data.

	unit	FR-40		FR-60	
		mean	SE	mean	SE
Prepupae weight (ww basis)	mg/PP	221	2.5	224	2.9
Prepupae weight (TS basis)	mg/PP	90.6	1.0	91.9	1.2
Survival Rate	%	69.9	9.9	99.7	18.9
Material Reduction (ww basis)	%	84.5	NA	85.5	NA
Material Reduction (TS basis)	%	68.8	NA	68.4	NA
Material Reduction (org. con. basis)	%	71.1	NA	70.8	NA
Biomass Conversion Rate	%	19.0	2.3	18.9	3.4

5. Discussion

5.1. Waste processing

The material reduction was calculated using Equation 4, but the values were calculated based on the ash balance because of the accumulation in the systems (Figure 5 -Figure 6). The accumulation was assumed to be part of the residue that would normally have left the system. The accumulation indicated that the systems had not reached steady state. The residue that was recycled in order to support the microbial community did not reach the last unit again when the measurements started. It took 24 days to fill the 17 units of both Larveros with substrate and larvae (in the beginning, the food added was less than 6.8 kg/day and was therefore added in the same unit). From that day, the last unit containing residue was taken out and the residue was (partly) recycled. Thus, only 17 days after this moment was there a steady-state in the system.

With a material reduction (total solids basis) of around 70% for both regimes, this continuous BSF composting system had a high waste reduction compared to what has been reported previously, ranging from 40-55% (Diener et al. 2009, Zheng et al. 2012, Lalander et al. 2014). Although Diener et al. (2009) and Lalander et al. (2013) used different feeds for the larvae, which could explain the lower reduction, Zheng et al. (2012) worked with restaurant waste too. However, in the experiments of Zheng et al. (2012), the grease was separated from restaurant waste in and this could have influenced the material reduction. Diener et al. (2011) fed larvae with organic solid waste. Two different feeding regimes and two different feeding methods (on top of the residue or mixed in with the residue) resulted in four trials. A similar reduction was reached in the system when applying a high amount of organic waste on top of the residue. The prepupae in that trial had the same weight as the prepupae in this experiment. Although a high reduction was realised in this system, the material was not stabilised (Table 3).

Anaerobic digestion, a technology where organic waste can be transformed into biogas, achieves a higher reduction of the organic content (Grimberg et al. 2015), around 95%, than the organic content reduction in this experiment (70%) and has a similar dry weight reduction of around 60-75% (Alfa et al. 2014). The difference in organic content reduction is probably due to the preference or ability of the consumer organism to process various compounds in the waste. The micro-organisms in a digester are able to convert nearly everything that can be converted while the BSFL associated micro-organisms does not appear to be able to: the organic content of the material does not change significantly in either of the two regimes (Figure 4d). However, other BSF composting systems have shown to be able to affect the organic content in the system; e.g. Lalander et al. (2014) realised a significant decrease of 14 percentage points in the organic content in a system that fed larvae with a mixture of pig manure, dog food and human faeces. This difference is probably related to the lower feeding regime (less food per larva) used in that system and the material retention time. In addition, anaerobic digestion requires the material to be transportable through pipes in order to maintain a closed system. Therefore, the material has to be diluted to sludge in order to make

it suitable for transport (Alfa et al. 2014). This causes the digestate (residue of an anaerobic digester) to be very wet (Owamah et al. 2014) and the wet weight reduction low. In this system, a wet weight reduction of 85% was achieved. This is of particular interest from a waste management perspective, where the total weight and volume reduction are indicators of a desirable waste processing technology.

