

**BIOCONVERSION OF ORGANIC FRACTION OF SOLID WASTE USING THE
LARVAE OF THE BLACK SOLDIER FLY (*Hermentia illucens*),**

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DECLARATION

I hereby certify that I am the sole author of this thesis and that no part of this thesis has been published or submitted for publication, or submitted for a higher degree to any other University or Institution.

I certify that, to the best of my knowledge, my thesis is a genuine reflection of work done and that any ideas, techniques, quotations, or any other material from the work of other people included in my thesis, published or otherwise, are fully acknowledged in accordance with the standard referencing practices.

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ABSTRACT

Ghana like many other developing countries is battling with challenges pertaining to effective ways of managing solid waste. Usually, waste management companies are unable to collect efficiently the waste produced in various municipalities and thereby always leaving a backlog. The larvae of the black soldier fly, *Hermentia illucens* (Diptera: Stratiomyidae) has been propagated as a feeder of organic material and may be used in simple engineered systems to reduce organic waste in developing countries. This research sought to assess the use of black soldier fly larvae to digest and degrade organic food waste collected from a restaurant – *Obaapa* – on the KNUST campus in a small-scale laboratory experiment in Ghana.

To cultivate the BSF larvae, one has to apply an attractant (carcass of rat meat, fresh fish, chicken feed and human faeces) to attract matured flies. The BSF is ascertained by observation and separation. Afterwards, the larvae are generated by providing ideal moisture conditions for the larvae to hatch from the eggs.

In the laboratory experiment, 5 feed classes code named B25, B75, B100, B125 and B200 with feeding rates of 0.25, 0.75, 1.00, 1.25 and 2.00 g/larvae/ day were fed in a small experiment box (in triplicates). These were inoculated with 50 larvae each of 5-6 day old BSF larvae. The research concludes that, a daily feeding rate of 1.00g of food waste per larvae per day is able to meet high organic matter extraction within a shorter period to produce better prepupal yield. Thus, for a 1 tonne waste, a million larvae are needed to successfully reduce it by 91.25% in 5 days. This research proves that the larvae of the Black Soldier Fly (*Hermentia illucens*), are able to digest and degrade municipal organic solid waste in Ghana between 44-94%.

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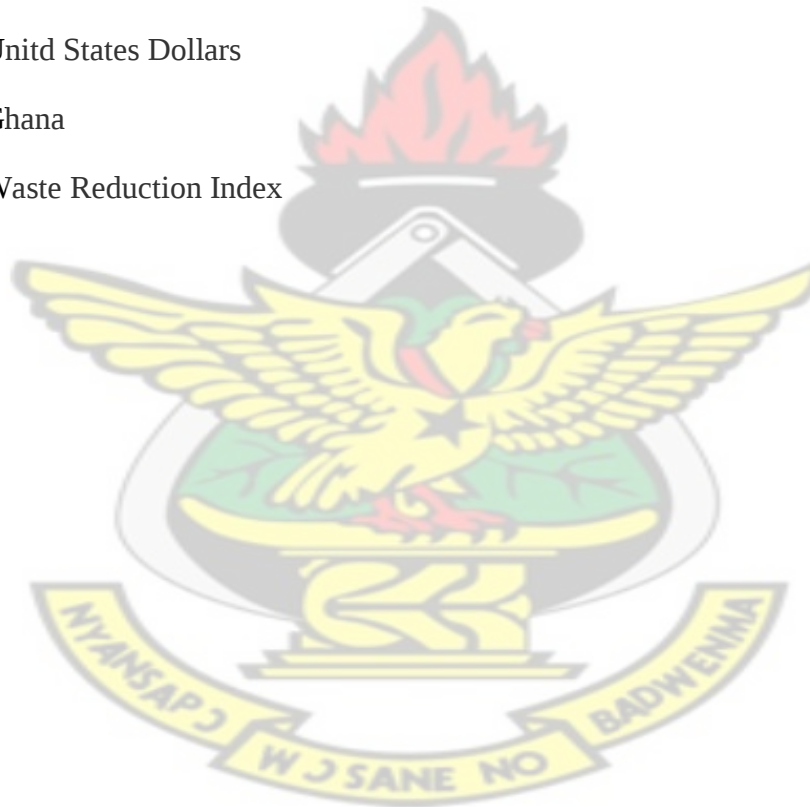
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ABBREVIATIONS

AMA	Accra metropolitan Assembly
KMA	Kumasi Metroplolitan Asembly
BSF	Black Soldier Fly
MSW	Municipal Solid Waste
CR	Conversion Rate
DME	Dry Matter Extraction
KNUST	Kwame Nkrumah University of Science & Technology
USD	Unitd States Dollars
Gh	Ghana
WRI	Waste Reduction Index

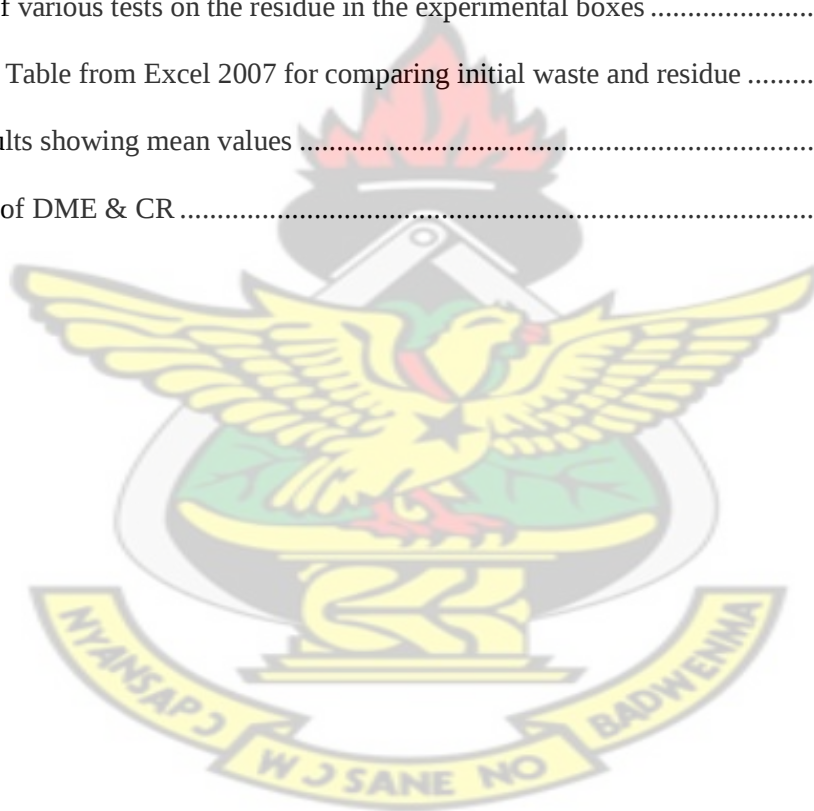


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1.0 INTRODUCTION

1.1 BACKGROUND

As the world's solid waste piles up, it is raising serious concerns among municipals, policy makers and other stakeholders, especially in developing countries. Modern technology, adept as it were in producing the waste, seems quite at a loss when it comes to getting rid of it. A United Nations Development Program (UNDP) survey, conducted in 1997 in 151 cities revealed that, poor solid waste disposal is the second most serious problem after unemployment. This affects negatively human health, the environment and is a major set-back to economic growth (Diener et al., 2009). On the contrary, a well managed solid waste system results in job creation and income generation both for the formal and informal sector (Diener et al., 2009). Thus, several cities all around the world are increasing their efforts at finding sustainable solid waste management solutions.

According to UN-Habitat's Third Global Report on the State of Water and Sanitation in the World's Cities (UN-HABITAT 2010), using technologies such as high-tech compaction trucks, fully automated collection systems, incineration and advanced waste-to-energy is unlikely to meet the needs of cities in most middle or low-income countries over a medium-term time horizon. The report explained further that, so-called waste-to-energy plants would, at a minimum, need major redesign to avoid the need for additional fuel to support combustion. This is because of the often high tropical rainfall, and high organic content of waste in middle and low-income countries which increases the moisture content of waste. Thus, for developing countries, it is important to identify simple, appropriate and affordable solutions that can be

implemented progressively, giving the inhabitants the best system they can afford and laying the basis for future improvements.

Solid waste in general has both organic and inorganic materials. The recycling of inorganic material (e.g. paper, glass, plastic, metals) from municipal solid waste has become an important source of income and employment of the informal sector, especially in developing countries (e.g. Ali 1999, WASTE & Skat 2006). In Ankara, Turkey, for example, scavengers collect and sell to middle men 50% of the recyclables produced by households, commerce and trade, yielding a total of about USD 50,000/day (Ali 2002; Diener et al 2011a), and at least 150,000 waste pickers throughout Delhi's waste management system divert more than 25% of all waste generated into recyclables, thus saving the municipal authorities substantial costs (UN-HABITAT 2010). However, reuse of organic waste which forms about 50% of municipal solid waste (Henry et al., 2006) is minimal despite its high recovery potential. For example a report by UN-HABITAT (2010), revealed that in Quezon City, Philippines, 33% (240,000 tonnes) of the total annual waste generated is reused by the formal and informal sectors. But, the recycled organic fraction accounts for only 6% (14,000 tonnes) – an example typical of countless municipalities in developing countries. Because of the inadequate reuse of the organic waste fractions, most end up on landfills. These organic components represent a large amount of stored chemical energy, and mainly responsible for the production of biogas and leachate (Alvarez 2012). If this portion of the waste stream could be partially or fully diverted from landfills and put to use on a significantly shorter time scale, landfill life spans could be increased and pollution issues stemming from leachate and gas production could be reduced (Alvarez 2012). With the world's population increasing, municipal solid waste and organic waste is accumulating, raising concerns about the green house gas they inevitably emit (Holmes 2010).

Waste diversion techniques currently include, the reduction of resource consumption, composting of organics, the reuse and recycling of a resource, anaerobic and aerobic digestion (Alvarez 2012). Composting forms the largest alternative for diverting organics from a landfill but has several setbacks; the tedious process in manually or mechanically mixing of heap to keep it aerobic, it cannot easily accept animal protein based wastes, and nutrient ratios must be planned and maintained for a successful run (Alvarez 2012).

In most recent times, black soldier flies, *Hermetia illucens* (L.) (Diptera: Stratiomyidae), have been studied in the Americas and parts of Asia demonstrating their potential use in waste management systems, including large confined animal feeding operations and bio-conversion in food waste (Sheppard et al., 2002; Barry 2004; Canary 2009; Diener et al., 2011a; Flechet 2008). This study seeks to explore the potential of this developing technology of bioconversion in Ghana.

1.2 PROBLEM ANALYSIS

In Ghana, all waste collected by waste collection companies are transported to the landfill for disposal (Hamdu 2009) or on a more or less uncontrolled dump site (“landfills”) where the material decomposes in large heaps under anaerobic condition. Thus, in no time, the “landfill” gets to its capacity and a new site has to be sought for dumping refuse coupled with its many attendant problems of environmental pollution.

Studies conducted by the two biggest metropolitan assemblies – Accra Metropolitan Assembly (AMA) and Kumasi Metropolitan Assembly (KMA) – revealed that the organics (vegetables,

fruits, greens) in the total solid waste collected are 65% and 44% respectively (KMA 2012). There is little or no reuse for the organic fraction and thus, a large portion of the organic waste end up on landfill sites which reduces the life span of these facilities. Financial constraints have made incineration not a viable venture for developing countries like Ghana. Organic waste that has not been landfilled has traditionally been treated through composting. This is usually done on small scales, the processes involved is long and the market prizes for this produce is uncompetitive as compared to inorganic fertilizers used by crop farmers.

These challenges have inspired the search of alternate ways of disposing organic waste. One of such is, bioconversion using the larvae of *Hermetia illucens*, better known as the black soldier fly. They have been propagated not only as converters of organic waste but also a nutritious feed for chicken, pig breeding and aquaculture (Barry 2004, Diener et al., 2011a, b; Sheppard et al. 1994; Newby 1997). The prepupae harvested can produce biodiesel (Newton 2005). This study looks at application of the bioconversion using BSF larvae – in Ghana.

1.3 JUSTIFICATION

Though the ability of the BSF larvae to digest and degrade organic waste is known, little information is available on the capability of the larvae to digest municipal organic waste and most of the researches done were in the Americas and Asia (Barry 2004, Diener et al., 2011a, b; Sheppard et al. 1994; Newby 1997; Flechet 2008). The potential use of bioconversion has not been investigated in most developing countries and in Ghana. This research seeks therefore to explore the practicality of this new technology in Ghana (a developing country) to help in the

disposal of the high organic waste generated in its municipal cities and benefit from the economic value of the products obtained.

