Rural R&D for Profit Program

Closing the Loop: Black Soldier Fly technology to convert agricultural waste

Final Report

Australian Pork Limited

18th April 2019 – 28th February 2023

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Plain English summary

Objectives

This project investigated BSF waste treatment technology to provide a new waste management options that is more sustainable, productive and profitable for the primary industries. It explored the conversion of low-value agricultural waste products into high quality, innovative fertilisers and soil improvers. This will potentially create new markets for primary industries leading to decrease primary production costs. The project also sought to overcome key barriers to adoption of novel fertilizers/soil improvers by engaging with regulatory bodies via project activities.

Methods and Outputs

The effectiveness of BSF waste processing technology for processing manure and waste at treating manure and waste whilst producing high-value by-products was developed and optimised during this project. This was done at (a) small laboratory scale and (b) larger pilot scale at a partner site (FGS), using a range of wastes and manures from agricultural industries.

The evaluation of BSF co-products (insect meal and frass) as novel fertilizers and soil improvers were tested in laboratory screening trials, glasshouse trials, and field trials. The economic, environment and agronomic value of these products were determined. Greenhouse gas (GHG) emissions, odour and leaching potential were also quantified.

During this project UWA and DAFQ have been working with FGS to develop innovative BSF waste treatment technology for processing manure and waste into high quality fertilisers and soil improvers. The BSF meal performed as well as synthetic fertilizer in pot screening trials run by DAFQ. During pot and field trials conducted at UWA the frass showed potential as a soil conditioner to ameliorate soil constraints (acidity), increase nutrient supply and crop performance. However, further R&D into biosecurity, social license and regulatory stakeholder engagement is required to address barriers to adoption before the technology can be fully commercialized.

Outcomes and conclusions

Our research conducted in WA and QLD on processing poultry, dairy, piggery and abattoir wastes using BSF technology has produced the following benefits:

- Reduction in manure volumes of 70-80%*
- Novel fertilizers (insect meal). BSF insect meal can be converted into a high quality synthetic fertiliser via process of protein and amino acid denaturation. In its denatured form it can be substituted for synthetic fertiliser (urea, DAP) leading to increase crop yield and reduced leaching potential. Inclusion of BSF insect meal derived from manure in fertiliser formulation increases plant growth and N/P utilisation efficiency.
- Soil conditioner (frass) raised the soil pH and stimulated plant growth promoting rhizobacteria leading to better soil function and stability
- Demonstrated low pathogen and heavy metal contamination risk in BSF co-products generated from dairy manure.
- Reduced stable fly emergence*

- Reduced greenhouse gases (GHG) and nutrient leaching*
- Produces a stable product with limited odour
- Further modification of the frass via coating improved the nutrient retention and release to crops
- Can be granulated to reduce transportation, application and handling cost.

*Compared to raw manures

Using BSF technology to turn manures and waste products into highly valuable products that can be recycled back into the production on the farm, provides the potential for a closed loop approach that could increase productivity and profitability, reduce waste, labour and transport costs, improve environmental impacts, and contribute to enhancing the sustainability of agriculture in Australia.

Recommendations for future research and adoption

- Full economical evaluation of BSF technology as a waste treatment option for manure
- Demonstration that the bulk replacement of synthetic fertiliser with insect meal fertilisers is feasible and commercially viable is urgently needed before commercialisation and adoption.
- Scale up and deploy of BSF technology on farm. Pork production or poultry processing are reasonable sectors to target first. A commercialisation partner and a major primary industry partner would be required.
- Full life cycle analysis to determine if BSF technology could be included in the GHG abatement technology.
- Explore other benefits of BSF derived by-products, such as extraction of high-cost oils, vitamins, chitin and micronutrients, production of bioenergy and biofuels oils, and explore bulk feed replacement with other aquaculture species.
- Resolve uncertainty about quality requirements for wastewater-derived biomass used in fertilisers

Benefits of the project to industry/primary producers

- BSF technologies that treat agriculture waste significantly reducing waste volume by up to 80% whilst producing a high-value fertiliser (insect meal) and soil improver (frass). This offers commercial and revenue opportunities.
- Manure-derived insect meal is an excellent source of N and P and resulting fertiliser formulation is better than commercial synthetic fertiliser (Urea and DAP). Farmers (e.g. compost or waste-derived microbial biomass).This could lead to improved fertiliser efficiency and reduced input costs. Also, BSF meal provides an alternative options of P and N reducing economical and environment costs associated with P and N fertilisation production.
- Manure-derived BSF frass used as soil improvers could reduce input costs (liming, reduction in fertiliser application), increase N retention in soils, raise soil pH (liming effect), improve soil function, structure, stability and resilience to soil constraints (acidic soils, drought) leading to increased farming efficiency, reduced losses and lower GHG emissions.
- WA farmers could apply frass derived from poultry manure in broad acre and horticulture agriculture in banned Shires. An amended Biosecurity & Agricultural Management Act 2007, to include BSF frass derived from manures is sought. This would be a major cost saving.

Collaborations

Development of novel fertiliser using BSF meal derived from manures has led to an external alliance with (Matt Redding collaboration) and CSBP (a major fertiliser producer in WA), leading to applications for further external funding for future work. We collaborated with Prof. Stephen Rushton at Newcastle University, UK, for data analysis and modelling. The findings of this research has resulting in Future Green Solutions forming new collaborative relationships. We have gained support across the primary sector organisations including; Western Australia Broiler Grower Association (WABGA); West Australian Pork Producers' Association (WAPPA); Altona hatcheries; Western Dairies, South West Catchments Council (SWCC); Department of Primary Industries and Rural Development (DPIRD), Grower Group Alliance and Soil Alliance. The CBH group, a grain growers' cooperative and major freight distributor has expressed interest in the research for their grower client base. The project facilitated negotiations with DPIRD, Department of Environment, Regulation and Water, regulatory relevance group (that reports and makes recommendations to the Stable Fly Ban Act) and other policy makers and regulatory bodies. All provided considerable in-kind to the project.

This research was jointly funded Australian Pork Limited, Agrifutures, Dairy Australia Limited, Australian Meat and Processing Corporation, Eggs Australia, and the Department of Agriculture, Fisheries and Forestry (Commonwealth).

Abbreviations and glossary

Ammonium (NH_4^+)	is the inorganic form N commonly found in synthetic fertiliser which is converted to nitrate by soil microbes
Black Soldier Fly (BSF)	is a common and widespread fly of the family Stratiomyidae. Scientific Name: *Hermetia illucens*. It is an introduced insect to Australia and is non-invasive, cannot bite (no mouth) and is not known to spread disease.
Black Soldier Fly farming	is managing a Black Solider Fly facility, which uses the larvae to breakdown waste and then the larvae can be harvested to produce a range of profitable products.
Black Solider Fly Larva/Larvae (BSFL)	is the singular/plural term for a form of Black Soldier Fly between egg and pupa.
Carbon dioxide gas (CO_2)	a greenhouse gas and by-product of microbial and soil biota respiration
Carbon to nitrogen ratio (C:N)	a measurement that indicates how easily waste material and their by-products are degraded by microbes
Castings	excrement (or poo) of larvae insects.
Department of Fisheries Queensland (DAFQ)	key research partner
Future Green Solutions (FGS)	industry partner and BSF producer
Greenhouse gas (GHG)	a gas produced by microbes that contributes to global warming
Heterotroph	in this case, a microorganism organism that cannot produce its own food and instead uses carbon in wastewater and oxygen or nitrate as an electron acceptor.
Manure	dung of animals that can be used to fertilise land.
Methane gas (CH_4)	a greenhouse gas
Microcosm	an artificial, simplified ecosystem, in this case of soil, used to emulate under controlled conditions, the behaviour in natural soils
Nitrate (NO_3^-)	inorganic form N readily taken up by plants and commonly found in synthetic fertiliser
Nitrogen gas (N_2)	a gas produced by denitrifying microbes
Nitrous oxide (N_2O)	a gas produced by nitrifying and denitrifying microbes
Paunch	eviscerated belly area of an animal.
Substrate	a feedstock that insect larvae can be fed for biomass production
The University of Western Australia (UWA)	lead research partner
Waste streams	the complete flow of materials from domestic or industrial areas through to final disposal

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1 Project rationale and objectives

Overview

This project investigated whether Black Soldier Fly (BSF) larvae could be used to manage livestock wastes. The project aimed to develop Black Soldier Fly Farming (BSF) castings and larvae into high quality, low-cost, slow-release, granulated fertilizer products that are safe to handle, store, transport and apply. The project progressed options for more sustainable livestock wastes management (manures and paunch), and demonstrated opportunities for circular economy. Adoption of BSF technology and its products has potential to increase productivity and profitability via reduced input costs and generation of alternative revenue streams to a wide range of agricultural enterprises.

Background

Primary industries produce large volumes of waste by-products that contain significant amounts of macro and micro-nutrients but are typically in a dilute, nutritionally unbalanced form for agricultural crops. This makes its transportation and reuse off-farm impractical and uneconomic. Nevertheless, these by-products have long been recognised as an important soil amendment providing nutrients for crops and pastures as well as soil health attributes through increased carbon storage, soil structure and the development of robust soil biota. Australia's organic produce is a growing market, with the domestic consumer market predicted to be worth $2.5 billion by the end of 2023. Waste-derived fertilisers can play a key role in providing nutrients to this market whilst removing a by-product of production.

The Challenge

Unfortunately, poor livestock and waste management practices in the past have led to stable-fly (*Stomoxys calcitrans*) (as opposed to the Black Soldier Fly which is not a pest) outbreaks, odour, greenhouse gas emissions (GHG) and nutrient leaching and runoff into waterways. This has resulted in stringent application restrictions being imposed for manure application through Health (Poultry Manure) Regulations 2001, and more recently, through the Biosecurity and Agriculture Management Act 2007 (BAM Act). These regulations on manure disposal have led to loss of important marketing options causing significant cost increases (> $4 million annually), particularly to the WA poultry industries. Currently, composting to Australian Standards on-site is both costly and lengthy. Furthermore, the compost industry does not have sufficient scale, capacity or end market to process the entire allotment of manure. Consequently, large quantities of manure (225,000m^3 of manure per annum) are transported outside the banned Shires to broad-acre agricultural zones for pasture and crop fertilisation at a significant cost to producers.

The solution

Black Soldier Fly (BSF) (*Hermetia illucens*) is an emerging waste treatment technology that could significantly improve profitability and sustainability for livestock and cropping farms. BSF are non-biting, non-invasive, non-pest fly species which in nature consume many wastes. The flies themselves do not eat but the BSF larvae can be used to consume animal manures and other organic wastes such as abattoir and food waste. The mature larvae are typically harvested as high value protein and oil suitable for animal and fish feed (Schiavone et al., 2017; Wang & Shelomi, 2017). However, due to the potential biosecurity risks, insect meal is not permitted to be utilised in the food chain. One possible alternative use for the insect meal is to convert it into mineral fertiliser (ammonium nitrate) via protein and amino acid denaturation. In addition to the insect meal, BSF farming produces ½ tonne of fly larval castings for every ton of waste processed. The larval waste is known as frass. Previous studies have shown that BSF frass has a good macronutrient profile and an effective soil improver product (Nelson et al., 2004, 2011). Developing the frass as a high quality soil improver would open new markets and create new revenues for profit, making BSF more economically viable for the livestock industries.

Once fully commercialised BSF cultivation could process hundreds of tonnes of waste per day. Furthermore, it uses a very small amount of space (½ tonne per m^2 of factory floor). The BSF technology is both suitable for medium to large enterprises and may provide more flexibility for smaller enterprises or regional hubs. BSF larvae reproduce rapidly, has high feed conversion efficiency, and can be reared on a range of waste streams, including animal, slaughterhouse and cropping wastes (Moula et al., 2018). In addition, the BSF process has been shown to:

- significantly reduce the biosecurity (pathogen survival, stable fly)
- Significantly reduce environmental risks (odour, GHG emissions, nutrient leaching and runoff) associated with waste management by outcompeting black stable fly and decreasing pathogen loading and nutrient content (total N by 55 and P by 45%, respectively) (Lui et al., 2008; Erickson et al., 2004)

By using BSF technology to simultaneously handle and reduce the volume of objectionable wastes, and convert them to highly valuable products that can be recycled back into the production processes, BSF technology can both increase profitability, reduce waste and its associated environmental problems, and contribute significantly to enhancing the sustainability of agriculture. However, the BSF products cannot be developed further in Australia until biosecurity, environmental and food safety risks are addressed.

In addition, the agronomic and economic value of these alternative fertilisers and soil conditioners needs to be evaluated prior to roll out and adoption. It is also essential to provide fertilizer/conditioner products that are tailored to crop nutrient requirements, machinery and operations. Consequently, part of this research evaluated the usefulness of further modifying the frass/meal via granulation, palletisation, coating and encapsulation technologies. Finally, research is needed to understand the mode and mechanisms of delivery so that the frass can be developed as a slow release fertiliser to enable commercialisation and minimise surplus nitrogen fluxes into the soil.

Aim

This collaborative project between industry, government and researchers aimed to a) develop insect meal and frass as a low-cost, tailored, slow-release, granulated fertilizer and soil improver product that is safe to handle, store, transport and apply; b) quantify the biosecurity and environmental risks associated with applying frass to cropping and c) overcome some of the barriers to adoption in regards to biosecurity risks, environmental regulation and public perception by involving policy makers and farmers during trials and through extension activities.. Adoption of BSF technology and its products has potential to increase productivity and profitability via reduced input costs and generation of alternative revenue streams to a wide range of agricultural enterprises. This project required both direct research on biosecurity and environmental issues, together with the development of the larvae and frass as fertiliser/soil improvers product to ensure positive impacts on soil and crops.

How this project addresses the overall objectives RnD4P program

1. **Advanced technology (priority 1): Increased productivity and profitability through generation of new revenue streams, regain and expansion of markets and reduced costs.** The BSF technology will increase returns through the reduction of on-farm operational costs associated with waste management (handling, transportation and application) and will generate new income through the sale of waste to the BSF operator. The BSF technology will generate further income through the sales of high quality feed, fertilizer and soil improver products. On mixed livestock and cropping enterprises further economic gains will occur through improved crop production and decreased operational costs via efficient water, fertiliser, liming, and pest control usage.
2. **Biosecurity (priority 2): Identify the existence of and or quantify the absence of biosecurity threats and improve market access for primary producers.** This will be achieved by providing scientific evidence that BSF technology is a biocontrol solution and pathway for greatly reducing the impact of pests (stable fly and other nuisance flies) and diseases (*Camplyobacter, Salmonella, E.coli*) associated with waste management in agricultural communities.
3. **Soil, water and managing natural resources (priority 3): Sustainable fertilizer application practices that maximize fertilizer efficiency, decrease GHG emissions whilst improving soil health and water efficiency leading to increased crop productivity and profitability.** This will be achieved by the development of slow release fertilizer targeted to commercial and/or domestic consumer markets, plant species and soil types; soil conditioners that address soil constraints (acidity, compaction, water repellency) and improved application practices (soil inversion and deep placement vs surface application, granulated, encapsulated or pelletised) together with a better understanding of the plant and soil microbiome.
4. **Adoption of R&D (priority 4) Overcome the barriers to adoption in relation to manure management and reuse by working together with industry, consultancy, government agencies and policy/regulatory stakeholders.** This will be achieved by providing the scientific evidence, information and extension services to help obtain community, regulatory and planning support to approve new manure disposal methods and BSF technology.

Outcomes

New technologies, products, resources, tools and knowledge generated from this project will contribute to the following areas:

1. **Advanced technology:** Provide a sustainable pathway for manure and waste disposal enabling the livestock industries by developing BSF technology in Australia to increase productivity and profitability by generating new products, revenue streams and markets whilst reducing on-farm operational cost associated with manure management and fertilizer use.
2. **Biosecurity:** By developing an advanced waste management method for determining and quantifying biosecurity risks, including reducing stable fly and other nuisance flies, the

project should help primary producers reduce costs, increase productivity and competitiveness, and help industries manage risks or gain, maintain or regain market access.

3. **Soil, water and managing natural resources (priority 3):** Encourage the adoption of more sustainable management practices (deep placement, slow release fertilisers) by growers, producers and land users that improve soil health, water use efficiency and nutrient use efficiency and overcome soil constraints. Failure to adopt sustainable practices will lead to long–term soil degradation, reduced animal and crop productivity and food security, biosecurity and climate change risks.
4. **Adoption of R&D (Priority 4):** BSF technology is recognized as an approved method for waste management and applying products of this technology (fertilizer and soil improvers) to land align with current biosecurity and environment regulations. However, media science methodologies will be adopted to address social license issues around the use of BSF technology.

2 Method and project locations

This project involved multiple disciplines and activities as outlined in Section 3. Detailed research methods are described in Section 3 for each research activity. The project was organised as follows:

- **Australian Pork Limited** managed the overall project and communication with the Department.
- **The University of Western Australia** was sub-contracted as the primary research provider to deliver research in the following areas:
  - A team of agricultural economists delivered a survey to predict waste volume supply (Activity 5.1), assessed the economic feasibility, socio-economic costs and benefits, and market potential of using BSF for waste management (Activity 5.2); conducted a cost benefit analysis on using frass as a novel fertiliser / soil (Activity 5.5) and conducted consumer surveys to evaluate consumers' perceptions towards and willingness to pay for BSF-based fertiliser or soil improver products (Activity 5.9).
  - A team of soil biology and environmental microbiology experts to conduct pot trials, laboratory trials, and field trials to determine (a) the fate and persistence of pathogen in frass amended soil (Activity 5.3), (b) nutrient and heavy metal leaching potential of different frass products applied different soil types (Activity 5.4); (c) the Greenhouse Gas potential of different frass products applied to soil types (Activity 5.4); and (d) the mode of action of frass and its impact on soil biology (Activity 5.5).
  - A team of plant scientists and crop nutrition experts to conduct pot trials and field trials to evaluate the effectiveness of using frass as a fertiliser / soil improver on targeted pasture, broadacre, and horticulture crops (Activity 5.5).
  - A team of agricultural engineers to develop a granulated or pettelised fertiliser product, and evaluate the performance of BSF frass-based fertilisation products (Activity 5.6).
- **Future Green Solutions** was sub-contracted as an Industry Partner to produce Black Soldier Flies (BSF) larvae and frass. Future Green Solutions is a Perth-based innovator that uses BSF to upcycle low value organic waste into high value products such as animal feed ingredients and other agri-products. Future Green Solutions were sub-contracted to optimise frass production on livestock waste streams. Future Green Solutions also carried out experiments to calculate the growth, yield and survival rate of BSF larvae reared on livestock wastes (Activity 5.1).
- The **Department of Agriculture and Fisheries QLD** was sub-contracted as a research provider for Activity 5.7. Research Scientists at DAF conducted growth accelerator trials to develop tailored-release nitrogen fertiliser formulations with BSF frass, and assessed the effectiveness of nitrogen inhibitors in these formulations.
- The **WA Department of Primary Industries and Regional Development** was sub-contracted to conduct field trials to quantify Stable Fly emergence in cropping/horticulture soils amended with BSF frass fertilizer (Activity 5.3).
- **Scolexia** was sub-contract as a research provider to assess the biosecurity risks of Black soldier fly products (Activity 5.3)) and to determine whether BSF products could meet

livestock feed specifications. Scolexia is a widely respected animal and avian health consultancy firm. They conducted a combination of literature reviews and pathogen testing of the products.

Regular research meetings with all providers ensured that researchers were aware of the various ongoing activities in this project, and that research activities were well-aligned.

2.1 Geographic location of project activities

Name & type of site	Street Address	State	Postcode
Australian Pork Limited (RDC Headquarters)	2 Brisbane Avenue Barton	ACT	2600
The University of Western Australia (Research provider)	35 Stirling Highway, Crawley	WA	6009
Future Green Solutions research laboratory (Industry partner)	1 Underwood Ave, Shenton Park	WA	6008
Shenton Park Field Station (field testing site)	1 Underwood Ave, Shenton Park	WA	6008
Scolexia (research provider)	8/19 Norwood Crescent, Monee Ponds	VIC	3039
QLD Department of Agriculture and Fisheries (research provider)	41 Boggo Road, Brisbane	QLD	4102
Bogdanich Farms (field testing site)	Indian Ocean Drive, Chitna Road & Cowalla Road, Gingin	WA	4671

While project activities were carried out largely in Western Australia and Queensland, results are applicable to intensive livestock industries Australia-wide. The demo fertiliser products developed in this project could be used by any broad-acre cropping business, intensive dairy or pasture business, or horticulture business in Australia.

3 Project Outcomes

3.1 Project level achievements

In this section, we provide a detailed description of project achievements against each activity defined in Sections B and C of the grant agreement. Research methodologies, outcomes, and outputs will be detailed separately for each research activity.

3.1.1 Activity 1 - Project initiation

APL as the grantee under the agreement took on the role of Project Manager for the project. Agreements with partner and research organisations were made and executed in March 2020. A steering committee made up of a representative of each funding body and the key researchers was also convened and has met 16 times to date.

3.1.2 Activity 2 - Project planning and management

The Activity Work Plan and budget was developed and approved by the Department and was later updated to reflect the impacts of COVID-19 and associated lockdowns on the project. A risk management plan, communications and extension plan as well as a monitoring and evaluation plan was also completed to meet the governance requirements. The activity plan was regularly reviewed and used as a focal point for both steering committee meetings as well as research team meetings to ensure progress was occurring on all planned areas of work.

3.1.3 Activity 3 - Communication and extension

A communications and extension plan was developed at the commencement of the project, however significant revision was undertaken due to the impacts of COVID-19, particularly in WA where most of the project activities were based. Key communications activities were approved by the steering committee through a 'silent approval' process whereby if no comment was made in the given timeframe, it was considered supported for release. Simpler communications such as project updates were approved for release by the project manager.

Funding partners worked directly with the project manager and the research leads to develop their own industry specific communication pieces where needed, such as industry magazine articles and specific discussions between industry participants and research groups. Working closely with partners ensured the materials developed fit within their publication styles and needs.

Communication and extension activities included conference presentations, journal articles, field days, news segments, factsheets and research updates. We created two websites to share project information – a community of practice on the AgriFutures Extension Aus platform and a project activity website supported by UWA.

3.1.4 Activity 4 - Monitoring and Evaluation

A monitoring and evaluation plan was delivered with the first milestone and is reported against in this report (see Section 7.3) as a final evaluation. A mid-term evaluation was provided in January 2021, which included insights from funding partners on what was working well in the project which was taken into consideration for the remainder of the work.

The final monitoring and evaluation report showed that nearly all KPIs have been achieved, with only 6 of the 92 KPIs across the life of the project not been fully realised. In most cases this was because the KPI was more ambitious than what could be reasonably achieved in the project lifespan, such as securing waste classifications for BSF products in all states when these products have not been considered under the frameworks state environmental departments have previously. However, in all 6 cases, some progress has been made and nothing is considered 'not achieved' in its entirety.

Overall, the project delivered well on its objectives and the progress reporting monitoring was useful to identify emerging issues as they developed. Future projects may benefit from a co-designed monitoring and evaluation process done in partnership with the Department to support project staff development in these processes and to ensure that evaluation plans are understood by all.

3.1.5 Research Activity 5.1 - Screening and optimisation of waste streams

5.1(a) Characterise all waste inputs to provide a profile of nutritional value and properties

Waste was collected from a range of sources in the Perth-Peel region. The team tested the nutritional value for the following waste streams (KPI 1.7, KPI 2.4):

1. Dairy (pasture-based - separated manure solids, accumulated solids that had been stockpiled, fresh manure) from a Margaret River property;
2. Fish (offal and carcasses) from Cottesloe;
3. Pork (manure on straw, separated manure solids, separated manure liquids and mortality carcasses) from a farm located in Boonanarring;
4. Poultry broilers (chicken manure on woodchip) from a farm in Mariginiup;
5. Poultry layers (cage/barn-laid egg production - manure, carcasses, rejected eggs, egg protein from wash lines) from a farm located in Neergabby;
6. Paunch (fresh and stockpiled paunch waste) from a processing plant in Narrogin;
7. Rendering (abattoir waste) from a processer in Hazelmere.

