

# USING BLACK SOLDIER FLY LARVAE TO TREAT FAECAL SLUDGE FROM URINE DIVERSION TOILETS

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## ABSTRACT

Challenges in improving sanitation include access to water. Since water and sanitation access problems are mostly faced by people in rural areas, waterborne sanitation is not an option. Thus strategies that improve sanitation while conserving water must be identified and implemented. Urine diversion dehydrating toilets (UDDTs) can be a solution for rural and semi-rural settlements, since they do not require water to dispose of human excreta. The use Black Soldier Fly (*Hermetia illucens*) larvae (BSFL) has emerged as a possible means of treating faecal sludge from on-site sanitation systems such as UDDTs. For UDDTs as used in eThekweni Municipality, where faecal sludge contains a relatively high proportion of sand, it was assumed that mixing an organic substrate with the sludge would be required to make it suitable for digestion by BSFL. In this study, two candidate organic substrates were assessed: dog biscuits, representing dog food waste; and fruit salad, representing food wastes from a fresh produce market. These substrates were mixed with UD sludge at three ratios and larval growth at each was assessed. In addition to biomass production, protein and lipid content of larvae were measured. No significant differences in biomass, protein and lipid was observed among treatments in each experiment. This indicates that BSFL are resilient to a wide range of faecal sludge compositions.

There was no significant difference in the growth of the larvae among three different types of sludges.

## INTRODUCTION

Access to improved, affordable sanitation facilities, and sustainable management thereof, is a problem in many developing countries. Sanitation is generally defined as safe storage and disposal of human excreta, while reducing pathogen transmission (1). About 2.4 billion of the world population still do not have access to improved sanitation facilities, with 70% of those people living in rural areas. Of these, 1 billion of these people still practice open defecation, with nine of ten people in rural areas still defecating in open water bodies, behind bushes and in gutters (2).

South Africa with other developing countries in sub-Saharan Africa still has fewer than 50% of the population without basic sanitation, although this varies widely among urban and rural areas. Approximately 40% of the population living in the rural areas of the Limpopo and Eastern Cape Provinces still do not have access to improved sanitation (3). Research has shown that the same areas where there is lack of basic sanitation, there is also a challenge to access clean drinking water (4). Thus strategies for providing basic sanitation without the use of water must be employed.

eThekwini Municipality in South Africa, selected urine diversion dehydrating toilets (UDDTs) as the most appropriate means of providing safe, hygienic and dignified sanitation in semi-rural and rural areas around Durban, in view of the hydrogeological features and population density in these areas. UDDTs separate urine and faeces at source, thereby allowing faeces to dehydrate to an easily managed product which should pose low health risk from pathogens. Users are instructed to add sand to the faecal vault after each use. This assists in dehydration of the faecal sludge, but poses a problem for treatment (5) (4).

A benefit of UDDTs is that they allow recovery and recycling of nutrients from urine and faeces, which can be used in agriculture to improve food security. However, human excreta potentially contain more than 50 different species of pathogens (6). These include helminth eggs which are highly resistant to desiccation and inactivation, as well as a variety of viruses, bacteria and protozoa (1). Most of these are in the faecal sludge. Thus before human faecal sludge can be used in agriculture, it must be processed and treated in order to kill pathogens (6).

eThekwini Municipality does not presently allow for reuse of urine or faecal sludge from UDDT in the municipal boundaries because of uncertainties around health risks, especially transmission of soil-transmitted helminths (*Ascaris*, *Trichuris* and *Taenia*). However, problems with excavation and management of faecal sludge from UDDTs has recently come to the fore. The initial model of UDDT operation adopted by eThekwini involved excavation of the relatively small quantity of faecal sludge, which should be mostly dry, by the householder, followed by burial of the sludge on the property (7) (8). Ideally a fruit tree with high-bearing fruit would be planted about the buried sludge. However, eThekwini offers pit latrine users an excavation service, with pits being emptied free of charge once every five years (9). An unintended consequence of this policy was that users of UDDTs began demanding a free emptying service, too. This has created the problem of management of the collected sludge. Because users are required to deposit sand in the UDDT vault after each use, the faecal sludge is not suitable for treatment by the same methods as pit latrine sludge, or by other conventional means.

One emerging means of treating organic wastes, including – potentially UDDT faecal sludge – is digestion by the larvae of black soldier flies. Black soldier flies (BSF) (*Hermetia illucens*) belongs to the Order Diptera and the family called Stratiomyidae (10). This insect originates from the warm temperate zones of America, but is currently widespread between 45°N and 40°S (tropical and warmer regions (10)). The adult fly is black in color, with a length of 15-20mm. The black soldier fly larvae (BSFL) constitute the first life cycle stages. They are dull whitish in color, until they approach the prepupal stage when they become brown in color. During the pupa stage their color turns into black. The larvae feed voraciously on a range of organic substrates, but stop feeding as they reach the prepupal stage. Once they stop feeding, they rely on the stored fat from larval stage and migrate out of the food source in search of a dryer pupation site (10). The larval form takes a 2-3 weeks until it reaches the final larval stage, the prepupae.

Once the pupal form is reached, pupae pupate into flies, provided conditions are suitable. They reach adult form, mate, lay eggs near food sources, then die. After the eggs hatch, the larvae migrate into food source (10) and the cycle is repeated.

The larvae can feed on wide variety of food including rotting vegetables and fruits, fish offal, human excreta and animal manure. BSFL have been proposed as a solution to treat and dispose of a range of organic wastes (11) (12). They have been shown to have the ability of converting wastes into protein (40-44% crude protein) and fat (depend on type of diet) which can be used for animal feed (11) (12). BSFL can also deal with extreme environmental conditions such as drought and food shortage. (11). It has advantage over other insect

species for treating faecal sludge since the flies are not attracted to foods or to human habitats (12), and so not a carrier of diseases (11). The disadvantage is that the BSFL require a warm environment to grow, which can be a problem during winter. The time taken for completion of life cycle depends on temperature, the amount of food fed to the larvae and also the quality of food (12). The optimal conditions for growth of BSFL include temperature ranging between 29°C to 31°C, relative humidity of 50% to 70%, and food quantity of 25 to 500 mg of fresh matter/larva/day (13). They have a unique gut microbiota which enables them to feed on different food types (11) (1). Their ability to digest large volume of organic wastes very quickly can be used commercially to produce valuable products and also to reduce house fly transmission by making the manure or the wastes more liquid, which is not suitable for house fly. Thus BSFL can be used to control house fly in areas with poor sanitation, as well as reducing the faecal sludge associated with on-site sanitation systems. So BSFL help to inhibit the transmission of diseases since house flies are a vector of diseases (10).