Results obtained from this study would be useful in piloting and up-scaling the technology to effectively reduce over dependency upon landfills or open dump sites. It would help in the design of bioconversion systems. Again, the knowledge gained would be useful to policy makers and municipal authorities as they develop efficient and effective Integrated Sustainable Waste Management (ISWM) systems.

1.4 OBJECTIVES

The main objective of this study is to assess the use of black soldier fly larvae to digest and degrade municipal organic solid waste in a small-scale laboratory experiment in Ghana. The specific objectives are

- To outline the procedure and challenges in generating larvae for the application to organic solid waste degradation.
- To characterise feed used for the organic solid waste treatment.
- To assess the waste reduction capacity of BSF and the reduction time.

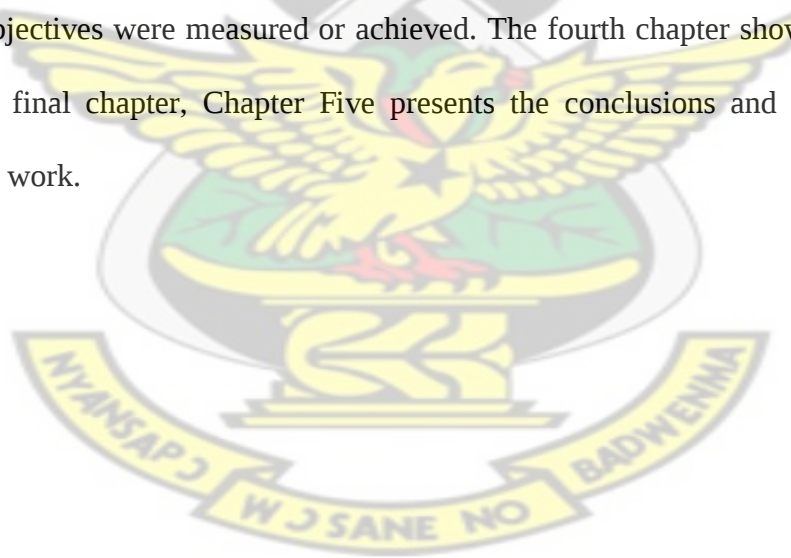
1.5 SCOPE OF WORK

Organic waste has a wide variety that includes sewage and agricultural waste. However, this study focuses on an aspect of the organic waste stream of municipals– food waste from the kitchen, specifically a restaurant. Details in biological behaviour of the adult BSF insect and the

larvae were not part of this work. Neither did it look into the abiotic factors, or toxicology of the BSF. The research work looked at what is involved (challenges) in getting larvae for inoculating a laboratory experiment and then examining the reduction potential of the larvae under normal environmental conditions in Kumasi, Ghana.

1.6 STRUCTURE OF REPORT

The report has been prepared into five chapters. Chapter One introduces the research, giving background information, the objectives and defining the scope of works. Chapter Two reviews literature in the area of bioconversion using BSF, considering what has been done and is still being done in the industry. Chapter Three focuses on the materials and methods used in this study, how the objectives were measured or achieved. The fourth chapter shows the results and discusses it. The final chapter, Chapter Five presents the conclusions and recommendations gathered from the work.



2.0 LITERATURE REVIEW

2.1 SOLID WASTE MANAGEMENT

Managing solid waste is one of the biggest problems facing most urban areas in the world no matter where, from the small towns and villages even to the mega cities. In almost all cases, waste quantities are increasing due to one or combination of these factors; increase in population, increase in waste per person; amount of waste generated by industry; and increase in the complexity and variety of waste (UNHABITAT 2010). Yet, Municipal Solid Waste (MSW) management remains a neglected key issue in developing countries (Diener et al. 2011b). Municipal Solid Waste is defined as “wastes generated by households, and wastes of a similar nature generated by commercial and industrial premises, by institutions such as schools, hospitals, care homes and prisons, and from public spaces such as streets, markets, slaughter houses, public toilets, bus stops, parks, and gardens” (UN-HABITAT 2010).

Usually, waste management companies are unable to collect efficiently the waste produced in various municipals, leaving a back log. These uncollected wastes on streets, and drains causes flooding and disease vector. In these developing countries, organic waste fraction forms about 80% of the total municipal waste and are often looked upon as a waste fraction with no market value and therefore ignored by the waste recycling sector (Ali 2002). Generally, disposal of solid waste has been traditionally carried out in one of these three methods: Landfilling, incineration, and composting.

2.1.1 LANDFILLING

Land disposal, in the form of landfills, surface impoundment, land application, and deep well injection are the most common form of waste management for nonhazardous and hazardous waste (Barry 2004). Municipal landfills consist of heterogeneous mixtures of wastes that are primarily of residential and commercial origin (Barry 2004). Food and garden wastes, paper products, plastics and rubber, textiles, wood ashes, and the soil used to cover the material are typically found in municipal landfills (Barry 2004).

In most developing countries, they are less if any, of properly engineered landfill sites. Most of the sites for disposing waste are actually open dump sites. In Ghana, there are currently only two engineered landfill sites in Kumasi and Tamale with construction going on in Accra and other parts of the country. The various dump sites -“landfills”- easily reach capacity and new areas have to be sought for waste disposal.

Though a modern landfill system is engineered to prevent leachate migration and subsequent groundwater pollution as well as control landfill gas, usually cracks and leaks in liners and piping system releases contaminants into the environment (Alvarez 2012). Strayed refuse, fire and explosions, vegetation damage, unpleasant odours, land settlement, and noise are other problems common to landfills that make it an unwelcomed neighbour. This increases transportation expenses as the landfills are located far from the waste generation sites (Alvarez 2012). Often, landfills have an active life of 25 to 30 years and are therefore a large investment in terms of capital and time. After the site closes, decomposition continues even though receiving waste has stopped. This causes a need to monitor the surrounding groundwater, surface water if present, geological site stability and potential gas migration (Alvarez 2012).

Landfills are the most widely used though associated with many problems. Other alternates are not cost competitive. In fact, the residues of other treatments eventually end up on the landfill site, making landfilling the most important of all treatment processes.

2.1.2 INCINERATION

The old concept of incineration basically means burning, setting fire to or destroying waste. Modern incineration practices however, involves the engineered process of using controlled flame combustion to thermally degrade waste materials. In some cases incineration is the preferred option as waste volume is reduced to about 5%; avoids contamination by reducing pollutant levels; easy to maintain; and the only solution for some waste types example medical waste. This practice is uncommon in dealing with MSW in developing countries mainly as a result of the high cost involved. Though incineration reduces volume by far, it does not completely solve the problem because the residue (ash) produced remains must still be landfilled. The main disadvantage of incineration is that emissions (dioxins, furans and trace metals) are released through the incinerator stack into the environment which are carcinogenic and can cause respiratory problems (Perry 1999). These emissions can be reduced through the use of baghouses and scrubbers. Compared to the background exposure, the emissions are relatively small from an incinerator but are still harmful (Perry 1999). Costs involved in controlling these emissions and setting up a modern incinerator for municipal use is high and hence not a viable option for most developing countries.

2.1.3 COMPOSTING

Composting is a very acceptable and viable option for handling most organic wastes presently being landfilled. During the composting process, microorganisms decompose organic compounds, which consist of carbohydrates, sugar, proteins, fats, cellulose and lignin. Many factors affect the composting process. Aerobic microorganisms need oxygen, water and nutrients for their metabolism and cell synthesis. As a result of microbial activity heat is released and, if retained within the composting mass, the temperature rises through the mesophilic phase into a thermophilic phase and then back in to the mesophilic phase (Terzin 2002). The microbial population changes during the course of these transitions, thereby affecting the rate of organic matter decomposition (Terzin 2002).

Although composting provides many benefits there are problems associated with traditional thermophilic composting. The time required to produce finished compost is highly variable and depends on the composition of the starting material, aeration frequency, proper nutrient ratios, moisture content and temperature (Alvarez 2012). Finished compost can be obtained in as little as one week in mechanical composting operation to as long as six months in a static pile. In addition, the composting end products uses are limited mainly to soil amending and erosion control applications (Alvarez 2012). In an effort to overcome some of the limitations of traditional composting, organisms have been introduced into the compost pile. The most common adaptation to this is vermicomposting.

Vermicomposting is using earthworms to breakdown organic material residuals reducing them into finer particles as they pass through their grinding gizzard (Ndegwa and Thompson 2001). However the problem with vermicomposting is that the process must be maintained below 35°C; otherwise the worms will die (Ndegwa and Thompson 2001). At these temperatures the organic

material never reaches a temperature to kill pathogens and pass Environmental Protection Agency (EPA) rules for pathogen reduction (Barry 2004).

The commercialisation and large scale application of composting in MSW treatment in Ghana has often not been employed. Most people do them on small scales in their backyards or on farms for personal use, not necessarily to sell. The long time and activities involved in preparation of compost discourages most farmers as well from its usage. The problem most operators of compost facilities mention is the market for their produce. Farmers are used to inorganic fertilizers which come at relatively cheaper prizes and even subsidized by government. The market value for compost is little and the industry remains stifled. Farmers do not understand the benefit of using compost whiles they can get other chemicals cheaper to serve the same purpose or for free. Currently, there is only one new compost facility - Accra Compost and Recycling Plant in Kotoku, Nsawam which is managed by Zoomlion Ghana Limited. This compost facility aims to commercialise and encourage compost use. Until government and others try to promote and pick up composting in the country, its usage will still remain embryonic and not help in landfilling diversion that much.

2.3 FOOD WASTE and FOOD WASTE RECYCLING

Currently, 30% of food produced globally is wasted or lost in between the farm and the final consumer (SIWI 2008). Thus, there is wastage on the farmer's land, as food is transported to distributor, during storage, from storage house to the market place and finally from the market to the consumer. Also, during the processing of food and cooking, there is wastage both at the commercial, industrial and residential level.

In developed countries, much of the food waste generated commercially is landfilled (Canary 2009). One alternative to divert waste and prolong life spans of landfills is to compost. Interestingly, this option has only survived in the United States as operators of compost facilities are able to charge a fee for receiving the food waste and more often than not, government provides subsidise (Canary 2009). Therefore, the return from this is actually not on the compost but on the waste received (Canary 2009). Thus, landfilling of food waste remains still the major option in food waste disposal.

As identified earlier, composting in Ghana is done on very small scales. Recently however, one company – Zoomlion Ghana Limited - is trying to enter into large scale production. The effect of poor compost facility operations has resulted in landfilling of food waste. This means that the nutrients available in them are all lost and not recovered.

In order to understand existing food waste collection methods, Canary (2009) classified food waste sources into six major categories: residential food waste; restaurant food waste; industrial processing food waste; crop agriculture waste; super markets & open air markets; and commercial food waste. In this research, restaurant food waste was used.

2.4 BIOCONVERSION BACKGROUND

Generally, bioconversion is described as the process which uses biological agents to transform a feedstock into desirable products. In recent times, the newest waste management technology is bioconversion using fly larvae - converting organic waste to insect larval biomass and organic residue. Bioconversion is a practice of recovering resources while simultaneously limiting the amount of organic material affecting landfill behaviour. Several organisms have been used in this

treatment process. However, for this research thesis we explore the use of the larvae of the black soldier fly (*Hermentia illucens*).

Bioconversion is not a source reduction management action, but may be considered a pretreatment technique (Barry 2004). A pretreatment technique is any process that alters the composition or other characteristics of the waste stream as generated prior to landfilling (Barry 2004). Barry (2004), who worked on the bioconversion of food waste using the black soldier fly larvae in contained systems in North Texas, United States, gave the following benefits of bioconversion of organic waste;

- A means of diverting food wastes from the Municipal Landfill providing greater disposal capacity
- Reducing potential landfill odour problems
- Reducing fouling problem for recyclables
- Reducing obstructed sewer pipes
- Reducing methane gas production from landfills as a result of anaerobic breakdown of organic materials
- Reduces the energy costs associated with transportation of food wastes
- Offers educational institutions from elementary to college by providing information to create a practical as well as educational tool, incorporating the fields of ecology, biology, economics and an essential lesson in sustainability

The biological process involved in bioconversion is natural and can occur everywhere. Little resources are wasted as the larvae are self-harvesting and studies indicate that the prepupae are an excellent source of feedstuff for fish, swine and chicken (Bondari et al. 1981; Hale 1973;

Newton et al. 1977). In bioconversion, the consumption of food waste and the healthy development of larvae rely on environmental factors that influence the physiology of the species and diet. Temperature, moisture content, density, physical and chemical properties of the medium, and competition are important variables; the relative influence of each factor can change given certain contexts and situations.