Details for each waste stream are also reported in a Technical Report that accompanies this final report (KPI 3.3 and KPI 4.4; Appendix A). Industry partner Future Green Solutions (FGS) collected waste substrates and used them within 48 hours (hrs) of collection, with any surplus frozen and defrosted when needed for further trials. All substrates were ground to particle size <5mm and mixed by hand where appropriate.

The moisture content for the waste substrates was calculated by difference after drying them at 70 degrees for 24 hrs. Waste samples were provided to *Eurofins* for laboratory analysis which included: total and available nutrients (including heavy metals); indicator pathogens; ash content; solids energy (calorific value); total oil, grease, and carbohydrates. Selected results for these analyses are reported below in Table 1, Table 2and Figure 1 to Figure 9 showing that our dairy waste was particularly high in ash while low in proteins and fat; piggeries and poultry layer manures were high in protein and energy, varying with the waste matrix; layer waste (other than the 100% manure), paunch, and abattoir waste (rendering) was particularly high in fat and high in energy. No pesticides or antibiotics were detected in any of the waste streams. All waste was tested for heavy metals (Arsenic, Cadmium, Lead). Traces of Lead were detected in Dairy and Poultry broiler waste, but not in any of the other waste streams. Arsenic nor Cadmium were detected. Water had to be added to some waste streams to achieve a better moisture ratio (between 65%–70%) for use as substrate for the Black Soldier Fly larvae.

Table 1 Crude analysis of multiple livestock waste streams

Industry Waste Stream	Moisture (%)	Dry Energy	Dry Protein	Dry Ash	Dry Carbohydrate	Dry Fat
		(g/100g dry sample)				
Dairy						
Holding Pond	66.7	597.6	3.30	66.37	30.03	0.21
Separated Solids	61.7	629.2	4.18	64.75	31.33	0.13
Pork						
Liquid Manure	97.3	1,925.9	59.26	44.44	0.00	0.00
Manure On Straw	64.4	1,500.0	24.72	13.48	61.80	0.28
Manure Separated Solids	73.1	1,565.1	17.84	10.04	70.63	0.86
Swine Carcass	79.8	1,668.3	91.09	6.44	0.00	2.13
Poultry (Meat)						
Manure On Straw	31.8	1,554.3	27.57	10.70	60.12	1.45
Manure On Wood Chip	28.4	1,550.3	30.59	12.43	55.87	1.82
Poultry (Eggs)						
Egg Waste	74.7	2,264.8	61.66	3.95	3.95	30.43
Hen Carcass	56.4	2,339.4	57.34	4.82	2.29	35.55
Manure 100%	59.0	1,068.3	50.24	41.46	7.32	1.90
Protein Waste	84.6	2,668.8	52.60	8.44	0.00	46.75
Beef						
Paunch Waste	82.2	2,758.4	8.99	6.18	28.09	56.18
Rendering						
Protein Solids	74.3	2,498.1	29.18	4.28	23.35	42.41

Table 2 Nutrient, metals, and mineral analyses of different waste streams (mg/kg)

Waste Stream	Total Phosphorus	Total Nitrogen	Aluminium	Boron	Calcium	Copper	Iron	Manganese	Magnesium	Molybdenum	Sodium	Sulfur	Zinc
Dairy													
Holding Pond	740	1,800	1,700	0	7,900	3	5,600	23	1200	0	330	680	55
Separated Solids	1,100	2,600	2,100	0	4,900	8	5,500	35	890	4	320	790	60
Pork													
Liquid Manure	170	2,600	5	0	850	30	35	12	280	0	1,400	240	64
Manure on straw	3,100	14,000	730	7	4,900	30	210	63	1800	0	2,500	1,400	100
Manure separated solids	2,100	7700	22	15	5,800	81	180	55	940	0	1,600	1,100	170
Swine Carcass	1,800	31,000	4	0	5,400	6	430	4	310	0	1,700	1,800	78
Poultry (meat)													
Manure on straw	3,400	30,000	160	25	8,000	70	430	190	2700	4	2,300	2,100	140
Manure on wood chip	5,800	35,000	350	30	15,000	100	880	270	3700	6	3,000	2,600	200
Poultry (eggs)													
Egg Waste	1,900	25,000	0	0	620	0	17	0	120	0	1,200	1,500	9
Hen Carcass	6,400	48,000	5	0	9,500	0	50	1	210	0	110	2,500	25
Manure 100%	7,100	33,000	190	10	49,000	12	330	130	3500	0	1,200	2,500	68
Protein Waste	2,500	13,000	1900	0	410	8	40	0	30	0	280	830	3
Rendering													
Protein Solids	580	12,000	70	0	1,500	4	320	4	90	0	530	740	31

Note: Cadmium, Cobalt not detected.

Laboratory tests of dairy waste Figure 1 and Figure 2 indicate:

- Relatively high ash residue content
- Relatively low total protein and fat content
- No pesticides, antibiotics, Arsenic, Mercury, or Cadmium detected
- No Salmonella detected
- Low levels of Lead (1 mg/kg)
- High levels of Aluminium (1700-2100 mg/kg)
- Low levels of Ammonia, Potassium, and Sulphur

Figure 1 Dairy waste crude analysis (HP = Holding Ponds, SS = Separated Solids)

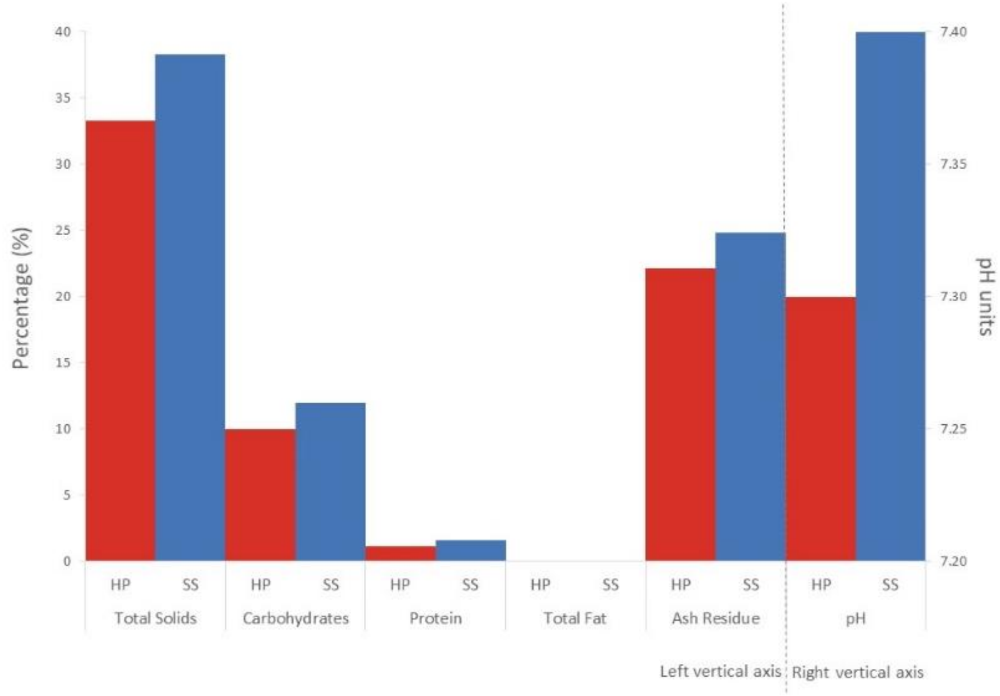
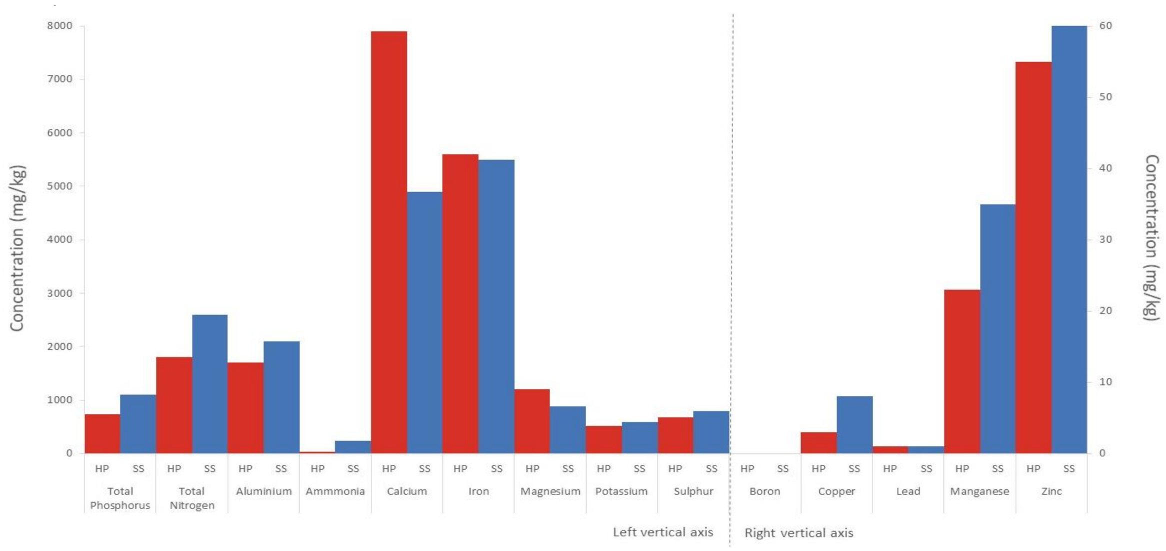


Figure 2 Dairy waste nutrient, metals, and mineral analysis (HP = Holding Ponds, SS = Separated



Laboratory tests of piggeries waste (Figure 3 and Figure 4) indicate:

- The proportion of solids, carbohydrates and protein varies with the waste matrix
- No pesticides, antibiotics, Arsenic, Cadmium or Lead detected
- Some Copper detected
- Relatively high levels of Aluminium in the Manure on Straw matrix
- High in Phosphorus, Nitrogen (esp. Ammonia), Calcium, Potassium and Sulphur

Figure 3 Piggery waste crude analysis (LM= Liquid Manure; MS = Manure on Straw; MSS = Manure Separated Solids; SC = Swine Carcass)

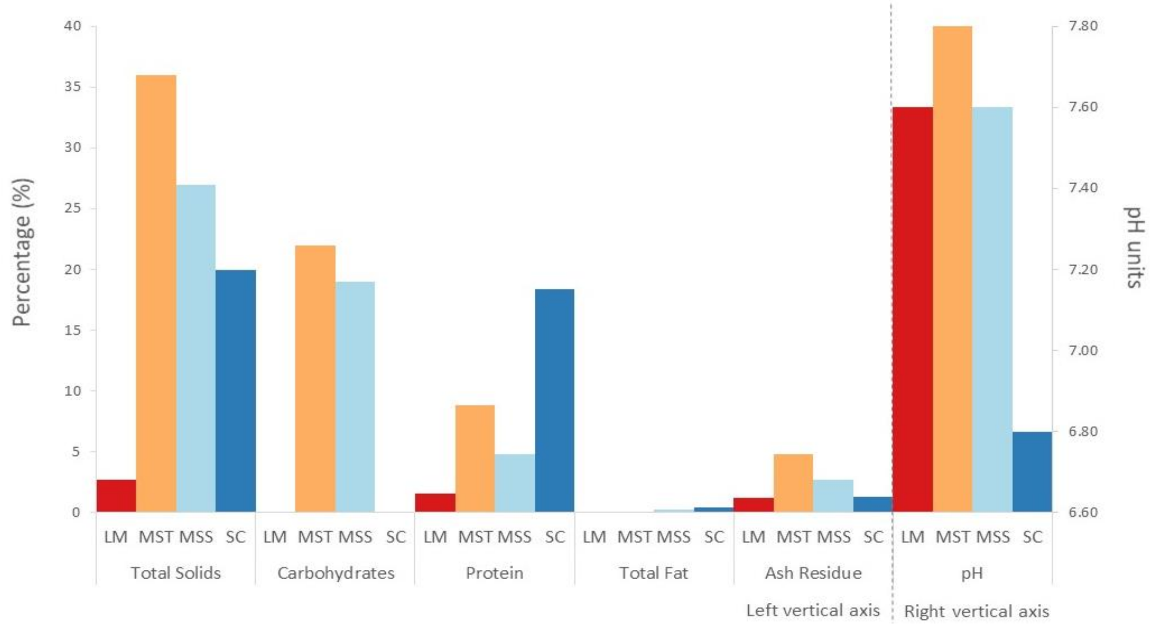
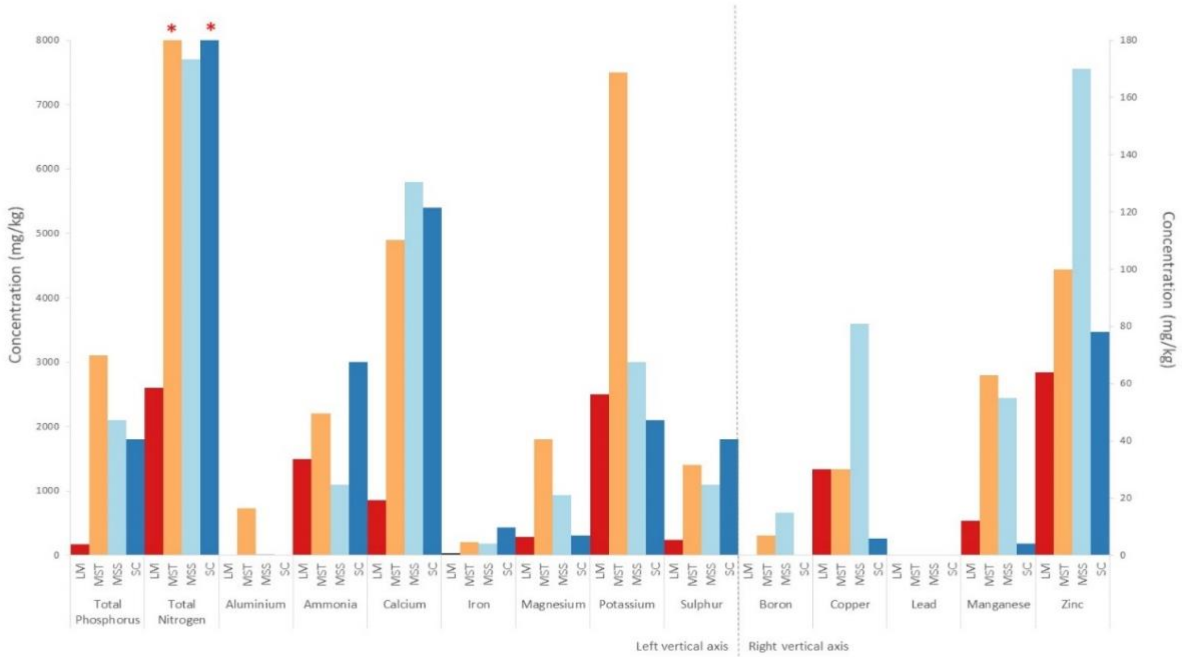


Figure 4 Piggery waste nutrient, metals, and mineral analysis (LM= Liquid Manure; MST = Manure on Straw; MSS = Manure Separated Solids; SC = Swine Carcass)



Note: * Total Nitrogen (MST) = 14,000 mg/kg; Total Nitrogen (SC) = 31,000 mg/kg.

Laboratory tests of poultry waste (Figure 5, Figure 6, and Figure 7) indicate:

- Waste from layers is low in carbohydrates and ash content, and high in protein and fats
- Waste from broilers is high in carbohydrates and proteins, and low in fats
- No pesticides, antibiotics, Arsenic or Cadmium detected
- Low levels of Lead (1 mg/kg) detected
- Copper, Boron, Zinc and high levels of Magnesium detected in some sample matrices

Figure 5 Poultry waste crude analysis (EW = Egg Waste; M100 = Manure 100%; PW = Protein Waste; MS = Manure on Straw; MW = Manure on Wood Chip)

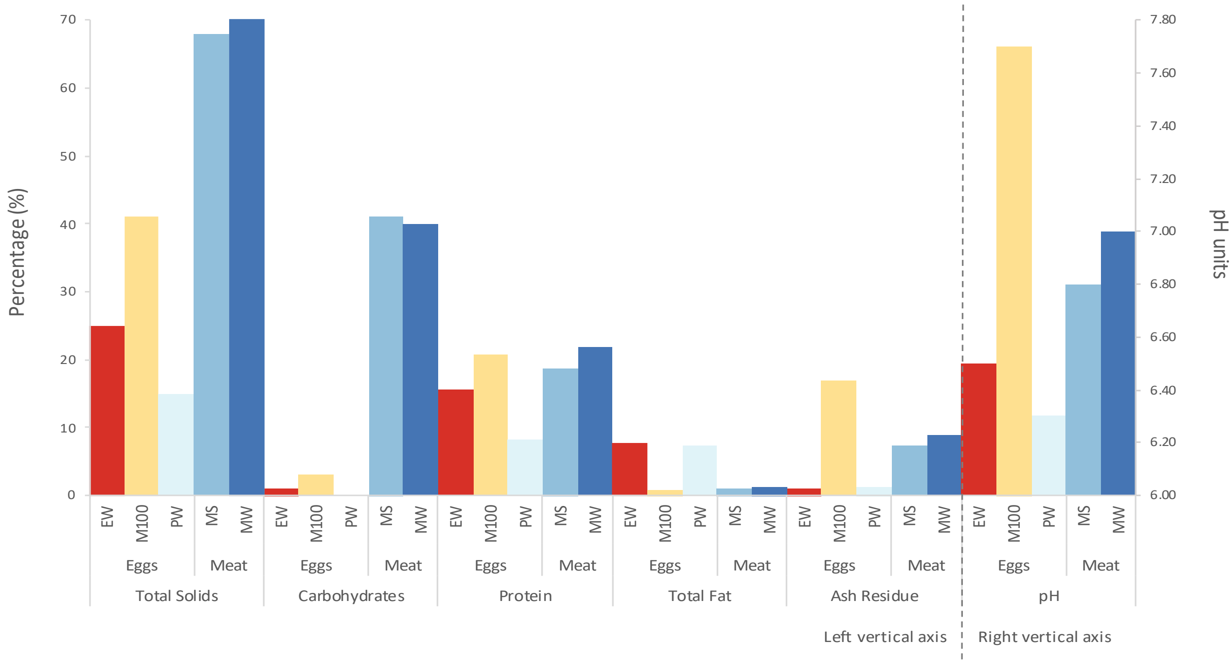
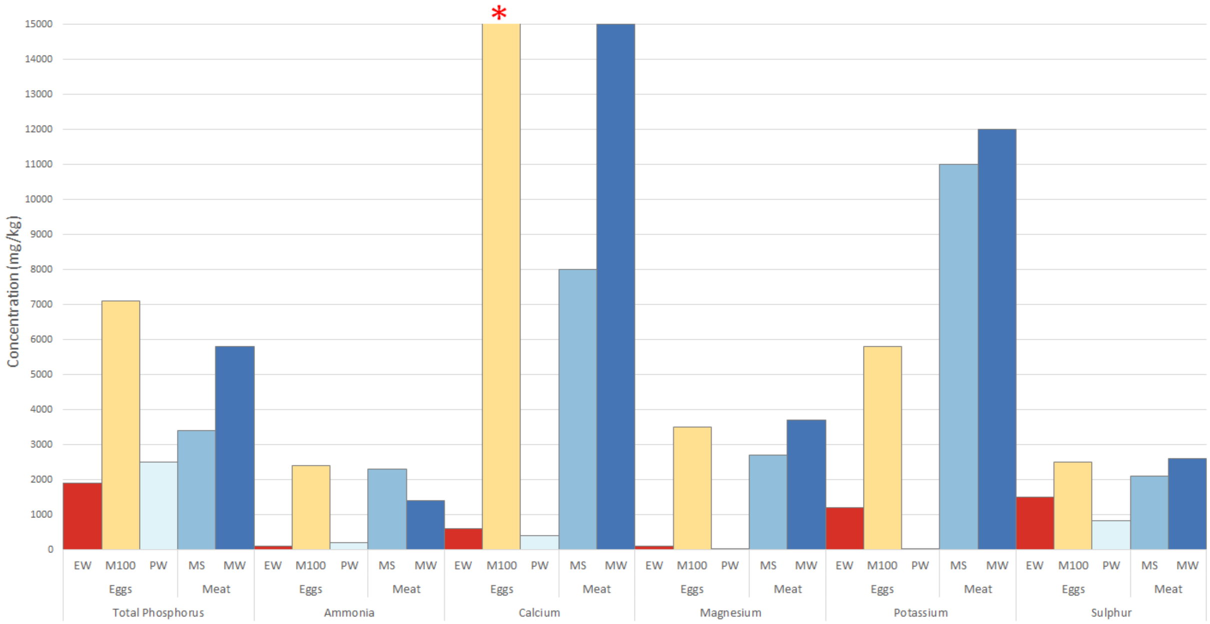
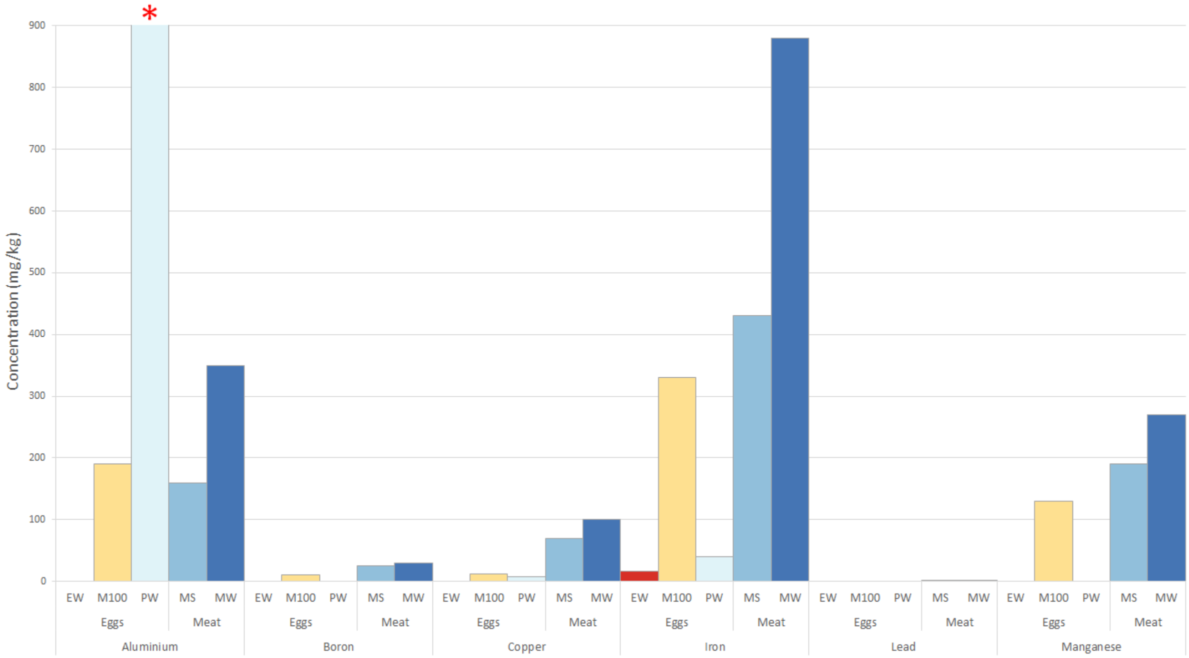


Figure 6 Poultry waste nutrient, metals, and mineral analysis in high concentrations (EW = Egg Waste; M100 = Manure 100%; PW = Protein Waste; MS = Manure on Straw; MW = Manure on



Note: * Calcium (M100) = 49,000 mg/kg.

Figure 7 Poultry waste nutrient, metals, and mineral analysis in low concentrations (EW = Egg Waste; M100 = Manure 100%; PW = Protein Waste; MS = Manure on Straw; MW = Manure on Wood Chip)



Note: * Aluminium (PW) = 1,900 mg/kg.

Finally, rendering waste analyses are presented in Figure 8 and Figure 9, showing that protein solids were not particularly nutritious in terms of proteins, fats, or minerals.