5.2. Product output

5.2.1. Organic fertiliser

The material over the whole length of both Larveros can be classified as raw, very active compost (Table 3 and Figure 4b). In Figure 4b the compost maturity development is shown. In FR-60, a significant increase of the ammonia emissions (higher grade is related to lower emissions) was found. This significant increase is in accordance with other studies, which mentions increased emissions of ammonia as one of the indicators of compost activity (Erickson and Islam 2004). Observations of the two Larveros are underlined by the observations of Wang et al. (2013), who fed pig manure to house fly larvae, and stated that there was a constant strong odour released by the systems related to the ammonia emissions. Two indicators, ammonia and carbon dioxide, combined result in the compost maturity (Table 2) and although it seems that ammonia was emitted according to observations and analysis, the carbon dioxide indicator did not show a strong increase (Figure 4b). This indicates that although the residue has an earth-like appearance, similar to residue described in literature (Kováčik et al. 2010, Diener, Zurbrügg, et al. 2011, Zhu, Wang, et al. 2012, Wang et al. 2013), it is still active and has a high water content (50%). In addition, it was found by a parallel experiment analysing the pathogen load in the system at various places, that pathogens were still present in the residue. The residue thus needs post-treatment before it can be used as a fertiliser (Cíčková et al. 2014). Post-treatment with thermophilic aerobic composting has shown to not only reduce the residue volume increasing the concentration of useful compounds for plants and reduce the overall phytotoxicity of the compost (i.e. toxic effect of a compound on a plant) but due to the high temperatures reached also reduces pathogenic microorganisms (Renčo et al. 2011, Zhu, Wang, et al. 2012). Renčo et al. (2011) tested the effect of four composts based on various manures on potato cyst nematodes (i.e. roundworms living on the roots of potato plants) and found that all four composts caused a significant reduction when compared to unamended soil. Zhu et al. (2012) inoculated 1.8 tons of pig manure with 9 kg of house fly larvae and compared the effect of a two-stage composting facility (i.e. house fly larvae and natural composting) to only natural composting. Not only did the two-stage composting achieve higher material reduction and pH, it did also realise a faster detoxification. Although the consistency of the material (various manures in the literature and mainly organic waste in this experiment) similar results were observed. It can thus be assumed that the same post-treatment technology will work for organic solid waste residue.

The pH of the material increased from 4.5 to 7.9 along the treatment. This development is in line with observations made in other studies (Erickson and Islam 2004, Zhu, Wang, et al. 2012) that fed BSFL with chicken manure and housefly

larvae with pig manure. However, a larger increase in the pH was observed in this experiment compared to the cited literature. This is probably due to the pH values of both studies starting at around 7.5. A more similar trend (increase from 5.5-8.0) was shown in the experiment by Lalander et al. (2014). An increase in the pH value is associated with curing of compost and therefore contributes to the fertiliser value of the residue as it needs to be inactive when applied (Wu et al. 2000).

5.2.2. Prepupae for animal feedstuff

The biomass conversion rate of nearly 20% on a total solids basis for both feeding regimes was high compared to the that described in the literature (8-12%) (Sheppard et al. 1995, Diener, Studt Solano, et al. 2011, Lalander et al. 2014). It was hypothesised that a continuous BSF composting system would have a positive effect on the biomass conversion ratio and the result was in accordance with this hypothesis. Both Diener et al. (2011) and Lalander et al. (2014) have used organic waste (or an organic waste replacement medium) as food for the larvae but both got a conversion of 12%. The difference in the larval feeding regime (100 mg wet compared to 160-240 mg wet), might explain the variation in the conversion rate. It could be that if the availability of food is higher, the biomass conversion rate will be higher as well.

The prepupal weight (around 92 mg dry weight/prepupa) found in this experiment is comparable to that of literature (Yu et al. 2011, Banks et al. 2014). Both Yu et al. (2011) and Banks et al. (2014) used excreta (chicken manure and human faeces respectively) and fed it to the larvae at a feeding rate of 2200 mg wet weight/larva and 100 mg wet weight/larva, respectively. Tomberlin et al. 2002 fed the larvae with two mediums; house fly larvae rearing substrate and regular chicken feed. The feeding rate was 90 mg wet weight/larva and this resulted in a higher prepupal weight of 111 mg (dry weight). Tomberlin et al. (2002) mentions that chicken feed, resulting in 111 mg larvae, is the best feed mixture when the aim is to produce heavy prepupae and that could explain why the weight of the prepupae in their experiment was higher. An overall comparison with the literature presented for this parameter shows that the continuous BSF composting system produces prepupae with a good weight. Although the protein and fat content of the prepupae was not measured due to time restrictions, literature is quite constant on the contents being around 40% and 35%, respectively (Hale 1973, Sheppard et al. 1995, St-Hilaire et al. 2007, Diener et al. 2009, Gobbi et al. 2013).