Thus, this small scale study on the use of organic waste treatment using the larvae of the BSF in Ghana is an important step in solving municipal waste disposal problems and creating economic opportunities for municipalities and small entrepreneurs in the MSW sector.

2.5 THE BLACK SOLDIER FLY

Studies of the Black Soldier Fly (*Hermentia illucens*) and its larvae date as far back as the 1950's. In 1959, Furman et al., found out that the larvae naturally controls populations of houseflies. In an attempt to find cheaper poultry feeds, the work on BSF larvae begun again in the 1970s as Hale (1973) investigated the use of the BSF larvae in poultry diet. In 1977, Newton, Hale, Booram and Barker examined their use as a feed supplement for swine. These at that time were known as “latrine” larvae and were naturally found in manure piles of large poultry, pig, and cattle operations. During the 1980's and 1990's, researches began to investigate what the larvae were doing there and it was noted that they reduced manure load in addition to naturally controlling populations of house flies which are a disease vector. Today, many researches have gone on about the larvae of the BSF.

BSF is usually found in tropical and warmer temperate regions between latitudes 45⁰N and 40⁰S (Ustuner et al. 2003) but it is believed to have originated in the Americas (McCallan 1973).

Although primarily adapted to these regions, it can tolerate wide extremes in temperature (McCallan 1973) except when ovipositing. In this research, it was found to be native in Kumasi, Ghana.

2.5.1 LIFECYCLE AND PHYSICAL REQUIREMENTS

The BSF has five stages in its lifecycle: egg, larvae, prepupal, pupae and adult. The larval stage is further divided into phases called *instars*. An instar is defined as the period between each moulting of their exoskeleton (Alvarez 2012). The number of instar stages varies for different fly species: *Hermetia illucens* has five instar stages. All of the adult fly's nutritional requirements are obtained during its larval stage and adult flies survive on their fat reserves obtained as maggots. When this fat reserve is depleted the adult dies (Myers et. al. 2008).

BSF has an estimated life cycle of 40 days (from egg to adult) but this length depends on the environmental conditions present and the rearing diet. Waste consumption rates appear to depend on the size of the maggot and the type of food being consumed (Diener et. al. 2009).

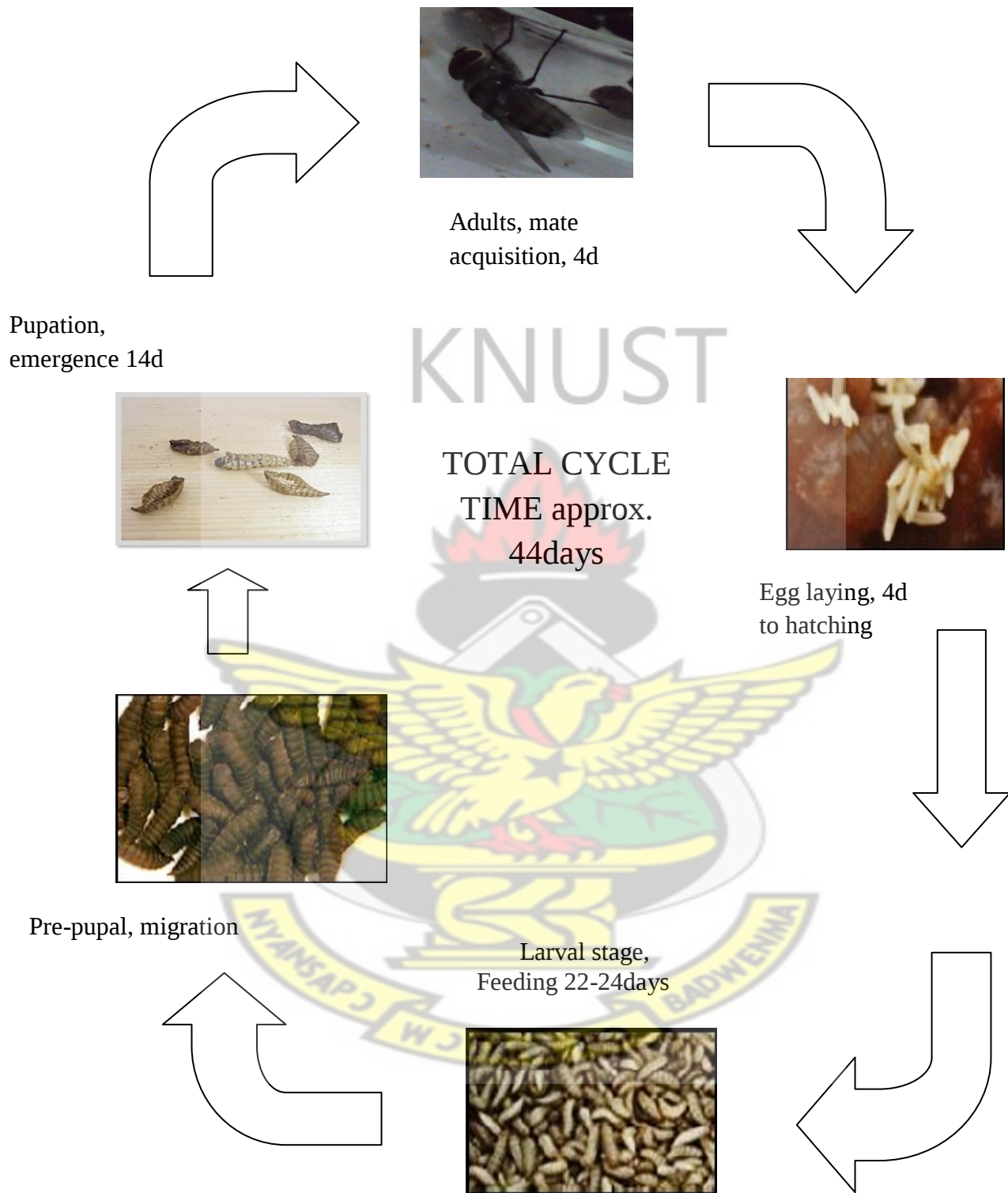


Figure 1 Life cycle of the Black Soldier fly

2.5.2 LARVAL STAGE

Newly hatched larvae are particularly beautiful with translucent bodies and a black eye spot. The larvae can mature in two weeks if conditions are ideal; however, food shortages may extend this period to four months (Sheppard et al. 1995). Immediately after hatching the new nymphs seek a food source and begin to feed. The maggots do not sleep but they do not consume waste continuously. For efficient food processing, optimal temperatures range from 27 to 33°C (Sheppard et. al. 2002). Lower temperatures are most likely tolerable because the maggots generate heat as they consume food through their writhing motions. The maggots secrete enzymes that make the food digestible prior to ingestion by liquefying the waste as they consume it (Alvarez 2012). Like temperature, unfavourable humidity affects growth, resulting in problems when estimating development times (Barry 2004). Laboratory studies with *H. illucens* range from 50 to 99% relative humidity (Bradley and Sheppard 1983). The moisture content of the resource is important and if the moisture content is outside the optimum range it affects BSF development (Alvarez 2012). This ranges, from 60% to 90% (Myers et. al. 2008). Too much moisture will force the maggots to migrate the food/resource matrix they are feeding in; and not enough will prevent efficient consumption. However, these moisture circumstances can be exploited to engineer a BSF waste management system: the moisture preferences of the larvae could provide a simple method to control and direct the maggot's location in the system (Alvarez 2012). The feeding site can be kept moist, and a path to a drier area (pupation medium) can be provided to encourage migration to the desired location. The development rate can be controlled, if required, by modifying the moisture content in the resource (Alvarez 2012). It also appears that soldier fly maggots secrete chemicals that warn other fly species that a food source colonized by soldier fly maggots is not an ideal egg laying site leading to effective reductions of the common housefly (Bradley and Sheppard 1983).

The range of diet of the larvae is wide: they can consume animal faeces, rotten and fresh flesh, fruits, restaurant waste, kitchen waste, and possibly a variety of other organic wastes (Holmes 2010, Sheppard et. al. 2002, Tomberlin et. al 2002, Canary 2009). The nutrition source used in the larval stage can also affect adult fly characteristics (Tomberlin et. al. 2002). Thus in a full scale waste processing facility the nutritional content of the incoming waste stream may need to be monitored to ensure that the larvae are eating a balanced diet. This would be important because the facility would require healthy adults to maintain egg production at required levels. Upon reaching maturity, the prepupal larvae are about 25mm long, 6mm in diameter, and they weigh about 0.2grams (www.esrint.com).

2.5.3 MIGRATION AND PUPATION

Prior to reaching the pupation stage the larva will leave the feeding site to find an adequate pupation site. Once the larvae have consumed enough food they begin to migrate away from the food source (Sheppard et. al. 1994). If the maggots that are ready to pupate can be directed to a location selected by a human operator, then the pupae are effectively harvesting themselves. The maggots have entered a wandering stage searching for a drier and darker location than the feeding site to continue their life cycle. Successfully exploiting this instinct means that no additional effort is necessary to remove them from the feeding site. Maggots have been shown to migrate considerable distances in order to find an adequate pupation site. The maggots will also change colour, a circumstance that could be exploited to further sort them for quality purposes. However, the maggots are accomplished burrowers and can enter small spaces and crevices with ease and when they are wet they can attach themselves to a wide variety of surfaces including plastics, wood, rubber and metals. This can pose problems trying to contain them in specific

feeding areas. The time spent by each maggot in the migration stage varies but appears to be dependent on the maggot's ability to locate an ideal pupation site. A study suggested that the maggots favour drier conditions for a pupation site but require ambient humidity levels of approximately 60% to emerge as adults (Holmes 2010, Sheppard et. al. 2002). Migrating maggots are suspected of leaving chemical trails that other maggots follow creating a migration path.

Another characteristic of an ideal pupation site is protection from predators and unfavourable environmental conditions, such as flooding. The pupation media itself should be porous and loose to allow for easy burrowing of wandering maggots. A medium with these properties should also provide adequate oxygen levels so the pupae can breathe. If the pupation medium is too fine the spiracles, or breathing structures, can become clogged possibly resulting in death. The depth of the pupation medium is also important. If it is too deep, the emerging flies will fail to reach the surface. If it is too shallow, the maggot may not deem the location adequate and continue to wander, wasting its fat reserves thereby reducing its harvest value or its chance to successfully mate. Wandering maggots have been shown to pupate without a pupation medium if no suitable medium is present (Holmes 2010). Studies have shown the ideal depth for a pupation medium is 15 to 20 centimetres. Pupation can last five to seven days depending on temperature and ambient humidity.

2.5.4 ADULT STAGE AND EGG LAYING

The adult fly measures up to 20 mm in length (Callan 1973) with a cylindrical abdomen easily recognized by “windows” of translucent cuticle (Oldroyd 1964). Adult flies vary in color from black, metallic blue, green or purple, to brightly colored black and yellow patterns (Drees 1998).

Adults engage in an aerial mating process and females oviposit near suitable larval medium (Sheppard et al. 1995). Two days after emergence, mating takes place and oviposition occurs two days after fertilization (Tomberlin & Sheppard 2002). Females seek out an area that is close to a food source to deposit their fertilized eggs. The mechanism for this action is believed to be the detection of volatile chemicals from rotting wastes (Sheppard et. al. 2002). Females will also leave chemical markers that attract other females to a suitable egg laying site. Females prefer not to lay eggs directly on a food source but near it (www.esrint.com). The ideal egg laying site should be maintained at 27°C with an ambient relative humidity of 60% or more: at these conditions egg hatching rates of 80% or more have been observed (Holmes 2010, Sheppard et. al. 2002). She produces about 900 eggs in a short life of 5-8 days (www.esrint.com).

Temperature and ambient light levels are important in order for the flies to initiate mating. One literature source suggests an irradiance of over 200 micromoles/m²/s as optimal (Sheppard et. al. 2002). There is limited data available on what specific wavelength or ranges of wavelengths are responsible for the initiation of mating and what time exposure is necessary. Mating levels of adults were highest under natural sunlight: the use of artificial lighting should therefore be considered supplemental when natural sunlight exposure is not adequate (Alvarez 2012).

In the majority of the studies reviewed where BSF colonies were reared, the egg laying and waste processing activities were conducted in separate locations. Eggs were collected in the corrugations of cardboard, or flutes, and then transferred to the testing apparatus (Tomberlin & Sheppard 2002). This is done because eggs are fragile, small and vulnerable to changes to environmental variables; the flutes provided protection for eggs and encouraged the female's ovipositor to lay eggs in the confined space. In a waste management facility, it would be preferable to automate or remove the need for egg handling.

The adult fly lives for only a few days or weeks, and does not bite or engage in pest-like behaviour. It does not seek to enter homes or restaurants, but lives its short adult lives remote from humans, maturing and mating primarily in wooded areas (Sheppard et al. 1995). Further, the adult *H. illucens* is not a pest, and actually drives off the common housefly usually associated with wastes and health hazards (Sheppard et al. 1995).