Figure 8 Rendering waste crude analysis

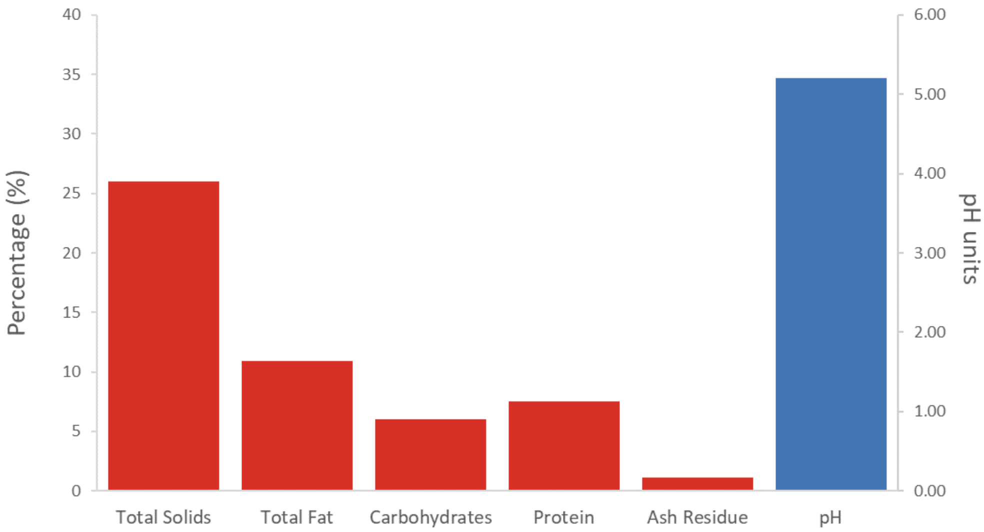
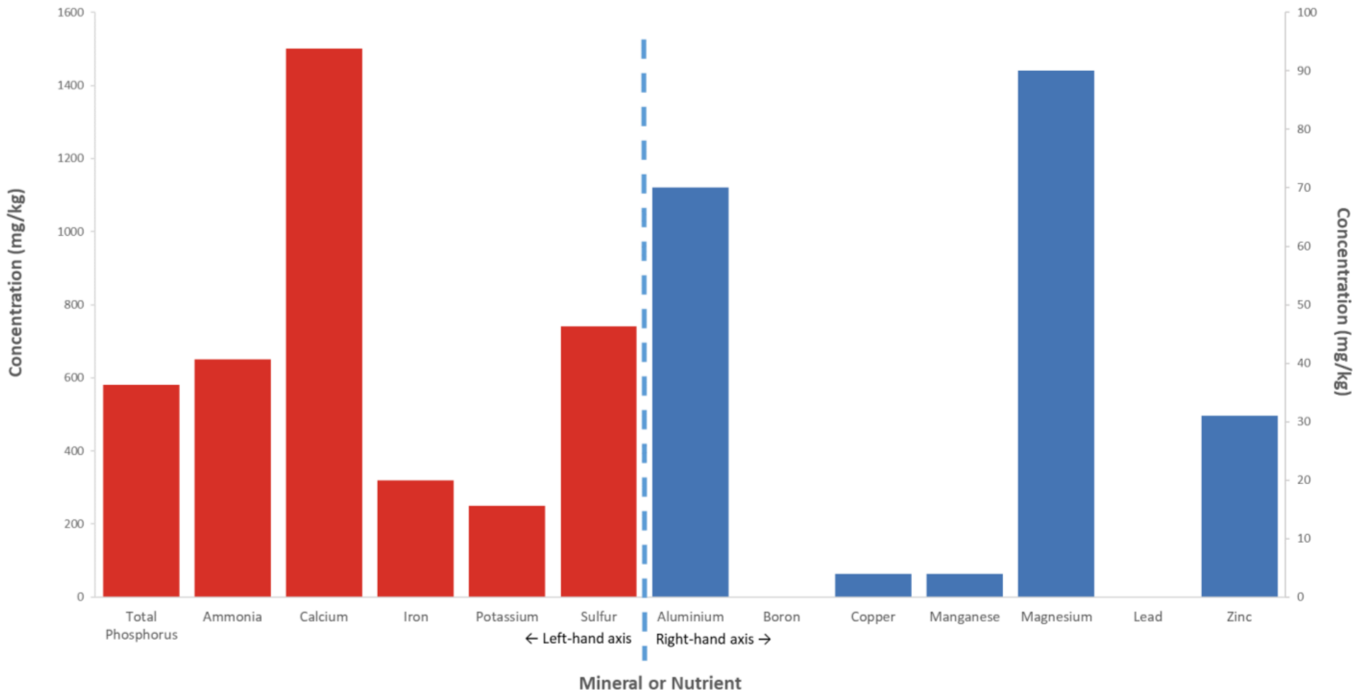


Figure 9 Rendering waste nutrient, metals, and mineral analysis



Any given waste stream is expected to have a highly variable nutritional value and mineral or (heavy) metal content. Assessing whether the waste from a particular property is consistent and appropriate as a BSF substrate would require repeated testing of the waste over time, after different livestock management changes (to test whether livestock management influences waste characteristics). Such tests were not conducted as part of this project. Nevertheless, Activities 5.1(c) and 5.1(d) provide a proof-of-concept that growing BSF larvae is feasible on different agricultural waste streams.

5.1(b) Conduct a survey to predict waste volume supply including variability and seasonality

To predict waste volume supply including variability and seasonality, we (a) collected information from existing data sources (literature and data repositories); and (b) conducted a national survey of livestock producers. Here, we describe the first set of data sources. The waste producers' survey is described under Activity 5.2 below.

Data was compiled for waste producers for all states based on publically available data from the AREMI website (https://nationalmap.gov.au/renewables/) (KPI 1.18). Information was extracted about the total residues and total dry residues of broiler litter production, dairy effluent, and feedlot waste data. The data is available at SA2 level for dairy effluent, and for LGAs for broiler litter and feedlot waste. The data shows where the 'hotspots' are for available waste. An internal report was presented with Milestone Report #2 (KPI 2.5).

The publically available data from AREMI was augmented and cross-checked through surveys with waste producers around Australia (Activity 5.2). Results are presented in a Technical Report (KPI 4.3) that is available at https://research-repository.uwa.edu.au/en/publications/spatial-analysis-of-farm-animal-wastes-in-australia.

This publically available technical report analyses the spatial availability of farm animal wastes in Australia, and presents the chemical analyses of different types of substrates (dairy, piggeries, and poultry).

Attempts were made to augment the publically available data with survey results. Unfortunately, the waste producers' survey (Activity 5.2) did not receive sufficient quality responses to provide a reliable prediction of waste availability across Australia.

5.1(c) Optimise frass production on single and blended waste streams based on predicted commercial supply ratios

5.1(d) Calculate the growth, yield and survival rate of BSF larvae reared on single and combined waste streams

Activities 5.1(c) and 5.1(d) are jointly reported in a Confidential Technical Report accompanying this final report (KPI 3.3 and KPI 4.4; Appendix A). Experiments were conducted between 2019 and 2022 with single and combined waste streams (Table 3). Over the course of these experiments, improvements were made to the rearing conditions of the BSF larvae, which significantly increased the technology's waste reduction potential, and much was learned about the larvae's ability to process livestock wastes.

Table 3 Experiments conducted by Future Green Solutions to produce frass and larvae on single and combined waste streams (Activity 5.1)

Trial	Substrates tested	Treatments
1	Individual waste matrices of Pork, Poultry broilers, Poultry layers, Rendering	Four feed rates per substrate (100mg, 150mg, 200mg, and 250mg dry feed/larvae
2	Mixed waste matrix of Pork, Poultry broilers, Poultry layers, Rendering	Four feed rates per substrate (100mg, 150mg, 200mg, and 250mg dry feed/larvae)
3	Industry Mix	Two liquid sources (liquid pork manures or water); Four feed rates per substrate (100mg, 150mg, 200mg, and 250mg dry feed/larvae)
4	Poultry layers, Industry Mix, Pork	Two moisture rates for poultry and IM (65% and 70%); Three feed rates for poultry and IM (150mg, 200mg, and 250mg dry feed/larvae); Two feed rates for pork (100mg and 150mg dry feed/larvae); Three seeding rates for pork (4.16, 5.20, and 6.24 larvae/cm^2)
5	Large scale IM	Upscaling trial of the Industry Mix (Trial 3) at 200mg dry feed/larvae
6	Pork mix	A. Fresh substrate (<24 hrs of collection) vs non-fresh (previously collected, frozen, and defrosted for this trial) B. Adding Clean Substrate to the fresh pork substrate at different rates. Varying processing times (from 7 to 19 days) C. Adding frass produced on Clean Substrate to the fresh pork substrate at different rates. Varying processing times (from 7 to 19 days). D. Supplementation with nutritional additives or bacteria inoculation (with probiotics)

Trial	Substrates tested	Treatments
7	Poultry broilers	Two moisture rates (66% and 70.4%); Varying processing times (7–19 days)
8	Poultry layers	A. 100% manure at two moisture rates (66% and 70.4%); Varying processing times (7–19 days) B. Layer mixed waste at 66% moisture; Varying processing times (7–20 days) C. Layer mixed waste at 70% moisture; Varying processing times (7–20 days)
9	Paunch	Two treatments (fresh and composted); Varying processing times (7–22 days)
10	Dairy waste	Five treatments harvested at Day 19: A. Fresh manure from the milking area B. Separated solids (Z-Filter products with flocculants) C. Solids from waiting area (3 wks stockpiled) D. Accumulated solids (3 months stockpiled) E. Mixed 50% separated solids and 50% fresh manures
11	Fish offal	Harvested at Day 21
12	Large scale pork mix	Upscaling trial of the Pork Mix (Trial 6), harvested at Day 17

* Pork = separated solids, manure on straw, swine carcass; Poultry = broiler manure on woodchips, layers 100% manures, layers broken eggs, layers protein wash-down, hen carcass; Rendering = Abattoir waste

This project demonstrates that BSF larvae can be used to reduce the volume of livestock. Treating livestock wastes with BSF larvae could reduce the volume of waste by around 80% over a 19 day period (Table 4**Error! Reference source not found.**). This means that, by using BSF technology, one tonne of agricultural (manures) wastes could produce around 200kg of frass and small amounts of larvae that can be used in the food-supply chain. No other technology can currently achieve such results.

Note that achieving over 85% reduction in manure waste volumes takes approximately three times longer than larvae on clean substrates only. This has implications for the costs of operating a BSF facility that will need to be analysed.

Table 4 Summary results for waste reduction (%) and larvae weight (g/ind) for the 'optimised' trials (Trials 5–12). n/a = no results available.

Trial	Waste reduction (%)	Average larvae weight‡ (g/ind)
Clean substrates (control) (Trial 9, Day 8)	82.7–93.0	0.191
100% Pork manures (Trial 6B, Day 8)	65.0	0.048
50% Pork manure : 50% CS (Trial 6B, Day 8)	80.2	0.074
Pork large scale (Trial 12, Day 19)	82.6	n/a
Poultry broilers -(Trial 7, Day 7)	75–81	0.055
Poultry layers (Trial 8, Day 7)	79–80	0.046
Paunch (Trial 9)	47–49	0.064
Dairy (Trial 10)	54.2–85.2	0.063
Fish (Trial 11)	n/a	n/a

‡ This column reports the maximum average larvae weight found for each substrate across trials.

Initial experimentation to produce BSF frass found that larvae growth and food conversion rates were significantly lower on manures than for insects reared on fruit, vegetable, and grain-based waste streams (the 'clean substrate' control treatment). This is likely due to the poor texture and nutritional value of manures, which tend to be high in protein but deficient in carbohydrates and total energy.

Inclusion of clean substrates into manure-based substrates showed some improvement in waste reduction, larval growth, and food conversion rates. Several enzymes were also trialled in the hope of unlocking greater nutrient potential within the substrate, particularly those locked in long chain carbohydrates, however no positive results were found using these additives.

Our analysis showed that the larvae grew better on blended substrates rather than single waste streams. It is thus recommended that manufacturers aiming to produce high quality frass or large quantities of larvae conduct further trials into the 'optimal' blend of wastes to generate the desired frass product. What constitutes an 'optimal' blend will depend not only of the nutritional value of the wastes but also the availability of different waste streams in the vicinity of the production site. For example, one may wish to proceed with a less 'optimal' substrate blend if particularly wastes are freely available in the area.

5.1(e) Determine the quality and quantity of BSF larvae meal and oil for feed trials (reared on approved wastes)

This has been integrated into Activity 5.8 (5.1e; KPI 3.4).

3.1.6 Research Activity 5.2 - Economic feasibility, socio-economic costs and benefits, and market evaluation

a) Cost assessment of current and future potential waste treatment options (including business as usual scenarios and BSF treatment options) for waste producers (i.e. costs associated with waste treatment, storage, transport, and disposal).

b) Evaluate the spatial distribution of waste producers, their volume, and supply and assess ramifications on transportation costs to BSF facility. Compare centralised facility against decentralised nodes on individual farms.

c) Determine the socio-economic costs and benefits of insect farming and alternative waste treatments (including environmental benefits such as reduced manure storage and smell)

Activities 5.2 a-c were completed as one package of work. A Postdoctoral Fellow was appointed to complete Activity 5.2 (KPI 1.10).

The spatial distribution of waste producers, the volume and supply of potential livestock waste for BSF processing was collated from the AREMI website (KPI 1.11). As reported in Activity 5.1, results were presented in a Technical Report (KPI 4.3) that is available at https://research-repository.uwa.edu.au/en/publications/spatial-analysis-of-farm-animal-wastes-in-australia (KPIs 1.11, 2.9, and 4.5).

Information about the types of socio-economic costs and direct financial costs for current treatment options for agricultural waste was collected through an in-depth review of the academic and grey literature. Various cost calculators are available that could be used to estimate the waste treatment costs for primary producers, however many of these are developed for the USA: Leibold & Olsen (2007), Hadrich, Harrigan & Wolf (2010), The Beef Feed Nutrient Management Planning Economics (BFNMP$) tool (Watson et al. 2019), the Waste Management Cost Estimator (Bullock et al. 1995). More detailed data about the waste treatment costs incurred by agricultural producers in Australia was therefore collected through stakeholder interviews with Australian producers (UWA Human Ethics approval RA-4-20-6192).

Interviews were conducted with industry experts and waste producers across WA, Queensland, and Victoria. A total of seven interviews were conducted via zoom during the months of October and November 2020 (KPI 2.8). Approximately 20 minutes was allocated for each interview. All interviews were conducted by the same project member. These interviews aimed to determine: (1) Stakeholders' level of understanding about BSF treatment; (2) Indicative interest in using a BSF facility for the management of livestock wastes; (3) Stakeholders' perception of the non-market costs or benefits (such as health impacts, biosecurity risks, or odour nuisance) of BSF farming, but also of current waste management treatment options; and (4) How agricultural producers currently measure their waste management and treatment costs (KPI 2.8).

The stakeholder interviews were used to create a nationally representative survey of agricultural producers (KPIs 2.10, 3.6 and 3.7). A particular challenge in the survey development was to create a questionnaire that could be applied to a wide range of producers (from intensive poultry or piggeries to beef cattle to meat processers), and that would capture the heterogeneity in waste management strategies across sectors. The final online survey (Appendix B) aimed to collect individual data on waste volumes, management practices, financial and non-financial costs of waste management, types of disposal practices, likely adoption of BSF technologies, and perceptions towards insect farming.

The survey was distributed through the project partners (APL, AMPC, Agrifutures, Eggs, Dairy) and other organisations, such as Nuffield Australia and grower group contacts of the research team. The research team tried to engage the project partners through repeated emails and follow-up phone calls, directly approached industry groups to distribute the survey link to potential respondents, and advertised the survey link on our social media pages. The initial survey incentive (a prize draw) was change to a $20 reimbursement for every completed response. Unfortunately, the survey link was picked up by survey 'bots' sometime around the 17th of September. Survey 'bots' are people or automated responses who do not fall in the target group of respondents, but who lie when giving their answers to qualify for the survey incentive. This resulted in a total of 4,368 responses to the survey, of which only the answers received before the 17th of September (22 responses) could be reliably identified as true answers. Due to the low quality of responses, we could not reliably predict waste supplies in geographical locations.

Despite the survey challenges, we were able to identify the various *types* of costs that producers incur (Table 5). Respondents reportedly incurred labour and operating costs for collection, treatment, storage, and disposal of their waste. Overheads and capital infrastructure costs were incurred primarily for waste collection, storage, and disposal. Only four farmers (two broiler farms, one dairy, and one pork) mentioned 'machinery' costs as an item. It became evident from the interviews and survey responses that producers do not typically collect information about their waste management costs. For example, only four respondents were able to provide an estimate of their freight costs (typically associated with disposal of waste), ranging from $0.60

per pig, to $10/tonne for egg producer, to $200/tonne for a pork producer (Table 6**Table 6**). With only four observations, no generalizable conclusions can be drawn around the transport costs of waste for the average producer.

Table 5 Costs associated with animal waste management practices (source: Interviews and survey of Australian livestock producers and processers)

Cost type	Animal waste management practice
Overhead (labour)	• Composting • Biogas and thermal treatment • Direct land application • Solids separation • Shed/ barn wash down • Stockpiling
Overhead (maintenance)	• Solids separation • Effluent ponds • Shed/ barn wash down
Variable (operating)	• Biogas and thermal treatment • Solids separation • Effluent ponds • Shed/ barn wash down • Screening and dewatering
Variable (freight, contracting)	• Composting • Biogas and thermal treatment • Shed/ barn wash down
Capital	• Composting • Biogas and thermal treatment • Direct land application • Solids separation • Effluent ponds • Shed/ barn wash down • Stockpiling • Conveyer belt • Screening and dewatering

Table 6 Locations and industry types covered in the producers' survey and interviews. Results are reported for the estimates of the freight cost associated with waste disposal (only reported by four respondents).

State	Industry	Freight costs	Freight (unit of measurement)
Western Australia	Dairy cattle	-	-
Victoria	Eggs	-	-
Western Australia	Pork	$0.60	per pig
New South Wales	Pork	-	-
Queensland	Broiler	-	$ per km per m3
Tasmania	Broiler	-	-

New South Wales	Pork	$200	per tonne
Western Australia	Dairy cattle	-	-
Queensland	Eggs	$10	per tonne
New South Wales	Eggs	-	-
Victoria	Dairy cattle	-	-
Victoria	Dairy cattle	-	-
Tasmania	Eggs	-	-
Western Australia	Dairy cattle	-	-
Western Australia	Broiler	$15	per tonne
Western Australia	Pork	-	-
Western Australia	Dairy (industry advisor)	-	-
Western Australia	Meat Processing	-	-
Victoria	Piggery (consultant and research advisor)	-	-
Queensland	Eggs	-	-
Victoria	Dairy processing	-	-
Western Australia	Broiler	-	-

A discussion of the different market- and non-market costs associated with livestock waste management (as shown in Table 5) and a framework for analysis could be written up as a scientific paper (KPI 3.8). A draft paper was being prepared, however the first author was on maternity leave at the time of writing this final report. We anticipate that the paper will summarise the financial costs, as well as the socio-economic costs (including the non-market costs) of waste management for different types of producers. We also analyse the perceptions of producers towards insect farming, to assess whether producers perceive there to be any negative impacts (costs, e.g. odour nuisance) or positive impacts (benefits, e.g. environmental benefits) from insect farming. At the time of writing, we have paused further progress on the paper until the first author returns from parental leave in March 2023.

The report on the spatial distribution of waste supply and implications for transportation costs to a BSF facility (KPI 4.5) has been reported under Activity 5.1(b). We did not compare centralized and decentralized nodes on individual farms, as the technology is still at an experimental stage. Using BSF larvae to manage agricultural waste is a specialized activity and therefore the adoption potential on individual farms was considered low. Feedback from interviews with waste producers also indicated that farmers viewed rearing insects as a complex operation, akin to operating another farming enterprise, and hence individual, decentralized facilities are unlikely to be adopted. Furthermore, experiments in this project have shown that BSF larvae grow better on mixed waste streams, which means a centralized facility is likely to produce a better product for the industry.

d) Market analysis of product types, volumes, potential demand and revenues across fertiliser markets (horticulture, broad acre, pasture and home garden).

A market analysis of organic fertilisers for the agricultural sector was conducted by means of a literature review, review of providers' websites, and personal communications with compost and fertiliser producers. Thirty fertiliser competitor products were found (not necessarily insect-based), including comparable organic fertilisers such as manures, insect frass, and compost. Prices vary widely across products. A report was written that summarises the nutrition composition and claimed benefits of existing product. The market analysis report (KPI 4.7) was approved by the project team and is publicly available online at: https://research-repository.uwa.edu.au/en/publications/market-potential-for-black-solider-fly-fertiliser-products

Previous analyses of BSF frass products conducted within the project indicate that BSF frass fertiliser has a higher carbon content (>35%), a higher Potassium content, and comparable Nitrogen and trace elements compared to competitor products.

An information fact sheet about Black Soldier Flies was produced for consumers (KPI 4.8) called What are Black Soldier Flies: some mythbusters.

e) Assess the economic and environmental value of further processing frass and/or larvae to create granular, pelletised, powder, frass tea, encapsulated or slow release high quality fertiliser for different target markets.

We evaluated the effectiveness and value of using BSF frass as a fertiliser. Potential environmental values include improved microbial activity in the soil, crop yield, or increased soil carbon. Environmental benefits and their economic values were estimated as part of Activity 5.5(d). Progress on Activity 5.2(e) is therefore reported under Activity 5.5(d).

A review of the academic and grey literature provided some information about the potential avoided costs if livestock waste were treated with BSF technology. At the time of writing, no studies exist that have estimated these avoided costs, and hence no estimates are available for the potential value of using BSF to treat livestock waste. **Table 7** summarises the types of non-market benefits that could be generated if impacts were avoided through treating manures with BSF technology.

Table 7 Non-market costs/benefit types arising from animal waste management practices

Cost/ benefit type	Example changes (improvement under BSF technology)	Example valuation approaches
Physical health	Number of waste-management related illnesses	Health impacts from pathogen contamination (e.g. to contaminated drinking water) or air pollution; Avoiding health impacts from consumption or exposure to pollutants (e.g. protection against contaminated drinking water by purchasing water filters); WTP to avoid health effects from contaminated drinking water or air pollution
Mental health	Reported cases of stress and anxiety	Mental health impacts on workers from stress or illness; WTP to avoid mental health effects
Safety	Perceived safety of waste management	WTP to avoid risk of fatality or illness from animal waste management practices
Annoyance	Levels of annoyance due to odour	Purchasing equipment e.g. face mask to avoid effect of odour; lower property prices in odour prone areas; medical costs related to odour
Ecosystems	Health of soils	Market impact to crop productivity from reduced soil health; Cost to restore contaminated soil; WTP to improve soil health through improved waste management or WTP to reduce the risk of soil contamination

	GHG emissions	Income from trading carbon credits in market interactions; WTP to reduce GHG emissions from implementing alternative waste management practices
	Abundance of invasive pests	Improved animal productivity from reducing risk of pest and disease outbreak
Water quality	Condition of waterways from reduced leaching of nutrients in manures	Improved animal productivity from access to clean drinking water; WTP to avoid poor water quality (see also 'recreation')
Recreation	Recreation activity in the area	Observations or surveys of (potential) recreational use; WTP to avoid impact to recreation sites at nearby waterways

Note: WTP = willingness to pay

f) Cost-benefit analysis of BSF fertiliser production at different scales of production and product types (e.g. granulation) to evaluate production costs versus revenues and environmental benefits (including odour and human health benefits)

A review of the waste management literature and interviews with waste producers was used to compile a list of the potential impacts of agricultural waste on the environment and human health. We also included social costs i.e. perceived safety of the waste management process and animal welfare resulting from pathogen disease incursions. Negative impacts are a cost (economic value) of agricultural waste, while positive impacts would be considered a benefit. Our intention was to augment the review with data collected through the waste producer's survey. However, due to data issues (as reported above), survey respondents could not be used.

The various types of environmental, health, and social costs are summarized in **Table 7**. A spreadsheet model was initially developed (KPI 4.6) to provide quantified estimates of costs. However, due to the limited availability of studies on this topic, no generalised conclusions could be drawn to create a robust benefit-transfer model for the environmental and health cost of livestock waste management strategies. The team has started a systematic review of the literature on this topic. This paper will be completed when the first author returns from parental leave.