There is therefore potential for BSFL to be used in degrading difficult faecal sludges, specifically dry sandy sludge from UDDTs, while simultaneously yielding valuable products in the form of animal feed. Dry, sandy UDDT sludge can be made more suitable for BSFL digestion by mixing it with organic wastes such as food fruit and vegetable waste. Studies to date have demonstrated the ability of BSFL to digest fresh faeces and pit latrine sludge (1) (14).

This study assessed how well BSFL are able to grow on faecal sludge when supplemented by an organic food source. The ability of BSFL to grow on three different sludges – digested activated sludge, pit latrine sludge and UDDT sludge was compared. Thereafter, the digestion of UDDT faecal sludge was compared at three different ratios of UDDT sludge to food source. The implication for the flexibility of BSFL to degrade different compositions of UDDT sludge, and hence to implement the technology under a range of conditions, was assessed.

## **MATERIALS AND METHODS**

### Construction of the growth chambers, experimental setup and feeding

The BSFL growth chambers were constructed from plastic containers of dimensions 40×25×30cm. A foil container 38× 23 cm was placed in each container to collect any leachate draining from the faecal sludge during digestion by BSFL. A plastic grid (34×22.5 cm) was placed over foil container supported by three wood dowels of length 34 cm. Shade cloth (50×65 cm) was laid on top of the grid. Potting soil and woodchips were mixed in a 1:1 ratio and placed on top of shade cloth to serve as bedding for the larvae. Electric cable trunking was split in half to make a ramp, angled at a slope of 20°, to allow prepupae to escape the food source at the appropriate life stage. Black soldier fly larvae were supplied by a local breeder, breeding wild flies. Larvae were hatched onto moistened dog biscuits. At the age supplied (10-14 days after hatching), the larvae were mostly too small to count individually without risking harm to them. Instead the dog biscuit/larvae mix supplied by the breeder was thoroughly mixed, weighed and portions of equal mass were added to each of the growth chambers. The breeder advised changing the diet of the larvae from moistened dog biscuits to fruit and vegetable scraps as the larvae developed. A qualitative observational trial was conducted to establish a suitable organic substrate for subsequent experiments. Similar numbers of larvae were placed on moistened dog biscuits, fruit salad from a local fresh produce shop, or a 1:1 mixture of moistened dog biscuits and fruit salad.

Larvae grew best and were most active when fed the dog biscuit and fruit salad mix, hereafter to as control mix.

During the first two days of each experiment, the larvae were allowed to grow on the moistened dog biscuit substrate on which they had been hatched, to acclimatize them to the growth chambers and allow them to overcome stress associated with transfer.

#### Experiment 1: Response of BSFL to different sludge types

The growth of BSFL on different types of sludge was assessed. At initiation, larvae were 14 days post hatching

Three types of faecal sludges were tested – UDDT sludge, sludge from a ventilated improved pit latrine (VIP) and digested activated sludge. All treatments were performed in triplicate, and the whole experiment was duplicated (Experiments 1A and 1B) because of concerns that larvae in the first experiment were not fed sufficiently. All treatments comprised a 1:1 mix of the relevant sludge type to control mix (1:1 dog biscuits: fruit salad). The control treatment was the control mix. The larvae were fed 10g per treatment initially and further amounts of 10g once the feed mixture was almost completely depleted (Experiment 1A). After two weeks of the trial, it was realised that the food supply was insufficient and the amount of food fed to larvae was increased 30 g per treatment every second day (Experiment 1B). Growth was measured by removing 25 larvae at random every 2-3 days, weighing them in a preweighed container, and then returning them to their respective growth chambers. Experiment 1A and 1B were terminated when migration of the prepupae out of the feed slowed noticeably. Experiment 1A was run for 11 days and Experiment 1B was run for 12 days.

#### Experiment 2: Response of BSFL to different proportions of UDDT faecal sludge to two organic substrates

Experiment 2 was hampered by availability of larvae because of minimal breeding of wild BSFL in winter. Because of the mild climate in Durban, this problem had not been anticipated. The only batch of BSFL available was stored cool after hatching because numbers were low and it had been hoped to get a larger batch. Consequently, at initiation of the experiment, the larvae were larger than in Experiment 1, and there were fewer larvae per treatment than in Experiment 1.

Two organic substrates were tested at three ratios of substrate to UDDT sludge. The two substrates were moistened dog biscuit (representing dog food waste) and fruit salad (representing vegetable-based food wastes). These were representative of possible substrates for use in digestion of UDDT in eThekweni Municipality. The combinations of substrate and UDDT sludge are shown in Table 1 below.

Table 1: Composition of feed for testing BSFL growth on two organic substrates at different ratios of substrate to UDDT sludge in Experiment 2.

Organic substrate	Composition of feed
Fruit salad	10% fruit salad:90% UDDT sludge
	25% fruit salad:75% UDDT sludge
	50% fruit salad:50% UDDT sludge
Moistened dog biscuit	10% dog biscuit:90% UDDT sludge
	25% dog biscuits:75% UDDT sludge
	50% dog biscuits:50% UDDT sludge
No substrate, UDDT sludge only (control)	100% UDDT sludge

Since the larvae in Experiment 2 were older than in Experiment 1, they were large enough to count. Each replicate had approximately 100 larvae, whereas approximation of the number of larvae per replicate in Experiment 1 was in excess of 300 larvae per treatment. The larvae were fed 20g per treatment every day, based on a feeding rate of 10mg/larval/day (M Lewis, Agriprotein, pers. comm.). Every second day, 25 larvae were removed from each growth chamber at random, placed in a preweighed container, and weighed. Weighed larvae were returned to the growth chambers from which they were removed. The experiment were run for only 7 days because the larvae reached the pre-pupae stage much quicker than those in Experiment 1, presumably because they were larger at the outset. However, far fewer prepupae crawled out of the food source; consequently most of the larvae were picked out of the growth container by hand. It is thought that this may have been related to colder winter weather since larvae were also more sluggish overall than in Experiment 1.

