http://www.lrrd.org/lrrd31/2/patri31022.html

Livestock Research for Rural Development 31 (2) 2019 | Guide for preparation of papers | LRRD Newsletter | Citation of this paper

Optimization of housefly larvae production on pig wastes and brewers' grains for integrated fish and pig farms in the tropics

P Mafwila[1,2], D Bwabwa[3], N Nyongombe[3], B Kambashi[1], J Mafwila[1], D Dochain[4], J Bindelle[2] and X Rollin[5]

[1] Université de Kinshasa, Faculté d'Agronomie, Kinshasa, Républic Democratique du Congo
patrickmaf@yahoo.fr
[2] Liège University, TERRA Teaching and Research Centre, Gembloux Agro-Bio Tech, Gembloux, Belgium
[3] Université pedagogique nationale, Route de Matadi Quartier Binza UPN Kinsasha, Républic Democratique du Congo
[4] Université catholique de Louvain, ICTEAM, Louvain-la-Neuve, Belgium
[5] Université catholique de Louvain, Marcel Huet Centre, Louvain-la-Neuve, Belgium

Abstract

Lack of appropriate animal waste management methods in many smallholder farms in the tropics often leads to environmental problems, especially in locations with high population density such as urban and peri-urban areas. On farms integrating pig production to fish farming, manure can be turned into a valuable feed source of high quality protein for fish through housefly larvae and contribute to intensify fish production and reduce cost of fish feed. Three experiments were carried to optimize operating conditions for maggot production on animal wastes and industrial byproducts found in Kinshasa, the capital city of the Democratic Republic of Congo (DRC). The comparisons were: (1) production on pure substrates (manure or brewer's grains) or mixtures with lysine or blood; (2) exposure time to flies for the insemination of the substrates: and (3) dynamics of larvae production. Mixing brewers' grains with Lysine or manure and/or blood more than doubled the amount of larvae that were harvested. Brewers' grains are a good source of energy, but are probably deficient in essential amino acids to support the growth of maggots. It also appears that only the first days of laying eggs are important since no difference was observed between temporary and permanent exposure of the substrates to houseflies. The peak of larvae production was reached 6 days after exposure. The addition of cow blood in increasing doses to a mixture of brewers' grains and manure linearly increased the production of maggots.

Key words: *maggot, mixture, nutrient, production, substrates*

Introduction

In the tropics, stored wet agricultural and agro-industrial by-products as well as on-farm wastes such as manure are prone to spoiling due to the quick proliferation of housefly (*Musca domestica*) larvae (maggots) and are also a biohazard for human and animal populations. In pig farms, the problem is exacerbated by the concentration of these facilities in peri-urban areas with a limited acreage, exceeding the assimilation capacity of the environment (Čičková et al 2012). Several technologies of on-farm wastes recycling have been proposed to reduce their impact on the environment but most of them are too expensive for smallholders (ková et al 2012). Interestingly, in Kinshasa (Democratic Republic of Congo) where crops and pig production are integrated with freshwater aquaculture (IAA), manure is collected and used to fertilize directly the ponds and/or the vegetable crops (Kinkela et al 2017). For the fish subsystem, this method stimulates the primary production of the pond to produce phytoplankton and zooplankton, which will be an important protein source in the fish trophic chain. This practice is however controversial. According to Nuov et al (1995) direct use of pig wastes as inputs into fish culture systems may be unacceptable or an inferior use of valuable inputs because non-filter feeding fishes, such as African catfish (*Clarias gariepinus*), may be unable to recover nutrients efficiently through the pond food web and require complete diets. Moreover, organic matter with high nitrogen content typically decomposes quickly with release of appreciable ammonia nitrogen ($NH3+NH4^{+}$), that can lead to water eutrophication. Microbial oxidation of ammonia nitrogen ($NH3+NH4^{+}$) usually called nitrification, removes dissolved oxygen from the water and produces acidity. Excessive nitrogen gas in water can cause gas bubble trauma in fish and some other aquatic animals (Boyd 2015).