From our work on Activity 5.2, it is evident that little information exists in the peer-reviewed literature on the costs and benefits of alternative livestock waste management strategies. It is strongly recommended that additional data is collected on the costs that producers incur when collecting, treating, storing, and transporting their waste. Given the challenges associated with farmers' surveys, it is recommended that such data is collected through interviews with livestock producers and participation in farm- or field-days (which was not possible during the Western Australian COVID travel restrictions). A better understanding of the costs of current waste management would provide information about the potential avoided costs when treating livestock waste with BSF larvae.

3.1.7 Research Activity 5.3 - Assess the biosecurity risks of Black soldier fly products (frass and larvae)

5.3(a) Provide a literature review on the biosecurity risks with BSF products

A review on of the Biosecurity Risks of Using Black Soldier Fly (*Hermetia illucens*) in the Australian Agriculture Sector (KPI 1.13, 2.11, 3.12) has been produced (Appendix C).

Summary of report

There has been increasing interest in farming Black Soldier Fly larvae (BSFL) commercially in Australia and abroad. Feed resilience, circular economy principles, and environmental benefits are widely considered important reasons for the use of insects for farming. Black soldier flies (BSF) are highly adaptable with respect to farming and processing methods, but this complexity creates challenges from a biosecurity risk control perspective. For improved control of biosecurity and hygienic risks it is vital to consider the whole production process, in addition to the current regulatory focus which targets controlling the substrates used to rear BSF.

Figure 10. Biosecurity risks within the black soldier fly production chain

Overall, BSF larvae appear to have low inherent biosecurity risks both to the environment and as an animal feed (Figure 10). Bacterial pathogens have been identified in various feed substrates given to BSF, however, the abundance of majority of these bacterial strains are significantly reduced during BSF production cycle and therefore have limited downstream risk. Rearing conditions and larval factors are believed to be very important for controlling bacterial pathogen survival. These factors are complex and poorly understood. Fungi, viruses, parasites and prions could pose a significant biosecurity risk but have not been thoroughly investigated.

Currently, there are no Australian State or federal regulations, microbial standard or rearing requirements that BSF farming enterprises must follow to reduce the biosecurity and hygiene risks. Internationally, biosecurity control has focussed on selecting low risk substrate to feed the insects. Insect specific regulations are available In the European Union (EU) and the United States of America (USA) where insect-derived feeds have to be recognised as GRAS (Generally Recognized As Safe).

Passive transfer of pathogens from the substrate by larvae to the BSF farming product is a major risk factor if the product is to be used as animal feed. Vegetable based substrates are considered much safer than manure based substrates and certain BSF rearing processes can be used to mitigate these risks further. Post-harvest processing methods of BSF larvae and frass can also impact on the biosecurity and safety risk of BSF products. A major challenge for insect farming is how to process insects without destroying their nutritional value. Traditional Australian rendering standards and methods may not be suited for insect farming if the products are to retain high nutritional value.

In Australia there are restrictions on what substrates can be used to rear insects. These regulations are state based and hence there is variation across Australia (Figure 11). Some animal feeds are regarded as either "swill", "Prohibited Pig Feed" (PPF), or "Restricted Animal Material" (RAM). Individual state biosecurity and animal feed guidelines must also be consulted for heavy metal, herbicide, antimicrobial and pesticide levels. Currently, there is no mechanism for approval or accreditation of insect rearing system, inclusion/exclusion of specific stock feeds and production methods in Australia. The lack of a clear regulatory framework regarding insect farming, including input and product testing requirements has been an impediment to potential start-up businesses.

	Aquafeed	Petfood	Poultry	Pigs	Ruminants
Aust Capital Territory	☑	☑	☑	☑S	☑S
New South Wales	☑	☑	☑	☑S	☑S
Northern Territory	☑	☑	☑	☑S	☑S
Queensland	☑	☑	☑S	☑S	☑S
South Australia	☑	☑	☑	☑S	☑S
Tasmania	☑	☑	☑	☑S	☒
Victoria	☑	☑	☑	☑S	☒
Western Australia	☑	☑	☑	☑S	☒

☑ All substrates approved for use

☑S Substrate approved for use if not classified as swill and/or RAM

☒ Must never be fed regardless of insect substrate used.

Figure 11 Current Australian State animal feed regulations applied to insect rearing

To conclude, the biosecurity risks of farming insects are measurable and controllable, however, many knowledge gaps in how to best manage these risks remain unknown. There is considerable uncertainty regarding the economic value of BSF larval farming and mitigating the biosecurity risks of farming insects. Future work should focus on developing of clear and consistent regulatory guidelines for BSFL farming including input and product hygiene and biosecurity requirements. Research should be directed to answering specific questions raised in the development of those guidelines.

5.3(b) Liaise with government regulatory bodies to ensure research addresses biosecurity concerns

Activities 5.3 (b) and 5.4(b) have been integrated.

Communication was initiated with the following State Environmental Regulators to identify the most relevant person/people to discuss the manure derived black soldier fly products (KPI 1.14). The following State Regulators were contacted and provided a response (Table 8) .

Table 8. State regulatory bodies involved in the consultation

State	Department	Role/Area
Western Australia	Department of Water and Environmental Regulation	Manager Licensing Waste Industries Industry Regulation - Regulatory Services
South Australia	SA Environment Protection Authority	Senior Environmental Adviser Circular Economy and Waste Operations
New South Wales	NSW Environment Protection Authority	Hazardous Risk Monitoring Management and Advice Regulatory Practice and Environmental Solutions Division
Queensland	Department of Environment and Science	Principal Environmental Officer Waste Assessment Waste Operations, Waste and Enforcement
Tasmania	Environment Protection Authority Tasmania	Senior Environmental Officer Waste and Wastewater, Waste and Contamination
Victoria	Environment Protection Authority Vic	Policy Officer Waste and Contaminated Land Unit, Policy and Regulation Branch

Discussions were held either via a teleconference or zoom/teams meeting with individuals or teams from each of the regulators who were generally from waste policy or permissions teams. It should be noted that the discussions were based on generalised processing and product information and therefore provide the initial thoughts and interpretation by the regulators. Their positions expressed are not definitive and are subject to change based on more detailed data (such as justification of fit for purpose status).

Additionally, an information fact sheet on Biosecurity Risks of Black Soldier Fly (BSF) Larvae as an animal feed was produced (Appendix C).

5.3(c) Determine the pathogen loading at the manure input, larval and frass stages of production

A technical report on Pathogen and Heavy Metal Loading at the Manure Input, Larval and Frass Stages of Black Soldier Fly Production (KPI 4.9; KPI 1.15, 2.12, 3.13) was produced (Appendix D).

Summary of report

This investigation was undertaken to determine pathogen and heavy metal loading at the manure input, larval and frass stages of Black Soldier Fly (BSF) production. Other biohazards were included in the investigation. For the purposes of this review biohazards are considered to be heavy metals, plant propagules and *entero* viruses (as per AS4454) as well as some insecticides and herbicides.

The analysis focused on organic wastes from Australian farms. A representative dairy, pig and poultry farm were selected from which manures were gathered. These results were compared to a normal vegetable mix used for rearing the BSF larvae. Table 1 shows that the faecal coliform loading varies between different manure types and stages of the BSF production cycle. Processing vegetable waste and dairy manure via BSF technology appears to increase faecal coliform abundance in the frass and larvae. In contrast, lower pathogen loads were observed in larval and frass samples reared on poultry and piggery manures.

Table 9. Faecal coliform counts from four neonatal BSF larval samples

	Faecal Coliforms (000's CFU/g)			
Substrate / Batch	**Input Substrate**	**Output Processed Larvae**	**Output Frass**	**Output Substrate no BSF Larvae**
Vegetable Mix 1	0	48,000	20,000	190
Vegetable Mix 2	0	0	84,000	
Vegetable Mix Avg	*0*	*24,000*	*52,000*	*190*
Dairy Manure 1	1,300	3,400	66	1,900
Dairy Manure 2	360	8	11	
Dairy Manure Average	*830*	*1,704*	*39*	*1,900*
Chicken Manure 1	1,000	0	590	1,200
Chicken Manure 2	750	2	640	
Chicken Manure Average	*875*	*1*	*615*	*1,200*
Pig Manure 1	1,500,000	0	20	450
Pig Manure 2	1,500,000	0.1	110	
Pig Manure 3	1,500,000	0.2	80	
Pig Manure Average	*1,500,000*	*0.1*	*70*	*450*
Manure Total Average	643344	487	217	1,183

Black soldier fly (BSF) larvae altered pathogen loads in manures and vegetable substrate. There was a consistent pattern of reduced bacterial pathogen loads during the rearing regardless of the feed substrate, but not total elimination of risk. Spore forming bacteria *Bacillus cereus* and *Clostridium perfringens* levels were markedly reduced during BSF rearing whereas *Enterococcus* were more resistant. Pathogen reduction was dependent on manure type, with *Salmonella* reduced to undetectable levels in the processed larvae reared on pig manure but not poultry or dairy manure. Interestingly, the abundance of *Escherichia coli* (*E. coli*) increased in the larvae reared on vegetable mix substrates but not in larvae reared on piggery manure (Table 10).

Table 10 Pig Manure larvae bacterial counts CFU/g on larvae before and after processing

Bacteria Species	Output Fresh Larvae Pooled All Batches (n=1)	Output Processed Larvae (n=3)
Faecal Coliforms	36000	100
E. coli	3300	107
Salmonella	1/1	0/3
Clostridium perfringens	1000	0
Bacillus cereus	0	0
Enterococcus	0	180
Listeria monocytogenes	0	0
Campylobacter	0/1	0/3

Yeast and fungi loads can also be high in the outputs from BSF rearing. Larvae and substrate type have marked influence on the species present. Opportunistic fungal pathogens were present in product outputs, but no mycotoxins were detected. Therefore, BSF outputs (products) from higher risk substrates such as manures may require additional processing such as rendering and composting to make them safe from biological risks.

5.3(d) Assess the stable fly emergence from the frass

An information fact sheet on stable fly emergence in cropping and horticulture soils amended with BSF frass fertiliser (KPI 4.10) was produced.

Stable Fly is a declared pest that grows in many types of rotting organic materials including animal manures, vegetable waste and plant residues. In Sept 2012, the Stable Fly (SF) was declared a pest under the Biosecurity & Agricultural Management Act 2000. Consequently, stockpiling, land application and movement of manure was banned in 13 Shires around Perth. Transporting the litter outside the banned Shires is very costly. Regulation of manure disposal options have led to loss of important manure marketing options causing significant cost increases (> 4 million), currently it costs producers \$8 per m^3 to dispose of litter. As a declared pest, it was important for this research to measure Stable Fly emergence after applying BSFL frass to agricultural soils.

Field trials were conducted around Gingin, 90km north of Perth (KPI 2.13, 3.14). This area has a long history of Stable Fly outbreaks. Stable Flies have developed in large numbers due to the combination of livestock production and increased horticulture with overhead irrigation. The porous, sandy soils have also enabled Stable Fly larvae to develop unhindered. Five field trials were conducted on three commercial vegetable farms during 2021 and 2022. Frass from Black Soldier Fly farming was applied to soil at rates of 1^{-10} tonnes per hectare. The treatments were conducted on replicate strips of 100 cm squared (Figure 12) across one or more sites.

Figure 12. Application of BSFL frass to irrigated horticulture near Gingin, Western Australia. The emergence of Stable Fly is promoted by frequent overhead irrigation and porous, sandy soils

Overall, the research findings showed that stable fly emergence either nil or negligible from any of the frass treatments and the emergence of other nuisance was also very low. Applying frass from Black Soldier Fly farming at recommended rates (up to 5 tonnes per hectare) produced no adult Stable Flies from 144 plots. At high rates (7.5 and 10 tonnes per hectare) it only produced 4 adult Stable Flies from 96 plots. In comparison, applying raw (uncomposted) poultry litter across the same five field trials produced 146 adult Stable Flies from 48 plots demonstrating that Stable Flies were active at the field trial sites (see Table 11).

Table 11. Stable fly production after applying soil amendment from BSF farming or raw poultry manure (positive control) to horticultural soils across five trial sites near Gingin, Western Australia.

Application rate (t/ha)	Number of Adult Stable Fly emerged
BSF frass	
1.0	0
2.5	0
5.0	0
7.5	1
*10.0	3
Poultry manure	
20.0	146

*Twice the recommended rate

Overall, the findings clearly showed that stable flies (and other nuisance flies) are unable to develop in soil amended with manure frass applied directly to non-irrigated pastures and cropping land at rates up to 10t/ha. This research shows that manure frass can be used in agriculture without being associated with fly breeding. These findings could be used to amend the Biosecurity & Agricultural Management Act 2007 to permit the application of animal manure processed by Black Soldier Flies in WA shires where animal manure is currently banned. This would be a major cost saving to WA livestock producers and may create new markets for Black Soldier Fly products, including frass and soil improvers or fertilisers.

5.3(e) Conduct pot and/or laboratory trials to determine the fate and persistence of pathogen in frass amended soil

Experimental design

This study aimed to understand the effect of waste processing with BSFL on the fate and persistence of bacteria pathogens significant to the Australian livestock and finfish industry (KPI 2.14, 3.15 and 4.11). This was achieved by comparing pathogen abundance (16S r RNA sequencing normalised with ddPCR) in different waste streams before (substrate) and after (frass) processing with BSFL. Three common waste streams were chosen as treatment substrates for comparison 1) plant-based waste, 2) fish waste, and 3) livestock manure. The composition of each substrate is outlined in Table 12. Bacteria pathogens of interest are outlined in Table 13.

BSFL rearing

Black soldier fly (*Hermetia illucens*) larvae (BSFL) were reared in three replicate plastic containers for each treatment substrate. Containers were seeded with 5-day old larvae at a rate of 1.5 larvae per gram of substrate. Larvae were kept at a constant 28˚C and 70% relative humidity for the duration of the incubation process. The fish offal and plant-based treatments were harvested after 7 days. As BSFL larvae have slower growth rates on manure substrates the livestock manure treatment was harvested after 9 days to allow larvae to reach the same life stage as the other treatments. Frass was separated from the larvae by passing each container through a 5 mm sieve.

Sample collection and DNA extraction

Substrate samples were collected just prior to larvae seeding and frass samples were collected immediately after separation from BSFL. Samples were stored in sterile Whirlpak bags at -80ºC until DNA extraction. DNA was extracted from 0.1 to 0.25 grams of sample using a QIAGEN DNeasy PowerSoil Pro Kit following the manufacturer's instructions.

Bacterial pathogen concentrations

Pathogen concentrations were inferred by taking the product of bacteria taxa relative abundance (assessed by 16S rRNA gene amplicon sequencing) and bacterial gene copy numbers (measured by digital droplet PCR). The relative abundances of bacterial taxa in each sample were measured using PCR amplification and next generation sequencing (NGS). The V3-V4 region of the 16S rRNA gene was targeted with the primer set 341F (5-CCTAYGGGRBGCASCAG-3) and 806R (5-GGACTACNNGGGTATCTAAT-3) (Yu *et al.*, 2005). PCR amplification and NGS was conducted by the Australian Genome Research Facility (AGRF) using the Illumina MiSeq platform (Illumina, San Diego, CA). All raw gene amplicons were processed using DADA2 with recommended settings (Callahan *et al.*, 2016). Taxonomic classification of sequence variants were classified based on SILVA version 132 (Quast *et al.*, 2013). Reads identified as chloroplast, mitochondria or Archaea were removed. For genera of interest, species level taxonomic assignment was confirmed using the NCBI BLAST tool (https://blast.ncbi.nlm.nih.gov/Blast.cgi).

16S rRNA gene copy numbers for each sample were determined using modified 515F (5-GTGYCAGCMGCCGCGGTAA-3; Parada *et al.*, 2016) and 806R (5'-GGACTACNVGGGTWTCTAAT-3; Apprill *et al.*, 2015) primers with a BioRad QX200 digital droplet PCR (ddPCR) system (Bio-Rad Laboratories, CA, USA). Gene copies were normalised per gram of sample used for DNA extraction.

Inferred concentrations of pathogens were calculated using the following equation:

$$IC\ (16S\ rRNA\ gene\ copies/g\ sample) = RA\ (\%)\ \times TB\ (16S\ rRNA\ gene\ copies/g\ sample)$$

where IC is the inferred concentration, RA is the relative abundance, and TB is the total bacterial concentration. Differences in the total bacteria load and inferred pathogen abundance (when pathogens were detected in both the substrate and frass) between substrate and frass sample types were determined using a student's t-test for each treatment (α=0.05). Data were log+1 transformed where necessary to meet the assumptions of normality and homogeneity of variances.

Table 12. Bacteria pathogens associated with the products and/or waste of fish, dairy, pork, and poultry industries (Birchall *et al.*, 2008; Chinivasagam, 2019; Novoslavskij *et al.*, 2016; Runge *et al.*, 2007; Sobsey *et al.*, 2011).

Pathogen		Fish	Dairy	Pork	Poultry
Brachyspira	*Pilosicoli*				✓
Campylobacter	*spp.*	✓	✓	✓	✓
Clostridium	*spp.*	✓			✓
Erysipelothrix	*Rhusiopathiae*	✓		✓	✓
Escherichia	*Coli*	✓	✓	✓	✓
Lawsonia	*Intracellularis*			✓	
Leptospira	*spp.*		✓	✓	
Listeria	*spp.*	✓	✓	✓	✓
Mycobacterium	*spp.*	✓	✓		✓
Pasteurella	*Multocida*				✓
Salmonella	*spp.*	✓	✓	✓	✓
Serpulina	*hyodysenteriae / pilosicoli*			✓	
Staphylococcus	*Aureus*	✓			✓
Vibrio	*spp.*	✓			
Yersinia	*spp.*	✓		✓	✓

Table 13. Percent composition of the three treatment substrates (fish waste, plant-based, livestock manure).

Waste type	Ingredient	Fish waste	Plant-based	Livestock manure
Animal	Barramundi frames	2.5%		
	Red Emperor Snapper frames	8.8%		
	Bar Cheek Coral Trout frames	7.5%		
	Spangled Emperor frames	6.3%		
	Rendered stick water waste			1.4%
	Poultry (layers) carcass			1.4%
	Poultry (layers) protein			1.4%
	Poultry (layers) egg waste			1.4%
	Swine - Carcass			1.1%
Plant	Apple	11.3%	15.1%	
	Carrot	6.6%	8.8%	
	Whiskey Mash	9.4%	12.6%	
	Rye	11.3%	15.1%	
	Ground Malt	13.2%	17.6%	
	Non-ground Malt	17.0%	22.6%	
	Bran	6.1%	8.2%	
Manure	Poultry (meat) manure on straw + wood chip			48.8%
	Poultry (layers) manure			20.9%
	Swine manure separated solids			9.4%
	Swine manure on straw			14.3%
	Total	100.0%	100.0%	100.0%

Results

Total bacteria loads were, on average, greater in BSFL frass compared to waste substrates however this was only significant for plant-based waste (p<0.01, Figure 13). Of the list of pathogens (Table 12) only *Campylobacter*, *Clostridium*, *Erysipelothrix, Escherichia, and Staphylococcus* were detected in samples (Table 14).

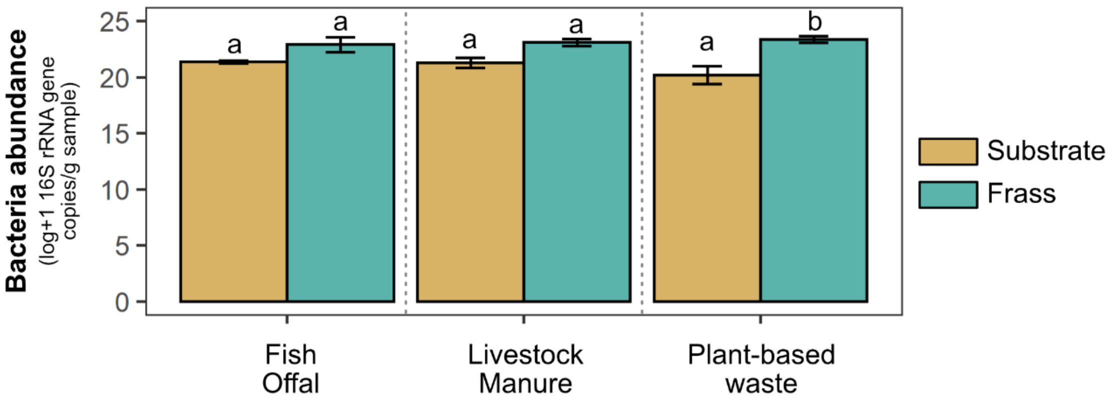


Figure 13. Total bacteria load in substrate and BSFL frass derived from fish offal, livestock manure and plant-based waste (n=3). Lowercase letters indicate significant differences.

Table 14. Mean (±SE) inferred abundance of bacterial pathogens in substrate and BSFL frass derived from fish offal, livestock manure and plant-based waste (n=3).

Group	Treatment:	Fish Offal		Livestock Manure		Plant-based	
	Sample type:	Substrate	Frass	Substrate	Frass	Substrate	Frass
Campylobacter	Mean	**-**	**1.1E+06**	**-**	**6.8E+06**	**-**	**-**
	SE	-	1.1E+06	-	2.5E+06	-	-
Clostridium	Mean	**3.5E+05**	**6.0E+06**	**2.4E+07**	**4.1E+07**	**1.5E+04**	**-**
	SE	2.2E+05	2.2E+06	1.0E+07	1.4E+07	1.5E+04	-
Erysipelothrix	Mean	**-**	**2.3E+08**	**1.2E+06**	**1.6E+07**	**-**	**-**
	SE	-	6.7E+07	7.4E+05	5.4E+06	-	-
Escherichia	Mean	**-**	**9.6E+05**	**3.4E+06**	**-**	**-**	**8.1E+07**
	SE	-	8.6E+05	1.7E+06	-	-	2.6E+07
Staphylococcus	Mean	**7.1E+04**	**1.9E+06**	**2.0E+07**	**6.7E+06**	**-**	**2.6E+07**
	SE	7.1E+04	1.4E+06	1.0E+07	2.0E+06	-	1.1E+07

Clostridium and *Staphylococcus* both had greater abundance in the fish offal frass samples compared to the substrate. However, this increase in abundance was not statistically significant for either group (Table 14, Figure 14). *Campylobacter*, *Erysipelothrix* and *Escherichia* were detected in the frass but not the substrate (Table 14, Figure 14). *Clostridium, Staphylococcus* and *Erysipelothrix* were detected both the manure substrate and frass (Table 14, Figure 14). The abundance of *Erysipelothrix* and *Staphylococcus* increased from substrate to the frass, however, this effect was only significant for *Erysipelothrix* which was 1343% greater in the frass compared to the substrate ($p<0.05$, Table 14, 15). In contrast, *Staphylococcus* abundance decreased by 2740% from the substrate to the frass but this was not statistically significant (Table 15). *Escherichia* was detected in the substrate but not the frass and *Campylobacter* was detected in the frass but not the substrate (Table 14, Figure 14). Plant-based waste had the least number of pathogen groups detected. *Clostridium* was detected in the substrate but not the frass while *Escherichia* and *Staphylococcus* were detected in the frass but not the substrate (Table 14, Figure 14).