#### Harvesting, drying and chemical analysis

When the larvae began to turn brown, a ramp with a collecting container was inserted in the growth chamber. Some larvae climbed the ramp and accumulated in the collecting container, with larvae from Experiment 1 climbing out of the growth chamber more effectively than those from Experiment 2. The rest of them that could not climb were picked out of the growth chamber by hand. Wet mass was taken from 25 larvae from each of the treatments of each of the experiments. Larvae were killed by immersion in boiling water for 5 minutes, then taking them out of the water and blotting dry using paper towel. Dry mass was determined by drying pre-weighed batched of larvae in an oven at 80°C in the oven for 72 hours, and then measuring their dry mass. The rest of the larvae were killed using the same method and were then stored in a fridge at 18°C until used for chemical analysis.

Since experiment 2 was run for shorter period of time and larvae masses were similar, the length of the larvae was also measured at the end of the experiment as an indicator of growth, using Vernier calipers. Protein content of feed samples was measured as total Kjeldahl nitrogen, in the laboratories of the Pollution Research Group at the University of KwaZulu-Natal. Protein content of larvae was analysed at the laboratories of the KwaZulu-Natal Department of Agriculture in Cedara, using the official Dumas method (AACC, 1999c) using a LECO TruMac N. Fat content of feed and larvae was analysed at the laboratories of the KwaZulu-Natal Department of Agriculture in Cedara, using ether extraction (AOAC, 1980 Ed) followed by gas chromatography.

### Statistical analysis for continuous data (growth expressed as BSFL mass)

IBM SPSS version 23 was used to analyse continuous data. To determine the effect of treatments at different time interval on larvae growth a mixed ANOVA was used. To test for normality of studentized residuals, the Shapiro-Wilk test was run. Mauchly's test was used to test for sphericity. Levene's test was run to test for equality of variances between treatments. The assumption of equality of variance was met with  $p > 0.05$ . Post hoc multiple comparisons using Bonferroni and Tukey tests were also used to compare the difference between treatments. Profile plots and mixed ANOVA was used to test for the interaction term.

### Statistical analysis of discrete data (dry mass, length, protein content and fat content)

IBM SPSS version 23 was used to analyse discrete data. Dry mass, length, protein content and fat content were compared among treatments at the end of the trial using a one-way ANOVA. To test for normality of the residuals, a Kolmogorov-Smirnov test was used. Residuals were normally distributed, with a  $p > 0.05$ . Levene's test was used to test for equality of the residual. The residuals were equally distributed among treatments with a  $p > 0.05$ . Post hoc multiple comparisons using Bonferroni and Tukey tests were also to compare the difference between treatments.

## **RESULTS**

### Experiment 1: Response of BSFL to different sludge types

#### Experiment 1A

Growth of BSFL, expressed as mass (g) per larva (Figure 1, showing results for Experiment 1A) showed no difference in growth on different sludge types until day 9. After day 9, in the yield for the control treatment (fed only control mix, 1:1 dog biscuit:fruit salad, as described in Methods) a slight difference was observed. The larvae from the control treatment had a higher mass compared to other treatments. The effect of sludge type fed to larvae had a significant effect on the growth of the larvae over time ( $p = 0.019$ ;  $F = 2.494$ ;  $DF = 12$ ). From day 9 the amount of feed provided to larvae was increased, as described in Methods.

The dry mass of BSFL harvested in Experiment 1A (Figure 2) was highest for the control treatment, as observed for growth (Figure 1). The dry mass of BSFL from the control treatment was 0.08 g/larvae which was the highest compared to others. The larvae from 50%C: 50%AS treatment had the lowest mass. However, the difference among treatments was not significant ( $p = 0.139$ ;  $F = 2.455$ ;  $DF = 3$ ).

Fat analysis of BSFL from Experiment 1A (Figure 3) showed a significant effect of type sludge fed on the content of fat (expressed as % dry mass). Larvae fed 50% control mix: 50% digested activated sludge had the highest fat content (36.1%) and larvae fed 50% control mix: 50% pit latrine sludge had the lowest fat content (34.2%). The difference was significant between treatments with a p-value of 0.009 ( $F = 7.801$ ,  $DF = 3$ ). Between the treatments, significant differences were observed between larvae fed 50% control mix: 50% UDDT sludge versus larvae fed 50% control mix: 50% pit latrine sludge, and larvae fed 50% control mix: 50% pit latrine versus 50% control mix: 50% digested activated sludge. Protein content analysis showed no significant difference among treatments with a p-value of 0.122 ( $F = 2.631$ ,  $DF = 3$ ) (Figure 3).

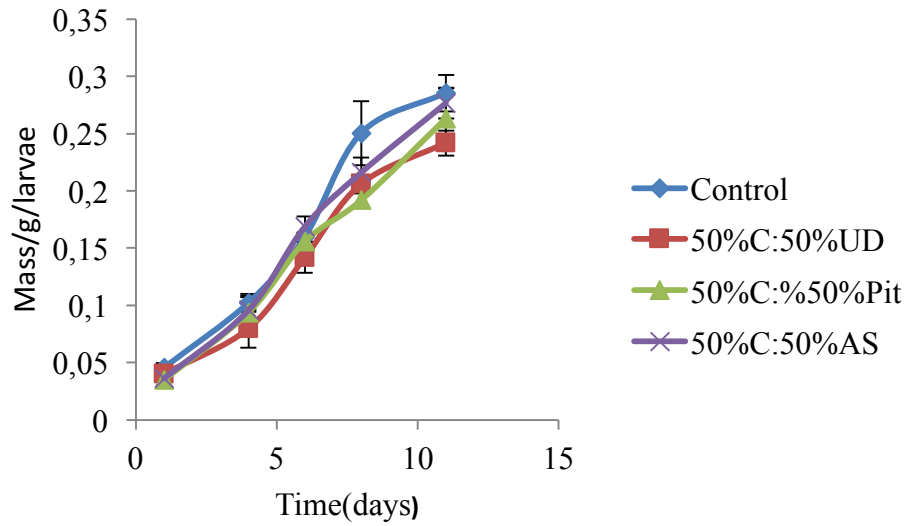


Figure 1: Experiment 1A; Mean larvae growth (mass/g/larvae) fed control mix (1:1 fruit salad and dog biscuit), or each of three sludges with a ratio of 1:1 to control mix over 11 days experimental period. (n=3). Error bars show standard deviation.