2.6 WASTE TREATMENT USING BLACK SOLDIER FLY LARVAE

Several studies have been done using the black soldier fly larvae to degrade organic material. Most of these studies focused on the degradation of cow, chicken or pig manure by *Hermetia illucens* larvae in cost- and maintenance-intensive systems of developed countries (Sheppard et al. 1995, Newton et al. 2005a, b). Sheppard et al. (1995) modified approximately 460 caged layer house fitted with a concrete basin below the cage batteries. The basin allowed easy harvesting and quantification of migrating prepupae. However, it did not allow controlling the feeding rate, and the waste reduction had to be estimated based on the depth of the accumulated chicken droppings. The larvae were able to convert chicken manure into a feedstuff, larval mass, containing 42% protein and 35% fats.

Newton et al. (2005b) employed the faeces of 12 pigs in an automated system using a dewatering belt to transport the material to the larva treatment bed, thus achieving a 56% of fresh material reduction. Newby (1997) investigated the activity of *H. illucens* in a compost type system at household level in Australia. Lardé (1990) tested the conversion of coffee pulp in a small-scale experiment. Though Bradley (1930) noted in 1930 that *H. illucens*-infested privies (i.e. pit latrines), thus indicating their potential to treat faeces, Diener (2011a) found out that not only

were the larvae of the black soldier fly able to survive and even develop in pure sludge but were also capable of significantly reducing the sludge biomass.

Additional research focussed on the limitation of this technology in waste management and in protein production. Diener et al. (2011a) highlighted the presence of heavy metals in the feed material which negatively influence life history of the fly population and can accumulate in the prepupae.

Though BSF's ability to digest other wastes including the organic portions of MSW, wastewater treatment sludge and fish rendering wastes has been studied, there are significant questions, such as: what are the optimum feed rates? This question is actually quite complex: the optimum applied rate depends on the food source and the age of the maggots because BSF maggots consume different wastes at different rates at different instar phases (Alvarez 2012).

In a study conducted by Nguyen (2010), five different diets, fish renderings, liver, fruits and vegetables, poultry feed and restaurant wastes were fed to different maggot groups. Each diet was shown to have a different waste consumption rate and the rate increased as the maggots increased in size.

In another study conducted by Myers et. al. (2008), BSF larvae were used to stabilize dairy manure in a controlled laboratory setting. BSF larvae were fed four different rates of manure to assess their development. Interestingly, the feed rate affected the development of the larvae: the larvae that received less manure did not weigh as much as their overfed counterparts and the adults of the underfed larva lived three to four days less. However, the larvae that were fed less manure turned out to be more efficient at reducing manure dry matter. Myers et. al. (2008) observed that larvae fed 27 g of manure daily reduced dry matter by 58% whereas the other test

subjects, receiving 70 g of manure per day, reduced dry matter by only 33%. There was also a higher incidence of mortality (29%) among larvae that were fed 70 g of manure when compared to the test groups fed 27 g and 54 g (<20%). Myers et. al. (2008) also found that the phosphorus content of the manure was reduced by 61% to 70%, and the nitrogen content was reduced by 30% to 50% respectively, across all treatments.

Diener et. al. (2009) studied the consumption rates of various organic municipal solid wastes. Their goal was to determine the maximum amount of waste (dry mass) that BSF maggots could process in a day while maximizing dry biomass production. Two hundred, six day old larvae were fed differing rates of various wastes. Quantitative nutritional aspects were based on the terminology outlined in Slansky and Scriber (1981). The following quantities were defined by Slansky and Scriber:

- B – the biomass that was gained (measured by Diener et. al. as the increase in pupae mass);
- I – the total food offered;
- F – the residue leftover in the containers (includes undigested food and excrement);
- M – the ingested food that was incorporated into biomass (calculated by mass balance in Diener et. al. (2009));
- AD – how much of the ingested food is digested; and
- ECD – how efficiently digested food is converted to biomass.

The values are related in the following fashion, with higher ECD values indicating a high food to biomass conversion efficiency.

$$ECD = \frac{B}{I - F}, B = (I - F) - M$$

Diener et. al. 2009 used these values, along with a reference food source, to estimate the amount of a waste type that could be added to a given area density of larvae. Diener et. al. (2009) showed that, an optimal feeding rate of 100 mg of chicken feed with 60% moisture (the reference food source) per larvae resulted in maximum tradeoff between nutrient rich prepupa and high waste consumption in the shortest time span. Using digestible energy as reference point, Diener et. al. (2009) projected the results to different kinds of waste. The actual calculations used to obtain the conversions were not described in the paper and the results obtained depend on a variety of factors including the fiber content of feed, moisture content of feed and ambient temperature which was not defined. Diener et. al. (2009) did not calculate the AD variable in their study (as suggested by Slansky and Scriber (1981)) which may have affected the accuracy of their estimates. Nonetheless, the projected conversion rates for additional waste types serve as useful starting points for operating a waste processing facility using BSF larvae. The estimates proposed by Diener et. al. (2009) are listed below for various feed sources. The table assumes a maggot density of 5 larvae/cm² but does not mention the age of the larvae.

Table 1 Waste Loading Rate – adapted from Diener et. al. 2009

Waste Type	Daily Feeding Rate (mg/larva/day)	Waste Loading Rate (kg/d/m²)
Chicken Feed	100	5.00
Kitchen Waste	61	3.05
Vegetable Waste	98	4.90
Green Banana	103	5.15
Pig Manure	158	7.90
Poultry Manure	175	8.75
Human Faeces	130	6.50

Although all the aforementioned studies showed promising results, the further development of this technology from an experimental scale into a practicable full-scale plant is yet to gain much ground. ESR International LLC in the United States has recently begun manufacturing the black soldier fly bioconversion unit in polyethylene (US patent 6,780,637). But due to cost and weight issues, they are not producing pre-cast concrete now. These units resemble garbage bins, but these bins are somewhat special in that they possess evacuation ramps that permit the larvae to self-harvest into a bucket (www.esrint.com). This 2-foot (0.61m) residential unit by ESR LLC has an average feeding surface area of 0.34 m^2 . At a disposal efficiency of $15 \text{ kgs/m}^2/\text{day}$, it can handle over 5 kgs of food waste per day. It can hold or contain over 144 liters of larval residue, and with a reduction in weight and volume of 95%, it must be emptied after receiving a total of



Figure 2 Residential biopods designed by ESR

(Source www.sprint.com)

2.89 m^3 of food waste. This unit serving a family of four people would have to be cleaned out approximately once every 8 years. With this larval bioconversion process, the costly transport of food waste to landfill is completely eliminated. However, ESR LLC is yet to produce relatively larger bins for municipal use and to explore the production of concrete bins in developing countries which would be cheaper as cost of labour and cement material is abundant.

Undoubtedly, organic waste continues to cause several problems in developing countries, as no valid solution has yet been identified. Development from experimental to full-scale waste treatment facilities, using the larvae of the black soldier fly, offers several advantages. Since such facilities can be developed and operated at low cost (low building and maintenance costs; independent from power supply), they are more adapted to the economic potential of developing countries (Diener et al. 2009). Furthermore, creating additional value chains and generating a surplus income through the sale of harvested prepupae or their use in animal husbandry can strengthen the economic resilience of farmers or small entrepreneurs to natural hazards or market fluctuations (Diener et al. 2009).

2.6 PRODUCTS FROM TECHNOLOGY

The diminishing global supplies of wild forage fish and rising market prices for fishmeal have prompted the animal feed industry in recent times to look for alternate protein sources (Diener et al. 2011a). The prepupae of BSF contain on average 44% crude protein and 33% fat (St-Hilaire et al. 2007). As a component of a complete diet they have been found to support good growth of chicks (Hale 1973), swine (Newton et al. 1977), rainbow trout (St-Hilaire et al. 2007) and catfish (Newton et al. 2004). Reviewed literatures reveal that prepupae meal can replace at least 25% of the fish meal in a diet with no reduction in gain or feed conversion ratio in a rainbow trout (St-Hilaire et al. 2007) and channel catfish. It is an appropriate alternative to fishmeal in animal feed with a potential market value of 330 USD/ tonne dry weight (Newton et al. 2005b). Bondari and Sheppard (1981) also revealed that there was no significant difference between diets using *Hermentia* larvae and commercial diets when blind taste tests with tilapia and channel catfish

was done. Hem et al. (2008) successfully applied the larvae of the black soldier fed on palm kernel meal as feedstuff in tilapia culture in the Republic of Guinea as well.

The residue the BSF treatment produces is another product that can be useful. During the bioconversion process, some nutrients like nitrogen or phosphorous is broken down (reduced) alongside the dry mass. A possible designated use of this residue is application in agriculture, similar to compost or subsequent processing in a biogas facility (Diener et al. 2011a). Newtons (Newton et al. 2005a) experiment with larvae residue from pig manure did not show promising results pertaining to performance of basil (*Ocimum basilicum*) and Sudan grass (*Sorghum sudanese*) grown on BSF mixtures of BSF residue with either clay or sand. Therefore more studies would have to be done if residue is to be used in agriculture.

Furthermore, fat can be extracted from the BSF larvae and prepupae for biodiesel production. Given the limited data on the extraction of fat from insects on large scale (commercial), more research is required to be fully explored.

2.7 RESEARCH GAPS

A typical waste stream is highly variable, being made of several different components. Its characteristic is dependent on the life style of producers. Therefore, the waste mix in a developed country is different from that of a developing country and generally from place to place. For example, the organic percentage of waste in Nairobi Kenya is 65% whiles San Francisco, United States is 34% (UNHABITAT 2010). However, most of these researches were done in developed countries with only few in developing countries. The only known applications of the soldier fly technology in the tropics are the bioconversion of palm kernel meal in the Republic of Guinea

and in Indonesia (Hem et al. 2008; Flechet 2008). There is the need to try the usage of this technology elsewhere to see its viability or sustainability.

The various studies mentioned all used homogenous organic materials and few studies centered on food scraps, household waste or faecal sludge as a feed source (Diener et al. 2011a; Barry 2004; Newby 1997). There is limited research in the use of non-homogeneous organic material. But, if this technology of using the BSF larvae is to help solve waste management problems, then there is a need to mimic actual waste to test the extent of bioconversion. For example, Diener (2011b) evaluated the feasibility of the black soldier fly to digest mixed municipal organic waste in a medium scale field experiment in Costa Rica, and explored the benefits and limitations of this technology. Average prepupae production of 252 g/m²/day was achieved and waste reduction ranged from 65.5 to 78.9%. This work however, did not give insight into the components of the mix.

Little information is available on the capability of the larvae to digest municipal organic solid waste or to process agricultural waste (e.g. coffee, banana), as well as excreta and faecal sludge from on-site sanitation facilities (e.g. pit latrines) – all waste products whose disposal and management cause serious challenges in low and middle-income countries (Diener et al. 2009).

Upscaling this technology to commercialise waste treatment using the *Hermentia* larvae is still in the embryonic stage. If the full potential of this new technology is to be exploited, then research more research has to be done. Thus, this research work was to examine the potential of the black soldier fly larvae to degrade food waste (from restaurant) on the campus of the Kwame Nkrumah University of Science and Technology (KNUST), Kumasi – Ghana, West Africa.

3.0 METHODOLOGY

3.1 INTRODUCTION

In response to the problem of disposing organic solid waste, the technology of bioconversion of organic solid waste using the larvae of the black soldier fly is becoming increasingly popular. Though the problems associated with solid waste management is more challenging in the developing countries, little work or no work has been done to explore the use of this technology in Africa. Thus, this research work seeks to explore the possible use of the technology in Ghana, Africa using very simple and low-cost materials and techniques in generating the larvae and degrade organic solid waste.

In order to assess the use of BSF larvae to digest and degrade municipal organic solid waste, organic waste was collected from a restaurant on KNUST campus and test were performed on the sample of the feed to determine the constituents. The BSF was attracted using various baits and the larvae generated from the eggs laid by the adult BSF were used to inoculate a weighed amount of solid waste to assess the waste reduction capacity of BSF larvae. The diagram (Fig. 3) shows an overview of the general methodology used in the research.