Mafwila et al (2017) observed that despite an optimal mix of subsystems, the majority of IAA farmers faced lack of commercial feeds such as pellets and high protein ingredients such as soya bean, blood meal and fish meal to formulate completed feeds due to their high cost (Charlton et al 2015). Moreover, in tropical Africa, smallholder farmers strive to trap nutrients, especially N, within their farms while it is a key limitation to sustain higher production levels that are required to reach food security (Rufino et al 2009). Kinkela et al (2017) showed that up to 90 % of farmers feed fish with agricultural and agro-industrial by-products such as brewer’s grains and wheat bran, that are low in protein quality. Therefore controlled production of housefly larvae by farmers on these substrates could be an opportunity to provide an important additional source of high quality protein and to intensify fish production by concentrating nutrients in a readily available and cheap ingredient. Housefly larvae meal contains good quality protein for poultry and fish (Bondari and Sheppard 1981; Aniebo and Owen 2010). Maggot flour has an amino acid profile comparable to that of fish meal It is also good source of minerals (Fasakin Balogun and Ajayi 2003; Téguia et al 2002). Moreover, this strategy could kill two birds with one stone. Insects and earthworms play a significant role in decomposing many types of wastes preventing hazardous release of harmful forms of nutrients in the environment, for example by reducing leaching and the production of volatile organic compounds through microbial fermentation (Hwangbo et al 2009). For several authors, the use of insects such as dipteran larvae as manure decomposers is a low-input sustainable waste management technique with potential for innovation to increase its efficiency (Pastor Velasquez Gobbi and Rojo 2015).

This study aimed to improve on-farm production methods of housefly larvae using as substrates local agricultural and agro-industrial by-products available (pig manure, brewer's grains, fresh blood) in integrated agriculture aquaculture farms in Kinshasa. To achieve this goal, we ask three research questions:

- what is the combination of on-farm available agricultural and agro-industrial by-products that lead to optimal production of maggots;
- does spawning time of houseflies affect the production of maggots;
- what is the optimal duration of the housefly rearing cycle to maximize the production of maggots in the main substrates and the cost / benefit ratio?

Material and methods

Three experiments investigating complementary aspects of the production and growth of domestic housefly larvae (*Musca domestica*) were carried out on a farm integrating vegetable production to pig and fish farming in ponds in the Funa valley of Kinshasa (D.R.Congo). The first experiment compared the cumulated level of larvae production on different substrates over 6 days: brewers' grains (BG), pig manure (M), an equiproportional mixture of BG and M (BG-M), BG with 1% lysine (BG-LYS), an equiproportional mixture of BG and cow blood (BG-B) and an equiproportional mixture of BG, M and B (BG-M-B). The second experiment compared the production of larvae on BG-M and BG-M-B with two exposure methods to the housefly: 18 days of permanent exposure (PERM) and 2 days of temporary exposure followed by 16 days of growth with no housefly access (TEMP). The third experiment compared increasing doses of cow blood (0%, 10%, 20%, and 30%) in an equiproportional mixture of BG and M during 9 days of larvae production. In the first experiment the presence of lysine among substrates was intended to verify that lysine was a limiting amino acid in BG. However, there is no practical value of its incorporation in the context of IAA farms.

For all experiments, all substrates were run in quadruplicate. The substrates were placed in plastic baskets maintained in the shade, covered in the bottom with a mosquito mesh screen through which the larvae migrated to a 30 × 26.5 × 32 cm^3 plastic bin placed below for collection (Photo 1). The substrates were daily adjusted to 77.5% of water content by addition of water in the morning to allow substrates to maintain humidity levels around 70% during the day. This is appropriate water content to limit fungal populations and to prevent desiccation of eggs (Lomas 2012). Holmes et al (2012) proved that with relative humidity around 70% the eggs eclosed faster, the egg eclosion rate was higher, the pupal mortality was lower, and the adult emergence and longevity were higher for the black soldier houseflies.

Photo 1. Growing bed and harvesting bin used in the experiments

The substrates were thoroughly mixed and an amount of 1,500 g for experiment 1, 2,500 g for experiment 2 and 3,000 g for experiment 3 were placed in homogeneous thickness of 3 cm (Lomas 2012). During the experiments, natural laying of eggs by houseflies on the farm was used.

Several parameters were measured: ambient temperature and substrates temperature were taken three times a day (7:30, 13:00 and 17:30). Weight and number of larvae were measured daily using Kern scale with 0.1 g of precision. After harvest from plastic bin during daily control, larvae were sorted and grouped into two categories (maggots and pupae) before being weighed. The method described by the Association of Official Analytical Chemist (AOAC 1990), was used to measure dry mater (DM), nitrogen, fat and gross energy content of larvae. Nitrogen content was determined by the Kjeldahl procedure using Kjeltec Auto Sampler System 1035 Analyzer (Tecator), and gross energy by the calorimetric procedure using the Adiabatic Calorimeter 1241 by PARR. Finally, in the third experiment the cost of larvae produced per kg of substrate used was calculated considering market prices of Kinshasa in 2016.