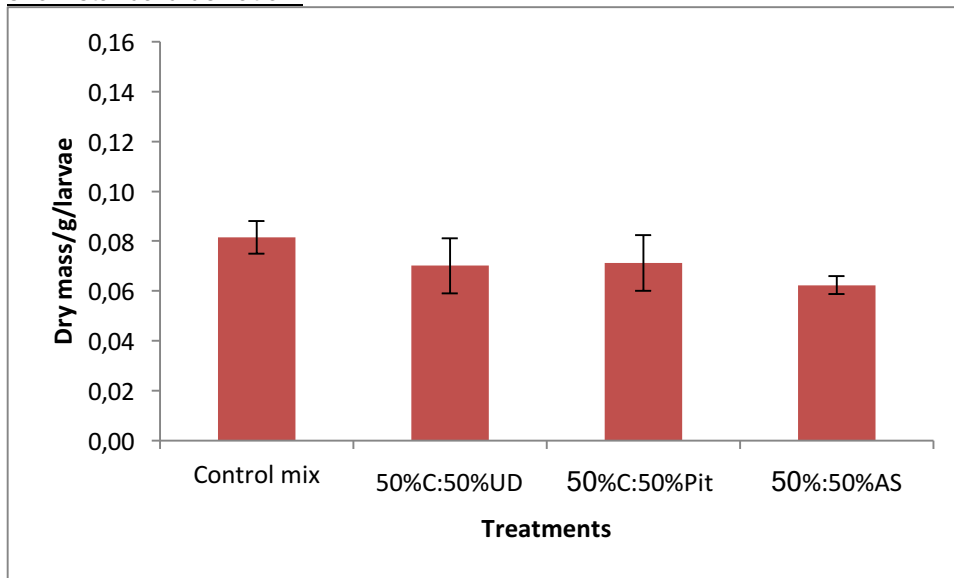


Figure 2: Experiment 1 A; Mean larvae dry mass(g/larvae) fed control mix (1:1 fruit salad and dog biscuit), or each of three sludges with a ratio of 1:1 to control mix over 11 days experimental period. (n=3). Error bars show standard deviation.

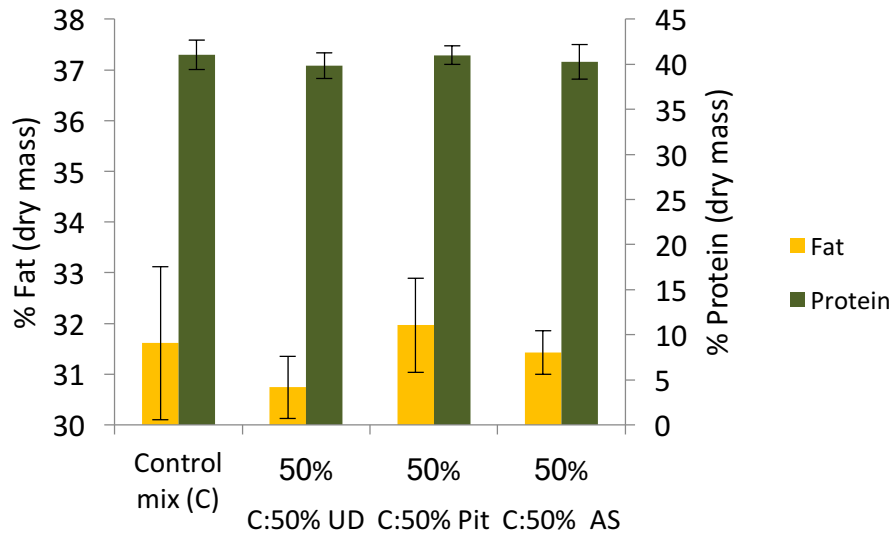


Figure 3: Experiment 1A; Percentage protein and fat content of larvae fed control mix (1:1 fruit salad and dog biscuit), or each of three sludges with a ratio of 1:1 to control mix over 11 days experimental period. (n=3). Error bars show standard deviation.

#### Experiment 1B

There was no difference in the growth of BSFL in Experiment 1B during the first 5 days (Figure 4). From day 6, the larvae from control treatment had a higher mass compared to the other treatments. The growth of the larvae fed different sludges was not affected by the type of sludge ( $p=0.161$ ;  $F=1.539$ ;  $DF= 12$ ). From figure 5, the BSFL fed control mix only had the highest dry mass of (0.13 g/larva) and the lowest dry mass was recorded for BSFL fed on 50% control mix: 50% pit latrine sludge(0.10 g/larva). The difference in dry mass/g/larvae was significant with a p-value of 0.029 ( $F=5.112$ ,  $DF= 3$ ). There were no significant differences in fat content or protein content among BSFL fed different sludge types ( $p>0.05$ ; Figure 6).

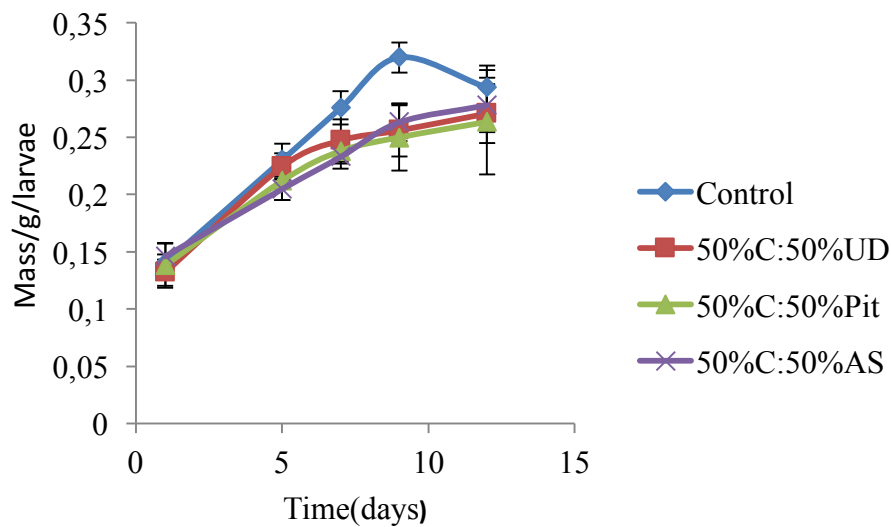


Figure 4: Experiment 1B; Mean larvae growth ( mass/g/larvae) fed fed control mix (1:1 fruit salad and dog biscuit), or each of three sludges with a ratio of 1:1 to control mix over 12 days experimental period. (n=3). Error bars show standard deviation.