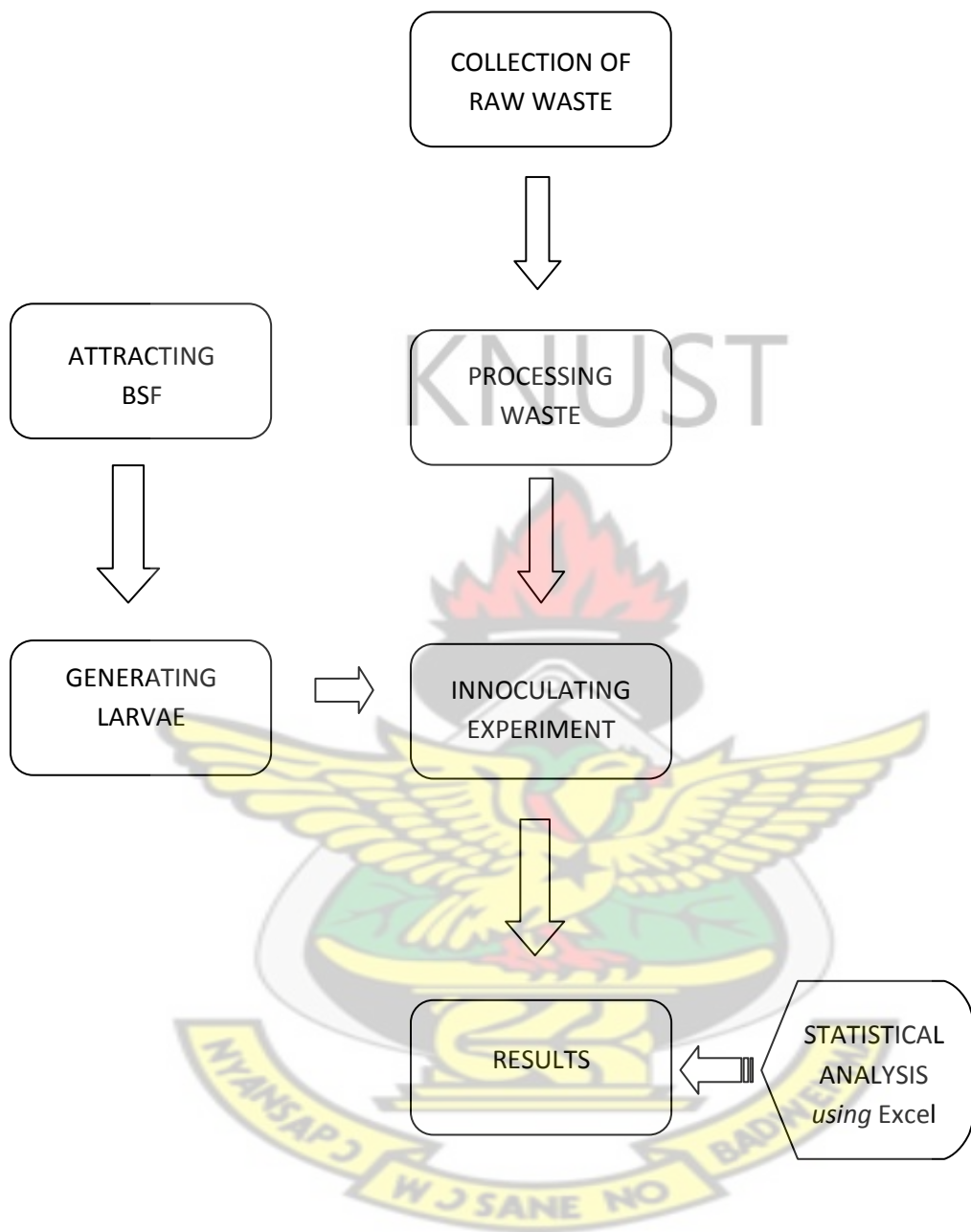


Figure 3 General overview of methodology

3.2 STUDY SITE

The research was mainly carried out in the Environmental Quality Laboratory of the Civil Engineering Department with aspects of it in the Zoology Laboratory of the Biological Science Department and the Chemical laboratory of the Faculty of Natural Resources, all of the Kwame Nkrumah University of Science and Technology (KNUST), in Kumasi, Ghana.

KNUST is located in Kumasi, the second biggest city and capital of the Ashanti Region of Ghana. KNUST is one of the Public Universities with a population of 36,000 whiles that of the entire city is 1,170,270 (2000 Census). Kumasi has attracted such a large population partly because it is the regional capital, and also the most commercialised centre in the region.

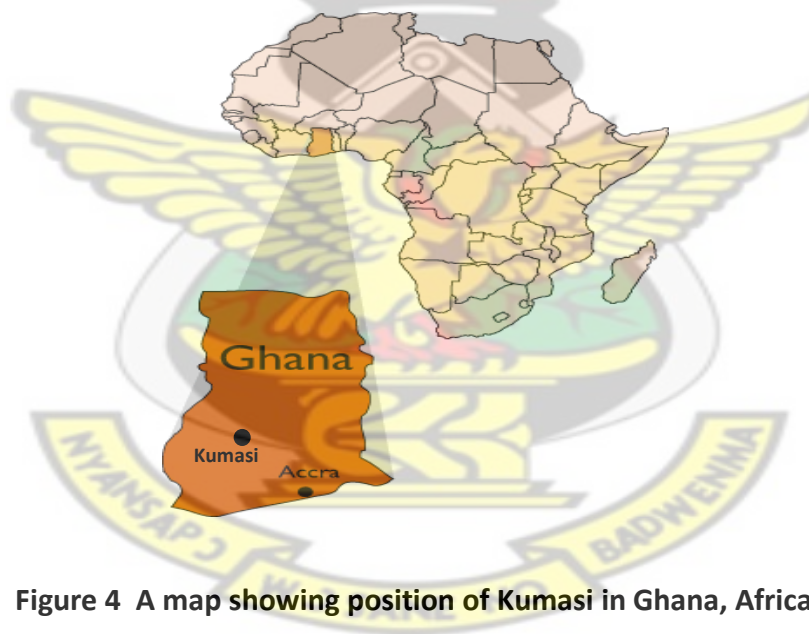


Figure 4 A map showing position of Kumasi in Ghana, Africa

Kumasi is located in the transitional forest zone and lies between latitude 6.35° – 6.40° and longitude 1.30° – 1.35° , having an elevation which ranges between 250 – 300 metres above sea level with an area of about 254 square kilometres. The average minimum temperature is about 21.5°C and a maximum average temperature of 30.7°C . The average humidity is about 84.16 per

cent at 0900 GMT and 60 per cent at 1500 GMT. The double maxima rainfall regime is 214.3mm in June and 165.2mm in September. (KMA 2012)

It has been estimated that the Kumasi Metropolitan Assembly (KMA) generates about 1500 tonnes of solid waste daily based on the current population. The Waste Department of KMA is overwhelmed by the task of hauling all the waste generated in the city – only 1300tonnes (86.67%) of the total waste generated is collected, leaving a back log everyday uncollected. This collected waste ends up at the Dompase Landfill Site, Kumasi. The composition of the waste includes organic material (40.19%); plastics (19.86%); glass/bottles (1.2%); paper and cardboard (7.04%); metals (2.23%); textiles (6.94%); inert (sand, ash, demolition waste, fine organics) material; and wood (1.71%) (KMA 2012).

3.3 THE FLY COLONY

3.3.1 ATTRACTING THE INSECTS

Several attractants were initially tested to ascertain the one that would attract more BSFs and at a faster rate. Wet chicken feed which was bran from a corn mill; fresh fish (head and gills); fresh human faeces; and finally fresh rat meat (carcasses from a Biology class) was tried.

In determining which attractant was more suitable, a selection matrix was used. Parameters used were chosen based on the advantages and disadvantages, characteristics of each treatment option and the effects on the environment. Four major parameters were considered in structuring the selection matrix: Cost; Sanitary; Availability; Ability to attract insects. The cost referred to how much in monetary terms it would cost to use that attractant in the process. Sanitary, was looked at in terms of odour (stench), how hygienic it is using that attractant and the risk of being infected with diseases when in contact with the attractant. Availability, meant how easily

accessible it is to obtain the attractant. Ability to attract insects was based on human observation, how quickly the insects came towards the attractant and how many they were.



Figure 5 Using fresh fish as an attractant

The adult black soldier flies, *Hermetia illucens* were attracted into small cages (30x30x45cm) built of wood and nylon netted side walls. It had two windows opposite the other to serve as opening to allow insects into the cage (fig. 3.3). The set up was placed near a farm situated next to a refuse dump site further behind the food canteen at the Biological Science Department of KNUST for a day to attract several BSFs. The stench from the meat set up was more and attracted faster more BSFs and was preferably used subsequently discontinuing the use of other attractants. This attracted other insects as well, like the house fly, *Musa domestica* into the cage. The BSFs were then separated from the other insects.



Figure 6 Insect trap for attracting the insects

3.3.2 ESTABLISHING BSF COLONY

The population of the adult BSF was established by separating them from houseflies and other insects. This was done by using cotton soaked with alcohol to drowse the insects in the cage (for a short while) and then transferring the BSFs into another insect trap of dimension 0.5x0.5x0.5m made of wood and a partially transparent white cotton cloth as side walls. The very minute holes of the cloth allowed sufficient air passage and prevented smaller insects from penetrating). The structure had an opening on one of its side walls fitted with a cloth which is tube-like in shape (figure 9).



Figure 7 Separating the BSFs into another insect trap



Figure 8 Transferring BSF into another insect trap after drowsing insects using alcohol

3.3.3 GENERATING THE LARVAE

Moistened fresh rat carcass placed in transparent plastic containers (13.5x13.5x4.5cm) was put into the cage for the adult flies to lay their eggs by it. The setup was placed on a raised platform to prevent ant and other predators. Because the work was carried out in the dry season (November – January) the setup had to be watered from time to time to keep conditions moist for the larvae to grow. The larvae hatched approximately 3-4 days after oviposition. Using forceps, 4 – 5 days old larvae were transferred to inoculate the experimental setup in the Environmental Quality Laboratory of the Civil Engineering Department of KNUST. Prior to inoculating the final setup, 10 randomly picked larvae were weighed and all the larvae as well to determine the average initial weight per larvae.

3.4 THE ORGANIC SOLID WASTE

3.4.1 COLLECTION OF RAW WASTE

The organic solid waste used was obtained from a restaurant – *Obaapa* – on the Campus of KNUST by means of a source separation done at their kitchen. Fresh waste (food waste) generated in a particular day was collected (1kg) in black polythene bags and taken to the laboratory for use immediately. This ensured that bacteria or fungus degradation had not started and flies infestation of waste was also avoided.

3.4.2 CLASSIFYING ORGANIC WASTE

The solid organic waste to be used was classified to give an idea of the nature of the mixed sample and inform future research. A simple classification of the organic food waste was

employed as there is an extensive variety in waste produced. The organic solid waste classification used was

- Cooked foods (rice, yam, meat, bones, etc)
- Uncooked foods (vegetables, peels of yam, cassava, plantain etc)

The total weight and the weight of each fraction were measured. Then a small sample (about 10g) of the thorough mixture was taken. It is weighed and placed in the oven for 24 hrs at 105°C to help determine the moisture and dry weight of the sample.

3.4.3 CHARACTERISATION

The food waste was shredded using a scissors and the hands to get the peels of raw food stuff like plantain and cassava into small fragments.



Figure 9 Waste collected from restaurant being separated into uncooked to be weighed

Thereafter, the mix was grounded to obtain a uniform paste by adding distilled water (about 0.7litre to 1kg of waste) and blending with a blender (Master Chef[®], speed of 2 at intervals of 15seconds for 6cycles). This was to increase the surface area to be fed on by BSF larvae.



Figure 10 Master Chef Blender getting ready to be used

A sample of the organic waste substance was taken to the Chemical Laboratory of the Natural Resources Faculty to test for the nutritional value of the waste. The Proximate test was done to obtain moisture, carbohydrate, protein, nitrogen and ash content. Samples were also collected for a Chemical Oxygen Demand (COD) and Biochemical Oxygen Demand (BOD) test to be done in the Environmental Quality Laboratory. Below is a table showing the tests done and the method used.

Table 2 Experimental parameters and analytical method

Parameter	Equipment	Unit of measure
BOD	Winkler Method	mg of O ₂ /l
COD	Closed Reflux method	mg of O ₂ /l
Temperature	Thermometer	⁰ C
pH	pH meter	-
Moisture	Oven heating at 105 ⁰ C	%
Volatile solids	Ignition at 550 ⁰ C	%
TKN or N%	Macro Kedjahl Method	%
Fats	Soxhlet aparatus	%

3.5 EXPERIMENTAL SETUP

Two similar experiments were conducted to assess consumption rates and determine optimal feeding rates. The problems encountered in the first, informing the setup in the second. Transparent plastic bowls (13.5x13.5x4.5cm) with lids were used for the feeding experiments in both cases. The lids had 5 holes of diameter 20cm hand drilled to allow for a circulation. In the first experiment, mosquito net was placed between the lids and the plastic bowls as a measure to control other insect intrusion (as suggested by Diener 2009). Five feeding rates were employed and classified as: B25, B75, B100, B125 and B200. The feed rates were 0.25, 0.75, 1.00, 1.25 and 2.00 g/larvae/day respectively. Each treatment had three replicates consisting of 50 larvae, 4-5 days old. The setup was placed in a dark corner to avoid light disturbances.