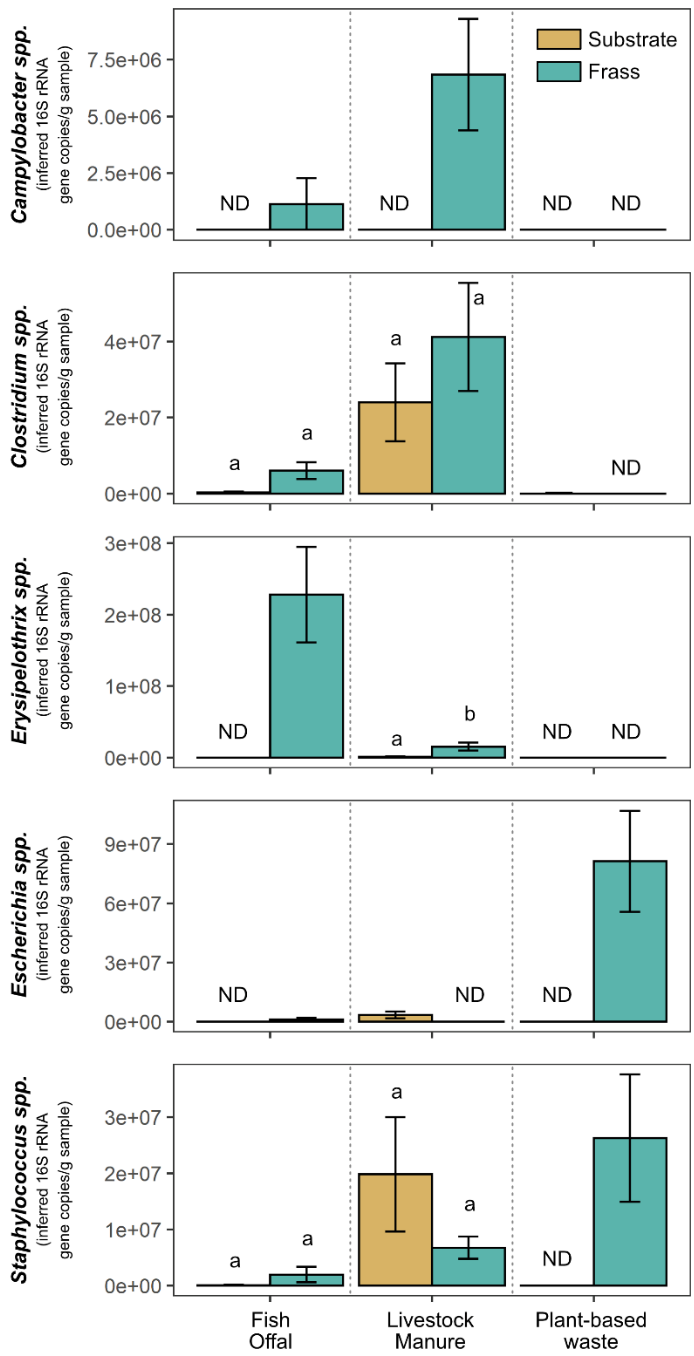


Figure 14. Inferred abundance of pathogen genera detected in substrate and BSFL frass derived from fish offal, livestock manure and plant-based waste (n=3). Lowercase letters represent significant differences. ND = not detected.

Table 15. Percent change in total bacteria load and inferred pathogen abundance from processing waste substrates with BSFL. Waste substrates were derived from fish offal, livestock manure and plant-based waste (n=3). Percent change is calculated as the change in abundance/inferred abundance between the waste substrate and BSFL frass. '*' = change in abundance was statistically significant; '+' = introduced (present in frass but not substrate); '–' = present in substrate but not frass; ND = not detected in substrate or frass.

	Fish Offal	Livestock manure	Plant-based
Total bacteria load	**+652%**	**+569%**	**+1365%**
Campylobacter spp.	**+**	**+**	ND
Clostridium spp.	**+1737%**	**+172%**	–
Erysipelothrix spp.	**+**	**+1343%**	ND
Escherichia	**+**	**–**	**+**
Staphylococcus	**-2704%**	**+295%**	**+**

Key findings

- Bacterial pathogen detection and abundance depended on the substrate waste stream.
- For all waste streams the total bacterial load of frass was greater than the waste substrate.
- Plant-based waste had the smallest number of pathogen groups detected.
- Most pathogens either increased in abundance after processing with BSFL or were 'introduced' by processing with BSFL. It is unclear if the introduction of pathogens to the frass is from the BSFL microbiome (e.g. gut) or from the environment. However, *Campylobacter* has been reported as comprising a large proportion of the BSFL gut microbiome when fed swine and manure, so their presence in the frass was likely derived from the BSFL gut (Ao *et al.*, 2021; Wu *et al.*, 2021). We also observed the highest abundance of *Camplylobacter* in the frass derived from livestock manure.
- Some pathogens can be reduced (fish offal substrate – *Staphylococcus* 2704% decrease) or eliminated (Livestock manure – *Escherichia* elimination; plant-based waste *Clostridium* elimination) by processing waste with BSFL. This supports previous work which shows BSFL can reduce Escherichia and *Salmonella* in livestock manure (Erickson *et al.*, 2004)**.**
- Longer processing times to allow inactivation of pathogens by BSFL (especially for livestock manure) or post-processing of BSFL frass, such as composting or drying, could be used to eliminate/reduce pathogen loads.

Conclusion

In conclusion, waste processing with BSFL has variable effects on the fate and persistence of bacteria pathogens significant to the Australian livestock and finfish industry. It is promising that some pathogens can be reduced or eliminated by processing waste with BSFL. However, as some pathogen groups were enriched or introduced by BSFL processing, further treatment (e.g., drying, composting, pelletisation) or longer processing times may be required to produce frass that is safe for use as a soil conditioner. Especially for frass derived from livestock manure or finfish waste.

5.3(f) Report findings as a scientific publication or technical report to stakeholders and government regulatory bodies so that BSF products meet the requirements to avoid classification as waste by-product.

Activities 5.3 (f) and 5.4(g) have been integrated into one technical report and a factsheet.

An information fact sheet on classification and regulation requirements for manure derived BSF frass as a product was produced (KPI 4.12). A technical report to stake stakeholders and government regulatory bodies so that BSF products meet the requirements to avoid classification as waste by-product has been produced (KPI 4.16; Appendix E).

Summary of report

Livestock industries in Australia produce significant amounts of manure that needs to be managed in a sustainable manner from an environmental and economic perspective. Manure management on Australian farms is a significant cost (estimated AUD$100-200 million annually) impacting on the productivity, profitability and sustainability of businesses. Offsetting the high cost of synthetic fertilisers with organic manure derived sources would also benefit the productivity and profitability of Australian agricultural production systems. Major constraints and barriers to solid manure use is the cost of transport and management of the materials (labour, storage, handling and reuse). Poor management of manures can also cause significant impact on the amenity and environment of the surrounding community. A potential waste management solution is the use of Black soldier fly(BSF) farming to convert agricultural manures and waste into high quality fertilizer whilst significantly reducing the volumes of manure to be handled and transported. Black soldier fly can be fed manures and spent bedding (which can include carcasses) and produce a product called frass which is a mix of insect excrement, food residue and exoskeletons. Research conducted as part of "Closing the loop: Black Soldier Fly technology to convert agricultural waste" in WA and QLD on poultry, dairy, piggery and abattoir wastes has produced the following results:

•Reduction in manure volumes of 70-80%

•Fertiliser/soil conditioner

•Demonstrated low risk of some pathogens

•Reduced stable fly emergence

•Reduced greenhouse gases (GHG) and nutrient leaching

•Produces a stable product with reduced odour

•Can be granulated to reduce transportation cost.

Using BSF technology to turn manures and waste products into highly valuable products that can be recycled back into the production on the farm, provides the potential for a closed loop approach that could increase productivity and profitability, reduce waste, labour and transport costs, improve environmental impacts, and contribute to enhancing the sustainability of agriculture in Australia. However, although the BSF technology exists and the products developed show promising results, it is necessary to understand how the regulatory framework in each State would classify and or view the products for reuse. This information is essential in understanding the potential adoption and reuse of BSF products for both regulators and industry. The objectives of this review were to liaise with regulatory bodies to identify potential waste classification of manure derived frass and identify environmental concerns, opportunities and barriers to the use of manure derived frass. The review focused on the reuse of the application of the manure derived products only. It did not focus on the licencing or permission requirements of the BSF processing facility or other feedstocks such as abattoir wastes (paunch etc) or clean substrates (FOGO, green wastes etc).

In order to understand the initial thoughts of the regulators in regard to the potential waste classification and opportunities/barriers associated with the use of manure derived frass products from BSF, discussions were undertaken with Luke Wheat from Future Green Solutions to gain a better understanding of the Black soldier fly process, feedstocks and products.

Communication was then initiated with the State regulators from Western Australia, South Australia, New South Wales, Queensland, Tasmania and Victoria. Discussions were held either via a teleconference or zoom/teams meeting with individuals/teams from each of the regulators who were generally from waste policy or permissions teams.

Discussions were based on generalised processing and product information due to confidentiality. This has implications on the limitations of the regulator's responses (not definitive) and therefore interpretation of policies/Acts are subject to change based on more detailed processing and product information being provided.(such as justification of fit for purpose status). Based on the provided information and initial regulator responses their appears to be no major regulatory barriers to the reuse and adoption of manure derived frass as a fertiliser and/or soil conditioner (Table 16).

Table 16. A summary of Australian State regulation in regards to manures and manure derived products

Product/State	Western Australia*	South Australia	New South Wales	Queensland	Tasmania	Victoria
Raw Manures	61A Solid waste >1000T/year need a licence to apply offsite or onsite (if not already a prescribed premise.	No restrictions- must meet general provisions of the *Act***	Resource recovery orders and exemptions (manures)- conditions must be met. General provisions of *Act***	Non regulated/general waste- apply to land no restrictions- general provisions of *Act***	No restrictions- must meet general provisions of the *Act****	Livestock Manure and effluent determination conditions must be met if over 20m^3 per month received/applied General provisions/GED of *Act***
BSF Manure derived products	Frass likely viewed as a product not a waste (Interim factsheet criteria)	Frass likely to be viewed as a product under Environment Protection Waste to Resources Policy 2010- demonstrate fit for purpose	Frass likely considered manure- Use as per Resource recovery orders and exemptions (manures)	Non regulated/general waste- apply to land no restrictions- general provisions of *Act***	Frass may not meet the definition of a waste under the EMPC Act and likely a product	Frass may be considered as manure-use as per Livestock manure determination if over 20m^3 per month received. General provisions/GED of *Act***

**Regulatory Reform in place*
***General provision means to not cause any environmental harm or nuisance under the relevant Act*
****Need confirmation*

In WA, SA and Tasmania there is potential under their respective policies and Acts for manure derived frass to be classified as a product rather than a waste. It can therefore be applied to land in a manner that meets the general provisions of the Acts i.e. does not cause environmental harm/nuisance.

In the remaining jurisdictions, frass may still be classified as manure. In NSW and Victoria reuse of manure is allowed under the resource recovery order/exemption and livestock manure and effluent determination respectively. This allows the application to land with conditions which basically specify that it must be used as a fertiliser/soil conditioner in a manner that does not cause environmental harm/nuisance i.e. general provisions/general environmental duty. In QLD will likely be classified as a non-regulated/general waste which means it can be applied to land with no restrictions, however as per all other jurisdictions it must not cause environmental harm/nuisance.

Regulators were generally very supportive and encouraged innovation in the waste management space. They were encouraged by the results of the "Closing the loop: Black Soldier Fly technology to convert agricultural waste" especially the reduction of waste volumes and were interested in learning more about the process and potential for BSF farming as a waste management option in Australia.

Open communication and transparency needs to be provided to the regulators and decision makers in order to facilitate and expedite the adoption of BSF products in Australia. Confidentiality can be maintained in the public area, however maintaining confidentiality and providing partial information will only create uncertainty, confusion and the likely mis classification or application of policy, guidelines and standards that are not commensurate with the level of risk of the product. Extension of the process, product and benefits to the Ag departments and industry will also likely create interest in the adoption of the product.

To facilitate a greater understanding of the process and the end product, further information should be collated on the potential process of BSF farming, feedstocks to be used, end product specifications, and data on product benefits (environmental, human and animal). To demonstrate general environmental duty and fit for purpose status information should be collated on Best Practice recommendations for reuse, duty of care statements developed as well as economic, agronomic and environmental analysis of BSF Frass compared to conventional fertilisers and raw manures.

3.1.8 Research Activity 5.4 - Assess the environmental risks of Black soldier fly products (frass and larvae)

5.4(a) Provide a literature review on the environmental risks with BSF products

A review on of the Potential environmental risk of Black Soldier Fly (Hermetia illucens) By - Products (KPI 1.16, 2.15, 3.17,4.13) has been produced (Appendix F).

Summary of report

The objective of the review was to explore the current literature about BSF and its potential impact in the environment and included bioaccumulation of heavy metals in the larvae and potential release of contaminants to the environment. This review also investigated the potential environmental impact of applying frass to land in terms of release of potentially toxic or harmful compounds for both humans and the environment. Overall, there are only a few studies that have reported the environmental impact of applying BSFL frass as soil improver or crop fertiliser. Environmental impacts of BSF was largely dependent on the substrate used to be fed to the insects. For example, chicken and cattle manure had a lower heavy metal content and leaching potential compared with other manure sources.

Heavy metals were identified as a particular risk associated with applying frass or insect products to land and the concentrations varied between initial waste substrates and batches of same substrate. Metals and metalloids include lead (Pb), arsenic (As), mercury (Hg), cadmium (Cd), zinc (Zn), silver (Ag), copper (Cu), iron (Fe), chrome (Cr), nickel (Ni), palladium (Pd) and platinum (Pt) are not biodegradable and tend to bioaccumulate. Unlike organic pollutants, which normally degrade into carbon dioxide and water, heavy metals can persist in the environment for a long period of time, so they have been regarded as a serious environmental concern. They cause several health conditions in humans, including cancer. The presence of heavy metals in the soil can seriously affect the edaphic fauna, causing a decrease in the biodiversity decreasing the number of sensitive organisms, selecting those that are naturally resistant to heavy metals. Hence their content in the environment must be checked and followed, and maximum limits established against standards and guidelines.

The maximum limit of heavy metals in compost for unrestricted use in Australia can be found in Table 1. The quantity of heavy metals found in the different BSF substrates inputs and by-products (frass and larvae) can vary. Studies have shown that frass derived from poultry and piggery manures can have high concentrations of copper and zinc. Other studies have shown that although heavy metals can accumulate in the BSF larvae they do not significantly alter larval growth rate. While other studies have reported that high concentrations of Zn, Cu, Cr, Cd, Pb, Ni and Hg reduced larval weight gain and the survival rate of the larvae. The accumulation of heavy metals in BSF is dependent of the growth stage and the type of feedstock.

Table 17. Unrestricted use upper limits for heavy metal contaminants. Source: Standard Australia (Standard, 2003).

Contaminant	Unrestricted use upper limits in mg kg^{-1}
Arsenic (As)	20
Cadmium (Cd)	1
Boron (B)	100
Chromium (Cr)	100
Cooper (Cu)	150
Lead (Pb)	150
Mercury (Hg)	1
Nickel (Ni)	60
Selenium (Se)	5
Zinc (Zn)	300

5.4(b) Liaise with government regulatory bodies to ensure research addresses environmental concerns

Activities 5.3 (b) and 5.4(b) have been integrated (KPI 1.17).

A summary of the communication is provided under Activity 5.3 b.

5.4(c) Determine the heavy metal and nutrient content at the manure input, larval and frass stages of production

A technical report on Pathogen and Heavy Metal Loading at the Manure Input, Larval and Frass Stages of Black Soldier Fly Production (KPI 1.18, 2.16, 3.18) was produced (Appendix D).

Summary of report

Samples were collected at different stages of BSF production and included the different manure inputs and their larval and frass outputs (co-products). Samples were submitted for heavy metal analysis using ICP standard methods. A trend of increasing heavy metal concentration was observed in all manure derived BSF outputs compared to manure inputs (Tables 18-20). When

BSF neonates were feed piggery manure (Table 18) the resulting frass and larval had copper and zinc levels above the Standard Australian AS44 54 recommendation (150 mg/kg and 300 mg/kg, respectively). Therefore, BSF frass and larval co-products derived from piggery manure may need to be diluted or mixed with another product prior to land application.

Table 18. Average heavy metal concentrations (mg/kg) during BSF treatment of piggery manure including input BSF neonates, input manure (feed stock), output larvae and output frass.

METAL	LOR mg/ml	INPUT NEONATES	INPUT MANURE	OUTPUT LARVAE	OUTPUT FRASS
Arsenic	5	0	0	13 (1)	0
Cadmium	0.1	0	0	0.3 (2)	0
Chromium	1	0	0	22 (2)	7 (3)
Copper	1	5 (4)	47 (3)	72 (3)	*167 (3)
Lead	1	0	0	2 (1)	0
Mercury	0.02	0	0	0	0
Nickel	1	0	0	0	3 (3)
Zinc	1	79 (4)	263 (3)	**600 (3)	*1133 (3)

LOR: number of batches above the limit of recording

Overall, when BSF were feed chicken manure mix the resulting frass and larvae had lower heavy metal concentrations that the piggery manure derived BSF co-products. Although, the poultry frass exceeded the Australian standard it was only just over the limit (Table 19).

Table 19. Average heavy metal concentrations (mg/kg) during BSF treatment of poultry manure including input BSF neonates, input manure (feed stock), output larvae and output frass.

METAL	LOR mg/ml	INPUT NEONATES	INPUT MANURE	OUTPUT LARVAE	OUTPUT FRASS
Arsenic	5	0	0	0	0
Cadmium	0.1	0	0.1 (2)	0.2 (2)	0
Chromium	1	0	2.3 (2)	0.5 (1)	2.5 (2)
Copper	1	5 (4)	33 (2)	16 (2)	34 (2)
Lead	1	0	0	0	0
Mercury	0.02	0	0.03 (2)	0	0
Nickel	1	0	2.6 (2)	0	2.8 (2)
Zinc	1	79 (4)	200 (2)	325 (2)	*355 (2)

Frass and larval co-products derived from dairy manure (Table 20) had the lowest heavy metal content and all metals were within standards for unlimited use as a compost and fertiliser. Larvae were also within NSW stock feed regulation requirements for cadmium, chromium and mercury. Therefore, land applications of BSF products derived from dairy manure show the least risk of heavy metal contamination.

Table 20. Average heavy metal concentrations (mg/kg) during BSF treatment of dairy manure including input BSF neonates, input manure (feed stock), output larvae and output frass.

METAL	LOR mg/ml	INPUT NEONATES	INPUT MANURE	OUTPUT LARVAE	OUTPUT FRASS
Arsenic	5	0	240 (1)	0	0
Cadmium	0.1	0	0.2 (2)	0.8 (2)	0
Chromium	1	0	6.3 (2)	1.6 (1)	0
Copper	1	5 (4)	25 (2)	32 (2)	3 (2)
Lead	1	0	2.2 (1)	0	0.7 (1)
Mercury	0.02	0	1.4 (1)	0	0
Nickel	1	0	1.6 (2)	2.3 (2)	0
Zinc	1	79 (4)	125 (2)	250 (2)	20 (2)

Potential zinc and copper contamination will need to be mitigated when using BSF products derived from piggery and chicken manures feed stocks.

5.4(d) Conduct pot trials to determine the nutrient and heavy metal leaching potential of different frass products applied to a range of soil types

A technical report on nutrient and heavy metal leaching potential of different frass products applied to a range of soil types (KPI 1.19, 2.17, 3.19, 4.14) was produced (Appendix G).

Summary of report

The leaching potential of nitrogen (N) and phosphorus (P) from granulated BSF frass products was benchmarked against mineral fertiliser and raw manures in two soil types: (i) sandy and (ii) sandy loam. The results should be viewed as the maximum leaching potential as no plants were used in the experiment: if plants were present their roots would have intercepted some nutrients and thereby reduced leaching. Results were also impacted by differing initial nutrient concentrations among treatments. There was a significant interaction of soil type and substrate and an effect of rate (Table 21). Nutrient leaching was generally greatest in the sandy soil. The leaching potential of NO_3-N from BSF frass did not differ from that of mineral fertiliser. Among BSF products, NH_4-N leaching was low. Notably, the granulation process increased NH_4-N if a binder was not applied; as may constitute an environmental risk. The leaching of P was a slower process than for N but was greatest for BSF products, particularly granulated types. For leaching of PO4-P there was a 3-way interaction between soil, amendment and rate (Table 21) which reflected minimal leaching in the sandy loam soil, but relatively high leaching in the sandy soil where there was also a large effect of application rate.

Major outcomes

- Soil type, type of organic amendment and application rate all affected leaching of nutrients.
- For sandy soil, the leaching of NH4-N was the greatest when amended with mineral fertilisers (but this may reflect an initially higher soil N concentration) and granulated BSF manures frass, particularly when no binder was included.
- For sandy loam soil, the leaching of NH4-N was negligible.
- For both soil types, the leaching of NO3-N did not differ among amendments.
- For sandy soils the leaching of P was greater for all BSF frass products (at high application rates) than mineral fertiliser but this likely reflects higher initial P concentrations in these products.
- Further work is merited on the leaching risk from all BSF manures products including the impact of soil type and the potential for binders to reduce leaching.

Table 21. Effect of soil type, soil amendment and application rate (see Table 2) on the total amount leached of NH4-N, NO3-N and PO4-P, and total leachate volume, over 84 days. Fisher's F value (F) as a measure of the strength of the factors in the model and significance at 95% confidence of the factor and interactions.

Factor		NH_4 - N leached (mg column $^{-1}$)		NO_3 - N leached (mg column $^{-1}$)		PO_4 - P leached (mg column $^{-1}$)		Leachate volume (L)	
		F	Pr > F	F	Pr > F	F	Pr > F	F	Pr > F
Soil type (ST)		115.91	**<0.0001**	6.43	**0.014**	1156.41	**<0.0001**	368.73	**<0.0001**
Substrate (S)		250.00	**<0.0001**	11.31	**<0.0001**	155.69	**<0.0001**	8.12	**<0.0001**
Application rate (AR)		287.91	**<0.0001**	20.48	**<0.0001**	931.12	**<0.0001**	Not significant	
ST*S		185.74	**<0.0001**	3.59	**0.011**	154.23	**<0.0001**	13.42	**<0.0001**
S*AR		103.88	**<0.0001**	Not significant		92.59	**<0.0001**	4.20	**0.004**
ST*S*AR		67.33	**<0.0001**	Not significant		92.29	**<0.0001**	Not significant	
Model	R^2	0.98		0.73		0.99		0.90	
	F	159.95		7.92		281.18		26.18	
	Pr > F	**<0.0001**		**<0.0001**		**<0.0001**		**<0.0001**	
	Fisher's LSD	0.465		5.285		0.663		0.342	

5.4(e) Conduct pot trials to determine the Greenhouse Gas potential of different frass products applied to soil

A draft scientific publication has been produced on the laboratory trials to determine the greenhouse gas potential of frass amended soil (KPI 4.15) produced from agricultural waste using Black Solider Fly technology (Appendix H).

Summary

Increasing world population and wealth have increased demand for food to feed the growing population leading to mass production of livestock and generation of large quantities of manure. Soil amended with manure has many benefits including increased nutrient supply, enhanced soil structure and improved crop yield. However, there are increasing concerns in terms of the environmental risks associated with applying manure to land especially greenhouse gas (GHG) emissions. Black Soldier Fly larvae (BSFL; *Hermetia illucens*) farming is an emerging technology that converts manure into protein for animal feed. Fly casting or frass, a major waste by-product of BSF farming, has been shown to have potential as a soil amendment. Nevertheless, the impact of amending soil with frass on GHG is not well understood and could limit its potential use due to strict regulations. **Activity 5.4(e)** aimed to assess the impact of processing manure with BSF technology on GHG emissions from soil (KPI 1.20, 2.18). Specifically, GHG emissions from soil applied with BSF frass derived from fresh poultry manure and unprocessed manure were compared. It was hypothesized that soil amended with BSF frass will emit cumulatively less N_2O, CO_2 and CH_4 compared to fresh poultry manure due to less availability of labile organic carbon, and, in the case of N_2O, inorganic nitrogen.

To investigate the impact applying BSF frass as a fertilizer for soil amendment on GHG emission compared to fresh poultry manure or chemical fertilizer, a laboratory microcosm experiment was conducted. The experiment consisted of four different treatments as follow: (1) soil amended with BSF frass (Frass), (2) soil amended with fresh poultry manure (Manure), (3) soil amended with urea (Urea) and (4) un-amended soil with no amendment supplement used as control (Control). All treatments except for the unamended control received a constant nitrogen rate of 100 kg N ha^{-1} and were replicated five times, resulting in 160 soil samples in total for analysis. The microcosm vials were placed inside unsealed 523 mL glass jars and incubated in the dark at 25 ºC for 2 weeks. The glass jars were unsealed, except during gas flux measurements when they were sealed with an air-tight lid fitted with a septum to trap expired gases for 2 hours. N_2O, CO_2 and CH_4 gas fluxes were analysed at 0, 2, 6, 10, 24, 72, 144 and 366 hours by gas chromatography. In addition, soil ammonium (N-NH_4^+), nitrate (N-NO_3^-) and dissolved organic carbon (DOC) concentrations were measured.