5.3. Outlook

The measured process parameters were very similar for the two regimes, except for the survival rate. FR-40 had a survival rate of 70%, but with a survival rate of 100% all larvae seemed to have survived in FR-60. Before analysing this difference, it should be noted that the values for the survival rate in both feeding regimes were higher when calculated with Equation 3. This means that for FR-60, the survival rate was over 100% and therefore, more larvae were put in the system than prepupae that crawled out. The larval enumeration procedure has probably not been accurate enough and therefore, it was decided to assume a 100% survival rate in FR-60 and look at the difference between FR-60 and FR-40. To do so, an

enumeration error of +30% was assumed. This means that the amount of larvae added to the system daily was not 4150 and 2750 but 5400 and 3600 for FR-40 and FR-60, respectively. In Table 7 the effect of the enumeration error and the survival rate on the variation of the larval density between the two feeding regimes is presented. The amount of larvae was calculated as a density in the material, using the volume 6.8 L, as this was the amount added to the unit each day. The result shows that an initial difference of 210 larvae per dm³ was reduced to 25 larvae per dm³. This means that although the two systems were inoculated with different amounts of larvae, the difference between the two regimes was neglectable when calibrating for the surviving number of larvae.

Table 7: comparison of larval density as provided, the larval density with the +30% enumeration error and the larval density with survival rate correction for both feeding regimes (FR-40 and FR-60) in the continuous BSF composting systems

	Unit	FR-40	FR-60	Difference
Intended larval density	Larvae/dm ³	610	400	210
Larval density (+30%)	Larvae/dm ³	795	530	265
Survival rate	%	70	100	30
Calibrated larval density (-S _r)	Larvae/dm ³	555	530	25

When comparing the survival rate of this experiment to the survival rate realised by experiments presented in literature (Myers et al. 2008, Tomberlin et al. 2009, Yu et al. 2011), it become clear that the survival rate greatly vary. Myers et al. (2008) used dairy manure and fed it with four feeding regimes (27, 40, 54 and 70 g) to 300 larvae. On wet basis, the 54 and 70 g feeding regimes of the study are comparable to the initial feeding regimes used in this study. The survival rate in the 54 and 70 g feeding regimes were 71-79% and thus comparable to the survival rate in FR-40. Tomberlin et al. (2009) fed larvae with a grain-based diet at three different temperatures (27, 30 and 36 °C). The variation in the results (74-97%) is comparable with the variation seen in the two feeding regimes of this study. However, the values produced by Tomberlin et al. (2009) include the pupal stage and therefore, the survival in the larval to prepupae stage would possibly be higher. Yu et al. (2011), using a very high feeding regime (2200 mg/larva) at 27-30 °C, got similar results (99%) to that of FR-60 (100%). Because the other parameters measured in this thesis project show comparable values between the two feeding regimes, the survival rate must have been influenced by a variable that was not related to the state of the material. When comparing the average temperature of the BSF composting facility (21 °C) with the temperatures Tomberlin et al. (2009) used for rearing larvae (27, 30 and 36 °C), the temperature in the BSF composting facility was much lower. Although Tomberlin et al. (2009) used growth chambers where the temperature could be adjusted and in general had a much smaller set-up; it still seems to mimic the natural environment better than the BSF composting facility used in this thesis project. The larval development speed in the experiment by Tomberlin et al. (2009) peaked at 30 °C and the prepupal weight peaked at 27 °C. It is not clear what the impact on the survival rate of a lower temperature could be, but Tomberlin's data shows an increase in the larval development time

with a lower temperature. If this is the case, the larval retention time in the current experiment might have been higher than 10 days. This would mean that the larval density at any given point in both Larveros would be higher. With a system of this size, a high density could cause a lower survival rate because of increased competition for food. In relation to the high density, another possibility could be that the feeding behaviour of the larvae puts restrictions on the maximum density in the system. The larval behaviour in the systems was similar. Large groups of larvae were observed feeding as a wall towards the fresh food. If this is their natural behaviour, it could present a possible limitation to the current set-up of the continuous system, making the feeding surface small and therefore the competition high. This might have affected the survival rate in FR-40, where more larvae were competing for the same amount of food that was provided to the larvae in the lower density regime FR-60.