In Dieners (2009) work, 200 larvae (6 days old) per treatment were used. Chicken feed was used as feed with feeding rates: 12.5, 25, 50, 100 and 200 mg /larvae/day. However, in this research, the production of larvae were in small batches, so 50 larvae were used. Thus, using a similar feed rate would be small, therefore the change in feed rates employed per larvae per day. Diener (2009) found out that 100 mg/larvae/day resulted in optimal bioconversion in a short time. Therefore, feed rates quite close to the optimal value found by Diener (2009) were chosen to see if there would be any variations. Hence, the addition of classes B75 and B125 as mentioned above.

After every two to three days, weights of the boxes are measured and five (5) randomly selected larvae are weighed to help in determining the residual weight. Transferring all larvae before measurements was seen as tedious and hence not employed. Three (3) larvae are weighed to find their wet weight and dried in an oven for 24 hours at 105 °C to determine both wet and dry weight of larvae respectively. The experimental diets were added at the same time following the above mentioned feed rates. During the same times, samples of the residue were taken and dried in an oven for 24 hours at 105 °C to measure dry matter.

Feeding was applied till about 50% prepupae were seen. This is identified by the change in colour from white to dark brown. The prepupae are weighed and the dried in an oven as mentioned previously for the dry matter. At the end of the experiment, all larvae left are taken out and the weight of the box with the residue is obtained. A sample of the residue is taken to the Soil Science Laboratory of the Faculty of Natural Resources to do a proximate analysis to compare with initial.

The second experiment was similar to the first with just some changes in materials and the addition of another container for each feed rate to serve as control (without larvae). The mosquito net was replaced with a cotton material cloth (held tightly between the box and the lid by a rubber band) with tinnier holes that still allowed sufficient circulation but small enough that the roving larvae in the box not to escape as in the first experiment. The inconsistencies in the first experiment due to migration of the larvae in the box and proliferation of other larvae resulted in rejecting the first experiment and not using it in the analysis. This is likely because of oviposition by other insects that managed to get into the setups.

Table 3 Feed classes and their feed rates

Feed Class	Feed rate (g/larvae)	No. Of larvae
B25	0.25	50
B75	0.75	50
B100	1.00	50
B125	1.25	50
B200	2.00	50
B25/C	0.25	-
B75/C	0.75	-
B100/C	1.00	-
B125/C	1.25	-
B200/C	2.00	-



Figure 11 Set-up for experiment 2

3.6 DETERMINATION OF VARIABLES

In bioconversion technology, the concern is raising the larvae to degrade the organic matter efficiently not only rearing the larvae. The following variables were employed therefore to examine food consumption: *waste reduction index (WRI)*, *dry matter extraction (DME)*, *conversion rate (CR)*. Statistical analyses were performed on the variables obtained using a one-way ANOVA to assess the differences between the feeding classes. The statistical tools used was Microsoft Excel 2007.

3.6.1 WASTE REDUCTION INDEX

Since the duration taken to degrade food varies per feed amount, it was factored in evaluating the amount of food reduced by using a waste reduction index (Diener 2009). This is given by

$$WRI = \frac{D}{t} \times 100, \quad \text{where } D = \frac{W - R}{W}$$

Where D is the overall degradation, t is the total number of days for larvae to feed on the food, and R is residue after time t .

3.6.2 DRY MATTER EXTRACTION

Dry matter extraction (DME) is the percentage of dried feed consumed by larvae. This is a more accurate measure of the larvae consumption as variations in moisture content due to evaporation are avoided (Barry 2004). This is calculated as follows

$$DME \% = \left(1 - \frac{\text{dry matter residue}}{\text{dry matter feed}}\right) \times 100$$

3.6.3 CONVERSION RATE

Conversion rate (CR) is the amount of dry matter feed used by the larvae to grow into dry matter prepupae. Therefore, the greater the CR value the more efficient the prepupae production (Barry 2004). This is calculated as

$$CR \% = \left(\frac{\text{dry matter prepupa}}{\text{dry matter feed}}\right) \times 100$$

4.0 RESULTS AND DISCUSSION

4.1 CHALLENGES IN GETTING LARVAE

4.1.1 SELECTING AN ATTRACTANT

The first challenge involved in this bioconversion procedure was getting the adult BSF only to establish a colony. There was a need to first attract the insects using bait (attractant). To test which bait would produce best results, different attractants were tried. The results obtained are shown below:

Table 4 Selection matrix for attractant

Parameters	Fish	Chicken feed	Human Faeces	Rat meat
Cost	GH ₵2 per 1	GH ₵1.2 per 1kg	free	GH ₵5 per 1
Sanitary	medium	medium	low	medium
Availability	medium	medium	high	low
Ability to attract insects	medium	low	high	High

From the results above, using the human faeces attracted more insects especially the BSFs. The faeces used were fresh stool of a young boy which was wetted with water. Obtaining this faecal matter was relatively easy and cheap as no cost was involved. The trial revealed on one hand, the potential of the larvae of the BSF to degrade faecal matter. When the setup was left in the open, eggs of BSF was laid on the faecal matter and later hatched into larvae which feed voraciously on the faecal matter.

Thoroughly wet chicken feed as proposed by Tomberlin & Sheppard (2002) and successfully used by Diener (2009) proved relatively slow in attracting insects. Though this did attract some BSFs together with other insects, by human observation, it was relatively not that much. The wet chicken feed was quite slow in fermenting to generate the kind of smell that attracts the flies. The chicken feed used was bran obtained from a corn mill. There was no means to classify the constituents of the bran obtained from the corn mill as did Diener (2009) who used an industry processed chicken feed. Thus, it cannot be said with certainty if that is the reason for the results obtained.

The fresh fish used was obtained from the Ayeduse market. The cost of the fish was GH¢2 per one medium sized fish. Three were used. The head and gills were isolated from the main body of the fish and both were thoroughly wet with water. A container bearing the head of the fish attracted more insects than the one with the other body parts.

The selection matrix showed that the rat meat thoroughly wet with water was the best attractant and therefore used chiefly in subsequent experiments to attract BSF insects. The rat meat used was obtained from the Animal House of the Biological Science Department at a cost of GH¢5 per medium sized one. It was thoroughly wet with water and did attract a lot of insects including the BSFs.

But from the experiment, it can be deduced that all the attractants can work well in getting insects of BSF. Thus, all of them could be used in attracting BSF insects. Any meat thoroughly wet can generate enough stench over a short while to attract a lot of insects.

4.1.2 ATTRACTING INSECTS

During the experiment, the setup to attract insects was placed near a refuse dump area with the notion that more insects would be attracted and at a quicker pace. However, it was noticed that irrespective of the place, the insects can scent the attractant and move around quickly from where, one cannot tell. This was noticed as the setup was placed near the Steven Paris Hostel on KNUST campus for a while and being moved from place to place. Similar results were observed as the setup was placed around the car park area of the Biological Science Block of KNUST. Thus, specifically citing setup near a dump site was found not to be that relevant. Once the stench generated by the attractant is not of public nuisance and considerably farther away from any other human activities, it can be placed anywhere and it would attract enough insects in a short while. The stench from the attractant is a lot and one has to be in nose mask to make this bearable. Lizards and ants were seen as the major predators. These were controlled by setting up on raised platforms and placing the attractant in insect traps.

The biggest challenge was separating the BSFs from other insects. This challenge was overcome by using descriptions and pictures from literature and the internet. It was challenging identifying the male and female adult species. This would have informed mating processes and other behavioural patterns of the BSF. However, doing so was not of much importance to this research and was thought of as unnecessary and tedious. The procedure of separating the insects by drowsing them was found to be a little technical as experience in doing this was essential in successfully separating them. Using alcohol was affordable and readily available. Cotton wool was partially dipped into alcohol during the usage.

4.1.3 GENERATING THE LARVAE

After using alcohol to help separate BSF from other insects, the substrate used to generate the larvae should always be kept moist not wet as it becomes difficult for the larvae to move about and feed properly. When conditions are more than moist (wet), it impedes movement of the larvae while dry conditions results in migration of the larvae out to look for feed. The setup should be kept under a shade to reduce evaporation and give a suitable environment.

The larvae needs a good circulation of air but if care is not taken and the right material chosen for covering the experimental box, they would migrate elsewhere with time. Thus, in this experiment, it was noticed that using a netted nylon mesh resulted in movement of some of the larvae out of the experimental box affecting results of the research work though this was successfully used by Diener (2009). Diener did not mention specifically in details the nature of the net used. The smallest net on the market is what was used in this work. Therefore, the possible explanation to this is that Diener (2009) used a net with tinier holes than the one available on the market in Ghana here. To address this challenge, another material was used which allowed adequate air circulation but prevented migration out of the experimental box. The material was a cotton lace-like material as shown in Plate 4.1a. While the experimental box with nylon net required tying with a rubber band (as seen in plate 4.1b) when used, that of the cotton lace-like material did not need that as it was firmly fastened between the box and its lid.



Figure 12 Materials used for covering box and lid. Right was used for the first experiment and the left for the second experiment

Again, one challenge involved being able to distinguish between the larvae of the BSF insect and others especially the housefly. Though a pure colony of BSF was established before larvae generation, knowledge of how really the larvae of the BSF looks like was seen as necessary. The research helped in determining the difference in larvae between the housefly larvae and that of the BSF larvae. This was based on observation as well as information gathered from the internet. Below is a table showing the difference between the larvae of the Black Soldier fly larvae and that of the housefly.

Table 5 Difference between the BSF and Housefly larvae

Difference	Housefly larvae	BSF larvae
Size	Reach length of approx 12mm	Grow to 19mm, bigger
Appearance	Light in colour, smooth	Much thicker, distinct ridges
Development	Hatch in a day	Requires about 4 days

4.2 WASTE CHARACTERISTICS

About 1 kg of food waste was collected from the Obaapa Restaurant. This source was chosen because of proximity to Laboratory area, the certainty of constant supply of organic waste when needed and giving a fairly similar waste content as food menu was quite similar on daily basis. The waste collected included cooked rice, bones, meat, fish, vegetables, peels of plantain, etc. The waste collected was a mix of cooked and uncooked. The ratio of cooked feed to uncooked feed was 1:1. Results of the proximate analysis of the initial waste feed sample before inoculating the experiment as well as other tests is summarised in the table below.

Table 6 Results of tests conducted on initial waste sample

PARAMETERS	MEASURING UNIT	RAW WASTE
MOISTURE	%	90.26
ASH	%	7.39
FAT	%	21.53
CRUDE FIBRE	%	2.97
PROTEIN	%	24.19
NITROGEN FREE EXTRACTS	%	43.92
CARBOHYDRATES	%	46.89
ENERGY	(KCAL/100G)	466.24
pH	-	3.9
TEMPERATURE	⁰ C	32
COD	mg O ₂ /L	4440
BOD	mg O ₂ /L	3020

The initial moisture content of the raw waste was high due to the amount of distilled water added (0.7 ml per 1kg) during the preparation of the waste before inoculating the experiment. The water added helped in blending the waste sample thoroughly to get a uniform paste. Nitrogen Free Extract (NFE) represents the soluble carbohydrate of the feed, such as starch and sugar.

Crude fiber represents insoluble carbohydrate. The ash fraction contains all the mineral elements put together. The initial pH is in the acidic range. Allatar, (2012) found out that BSF larvae are tolerant to initial pH levels ranging from 0.7 to 13.7 and are able to amend initial pHs of 2.7 to 12.7 to between 7.8 and 8.9 within one to two days. Assessment of pH was not a focus in this research but the generally high degradation recorded showed that whatever changes in pH that may have resulted, the larvae are able to adapt to it.

Results of the proximate analysis, COD and BOD of the residues left in each experimental box are presented in the table below. It was not possible to measure pH, temperature of the samples left as some were dry and small in number.

Table 7 Results of various tests on the residue in the experimental boxes

	B25	B75	B100	B125	B200
MOISTURE	-	40.29	26.73	16.8	78.29
ASH	-	13	15	10	8
FAT	-	8	10.5	28.5	32
CRUDE FIBRE	-	29.73	17.89	15.83	7.43
PROTEIN	-	22.49	28	20.78	18.59
CARBOHYDRATES	-	56.51	46.5	40.72	41.41
COD	-	14880	18560	19040	6480
BOD	-	5210	7800	10472	3954
BOD/COD	-	0.35	0.42	0.55	0.61

Residue from class B25 was unable to be analyzed as the residue sample left was too little for all the various tests (fig 13).