One-factor ANOVA with Average Comparison Test (Tukey Test) were used to compare mean values of larvae production per substrate in the first and second experiment. The GLM procedure of repeated measures Univariate Tests of ANOVA was used to compare mean values of temperature for different substrates in SAS for the first experiment. The three-time measurements were considered as repeated measurements for the experiment. Finally, a general linear model of regression was used to test the effect of blood doses in different mixtures for the third experiment.

Results

Result of maggot production over a 6-day period showed that brewers’ grains and pig manure used separately provided the lowest production, while highest production was reached by mixing BG-M-B (Table 1). Mixing brewers’ grains with lysine or manure and/or blood more than doubled the amount of larvae that were harvested. No differences were found in substrate temperatures during the 6 days of larvae production (*p*=0.97). Temperature greatly varied according to the time measured (*p* <0.0001) with highest mean temperature values at 13:30 (33.3°C). During the six days of maggot production temperature varied from 23.1°C up to 47.9 °C. The highest value was observed on the fourth day with an average value of 40.7 °C.

Table 1. Maggots and pupae production (g/kg of substrate) during 6 days (g/kg of fresh substrate) reared on different mixtures of brewer's grains (BG), manure (M), Lysine (LYS) and fresh cow blood (B) as substrates (N=4).

Substrates	**Maggot production (g/kg substrate)**	**Pupae production (g/kg substrate)**	**Temperature (°C)**
BG	53.8^{bc}	0.10	32.7
M	30.6^{d}	0.3	30.9
BG-M	89.3^{b}	0.10	32.8
BG-LYS	94.2^{bc}	0.00	32.6
BG-B	107^{b}	0.00	31.1
BG-M-B	180^{a}	0.00	30.8
SEM	10.3	0.05	0.57
p	0.001	0.52	0.79

^{abc} *Mean values in the same column without common letter are different at p<0.05*

During this first experiment not all the available substrate was consumed by the maggots (Figure 1). Disappearance of substrates started on the 1st day and increased from the third to the fourth day and then decelerated from the fourth to the sixth day. The mixture of BG and M was more consumed by maggots than the other ones (692 g of fresh substrate). BG and mixes of BG-LY were less consumed (455 and 439 g of fresh substrate). The second experiment showed that the peak of larvae migration was reached after 6 days (Figure3). Consistently with the first experiment, BG-M-B, whether in the TEMP or the PERM treatments presented the best maggot production, with 112 ± 30.3 g/kg and 111 ± 40 g/kg of substrate respectively, compared to BG-M which yielded 47.4 ± 6.73 g/kg and 53.6 ± 8.48 g/kg for the TEMP and PERM treatments respectively. No difference was observed according to the exposure method to the houseflies (PERM vs. TEMP) (*p*=0.515). Finally, experiment 3 that compared increasing doses of cow blood (0%, 10%, 20%, and 30%) in the mixes of BG-M-B during 9 days of maggot production showed that increasing doses of cow blood improved linearly the production of maggots (Figure 4) (*p*<0.05). No saturation or plateau effect was observed with the doses of blood that were used.

The addition of cows' blood indicated an optimum value between 10 and 20 % of blood into BG-M since such levels did not affect the production cost of maggots per kg. However, the addition of blood to 30% increased the cost of production.

Figure 1. Daily evolution of the mass of the substrates (g/bin)

Figure 2. Evolution of the production per kg of substrate as a function of time with two contrasting exposures to the houseflies (N=4)

Figure 3. Production of maggot according to the cow's blood content of a BG-M mixture (g / kg of fresh substrate)

Table 2. Cost of house maggot production according substrate use with increasing dose of cow blood

Substrates	**Substrates price ($)**	**Maggot production (g/kg fresh substrate)**	**CP ($)**
BG	0.15	30.6	1.82
BG-M	0.14	48.4	1.19
BG-M-B (10%)	0.21	75.9	1.12
BG-M-B (20%)	0.28	108	1.04
BG-M-B (30%)	0.35	127	1.10

CP: cost price to produce 1 kg of maggot.