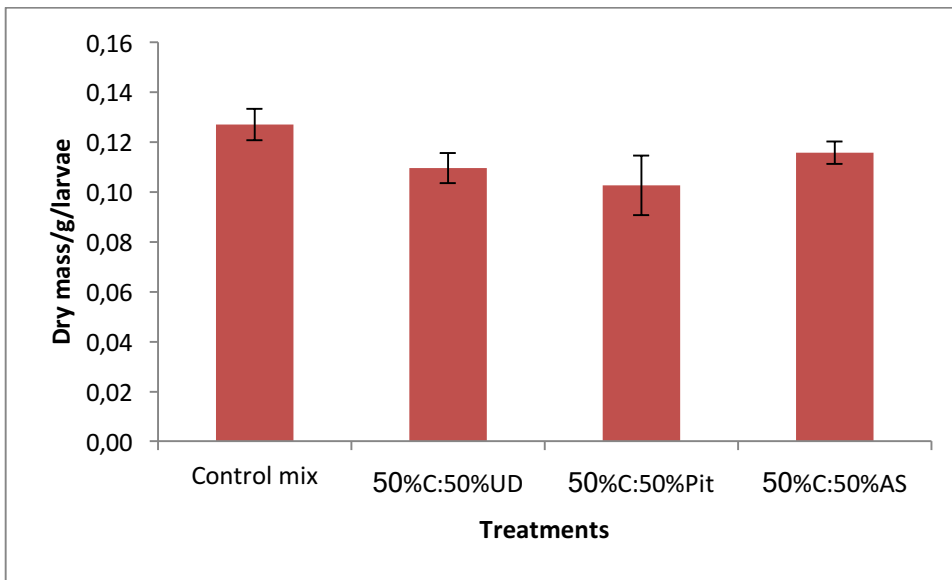


Figure 5: Experiment 1B; Mean larvae dry mass(g/larvae) fed control mix (1:1 fruit salad and dog biscuit), or each of three sludges with a ratio of 1:1 to control mix over 12 days experimental period. (n=3). Error bars show standard deviation.

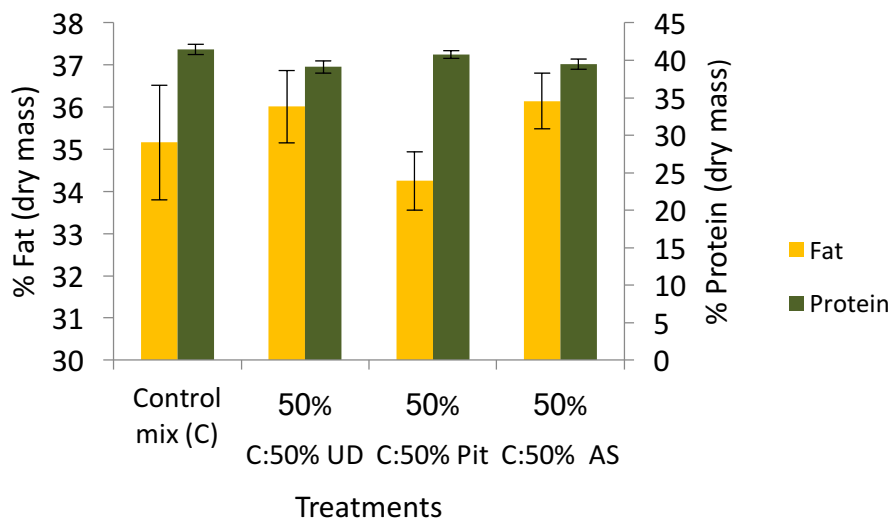


Figure 6: Experiment 1B; Percentage protein and fat content of larvae fed control mix (1:1 fruit salad and dog biscuit), or each of three sludges with a ratio of 1:1 to control mix over 12 days experimental period. (n=3). Error bars show standard deviation.

## Experiment 2: Response of BSFL to different proportions of UDDT faecal sludge to two organic substrates

Response of BSFL to different proportions of UDDT faecal sludge to dog biscuit as organic substrate

Varying the proportion of dog biscuits as organic substrate to UDDT sludge in feed given to BSFL had no significant effect on growth and yield measured as increase in mass ( $p=.209$ ,  $F=1.606$ ,  $DF= 6$ ; Figure 7); dry mass ( $p=0.295$ ;  $F=0.718$ ;  $DF= 2$ ; Figure 8); or length ( $p=0,702$ ;  $F=0.485$ ;  $DF=3$ ; Figure 9).

Fat content was lowest in larvae fed 100% UDDT sludge (27.31%) while larvae fed 50% UDDT sludge had the highest fat content (30.14%). As the proportion of UDDT sludge increased, the fat content of larvae decreased. Differences in fat content across the treatments were significant with p-value of 0.000(F=19.8, DF= 3). There was no significance in protein content across treatments (p=0.702; F=0.485; DF=3; Figure 10).

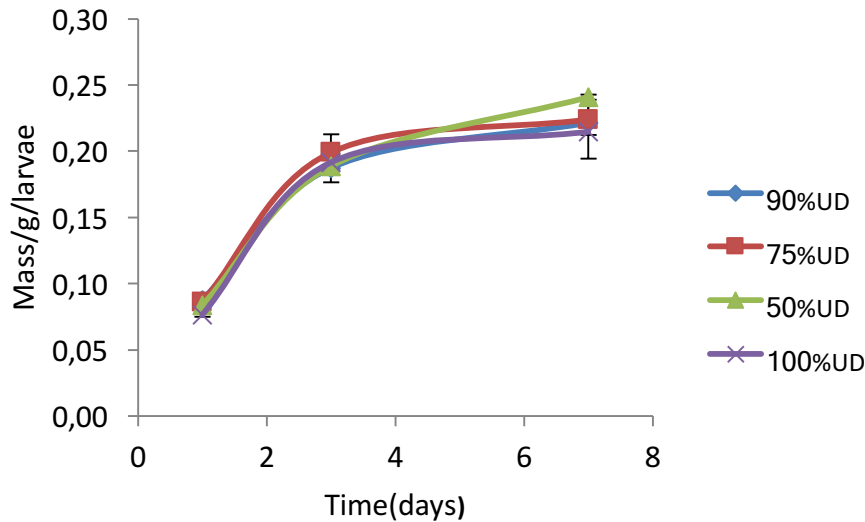


Figure 7: Mean larvae growth (mass/g/larvae) fed different proportions of UDDT sludge to dog biscuit over 7 days experimental period (n=3). Error bars show standard deviation.