Figure 13 Residue of one of the experimental box for class B25

Results obtained from the residue were compared with that obtained from the initial (fresh) organic waste. Shown below is a graph comparing the various parameters that were analyzed.

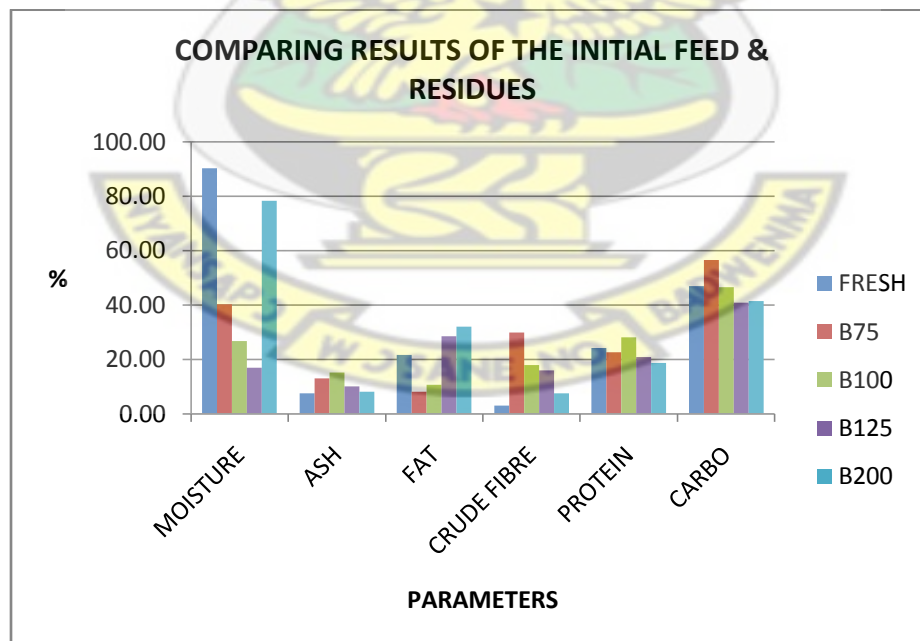


Figure 14 Graph comparing initial waste sample (fresh) to residue obtained after bioconversion

According to the BOD and COD value of the initial raw waste, we can say that the raw waste had a high degradability potential. Of course, this was expected as sample taken was a pure mixture of organic matter. The BOD and COD values of the residue and the initial raw waste were compared as well. Katarina, (1997) reported that the BOD/COD ratio, if lies below the range of 0.5 indicates that the sample contains less organic matter that are degradable and vice versa. BOD/COD of the residue lies in the range of 0.35-0.61. Food classes B25 had totally degraded and samples were unable to be taken for tests. B75 and B100 had been degraded to a larger extent hence the BOD/COD ratio being less than 0.5. That of classes B125 and B200 indicates that the waste can be further degraded as it was observed that there were some parts of the waste the larvae had not tempered as a result of anaerobic conditions developing likely due to a lack of drainage system in the experimental box setups.

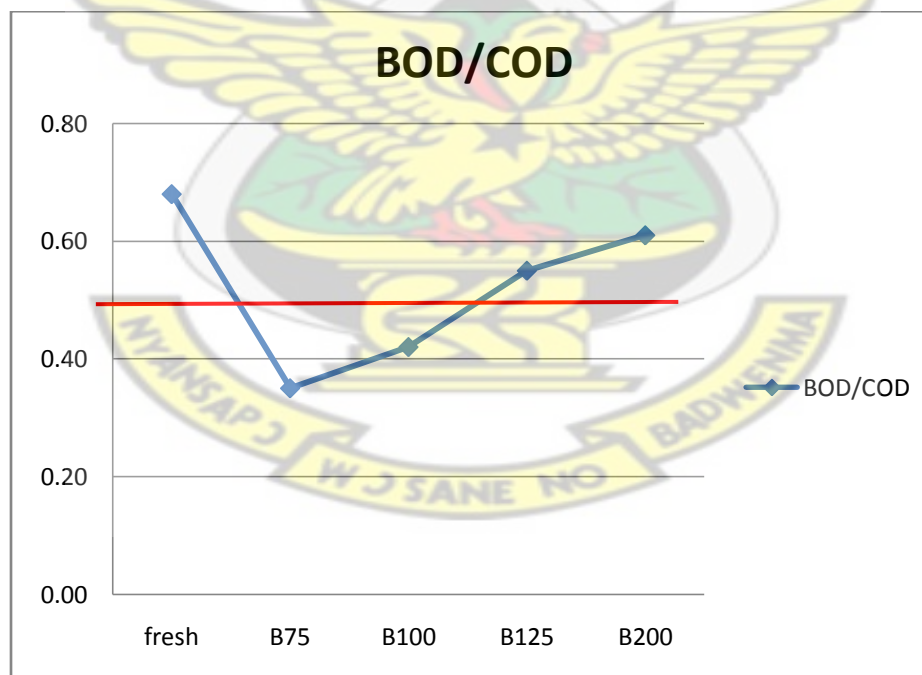


Figure 15 A graph showing the BOD/COD ratios of residue of the feed classes

Results obtained from the residues in the experimental boxes at the end of the experiment were compared with the initial waste added to see if there were any significant differences. The graph and results obtained after doing a One-way ANOVA in Microsoft Excel 2007 is shown below.

Table 8 ANOVA Table from Excel 2007 for comparing initial waste and residue

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	452.3957	4	113.0989	0.23812	0.91411	2.75871
Within Groups	11874.14	25	474.9655			
Total	12326.53	29				

where *SS* – sum of squares; *df* – degree of freedom; *MS* – mean squares; *F* – F score is the actual test statistic; *P-value* – significant level; *F crit* – critical F value.

There was not a significant effect of the different parameters on the residues of the waste sample as well as the initial sample (fresh) used. $F = (4, 25) = 0.238, p = 0.914$

There was therefore no pattern to establish. However, the results reveal the ability of the larvae to degrade a wide variety of waste including proteins and fats and oils. The presence of all these nutrients helped in the growth of larvae into the prepupae stage and finally the adult stage. The nutritional content of the residue shows that it could be explored to use in the agricultural industry as manure. However for this to be done successfully, much research must be done to see the effects on crop plant and its application to the soil.

4.3 WASTE REDUCTION

4.3.1 WASTE REDUCTION INDEX

Results obtained from the first experiment showed a lot of inconsistencies and thus was not considered in the final analysis. In the second experiment where an additional experimental box

was set up as a control, the percentage reduction for the classes with the higher feed rates was less. The control boxes, which involved another setup without any larvae showed similar trends (fig. 16).

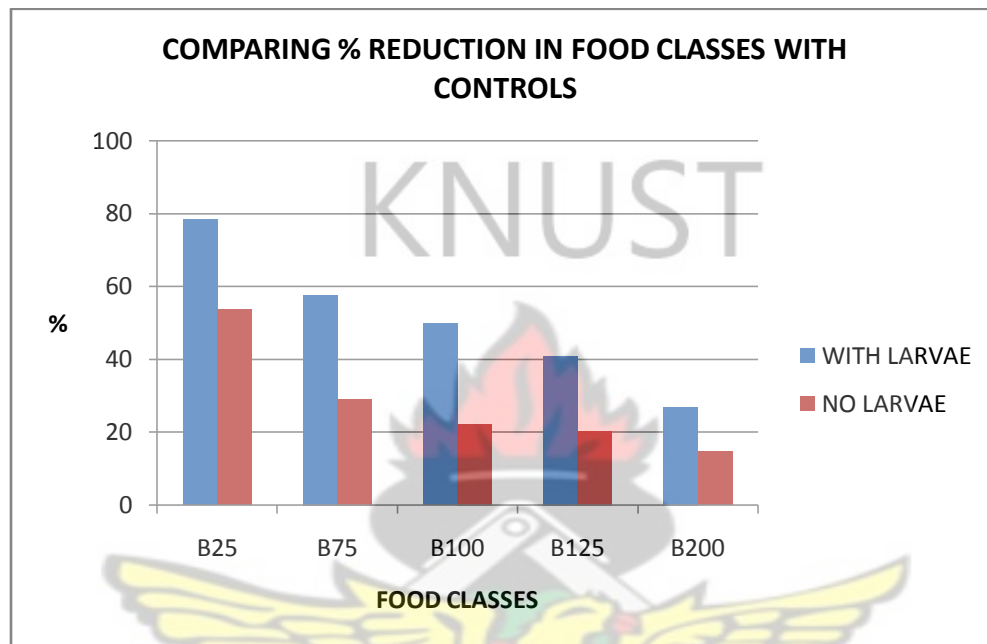


Figure 16 A graph comparing the waste reduction in the food classes and controls

The control boxes were placed at the same place as the other experimental boxes with larvae. Dehydration and the growth of mould (fig. 17) after a while on the feed in the control boxes likely resulted in the reductions. All the controls developed moulds and fungi. Since there are a large number of microorganisms in the environment, this did not come as a surprise. Moulds and fungi did not develop in the experimental boxes with the larvae till majority or if not all had died or become prepupae before showing signs as occurred in some boxes of B200. This observance suggests that the larvae have the ability to resist or suppress the growth of these microorganisms. It is likely they feed on them.



Figure 17 A Control box B75/C showing signs of mould/fungi infestations

It must be mentioned that the waste food added to the experimental boxes may have lost water during the experimental period. It cannot be determined for a certainty how much this loss influenced the results. It is likely that the smaller feed rates experienced a stronger dehydration due to unfavourable surface area to volume ratio, hence the large values in waste reduction.

To take into account the number of days each treatment used for the feeding stage, a Waste Reduction Index was calculated.

Table 9 WRI results showing mean values

	REDUCTION (%)	DAYS(d)	WRI(%/d)
B25	78.40	10	7.84
B75	57.61	8	7.20
B100	49.78	5	9.96
B125	40.88	5	8.18
B200	26.88	7	3.84

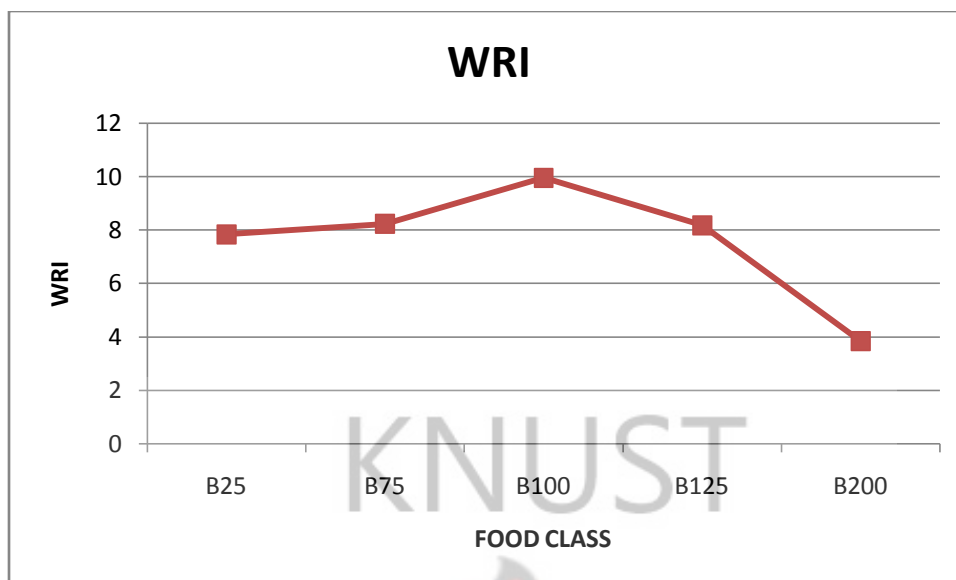


Figure 18 Waste reduction indexes for the different food classes

Applying the time taken for feeding of larvae in each food class showed that food class B100 had the highest WRI, indicating that 1.00 g of food per larvae per day is the most suitable feeding rate to efficiently reduce organic food waste.

4.3.2 DRY MATTER EXTRACTION & CONVERSION RATE

Bioconversion is not only about producing larvae but rearing them for efficient consumption of feed given. The DME and CR were therefore calculated as mentioned in Chapter 3.6. The table below shows mean values of results obtained.