Key findings

- Gas flux was greatest for CO_2 throughout the experiment ranging from 0.001 to 66.251 kg/ha/day. In contrast, N_2O (0.001–0.489 kg/ha/day) and CH_4 (-0.002-0.002 kg/ha/day) emissions were much lower.
- Soil amendment had no impact on CH_4 emissions and there was no difference in CH4 emissions over time (Table 22, Figure 15).
- Cumulative N_2O and CO_2 emissions increased over time (Figure 15).
- Soil amended with unprocessed poultry manure had the highest cumulative GHG fluxes followed by BSFL frass derived from poultry manure, urea (inorganic fertiliser) and the control treatment.
- Soil amended with BSFL frass derived from poultry manure emitted cumulatively less N_2O and CO_2 compared to unprocessed poultry manure. This effect is likely due to less labile organic carbon (DOC; Figure 16) and mineral nitrogen (DOC; Figure 16) in frass treatment the soil, thus less nitrogen or carbon available to soil microorganisms. Previous studies have shown that processing livestock manure with BSFL can decrease available nitrogen and phosphorus content was decreased by 30-80 % and 44-75 %, respectively, depending upon the manure type. Consequently, BSF frass provides a more balanced nutrient content and profile compared to the fresh manure potentially making it a suitable fertilizer.
- The outcome of this investigation shows that BSFL frass could be utilized as a high-quality fertilizer whilst mitigating GHG emissions. This is a significant result as manure storage an application has been identified as a major source of GHG (66%) across the supply chain for Australian livestock farms.

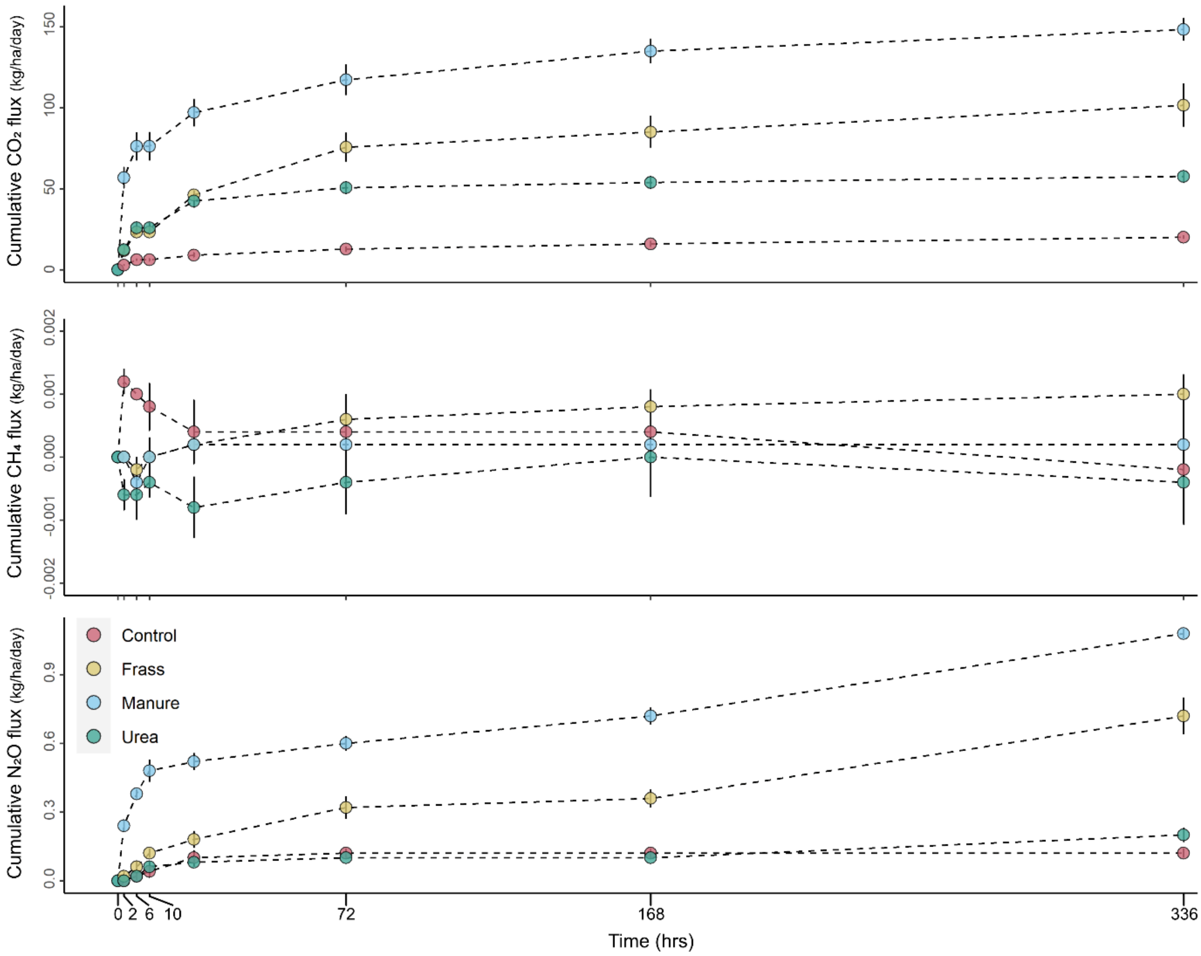


Figure 15. Mean (n=5) cumulative flux of CO2, CH_4 and N2O emissions over 336 hours following soil amendment with poultry manure derived BSFL frass, poultry manure, urea or an un-amended control. Error bars represent standard error.

Table 22. Mean (n=5) cumulative gas flux (kg/ha/day) of CO_2, CH_4 and N_2O following soil amendment with poultry derived BSFL frass (frass), raw poultry manure (manure) or urea over 336 hours. Ns = not significant. Significant post-hoc comparisons among treatments are indicated by lowercase letters.

GHG	Control	Frass	Manure	Urea	*p-value*
CO_2	20.14^{a}	101.44^{c}	148.21^{d}	57.56^{b}	<0.001
CH_4	-0.00	0.00	0.000	-0.00	NS
N_2O	0.11^{a}	0.74^{b}	1.08^{c}	0.20^{a}	<0.001

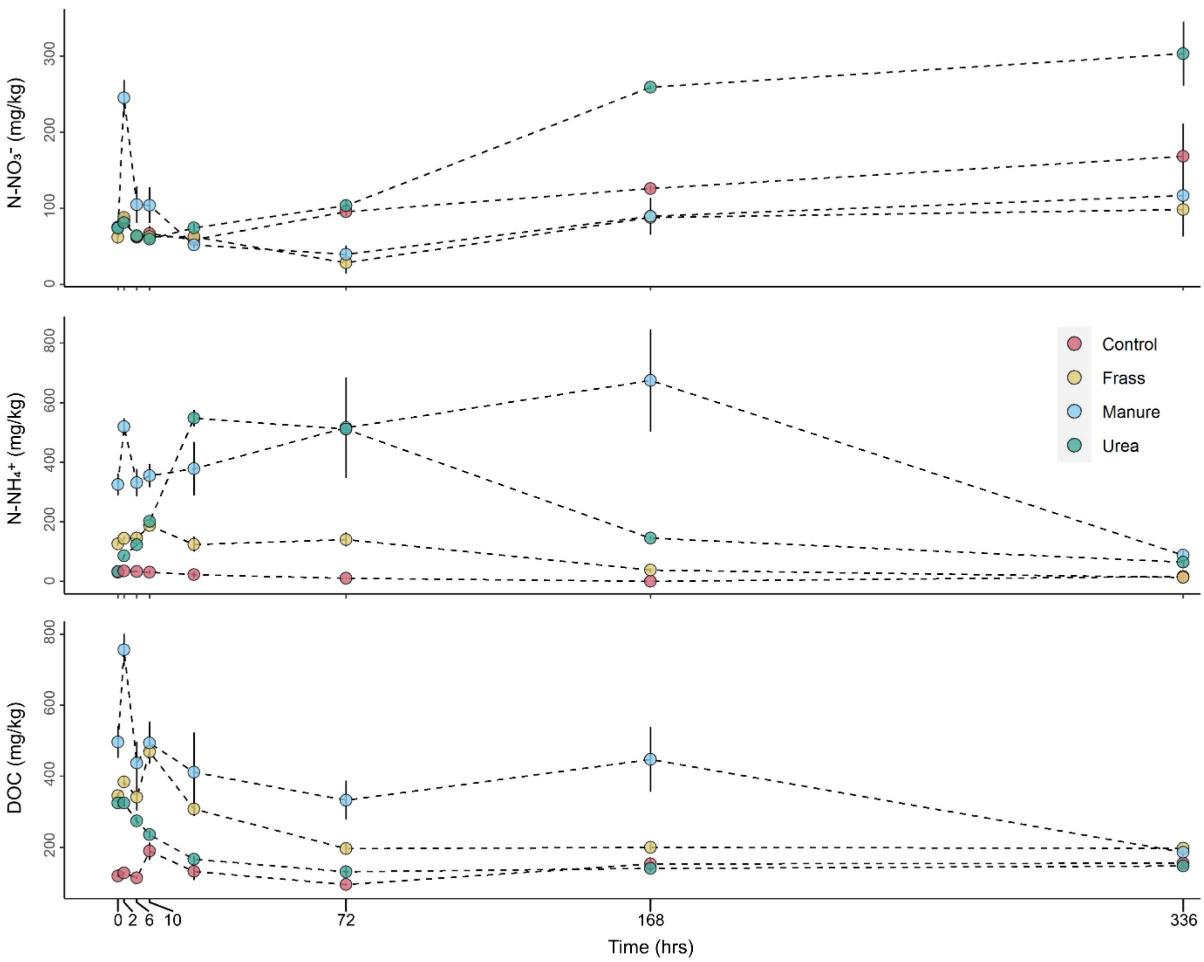


Figure 16. Mean (n=5) concentration of N-NH_4^+, N-NO_3^- and DOC over 336 hours following soil amendment with poultry manure derived BSFL frass, poultry manure, urea or an un-amended control. Error bars represent standard error.

Conclusion and significance

Our results suggest that the emissions of N_2O and CO_2 are decreased when soils are amended with frass. Therefore, BSFL farming is a potential strategy for mitigating GHG emissions while decreasing the volume of waste by up to 50% or more and producing a higher value nutritive fertiliser product. However, further field scale trials and a full life cycle analysis are required to determine GHG emissions across the entire BSFL production cycle compared to conventional manure stockpiling and spreading.

This information may be used to inform and encourage uptake of BSFL farm as a waste management strategy across the Wheatbelt region of Western Australia which could result in 1) in increased profits due to more efficient waste management 2) generation of alternative and sustainable fertilisers 3) decreased carbon footprint, and 4) adoption of additional sustainable agricultural practices. Furthermore, the present study suggests that BSFL farming could be an effective mitigation strategy at a national scale to decrease GHG emissions from soils amended with manure. Other benefits include less land required for waste disposal reduced pests and pathogens (**Activity 5.3(d)**) and decreased cost for manure disposal (around $8 per cubic meter). These are particularly relevant to the West Australian poultry industry which has strict regulations imposed on manure application. Ultimately, Australian livestock industries have the potential to be more competitive and sustainable if cost-effective mitigation strategies like the use of BSFL frass can be used without impacting on productivity and profitability.

5.4(f) Conduct odour perception surveys

Odour perception surveys were conducted as part of Activity 5.2 (with waste producers) and Activity 5.9 (Potential frass end-users). In both surveys, respondents were asked if they thought '*Organic fertilisers or soil improvers produced from livestock wastes using Black Soldier Flies would have an unpleasant odour'* and whether the thought that '*Producing organic fertilisers or soil improvers from livestock wastes using Black Soldier Flies is disgusting'.* Answer options were 'Disagree', 'Somewhat disagree', 'Neutral/Don't know', 'Somewhat agree', 'Agree'.

Due to challenges reaching survey completes in Activity 5.2, it could not be determined which responses were from legitimate Australian livestock farmers. Only three respondents answered the perception questions and their responses have been added to the results in Table 23.

The end-users' survey was completed by 112 respondents (see also Appendix). After removing incomplete responses (47), respondents who were not farmers (4), and respondents from outside Australia (13), there were 48 usable responses from across Australia. Respondents included livestock farmers, broad-acre cropping, horticulture, and viticulture farmers.

Similar to the producers' survey, the majority of respondents (52%) disagreed or somewhat disagreed with the statement that BSF products would have an unpleasant odour, while 36% did not know. Only 6 out of 50 'somewhat agreed' that BSF products would have an unpleasant odour (none agreed completely). Nearly all respondents (94%) either disagreed or somewhat disagreed with the statement that '*Producing organic fertilisers or soil improvers from livestock wastes using Black Soldier Flies is disgusting'*. Only 1 person agreed and 1 person somewhat agreed with that statement.

Table 23 Perceptions about Black Soldier Fly products and technology (number of respondents). Statements measured on a 5-point Likert scale from 1= Disagree to 5 = Agree. Source: waste producers' and end-users' surveys (n=51)

Statement	Disagree	Somewhat disagree	Neutral	Somewhat agree	Agree
Organic fertilisers or soil improvers produced from livestock wastes using Black Soldier Flies will have an unpleasant odour[a]	13	13	18	6	0
Producing organic fertilisers or soil improvers from livestock wastes using Black Soldier Flies is disgusting	43	5	1	1	1
Producing organic fertilisers or soil improvers from livestock wastes using Black Soldier Flies is a better alternative to handle agricultural waste compared to landfilling	0	4	3	5	39
Organic fertilisers or soil improvers produced from livestock wastes using Black Soldier Flies will contribute to improved environmental sustainability of the agricultural industry	1	0	10	11	29

[a] One respondent did not answer this question (n=50)

Despite the small number of respondents, these results show that there is unlikely to be a perceived nuisance from BSF products amongst agricultural producers. Respondents did not perceive organic fertilisers or soil improvers produced from livestock wastes using BSF as disgusting, and only some agreed that the product may have an unpleasant odour. Combined with the majority of respondents agreeing that '*Organic fertilisers or soil improvers produced from livestock wastes using Black Soldier Flies will contribute to improved environmental sustainability of the agricultural industry*', our results demonstrate that farmers have, in general, a positive perception of Black Soldier Fly technologies.

5.4(g) Report findings as a scientific publication or technical report to stakeholders and government regulatory bodies so that BSF products meet the requirements to avoid classification as waste by-product.

Activities 5.3 (f) and 5.4(g) have been integrated into one technical report and a factsheet.

An information fact sheet on classification and regulation requirements for manure derived BSF frass as a product was produced (KPI 4.12).

A technical report to stake stakeholders and government regulatory bodies so that BSF products meet the requirements to avoid classification as waste by-product has been produced (KPI 4.16, Appendix E) summary of the technical report is provided under Activity 5.3 f.

3.1.9 Research Activity 5.5 - Assess the benefits of using BSF frass and/or larvae as a soil improver

5.5(a) Conduct laboratory trial to determine the mode of action of frass

An information fact sheet and draft scientific publication has been produced on the laboratory trials to determine the mode of action of frass and creating a fertilizer and/or soil improver product (KPI 2.19; KPI 4.17) from agricultural waste using Black Solider Fly technology (Appendix I).

Summary

BSFL frass has potential to be used as an organic fertilizer in a circular economy. However, BSFL frass has a high ammonium content which could result in nitrogen (N) loss following its application to land via leaching, volatilization and runoff. One solution is to process the frass by combining it with another by product of BSFL farming, solid fatty acids (FA). FAs can be removed during the fat separation stages of larval protein recovery. Lauric acid, myristic acid and stearic acid are three major FAs found in BSFL. Currently, BSFL reared on manures are not allowed to be directly used in animal feed formulations according to Regulation (EC) No. 1069/2009 (European Commission, 2009) due to biosecurity risks (e.g., foodborne pathogens). However, the extracted larval FAs could potentially be used as a coating material in the production of slow-release fertilizer.

FAs have already been used successfully as a coating agent to manufacture slow-release inorganic fertilizers. Therefore, the aim of this activity was to evaluate the impact of combining frass with three FAs [lauric acid (C12), myristic acid (C14) and stearic acid (C18)] on the 'mode of action' (availability and concentration) of mineral N in soil and the ecological function of the soil bacterial community (KPI 1.22).

BSFL frass was derived from a poultry manure substrate collected from a poultry meat production farm in Gingin, Western Australia. Frass was produced by rearing BSFL under controlled conditions (aerobic, 25 ˚C) at a feeding rate of 150 mg of manure per larva per day. After 5 days the remaining organic matter was separated from the larvae with a 5mm sieve and stored at 4˚C. Due to the BSFL only being reared for 5 days the frass still contained particles of unprocessed manure.

Frass was either left untreated or combined with different FAs to form a new soil amendment product, FA-P frass. FA-P was created by grinding BSFL frass into a fine powder and then combining the powder with a lauric acid (C12), myristic acid (C14) or stearic acid (C18) solution at a ratio of 1:0.6 (frass:FA). The resulting product was then ground again and passed through a 2 mm sieve. The experimental design consisted of five soil amendments in triplicate and five sampling times over a 28-day soil incubation at 25°C. The soil amendments were (i) lauric acid processed frass (frass+C12), (ii) myristic acid processed frass (frass+C14), (iii) stearic acid processed frass (frass+C18), (iv) unprocessed frass (Frass), and (v) an unamended control. Each soil microcosm contained 40 g of topsoil sourced from Pingelly (Wheatbelt region, Western Australia) amended with either a BSFL frass product (applied at a rate of 46 kg of N ha^{-1}) or no amendment (control). The N application rate was selected to reflect district practice rate of conventional fertilizer urea (CH_4N_2O) application at 100kg of ha^{-1} (46 kg of N ha^{-1}) for pasture or grain cropping. Microcosms were destructively sampled at times 0 (T0), 1 (T1), 7 (T7), 14 (T14) and 28 (T28) days to measure soil respiration, soil chemistry and to characterise the soil microbial community.

Key findings

- The dominant form of N in cage-eggs-manure frass amended soils, regardless of fatty acid processing, occurred as N-NH_4^+ rather than N-NO_3^- (Table 24).
- Processing BSFL frass in combination with fatty acids slowed the release of N-NH4+ into soil compared to unprocessed frass (Figure 17). FA-P frass releases mineral N more slowly compared to untreated frass because hydrophobic fatty acids prevent solubilization of embedded N. However, there was no effect of fatty acid processing on N-NO3- release from frass (Figure 17).
- FAs with a shorter hydrocarbon chain length were most successful at slowing N-NH_4^+ release from BSFL frass. Lauric acid (Frass+C12) slowed N-NH_4^+ release to the greatest extent, followed by myristic acid (Frass+C14) and stearic acid (Frass+C18), respectively.
- FA processing of the frass resulted in a lag period for N immobilisation, as differences in N-NH_4^+ concentrations between frass treatments only occurred from 14 days of incubation onwards (Figure 17, 18). The C:N ratio for all three FA-P frass treatments were significantly higher than the unprocessed frass treatment and this would have favoured microbial N immobilisation over mineralization.
- Fatty acid chain length regulated the soil microbial community composition (Figure 19). Changes in community composition were linked to changes in TOC and total carbon over the incubation period and some bacterial groups were more strongly affected by certain frass treatments. Unprocessed and stearic acid processed frass became enriched in groups of slow-growing K-strategist bacteria (Bacteroidetes*; Chitinophaga* sp. and

Gammaproteobacteria*; Luteibacter* spp.) at the latter stages of the incubation, but not for lauric or myristic acid processed frass (Figure 19). The gradual increase in K-strategists at the later stages of incubation reflects their ability to degrade complex C materials such as lipids, cellulose, lignin and organic acids. Consequently, when frass is combined with FAs, FA chain length plays an important role in regulating the composition of r-/K-strategists in soil which likely impacts on N and carbon cycling.

Conclusion/Significance

In conclusion, combining BSFL frass with fatty acids slows mineral N release compared to untreated frass. Especially regarding the release of ammonia. In addition, amending soils with frass drastically changes the native soil microbiome which may play a role in N release. These results are promising and suggest that combining BSFL frass with fatty acids could potentially be developed into a slow-release fertilizer.

Differences in ammonia release only emerge 14 days after frass application to soil which could be particularly effective for cereals crops by delaying N release until mid-stem elongation, thereby ensuring optimal N uptake and lowering N losses. Using frass in this way could lead to improved fertilizer use efficiency and increased profitability for growers via improved crop yield and lower production costs. Furthermore, fatty acids are a by-product of Black Soldier Fly production (BSFL oil contains a large proportion of lauric acid), therefore combining BSFL derived fatty acids with BSFL frass could be a profitable new fertiliser product that also supports the circular ethos of BSFL farming.

Table 24. Physicochemical properties of Tenosol topsoil (soil) collected from the University of Western Australia Farm (Ridgefield, Pingelly), unprocessed BSFL frass (frass), combined unprocessed BSFL frass and soil (frass + soil), and combined fatty acid processed (FA-P) frass and soil (soil + frass + C12; soil + frass + C14; soil + frass + C18). Frass was derived from poultry manure. Values represent the mean and standard error in parenthesis (n=3). C12 = lauric acid, C14 = myristic acid, C18 = stearic acid.

Variable	Unit	Soil	Frass	soil + frass	soil + frass + C12	soil + frass + C14	soil + frass + C18
TC	%	28.94 (0.97)	29.90 (0.05)	32.20 (0.65)	33.44 (0.42)	34.52 (0.75)	34.43 (0.33)
TN	%	2.57 (0.04)	3.10 (0.01)	2.90 (0.05)	2.71 (0.01)	2.85 (0.07)	2.71 (0.04)
C:N	-	11.28 (0.27)	9.60 (0.03)	11.11 (0.05)	12.37 (0.17)	12.14 (0.10)	12.73 (0.10)
N-NO_3^-	mg Kg-1	59.91 (1.16)	14.30 (1.20)	57.23 (0.82)	56.05 (0.63)	55.28 (0.64)	55.52 (0.55)
N-NH_4^+	mg Kg-1	1.78 (0.49)	105.50 (17.1)	79.22 (7.31)	72.10 (4.59)	80.78 (2.07)	75.72 (7.78)
pH ($CaCl_2$)	pH	5.16 (0.03)	7.00 (0.04)	5.84 (0.30)	5.36 (0.03)	5.47 (0.04)	5.63 (0.05)

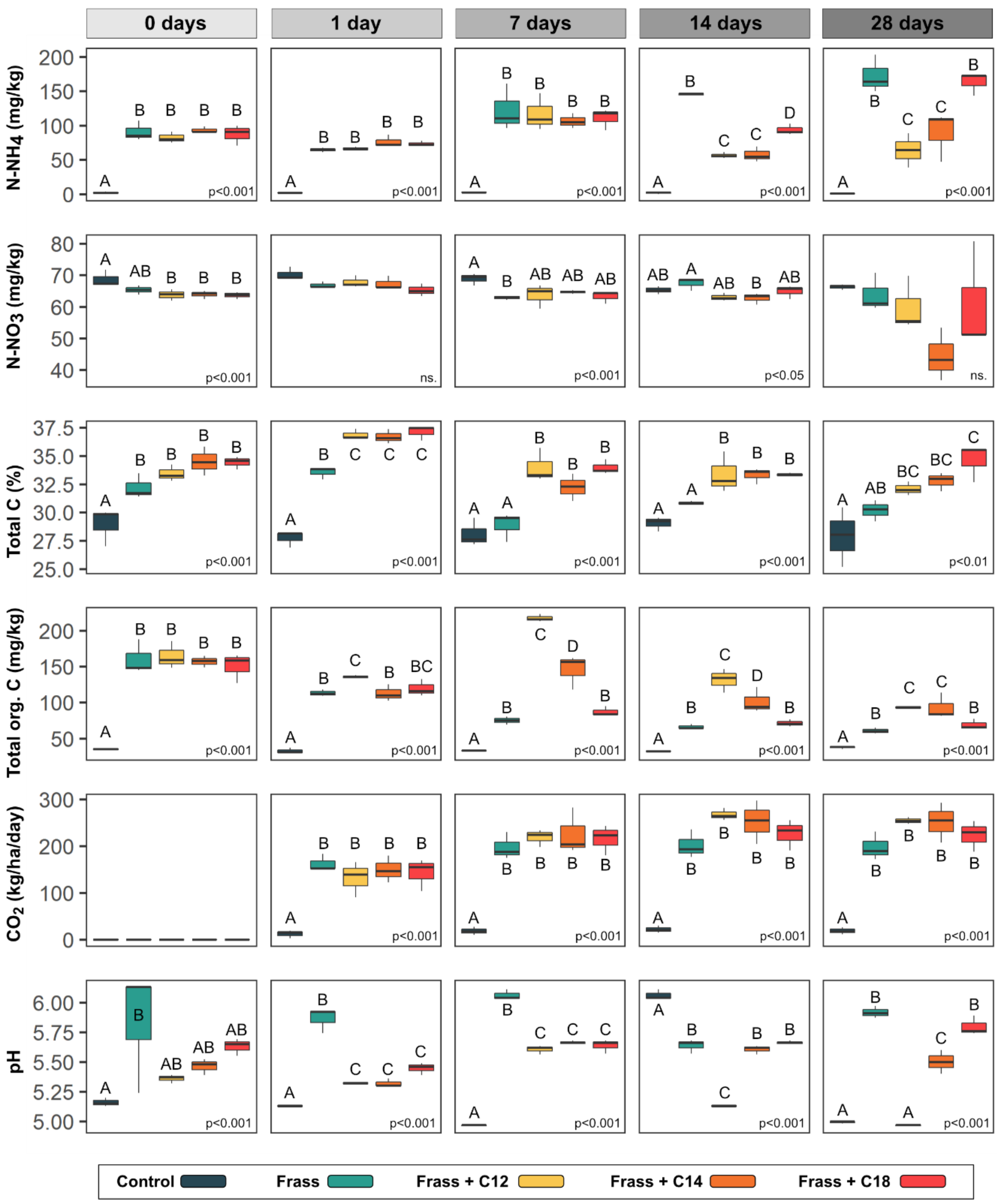


Figure 17. Amount of ammonium (N-NH_4 mg/kg), nitrate (N-NO_3 mg/kg), total carbon (%), Total organic carbon (mg/kg), respiration (CO_2 kg/ha/day), and pH measurements from soil microcosms amended with Black Soldier fly larval (BSFL) frass (frass), fatty acid processed BSFL frass (Frass +C12, lauric acid; Frass+C14, myristic acid; Frass+C18, stearic acid), or no frass (control). Microcosms were incubated over 28 days (in the dark at 25ºC) and destructively sampled at times 0, 1, 7, 14 and 28 (N=75). Significant post-hoc comparisons among treatments for each time point are indicated by uppercase letters.