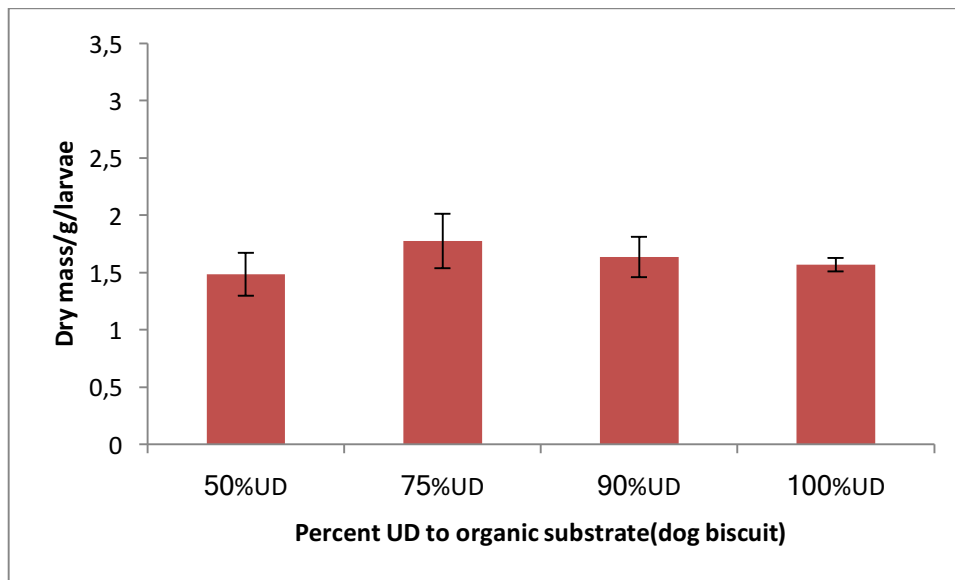


Figure 8: Mean larvae dry mass (mass/g/larvae) fed different proportions of UDDT sludge to dog biscuits over 7 days experimental period (n=3). Error bars show standard deviation.

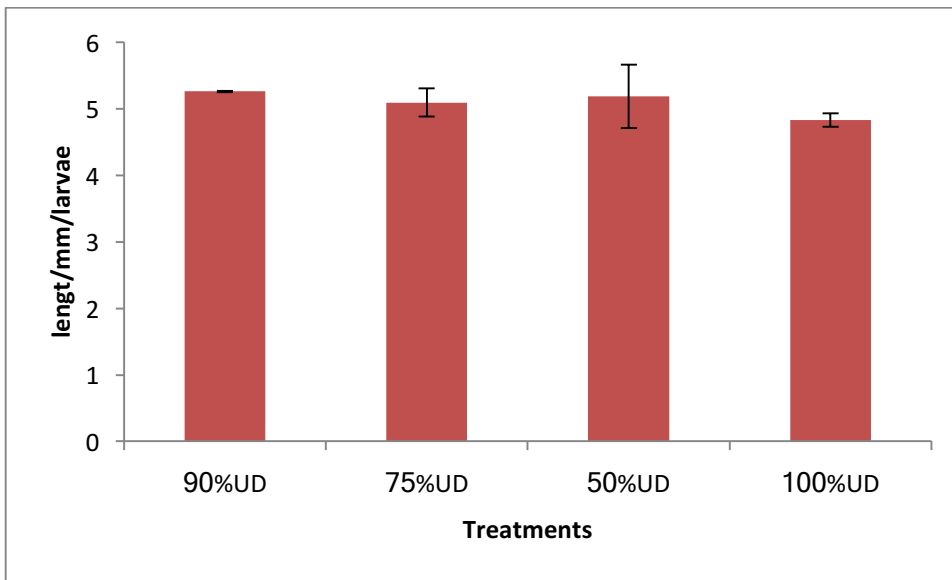


Figure 9: Mean larvae length (mass/g/larvae) fed different proportions of UDDT sludge to dog biscuits over 7 days experimental period (n=3). Error bars show standard deviation.

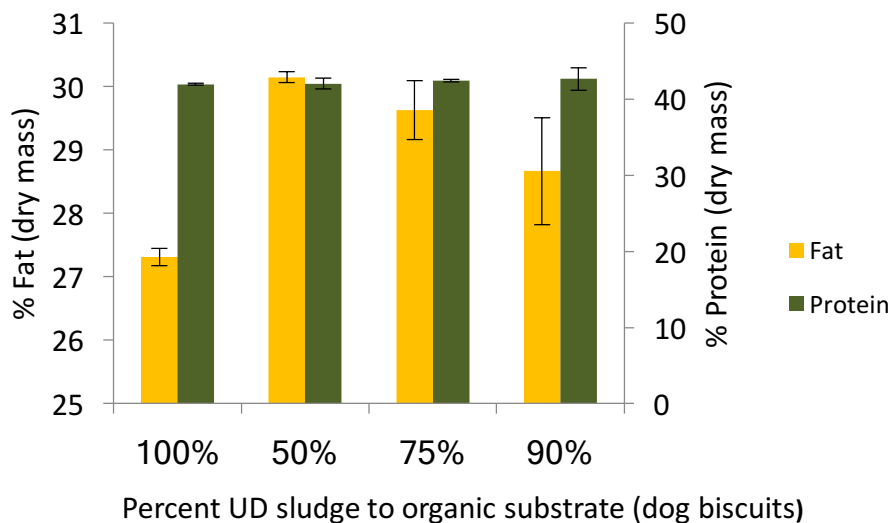


Figure 10: Percentage protein and fat content of larvae fed different proportions of UDDT sludge to dog biscuits over 7 days experimental period (n=3). Error bars show standard deviation.

### Response of BSFL to different proportions of UDDT faecal sludge to fruit salad as organic substrate

Varying the proportion of fruit salad as organic substrate to UDDT sludge in feed given to BSFL had no significant effect on growth and yield measured as increase in mass ( $p=.166$ ,  $F=19.56$ ,  $DF= 4$ ; Figure 11); dry mass ( $p=0.525$ ;  $F=0.718$ ;  $DF= 2$ ; Figure 12); length ( $p=0.915$ ;  $F=0.900$ ;  $DF=2$ ; Figure 13); fat content ( $p=0.495$ ;  $F=0.797$ ;  $DF= 2$ ; Figure 14) or protein content ( $p=0.251$ ;  $F=1.758$ ;  $DF=2$ ; Figure 14).