Table 10 Results of DME & CR

	Dry Matter Extraction (%)	Conversion Rate (%)
B25	93.05	4.27
B75	94.01	58.77
B100	91.25	33.92
B125	62.81	86.48
B200	44.91	17.51

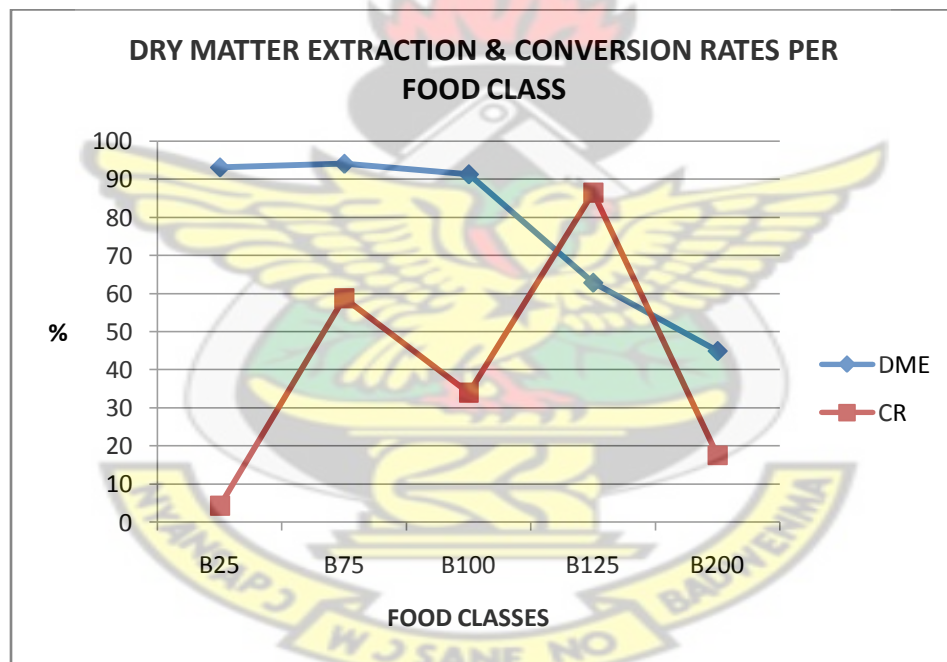


Figure 19 DME & CR of the Food classes

The dry matter extraction ranged from 44.91-94.01% with the average of 77.21%. The high values showed the ability of the BSF larvae to consume organic waste voraciously. On the other hand the conversion rates percentage followed a very different pattern than expected. B25 and

B200 recorded the lowest values, suggesting that the larvae were unable to benefit much from the food they consumed. Since the nutrients in diet have a direct bearing on prepupae development, it suggests that the nutrients available in B25 and B200 were quite not adequate. A look at the nutritional content of class B200 as revealed in the proximate analysis test done on its residue showed that, it had the lowest nutritional contents compared to all other classes except B25 where it is unable to tell as a proximate analysis was not performed on that due to inadequate sample availability after reduction.

4.4 DEVELOPMENT OF LARVAE

In the second experiment, the larvae were unable to escape as happened in the first experiment where nylon net was used in covering the experimental boxes. After more than 50% of the larvae turned into prepupae (identified by their change in colour from white to dark brown), one box each of each treatment (food class) was left unattended to see what would happen. It was interesting to note that, some of the larvae were able to go through the complete life cycle, turning into adult flies. This happened in all the different feed classes.

However, the adults died in no time with class B25 recording the largest amount of adult flies development. A total of about 42 were counted in the total time period of 18days since the set up was inoculated for B25. By the end of that time, B25 had 2 adult flies alive; B75 11 alive; B100 5 alive; B125 3 alive; and B200 1 alive.



Figure 20 Residue of box B25

The more development into adults in the B25 and B75 boxes (fig. 21) was possibly as a result of the higher reduction which resulted in emptying feed placed in them. Since the prepupae and pupa needs a dry environment to grow, those food classes were able to provide some dry areas to allow for pupation and subsequent emergence of adult (fig. 21).

5.0 CONCLUSION AND RECOMMENDATION

5.1 CONCLUSION

- This research proves that the larvae of the Black Soldier Fly (*Hermentia illucens*), are able to digest and degrade municipal organic solid waste in Ghana between 44-94% given a feeding rate of 0.25 – 2.00g per larvae per day.
- A daily rate of 1.00g of food waste (from kitchen restaurant) per larvae per day is able to meet high organic matter extraction within a shorter period, producing better prepupal yield. Hence, the most suitable feeding rate. Therefore, for 1tonne of organic waste, one million BSF larvae are needed to successfully reduce it by 91.25%.
- To cultivate the BSF larvae, one has to apply an attractant (carcass of rat meat, fresh fish, chicken feed and human faeces) to attract matured flies. The BSF is ascertained by observation and separation. Afterwards, the larvae are generated by providing ideal moisture conditions for the larvae to hatch from eggs.
- The proximate analysis of the test feed shows 90.26% of moisture; 7.39% of ash; 21.53% of fat; 2.97% of crude fibre; 24.19% of protein; 43.92% of nitrogen free extracts; 46.89% of carbohydrates; and has 466.24% KCAL/100g of energy.
- The larvae of the BSF can resist or suppress fungal or mould growth as it feeds on the waste.

5.2 RECOMMENDATION

- Nylon net should not be used to cover bowl and lid as the larvae easily escapes from the feeding area.
- A thermostat and heating element be installed in the feeding containers to maintain constant temperatures. Light intensity measuring device should be attached to monitor the effect of light intensities on the larvae's performance as well. Humidity tests should be conducted to know the ideal range for larvae.
- Studies should be conducted using faecal matter to explore the potential of BSF in treating organic faecal matter as it was observed during the use of it as an attractant that the BSF larvae is able to feed on it.
- Studies should be done to determine if substrates shredded prior to consumption are consumed more rapidly.

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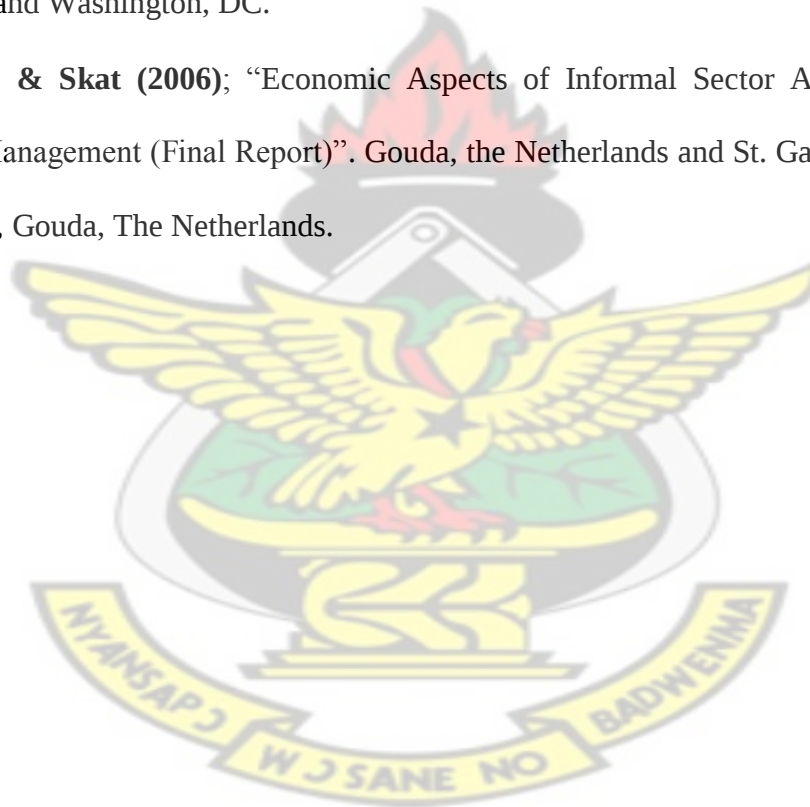
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APPENDIX

A. PICTURES FROM WORK



A-1 Nylon net used in Experiment 1



A-2 Entire setup of experimental boxes for experiment 2



A-3 Working in the laboratory



A-4 Residue of one of the boxes, B25



A-5 Some prepupae and adult flies that have developed



A-6 Weighing of experimental boxes(left) and residue (right)



A-7 Control boxes (B100/C left and B25/C right) showing signs of mould fungal growth



A-8 Residue after degradation and introduction of fresh feed sample (left)

B. RAW DATA

B.1 Results of initial raw waste characteristics

CODE	% MOISTURE	% ASH	% FAT	% CRUDE FIBRE	% PROTEIN	% NFE	% CARBO	ENERGY (KCAL/100G)
K1	90	6.9307	21.393	2.9851	24.063	44.629	47.614	467.302
K2	90.521	7.8431	21.675	2.9557	24.313	43.214	46.17	465.179

B-2 Results of DME

Dry matter extraction % = $[1 - (\text{dry matter residue} / \text{dry matter diet})] \times 100$.					
Calculating dry matter diet		Initial weight 14.6989	Final weight (dried) 13.0824	Dry matter diet 1.6165	
Feed class	Initial weight		After oven drying at 105°C for 24hrs	dry matter residue	DME
B25	15.0868		14.9745	0.1123	93.05289
B75	13.4554		13.3585	0.0969	94.00557
B100	13.0269		12.8855	0.1414	91.25271
B125	15.7179		15.1168	0.6011	62.81472
B200	14.8318		13.9412	0.8906	44.90566

B-3 Results of Conversion Rate

Conversion rate % = $[\text{dry matter prepupae} / \text{dry matter diet}] \times 100$						
Feed class	Container weight	prepupae weight	Final weight after drying	dry matter prepupae	DM of all prepupae	CR
B25	14.3149	0.0216	14.329	0.0075	0.075	4.639654
B75	13.0823	0.1508	13.1381	0.095	0.95	58.76895
B100	12.6809	0.0913	12.71737	0.05483	0.5483	33.91896
B125	14.8742	0.2068	14.9412	0.1398	1.398	86.48314
B200	13.3985	0.039	13.4092	0.0283	0.283	17.50696

B-4 Initial weights of 5 day old larvae collected before inoculation

larvae	batch 1	batch 2	batch 3
1	0.0169	0.0188	0.0166
2	0.0345	0.0179	0.0295
3	0.0291	0.0185	0.0081
4	0.0185	0.0154	0.0052
5	0.0136	0.0208	0.0166
6	0.0363	0.0254	0.0139
7	0.0233	0.0227	0.0166
8	0.0319	0.0269	0.03
9	0.0383	0.0226	0.0135
10	0.0346	0.0286	0.0185
average	0.0277	0.02176	0.01685



B-5 Calculating waste reduction (from excel)

	box weight	feed amt	all content		larvae wt	RES	l1	l2	l3	av-larvae	50 larvae	feed left	% red
B25/1	30.5	12.5	45	0.04	2	32.9	0.0273	0.0281	0.0281	0.027833	1.391667	1.008333333	91.93333
B25/2	30.5	12.5	44.4	0.028	1.4	33	0.0235	0.0315	0.0208	0.025267	1.263333	1.236666667	90.10667
B25/3	30.5	12.5	44.2	0.024	1.2	33.3	0.0185	0.0304	0.0227	0.023867	1.193333	1.606666667	87.14667
B75/1	29.4	37.6	68.6	0.032	1.6	49.3	0.028	0.0377	0.0295	0.031733	1.586667	18.31333333	51.29433
B75/2	29.4	37.5	68.8	0.038	1.9	49.4	0.0321	0.0344		0.03325	1.6625	18.3375	51.1
B75/3	30.4	37.5	69.4	0.03	1.5	50	0.0386	0.0285	0.0404	0.035833	1.791667	17.80833333	52.51111
B100/1	29.4	50	80.6	0.024	1.2	59.5	0.028	0.0443		0.03615	1.8075	28.2925	43.415
B100/2	29.4	50	81.2	0.036	1.8	64.1	0.0292	0.033	0.0378	0.033333	1.666667	33.03333333	33.93333
B100/3	29.4	50	81.4	0.04	2	64.1	0.0358	0.0346		0.0352	1.76	32.94	34.12
B125/1	29.4	62.5	93.5	0.032	1.6	70.1	0.0295	0.0291		0.0293	1.465	39.235	37.224
B125/2	30.5	62.5	94.7	0.034	1.7	69.5	0.0315	0.0388		0.03515	1.7575	37.2425	40.412
B125/3	29.4	62.5	93.6	0.034	1.7	70.6	0.0269	0.0334	0.0286	0.029633	1.481667	39.71833333	36.45067
B200/1	29.4	100	131.1	0.034	1.7	108.5	0.0378	0.035	0.0372	0.036667	1.833333	77.26666667	22.73333
B200/2	29.4	100.1	130.8	0.026	1.3	104.8	0.033	0.0365	0.0344	0.034633	1.731667	73.66833333	26.40526
B200/3	29.4	100	130.7	0.026	1.3	106.4	0.0317	0.0372	0.027	0.031967	1.598333	75.40166667	24.59833

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