Discussion

This study has shown that the nutrient content of the substrate is a determining factor to optimize maggot production. An ingredient such as brewer’s grains is a good source of energy but is deficient in several amino acids to support the growth of maggots as showed by Mussatto et al (2006). The addition to the growth substrates of ingredients that appear to be more balanced in amino acids (cow blood or manure) or that provide specific otherwise deficient amino acids (lysine) allowed doubling the production of maggots on BG. As well-known for other more conventional single stomached domestic animal species such as pigs or poultry (Pérez and Sauvant 2004), BG are deficient in lysine for the growth of maggots (Mussatto et al 2006). Cow blood is a source of many essential amino acids (NRC 2000). Nevertheless, it seems that lysine remains the most critical one since no further improvement as compared to pure lysine was observed. Pig manure can improve production on BG probably due to the presence of bacteria from the digestive tract of pigs that are also rich in esential amino acids, among which is lysine (Metzler et al 2005; Dai Zhang Wu and Zhu 2010). Interestingly, the combination of BG-M-B was able to sustain the highest levels of maggot production, almost doubling that of BG-LYS. The increase in maggot production following the increase in cow blood doses in the BG-M mixture supports this previous argument. Further investigation is needed to identify the optimum dose of cow blood in the mixture combining technical to social, environmental and economical parameters.

Stability in disappearance of substrates observed from the 4th day onwards shows that larvae migration begins on the 4th day of production. Consumption of substrates decrease because larvae are migrating to the plastic bin. The optimal duration of housefly larvae production is 8 to 9 days. This allowed harvesting of maximal housefly larvae without harvesting many pupae that are of lower nutritional value than larvae (Pieterse and Pretorius 2013). Not all the substrate is consummed by larvae, since only 30 to 40% are used. The remainder can thus be use for other purposes on the farm such as composting. Apparently, only the first days of laying eggs are important to reach the highest housefly larvae production. Indeed, no difference was observed between temporary exposure and permanent exposure modalities. Once the first eggs are laid, substrates are less atractive to houseflies due to the decrease in smell and nutrient content of the substrates. Temperature of the substrates during all experiments allowed a good development of maggots with a mean value of 31.3°C. The temperatures below 20°C tend to slow the development and the transformation of the larvae in adults while temperatures above 35°C tend to accelerate the whole process (Lomas 2012). Temperatures from 7°C to 43°C are suitable for houseflies, but *M. domestica* are most active at about 33°C that is close to the ambient temperature in the humid tropics.

The increasing price of maggot production per kilogram for 30 % of addition of cow blood suggests that the cost effectiveness depends strongly on the level of blood in the substrate. The most cost-effective substrate seems to be a mixture of BG-M with 20% of cow blood following the increase in production coupled with the lower cost of maggot production per kilogram. Nevertheless, in the case of cow blood shortage, the simple mixture of manure to brewer’s grains is already an interesting solution although its cost is higher than the mixture of BG-M with cow blood. Its strength is that it requires very little investment for farmers by using available substrates on the farm, especially those growing pigs in intergrated agriculture-aquaculture system. In general all ingredients used in the mixtures are locally available as agricultural by-products, however cow blood requires the extra investment of the cost of transportation and the time lost for purchasing which was not considered in this economic analysis. Including these economic parameters the optimal amount of cow blood to add in the mixture should be lower than expected.

Conclusions

- Housefly larvae production is a good alternative in integrated agriculture aquaculture production to provide high quality protein to produce fish.

- Larvae production using available agricultural and agro-industrial by-products can be further improved by research to investigate the nutritive and the microbiological value for pond fishes, well as the global economic balance of the whole operation for large scale production in the farm.

Acknowledgements

The authors sincerely ackowledge the excellent collaboration of Papa Kally for allowing us to run the experiments on his farm. This research was funded by the Academie de Recherche et d'Enseignement Superieur – Cellule de Coopération au Développement (ARES-CCD, Brussels, Belgium).

References

Aniebo A and Owen O 2010 Effects of age and method of drying on the proximate composition of housefly larvae (Musca domestica Linnaeus) meal (HFLM). Pakistan Journal of Nutrition, v.9, n.5, p.485-487. http://dx.doi.org/10.3923/pjn.2010485.487

Bondari K and Sheppard D C 1981 Soldier fly larvae as feed in commercial fish production. Aquaculture, v.24, p.103-109 https://doi.org/10.1016/0044-8486(81)90047-8

Boyd C E 2015 Nitrogen. In : Water Quality. Springer, Cham, 2015, p. 223-241. https://doi.org/10.1007/978-3- 319-17446-4_11