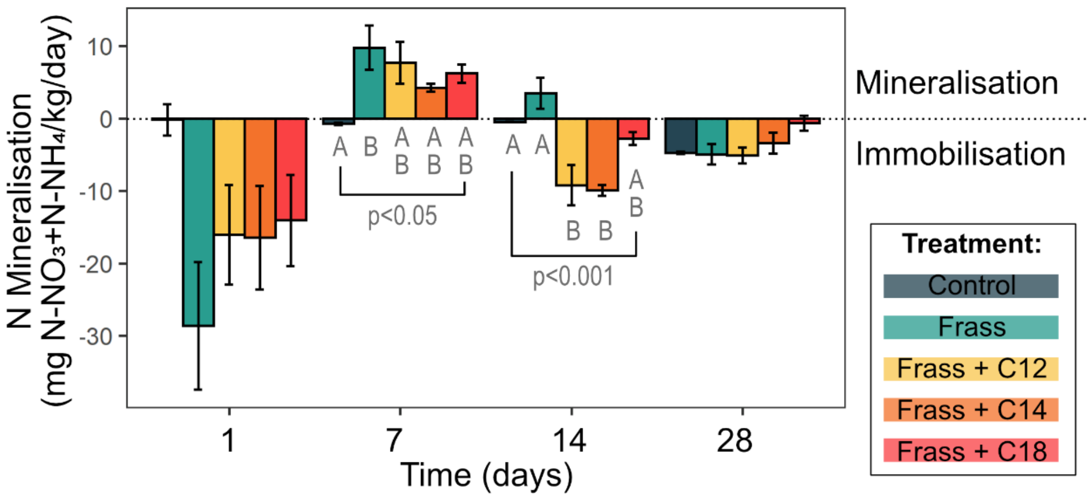


Figure 18. NET Nitrogen mineralization (mg N/kg/day) of soil microcosms amended with unprocessed Black Soldier fly larval (BSFL) frass (frass), fatty acid processed BSFL frass (Frass +C12, lauric acid; Frass+C14, myristic acid; Frass+C18, stearic acid), or no frass (control). Microcosms were incubated over 28 days (in the dark at 25ºC) and destructively sampled at times 0, 1, 7, 14 and 28 days (N=75). Significant post-hoc comparisons among treatments for each time point are indicated by uppercase letters.

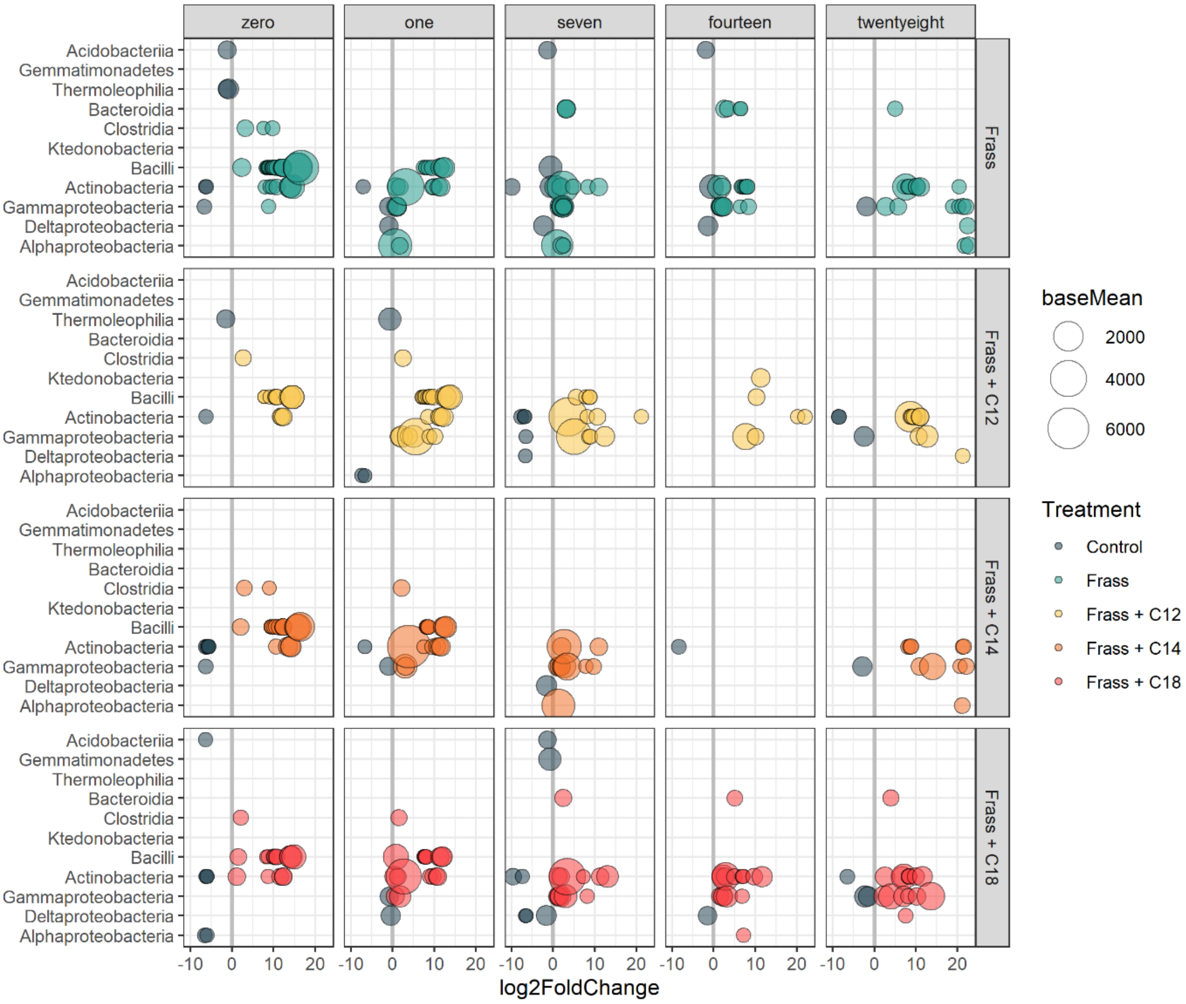


Figure 19. Differentially abundant ASV classes (significance of p < 0.001, agglomerated by genus) in soil microcosms amended with unprocessed Black Soldier fly larval (BSFL) BSFL frass (frass; > 0 log 2-fold change) or amended with fatty acid processed BSFL frass (Frass +C12, lauric acid; Frass+C14, myristic acid; Frass+C18, stearic acid; > 0 log 2-fold change), compared to control mesocosms with no BSF frass amendment (< 0 log 2-fold change). Microcosms were incubated over 28 days (in the dark at 25ºC) and destructively sampled at times 0, 1, 7, 14 and 28 days (N=75). ASVs are arranged by increasing significance of their adjusted p-values and the direction of the log 2-fold change.

5.5(b) Conduct laboratory trial to determine the impact of frass on soil biology

A PhD student was recruited (KPI 1.21) and they successfully carried out the research that contribute to 5.5b-c (KPI 2.20, 3.24). A draft scientific publication (KPI 4.18) has been prepared on the laboratory trials to determine the impact of manure derived frass on soil biology (Appendix J).

Summary

The black soldier fly (BSF) (*Hermetia illucens L.*) is an emerging waste treatment technology that could provide a circular economy approach to manure management by harvesting protein from manure fed BSF larvae as either feed or fertiliser. However, for every 5 tonnes of manure processed, a tonne of BSF frass is produced that constrains the scaling of this technology. Generally BSF frass has a low nutrient content and is not suitable as a fertiliser when applied singly. However, when applied as a top-up fertiliser following an initial basal dose of synthetic fertiliser it could enhance plant growth performance. Here, we determine the optimal application rate of frass to maximise plant growth. Secondly, we explore the impact of increasing frass amendments on rhizosphere bacterial community structure and function. Frass amendments provide nutrition, carbon and influence the physical and chemical structure of soil and thereby will alter the rooting matrix for plants, and the habitable space and proximity to resources for biological communities. The resulting improved structure and soil quality enhances biological processes that support plant growth such as C and N cycling.

A pot trial was conducted where chilli (*Capsicum annum L.*) plants were grown for 14 weeks in an agricultural sandy loam soil amended with seven different rates of manure-derived BSF frass (0, 5, 10, 15, 20, 25, and 30 g pot^{-1}). At harvest, plant growth parameters (shoot and root dry weight, shoot N content), soil quality measures (pH, EC, soil mineral N (NH4+, NO3-)), microbial C and N biomass and rhizosphere bacterial abundance were quantified. 16S rRNA sequencing followed by in silico PICRUSt analysis of functional gene prediction were used to interrogate the rhizosphere bacterial community and its functional component involved in nitrogen (N) cycling and soil carbon (C) degradation.

Key findings

- Amending soil with manure-derived frass was shown to enhance both the soil quality and chilli crop productivity (Figure 20 and 21).
- Shoot dry weight significantly ($P < 0.01$) increased with increasing rates of BSF frass applied until reaching a maximal growth was achieved at 15 g pot^{-1} BSF frass. Further additions of frass beyond this resulted in decreased growth (Figure 20a). Similar trends were observed with root dry weight, shoot N content and shoot N concentration (Figure 20). This suggests that the application of manure frass to Chilli plants could potentially provide half of the crop's nutritional requirements meaning that the grower could reduce their synthetic fertiliser inputs. However, the effectiveness and economic value of fertiliser replacement would need to be verified in the field and through a full cost benefit analysis.
- The soil pH, mineral N content, soil MBC and MBN significantly increased with increasing frass application rates ($P < 0.01$) (Table 25; Figure 21). This suggests the manure-derived frass contained a high concentration of carbon and nutrients that provides energy for the soil microbial community and their activities.

- There is a marked increase in soil pH at higher application rates of manure-derived frass (Figure 21). Soil pH has been identified as a key driver of microbial community structure and function.
- Analysis of the bacterial community structure revealed that the phyla Firmicutes and Bacteroidetes and some families of Proteobacteria were more abundant at higher frass application rates to soil (Figure 22).
  Further analysis (at lower taxonomic levels) revealed these phyla were largely dominated by plant growth promoting rhizobacteria (PGPR) genera *Bacillus, Luteimonas, Sporosarcina, Sporosarcina, Devosia* and *Flavabacterium* (Table 26).
- These microorganisms are essential for plant nutrient acquisition, disease suppression, resistance to various stresses, and plant development. *Bacillus* species are known to secrete polysaccharides that promote plant growth and support health. *Luteimonas* and *Sporosarcina* increased the acquisition of nitrogen (N) and phosphorous (P) by plants especially when growing in the marginal agricultural soil. *Sporosarcina, Devosia* and *Flavabacterium* produce siderophore and indole acetic acid (IAA) in the presence of plant exudates containing l-tryptophan. Some of these microorganisms also secrete polysaccharides that help bind soil particles and improve soil structure.

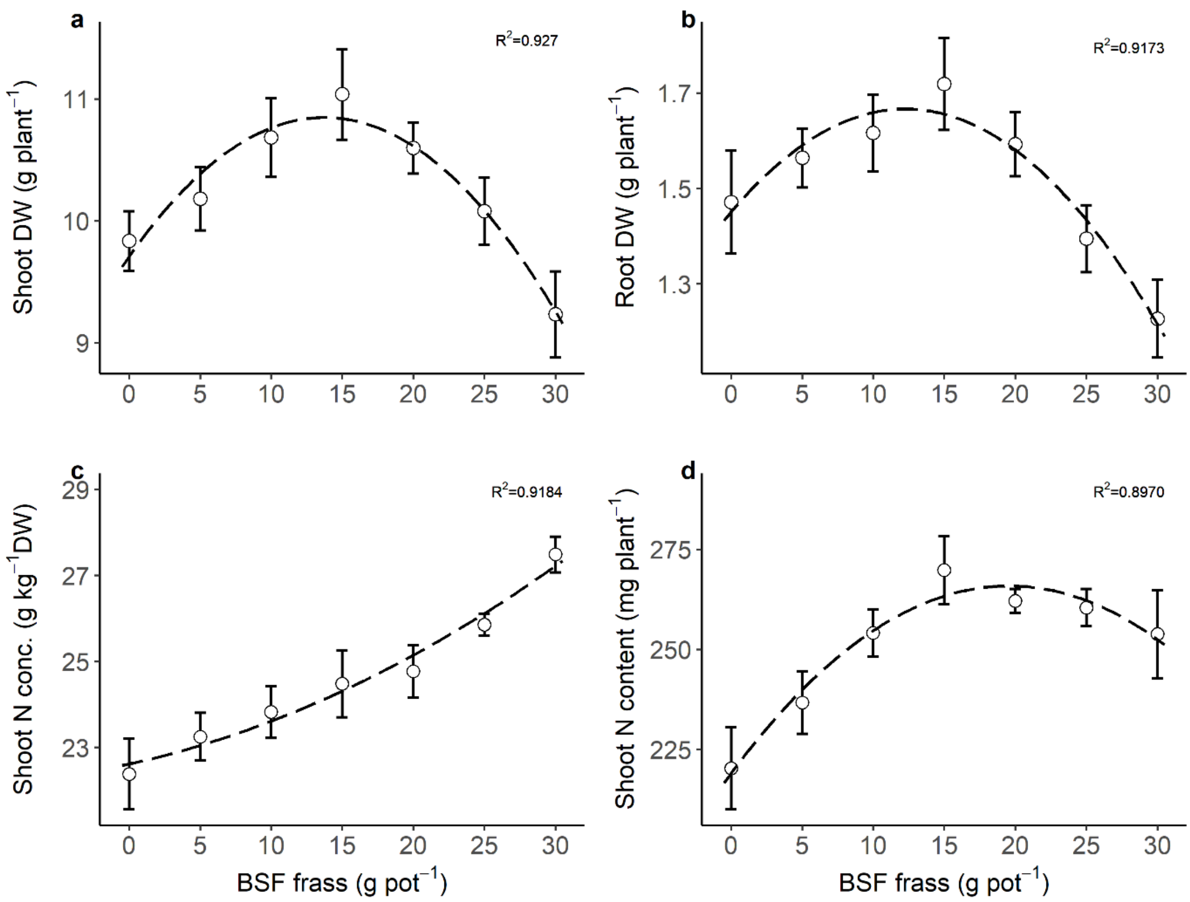


Figure. 20. The influence of increasing the rate of BSF manure derived frass on (a) shoot dry biomass (DW), (b) root dry biomass (DW), (c) shoot N concentration and (d) plant N uptake for chilli grown on a sandy loam soil after 14 weeks. The vertical bar indicates standard errors of the means (means ± SE, n=4).

Table 25 Soil mineral N in response to different rates of BSF frass (means ± SE). Means within a column not sharing the same letter differ significantly.

Treatment (BSF frass g pot^{-1})	NH_4^+-N (mg kg^{-1})	NO_3^-- N (mg kg^{-1})
0	0.153 ± 0.009c	0.72±0.13f
5	0.161±0.008c	0.78±0.08ef
10	0.162±0.011c	1.22±0.11de
15	0.180±0.009c	1.30±0.10cd
20	0.206±0.012bc	1.75±0.02bc
25	0.247±0.014ab	2.08±0.12ab
30	0.263±0.014a	2.29±0.12a

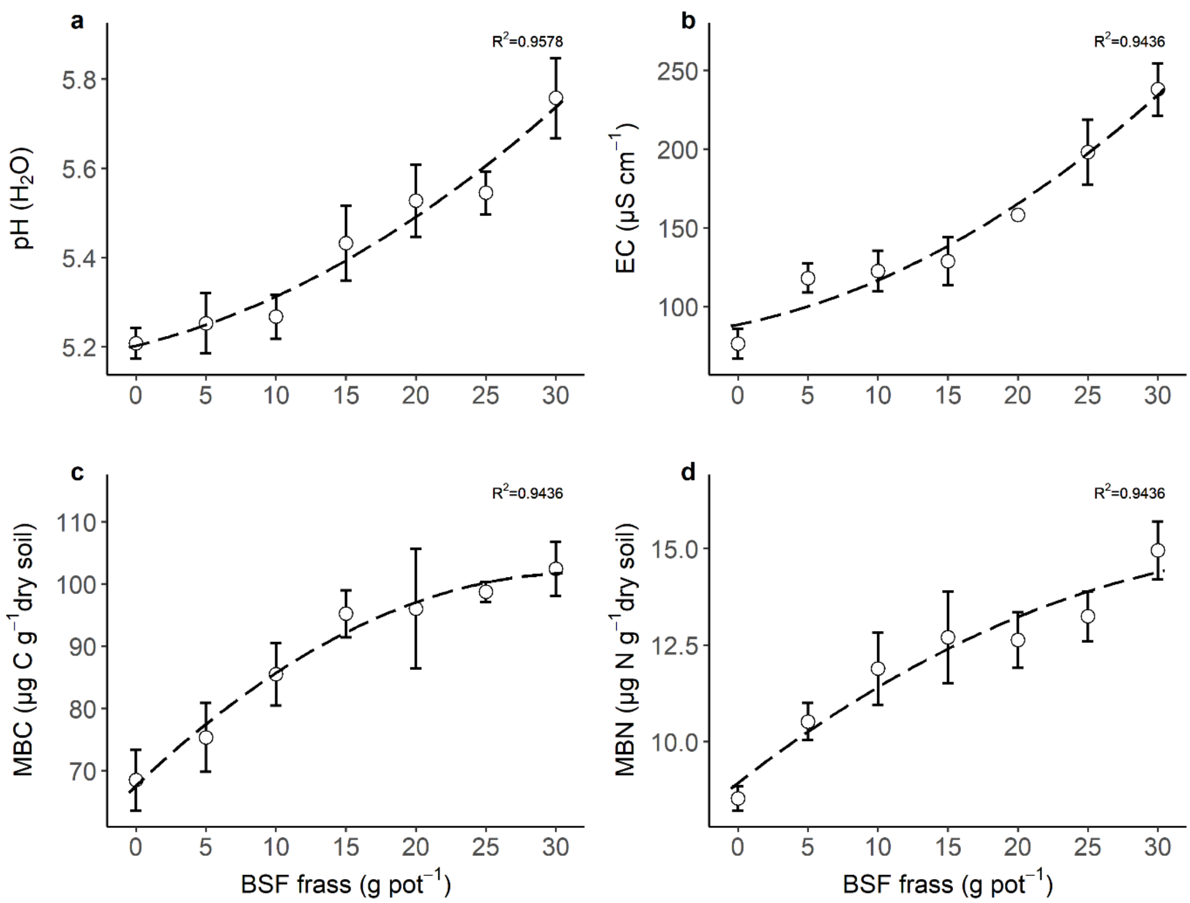


Figure 21. The influence of increasing the rate of BSF manure derived frass on (a) soil pH, (b) electrical conductivity (EC), (c) microbial biomass C and (d) microbial biomass N for chilli grown on a sandy loam soil after 14 weeks. The vertical bar indicates standard errors of the means (means ± SE, n=4).

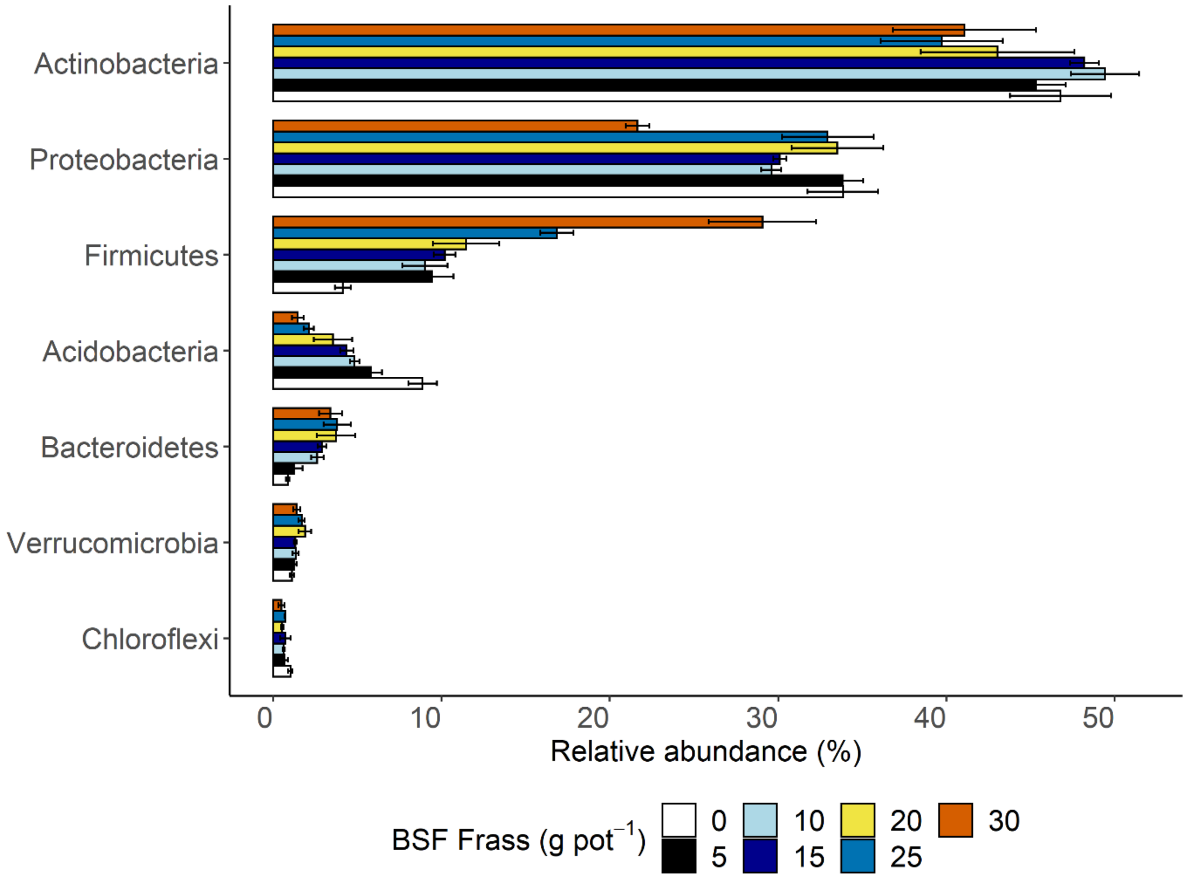


Figure 22. The influence of increasing the rate of BSF manure derived frass on the relative abundance of bacterial phyla for chilli grown on a sandy loam soil 14 weeks after transplanting. Bars represent the standard errors of the means (n=4; mean ± SE).