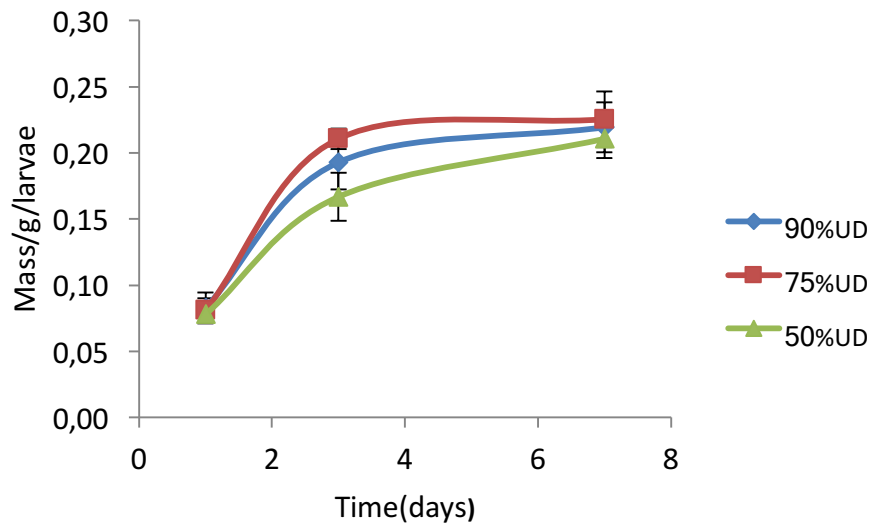


Figure 11: Mean larvae growth (mass/g/larvae) fed different proportions of UDDT to fruit salad over 7 days experimental period (n=3). Error bars show standard deviation.

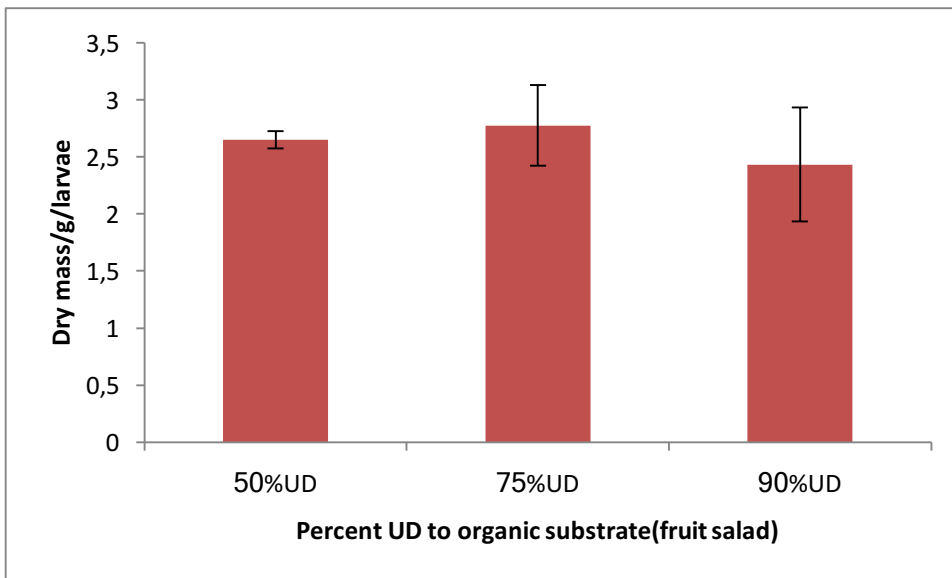


Figure 12: Mean larvae dry mass (mass/g/larvae) fed different proportions of UDDT sludge to fruit salad over 7 days experimental period (n=3). Error bars show standard deviation.

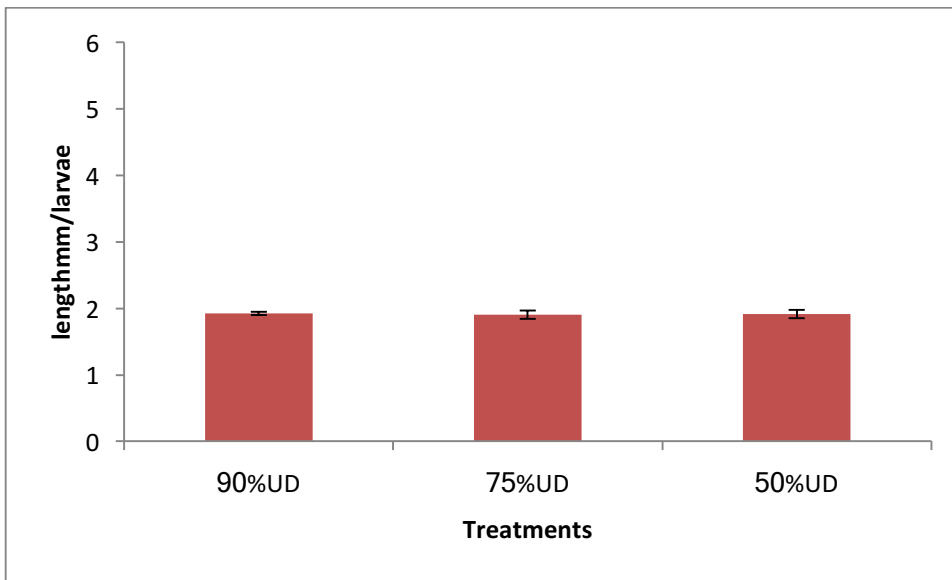


Figure 13: Mean larvae length (mm/larvae) fed different proportions of UDDT sludge to fruit salad over 7 days experimental period (n=3). Error bars show standard deviation.