Some changes to the experimental set-up might have given a different result. Firstly, it is important to assure that the system has reached steady-state when the aim is to produce a mass balance. Therefore, a longer start-up period and a steady flow of recycled residue are required. Secondly, the larvae provided for the Larveros came from various sources and were of different sizes nearly every day. During the last week of the experiment, hatchlings were added instead of older larvae. The hatchlings were more easily affected by the environment and might have died, for example from various types of mould growing on excess larval food (Tomberlin et al. 2002). A steady flow of larvae of the same size and from a single source would be a requirement before starting a new experiment in order to have data that is better to compare. Lastly, the temperature in the BSF composting facility should be the same as the rearing facility. Literature shows that 27 °C is an optimal temperature for larval development (Tomberlin et al. 2002, Diener, Studt Solano, et al. 2011).

Furthermore, the smaller difference between the two feeding regimes, caused by the difference in the survival rate, most likely is the reason why the process parameters did not differ between the two feeding regimes. Two new feeding regimes, possibly more extreme and with systems in steady-state, could give a clearer picture of the impact of different variables.

In a set-up such as this, a regime following FR-60 would be favourable comparable to FR-40. The material reduction – 70% on total solids basis and 85% on wet weight basis – was high in the two regimes, and though the larval input in FR-40 was higher, no other differences in the process parameters was observed. The production of larvae was costly as it was labour intensive and required resources (light and heat). Therefore, when the same results can be gained by adding fewer larvae, it is beneficial for the overall cost of the treatment. However, if the assumption that the larval density is restricted by the feeding surface is taken into account in a new design, the increased feeding surface could result in a higher survival rate.

In Figure 7 a possible solution to increase the feeding surface in the Larveros is displayed. Instead of adding all the new material in one unit and then adding the larvae on top (top figure), a new unit could be added before spreading the two- and three day old material equally over the new unit and the two next units (bottom figure). The new material would then not be added to a single unit but spread over

the first three units before adding the larvae spread over the three units. The feeding surface in this experiment was equal to the width of one unit multiplied by the material height (275 cm^2). The proposed spreading of the material would result in the same width, a smaller height but would then add the surface of the material as the larvae added in the days before can feed from below the newly added material.

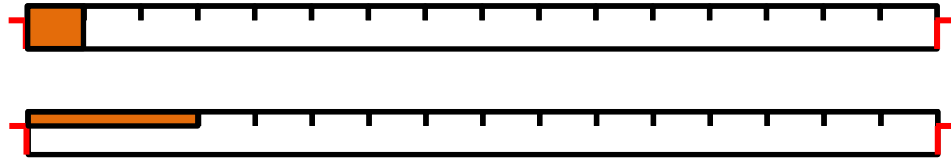


Figure 7: Schematic side view of one of the Larvero used in this experiment (top) with one unit that was filled with material (green surface) and one Larvero where the new material (green) is spread over three units instead of filling one unit (bottom).

If the feeding surface in the current system was indeed the cause of the lower survival rate in FR-40 and the system as shown in the bottom of Figure 7 could then increase the survival rate, the FR-40 regime, producing the same amount of larval biomass with less material, would be favoured in a system aimed at an optimum product output and could possibly increase the waste treatment capacity per area by increasing the material height or feeding rate.

6. Conclusions

Except for the survival rate, all parameters (pH, compost maturity, TS, organic content, material reduction, biomass conversion rate and prepupal weight) gave similar results for the two feeding regimes, where less food per larvae resulted in higher mortality.

The material reduction was high compared to literature with a dry weight reduction close to 70% compared to literature average of 40-55%. Of specific interest was the wet weight reduction around 85%.

The biomass conversion rate also achieved a high value (~20%) when compared to literature average of 8-12%.

The survival rate showed a large difference between the two feeding regime. The survival rate in FR-60 was adjusted to 100%, resulting in a survival rate of 70% in FR-40.

In this set-up, the FR-60 is the preferred regime in regards to optimum waste reduction and product output as quantity and quality of the products did not differ significantly from FR-40 but could be realised with less resource input as less larvae are required to attain the same result.

7. Bibliography

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