Charlton A J, Dickinson M, Wake field M E, Fitches E, Kenis M, Han R, Zhu F, Kone N, Grant M, Devic E, Bruggeman G, Prior R and Smith R 2015 Exploring the chemical safety of fly larvae as a source of protein for animal feed. Journal of Insects as Food and Feed, v.1, p.7-16. https://doi.org/10.3920/ JIFF2014.0020

Čičková H, Pastor B, Kozánek M, Martínez-Sánchez A, Rojo S and Takáč P 2012 Biodegradation of pig manure by the housefly, Musca domestica: a viable ecological strategy for pig manure management. Plosone, v.7, n.3, e32798. https://doi.org/10.1371/ journal.pone.0032798

Dai Z-L, Zhang J, Wu G and Zhu W-Y 2010 Utilization of amino acids by bacteria from the pig small intestine. Amino Acids, v.39, n.5, p.1201-1215. https://doi.org/10.1007/ s00726-010-0556-9

Fasakin E A, Balogun A M and Ajayi O O 2003 Evaluation of full fat and defatted maggot meals in the feeding of clariid catfish *Clarias gariepinus* fingerlings. Aquaculture Research, v.34, n.9, p.733-738. https://doi.org/10.1046/j. 1365-2109.2003.00876.x

Holmes L A, Vanlaerhoven S L and Tomberlin J K 2012 Relative humidity effects on the life history of *Hermetia* illucens (Diptera: Stratiomyidae). Environmental Entomology, v.41, p. 971-978. https://doi.org/10.1603/ EN12054

Hwangbo J, Hong E, Jang A, Kang H, Oh J, Kim B and Park B 2009 Utilization of house fly-maggots, a feed supplement in the production of broiler chickens. Journal of Environmental Biology, v.30, n.4, p.609-614.

Kinkela P M, Mutiaka B K, Dogot T, Dochain D, Rollin X, Mvubu R N, . . . Bindelle J 2017 Diversity of farming systems integrating fish pond aquaculture in the province of Kinshasa in the Democratic Republic of the Congo. Journal of Agriculture and Rural Development in the Tropics and Subtropics (JARTS), v.118, n.1, p.149-160. http://nbn-resolving.de/urn: nbn:de:hebis:34-2017032852295

Metzler B, Bauer E and Mosenthin R 2005 Microflora management in the gastrointestinal tract of piglets. Asian-australasian journal of animal sciences, v.18, n.9, p.1353-1362. https://doi.org/10.5713/ajas. 2005.1353

Mussatto S I, Dragone G and Roberto I C 2006 Brewers' spent grain: generation, characteristics and potential applications. Journal of Cereal Science, v.43, n.1, p.1-14. https://doi.org/10.1016/j.jcs. 2005.06.001

National Research Council 2000 Nutrient Requirements of Beef Cattle: Seventh Revised Edition: Update 2000. Washington, DC: The National Academies Press. https://doi.org/10.17226/9791 .

Nuov S, Little D C and Yakupitiyage A 1995 Nutrient flows in an integrated pig, maggot and fish production system. Aquaculture Research, v.26, p.601–606. https://doi.org/10.1111/j. 1365-2109.1995.tb00950.x

Pastor B, Velasquez Y, Gobbi P and Rojo S 2015 Conversion of organic wastes into fly larval biomass: bottlenecks and challenges. Journal of Insects as Food and Feed, v.1, n.3, p.179-193. https://doi.org/10.3920/ JIFF2014.0024

Pérez J M and Sauvant D 2004 Tables of composition and nutritional value of feed materials (No. 636.085 Sa89t Ej. 1 021121). INRA. https://www.cabdirect.org/ cabdirect/abstract/20073063703

Pieterse E and Pretorius Q 2013 Nutritional evaluation of dried larvae and pupae meal of the housefly (*Musca domestica*) using chemical- and broiler-based biological assays, v. 54. https://doi.org/10.1071/ AN12370

Rufino M C, Tittonell P, Reidsma P, López-Ridaura S, Hengsdijk H, Giller K E, Verhagen A 2009 Network analysis of N flows and food self-sufficiency: a comparative study of crop-livestock systems of the highlands of East and southern Africa. Nutr Cycl Agroecosyst, v.85, p.169-186. https://doi.org/10.1007/ s10705-009-9256-9

Téguia A, Mpoame M and Okourou Mba J A 2002 The production performance of broiler birds as affected by the replacement of fish meal by maggot meal in the starter and finisher diets. Tropicultura, v.20, p.187-192. http://www.tropicultura.org/ text/v20n4/187.pdf

Received 26 November 2018; Accepted 5 January 2019; Published 1 February 2019

Go to top