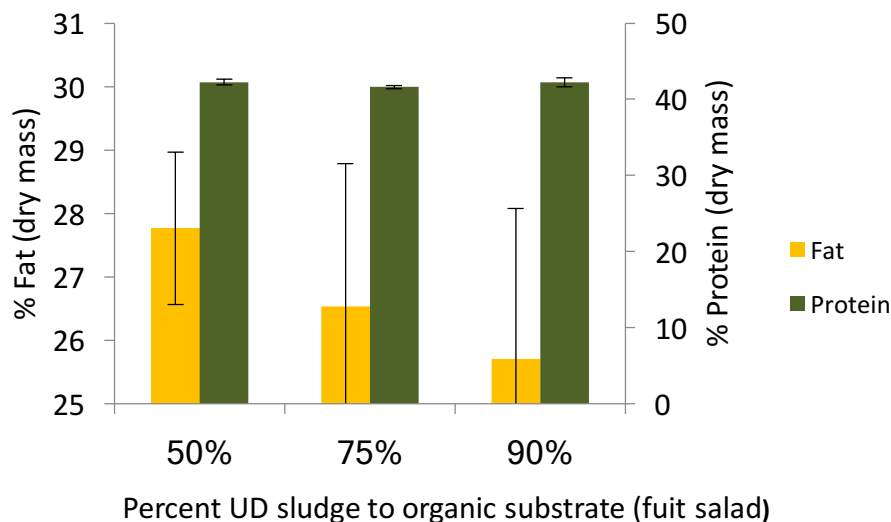


Figure 14: Percentage protein and fat content of larvae fed different proportions of UDDT sludge to over 7 days experimental period fruit salad (n=3). Error bars show standard deviation.

## DISCUSSION

Experiment 1A showed that BSFL growth (based on change in mass over time) is not affected by the type of the sludge fed to them. During the first weeks of the experiments, the larvae were accidentally exposed to starvation and during this time no difference in growth was observed. After the amount of food fed to the larvae was increased there was a slight difference this was because the larvae were already approaching the maturity stage. The larvae in Experiment 1A took 11 days to reach the prepupal stage. Experiment 1B was a repeat of Experiment 1A, with an increased amount of food from the outset. There was no significant difference in the growth of larvae fed three sludges. Larvae grew better on the control mixture. It took 12 days for the larvae to reach the prepupal stage, similar to the time taken in Experiment 1A. The highest dry mass in Experiment 1A, where the larvae were

starved, was 0.084g/larvae, while in Experiment 1B, where more food was added, the highest dry mass was 0.132g/larvae (Figure 2; Figure 4).

Other studies have shown that BSFL fed low amounts of food took longer to develop, as compared to the larval fed higher amounts of food. Food starvation prolongs the time that larvae take to reach maturity. However feeding the larvae more than 100 mg chicken feed/larva/day did not increase the rate of growth or completion of larval stages. When the larvae were not getting enough food; they crawled out of the food as soon as they have acquired the minimum energy they require for their development to pupal stage. The larvae also reach a stage where they cannot feed further (1). This is caused by the secretion of prothoracicotrophic hormone (PTTH), causing the larvae to stop feeding. Other studies have shown that PTTH stimulates the release of ecdysteroids; these steroids play a role in the larval development. In the absence of PTTH the larvae have prolonged feeding period without a change in growth rate, thus a delaying the larval development (15). Starved larvae have been shown to have reduced dry mass (16), corresponding with the observation made here that the maximum dry mass of BSFL from Experiment 1B was higher than that from Experiment 1A.

The secretion and functioning of PTTH and ecdysteroids, hormones mentioned above as central to the development of larvae, are influenced by temperature (17). Experiment 2 on the effect of ratios of UDDT sludge to organic substrate on BSFL growth were performed on the batch of larvae available in winter. These larvae grew little, matured quickly and did not move out of the food source efficiently. Thus, from the observation made during this experiment and also understanding the functions of PTTH, it is proposed that PTTH was not stimulating the release of ecdysteroids and that this was the main limiting factor in Experiment 2 in this study. It was observed that in all the treatments the larvae were alive, indicating that the larvae were able to feed and survive on different concentration of UDDT sludge supplemented with 2 types of organic substrate. The effect of these on growth needs to be repeated at warmer temperatures when better growth and development can be expected. The existing study on using BSFL for digestion of faecal sludge used fresh human faeces (1), thus information from this study on UDDT sludge and other sanitation sludges needs to be repeated and the results verified.

Protein content (% dry mass) varied only slightly among all treatments and experiments, ranging from 31% to 42%. Experiment 1 (three sludges) had a higher protein content compared to Experiment 2 (UDDT sludge and varying proportions of organic substrates). Other studies done on feeding fresh human faeces to BSFL; showed that the larvae had a protein content ranging for 28.2% to 44% (1) (10). The amount of protein required for fish meal range from 31% to 45 % (18). Thus the study reported here, using wild BSF rather than a captive population and with one experiment conducted under temperature conditions not conducive to larval development, still produced larvae of comparable quality to more controlled environments and of a quality that is suitable for use as fish meal.

Fat content in was low in sludges compared to substrates, ranging from 25% to 30%. Studies show that fat content depend on the type of diet fed to the larvae ranging from 15% to 35%. From animal manure it can range from 15% to 35% and in food wastes it can range from

42% to 45% (10). In treatments with greater proportion of UDDT sludge to substrates, fat content was low(table 2).

## CONCLUSION

Differences in growth, final dry mass, and protein and fat content of BSFL grown on three different types of faecal sludge, and on varying ratios of UDDT sludge to organic substrate were minimal. This suggests that BSFL are able to tolerate a high proportion of faecal sludge in feed, particularly dry UDDT sludge containing a high proportion of sand. Performance on 100% UDDT sludge was generally lower than on mixtures of UDDT sludge and organic substrates, but this difference was only rarely significant. This indicates good potential for the use of BSFL to treat UDDT sludge, and indicates that the larvae are resilient to feed composition and, to an extent, environmental conditions. A sensitivity to low temperatures is indicated. The protein content of BSFL produced from the wide range of relatively poor substrates was comparable to that reported elsewhere and suitable for use as fish meal, further pointing to the resilience of BSFL to grow and produce valuable product on sanitation residues under variable conditions.

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Table 2: Protein and lipid content for feed mixture

<b>Sludge to control(dog biscuit and fruit salad</b>	<b>Protein(% mass)</b>	<b>Dry Fat(%Dry mass)</b>
Control	16.00	29.75
50%C:50%AS	27.31	9.54



50%C:50%Pit	17.49	11.18
50%C:50%UD	14.73	8.61
<b>UD to substrate(dog biscuit)</b>		
90% UD	32.50	1.58
75% UD	15.20	2.13
50% UD	16.93	3.04
100% dog food	20.37	4.83
<b>UD to substrate(fruit salad)</b>		
90% UD	13.36	6.56
75% UD	13.18	14.58
50% UD	10.81	27.93
100% fruit salad	12.30	54.64
<b>Sludge only</b>		
100% UD	13.48	1.22
100% Digested activated sludge	38.68	3.07
100% pit latrine sludge	18.98	6.35