Table 26 Analysis of variance of the taxon of the plant growth promoting rhizobacteria at phylum, family and genus levels showing positive correlation with increasing rates of manure-derived frass. Significant codes: *p*<0 '***', *p*<0.001 '**', *p*<0.01 '*'.

Phylum	Family	Genus	Beneficial role in plants
Firmicutes (***)	Baciliaceae (***)	*Bacillus (***)*	Secrete metabolites that stimulate plant growth and health.
	Paenibacilaceae (***)	*Thermobacillus (***)* *Ammoniphilus (**)*	Plant growth-promoting rhizobacteria
	Planococcaceae (***)	*Psychrobacillus (**)* *Sporosarcina* (*) *Solibacillus (*)*	Plant growth-promoting rhizobacteria Phosphate and zinc solubilisation and plant growth hormones production
Bacteroidetes (*)	Flavobacteriaceae (*)	*Flavobacterium* (*)	Plant growth hormone production
Proteobacteria (*)	Devosiaceae (*)	*Devosia (*)*	Produce siderophore and indole acetic acid that promote growth
	Xanthobacteraceae (***)	*Luteimonas (**)*	Acquisition of nitrogen (N) and phosphorous (P)
	Azospirillales_Incertae_Sedis (***)	*Stella (***)*	Plant growth-promoting rhizobacteria
	Polyangiaceae (***)	*Sorangium (***)*	Plant growth-promoting rhizobacteria

5.5(c) Conduct pot trials on targeted pasture, broadacre or horticulture crops as determined from the market analysis and industry engagement

A technical report on the agronomic benefits of using manure frass as a soil amendment for pasture and broadacre crops has been produced (Appendix K). The market engagement did not yield any specific target market, we therefore used representative crops that fit the experiment.

Summary

The aim was to conduct pot trials to assess the growth of pasture, broadacre or horticultural crops using BSF frass (KPI 2.21, 3.25). There were difficulties encountered with the experiments designed to (i) develop the BSF frass N curve for plant species and (ii) determine accurate BSF frass (manures) rates, particularly for wheat. However, an additional pot trial was completed which examined the growth of ryegrass to BSF (manures) frass. This activity generated significant knowledge of the BSF (manures) frass that will improve its potential adoption as an agricultural or horticultural commodity. Finally, a third experiment examined whether BSF (manure) frass can be used as a soil conditioner for horticulture to ameliorate soil acidity and improve soil fertility. In Australia, about 50 % of Australian agricultural land has problems with soil acidity (Australian Agriculture Assessment 2001) and lime is used for remedy. Some organic amendments have been found to increase the soil pH but to what extent soil amendments with BSF frass can improve soil acidity is currently unknown.

A pot trial was conducted where Lettuce (*Lactuca sativa L.*) plants were grown for 10 weeks in an agricultural sandy soil collected from a vegetable farm in Gingin, Western Australia. The soil was amended with six different rates of manure-derived BSF frass (0, 5, 10, 15, 20, and 25 g pot^{-1}). There were four replicates for each treatment and two harvests (35 and 70 days after transfer(DAT)). At harvest, plant growth parameters (shoot and root dry weight, plant N content), soil quality measures (pH, EC, soil mineral N (NH_4^+, NO_3^-)) and rhizosphere bacterial abundance were quantified.

Major outcomes

- In the first experiment wheat was unable to grow with addition of milled BSF larvae and its shoots were heavily stunted by the addition of BSF frass >30 mg N kg^{-1}, presumably due to ammonium (NH4-N) toxicity (Figure 23).

- Post-experiment, soil contained high nitrate (NO_3-N) and low NH_4-N, consistent with the microbial process of nitrification.

- The optimum N application rate was not achieved as most wheat appeared to be N deficient, indicating the N rates, calculated on total frass N content, significantly overestimated plant available-N.

- A second experiment tested the growth response of ryegrass to commercially relevant application rates of BSF manures frass supplied in its raw form or granulated, compared to raw pork manures and mineral fertiliser application (Figure 24).

- BSF vegetable frass and granulated manures frass (+ molasses binder) at high rates of application (i.e., 10 Mg ha^{-1}) significantly improved ryegrass growth more so than did mineral fertiliser application (nil BSR frass).

- Root dry mass at application rates ≥5 Mg ha^{-1} was increased, except for raw manures, granulated manures frass (no binder) and granulated manures frass (potato starch binder).

- A third experiment found that applying BSF (manure) frass to lettuce plants showed a typical fertiliser growth response (increased dry shoot and root weight) with increasing application rates (Figure 25). A similar trend was observed with shoot N concentration and content.

- The results also showed a marked increase in soil pH with increasing rates of frass applied (Figure 26) implying the BSF (manure) frass has a liming effect. Agricultural manures and wastes often have a liming effect since they often contain high concentrations of ammonium and basic cations such as Ca^{2+} or Mg^{2+} which serve to neutralize soil acidity (Walker et al., 2004).

- An increase in the soil pH can have a significant impact on soil fertility by altering chemical properties of soil such as organic matter content and cation exchange capacity (CEC). Raising the pH can also increase the availability of macro-nutrient and micro-nutrient to plants and soil microorganisms.

- BSF manure frass has the potential to be used as a soil conditioner to ameliorate plant growth constraints caused by soil acidity thereby reducing the amount of liming required especially in acidic, semiarid areas such as Western Australia. However, the effectiveness and economic value of frass as soil ameliorant needs to be verified in the field and through a full cost benefit analysis.

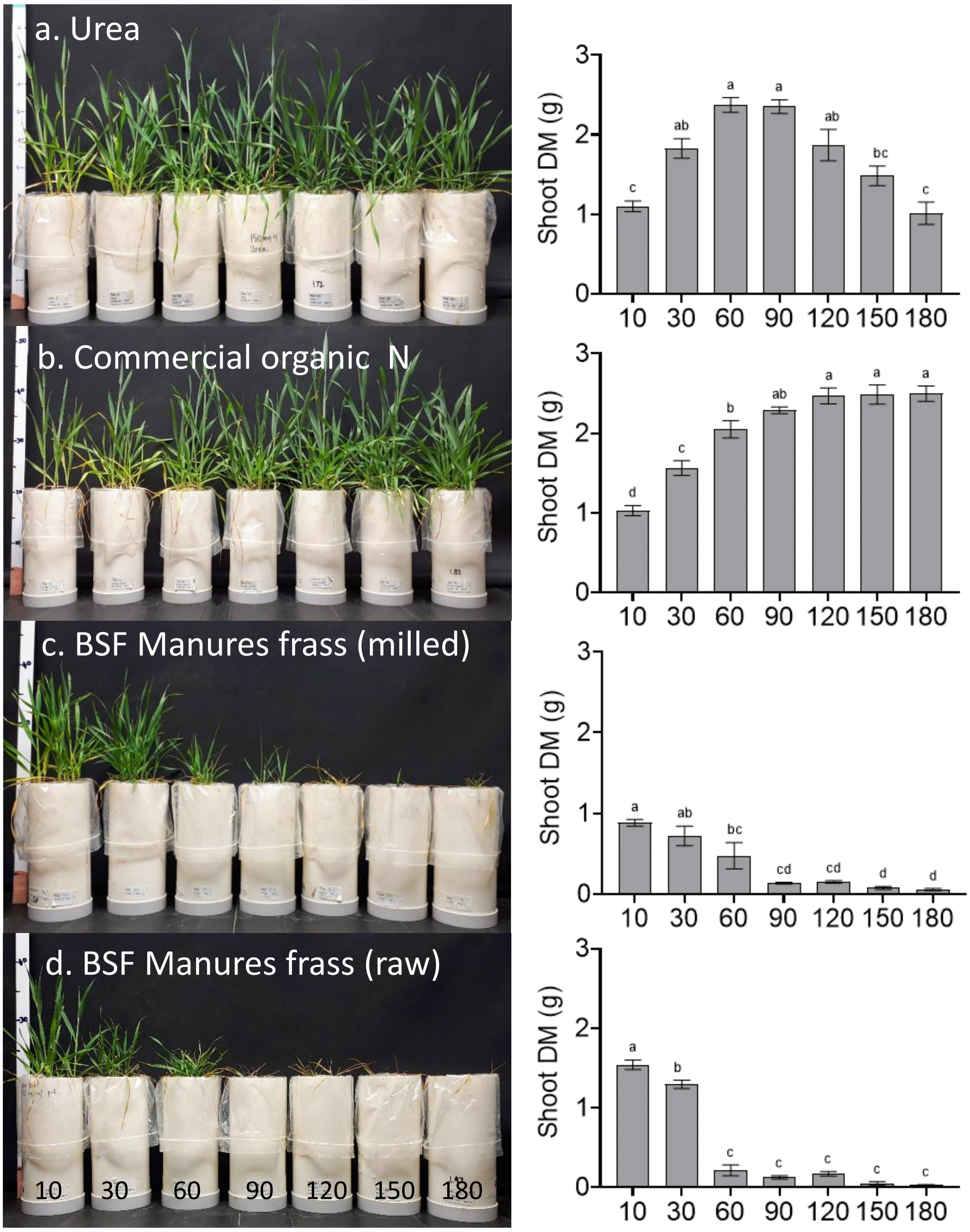


Figure 23. Mean shoot dry matter response of wheat (*Triticum aestivum* L.) to increasing soil nitrogen (N) concentrations (10, 30, 60, 90, 120, 150 and 180 mg kg^{-1}) provided as a) Urea; b) Commercial organic N; c) BSF manures frass (milled) and; d) BSF manures frass (raw). Values are the mean of five replicates (± s.e). Letters denote significant differences between N rates (Tukeys post-hoc test).

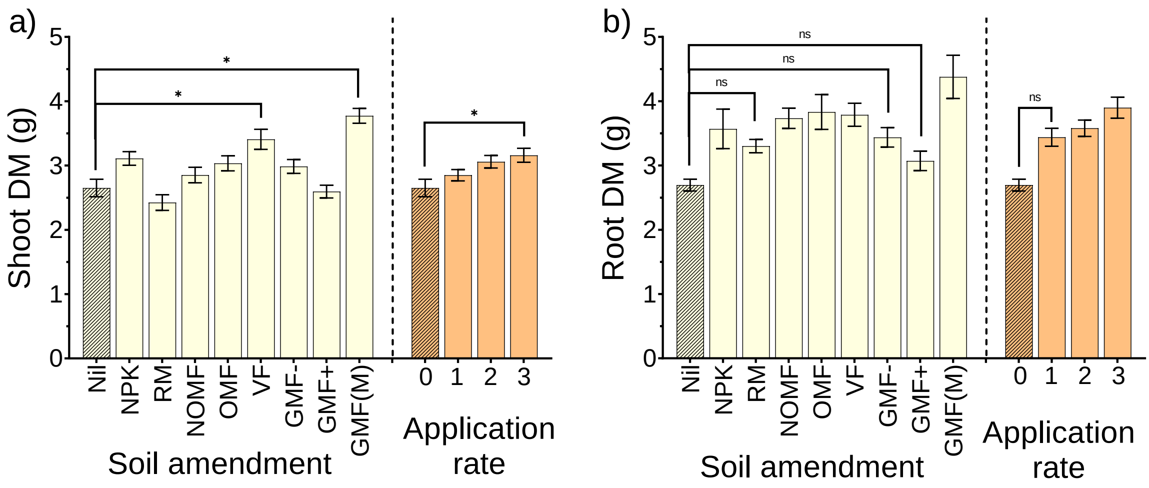


Figure 24. The effect of soil amendment and application rate on a) shoot dry mass (± s.e) and b) root dry mass (± s.e) of rye grass with Dunnett's post-hoc test. Treatments denoted by * in 7(a) significantly differ to the control (Nil) treatment (shaded). Treatments denoted by ns in 7(b) do not significantly differ to the Nil treatment. Soil amendment abbreviations: Nil (Control), NPK (mineral fertiliser), RM (Raw manures), NOMF (Non-optimised manures frass), OMF (Optimised manures frass), VF (Vegetable frass), GMF- (Granulated manures frass (no binder)), GMF+ (Granulated manures frass (with binder), GMF (Granulated manures frass (molasses binder)). Application rate abbreviations: 0 (control), 1 (2.5 Mg ha^{-1} frass products or 50 kg ha^{-1} mineral fertiliser), 2 (5.0 Mg ha^{-1} frass products or 100 kg ha^{-1} mineral fertiliser), 3 (10.0 Mg ha^{-1} frass products or 200 kg ha^{-1} mineral fertiliser).

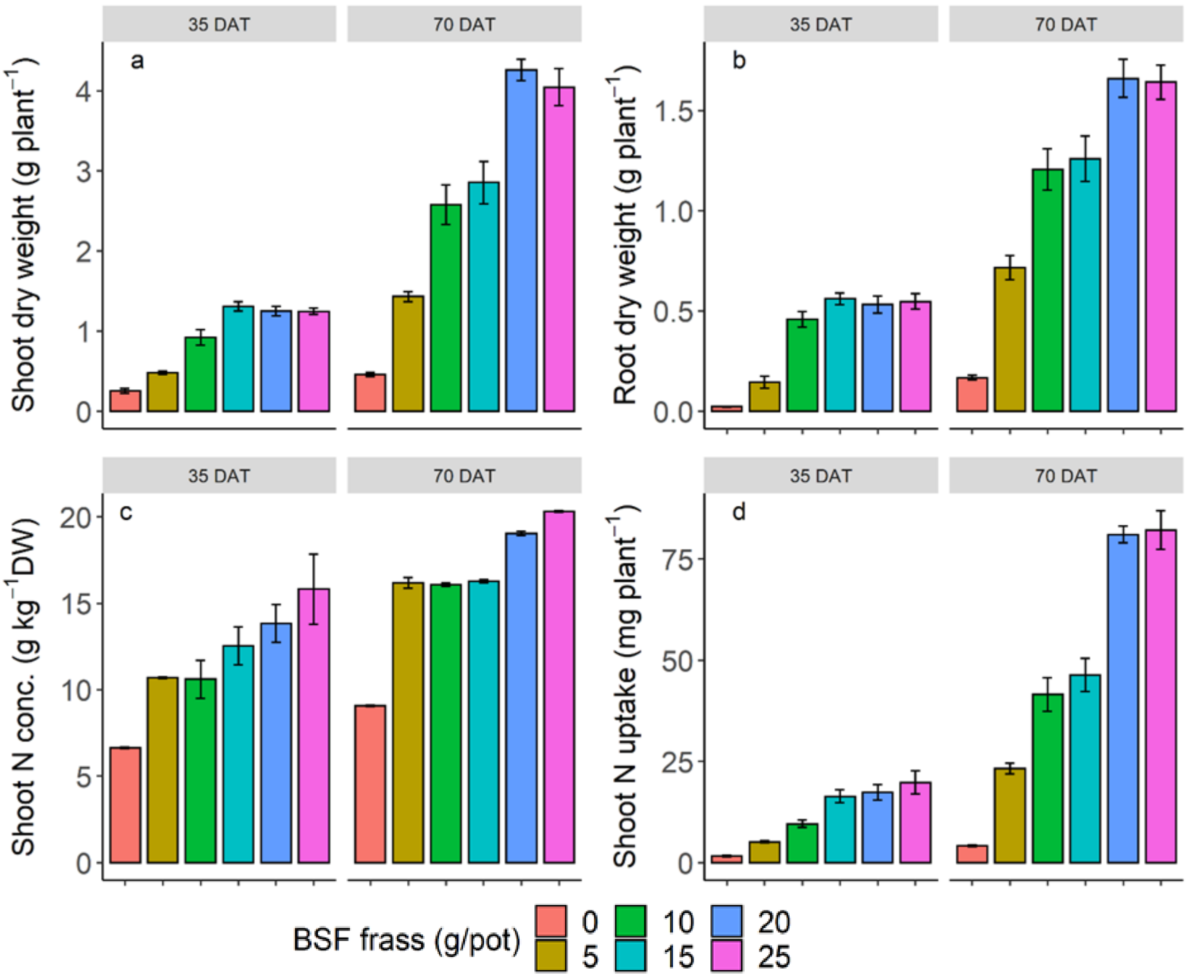


Figure 25. Changes in shoot dry weight (a), root dry weight (b), shoot N concentration (c) and shoot N content (d) of lettuce in soil amended with six different rates of BSF frass at 35 and 70 days after transplanting. Treatments include: 0, 5, 10, 15, 20 and 25 g BSF frass per pot (2.5 kg soil/pot). Bars represent the mean of each treatment and error bars are the ± standard error of the mean (n=4).

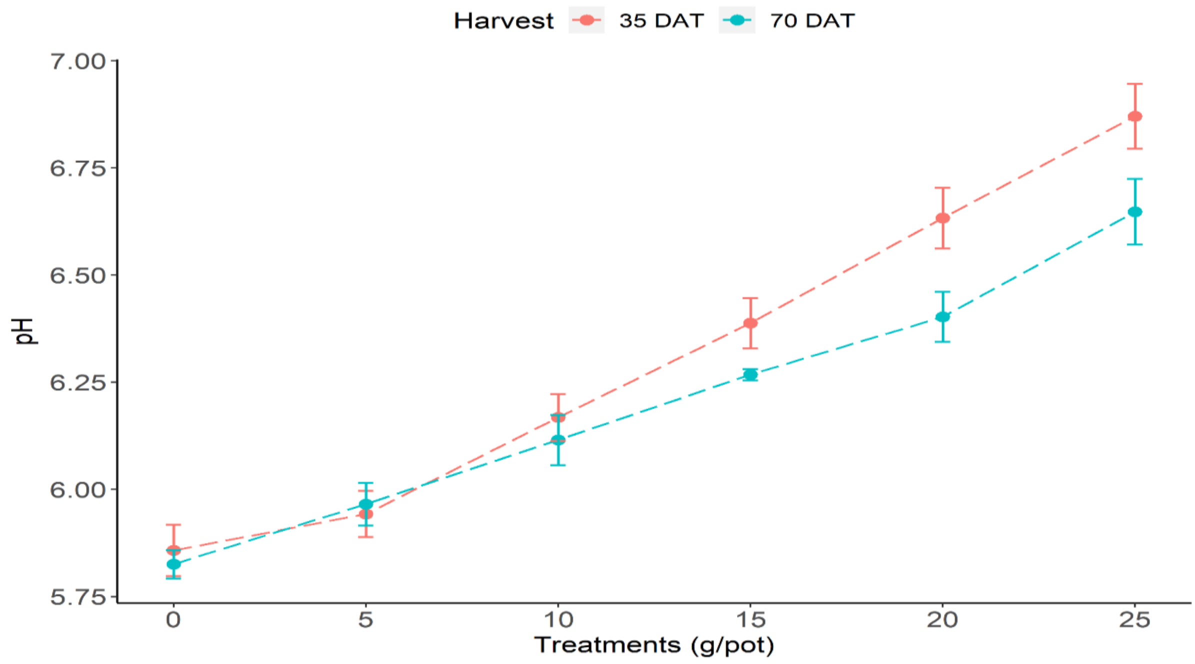


Figure 26. Changes in pH (a), and EC (b) in soil amended with six different rates of BSF frass at 35 and 70 days after transplanting. Treatments include: 0, 5, 10, 15, 20 and 25 g BSF frass per pot (2.5 kg soil pot-1). Bars represent the mean of each treatment and error bars are the ± standard error of the mean (n=4).

5.5(d) Conduct field trials to evaluate the effectiveness of using frass to ameliorate soil constraints (e.g. acidity, water repellency) and provide a full cost economic and environmental cost benefit analysis

Field trials to evaluate the effectiveness of using frass to ameliorate soil constraints

A technical report on the agronomic benefits of using frass as a soil amendment for pasture and broadacre crops in the field has been produced (Appendix L).

Summary

The use of Black Soldier Fly (BSF) frass as a soil amendment in field grown crops is little studied. The application of BSF frass is expected to improve soil structure and function which would contribute to improved growth of broadacre crops and pastures and will generate new knowledge of a product that will ultimately benefit primary producers. Two field trials (on a sandy soil type) were conducted to examine the effect of (i) five BSF vegetable frass application rates on four plant species (wheat, canola, rye grass, red clover) – Experiment 1; and (ii) five BSF manures frass application rates on two plant species (rye grass and wheat) – Experiment 2. The field trials were strategically located adjacent to the industry partner Future Green Solutions operational facility: this enabled regular communication and collaboration. These field experiments successfully demonstrated an improvement in soil nitrogen and carbon and reduced nutrient leaching from the application of BSF frass which led to improved nutrient uptake, vegetative growth and crop yields. Estimates of commercially appropriate application rates were also able to be estimated based on crop performance.

Major outcomes

- The combination of BSF frass and mineral fertiliser applications increased rye grass dry matter productivity 5-fold compared to the mineral fertilisers (nil BSF frass) in isolation. This equates to an increase of ~1 Mg ha^{-1} dry matter for every 2.5 Mg ha^{-1} of BSF frass.

- BSF frass application rate at > 5 Mg ha^{-1} were required to generate an increase in DM productivity of red clover (pasture legume able to fix its own N). This increase was approximately 0.5 Mg ha^{-1} compared to the nil BSF frass and no further improvements were evident.

- Wheat yield improved from 2.4 Mg ha^{-1} (nil BSF frass) up to 4 Mg ha^{-1} (10 Mg ha^{-1} BSF frass), with no further impact above application rates of 5 Mg ha^{-1} (Figure 27).

- BSF frass increased available soil nitrogen (NH_4-N and NO_3-N) and soil carbon compared to mineral fertiliser applications (Figures 28 and 29, respectively)

- BSF frass improved retention of applied mineral fertilisers in sandy soils.

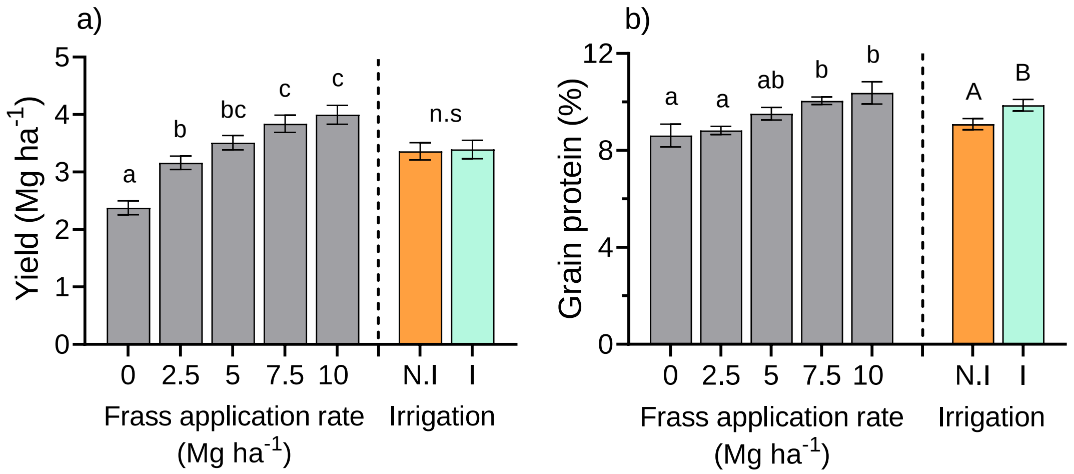


Figure 27. The effect of BSF frass application rate and irrigation (non-irrigated (N.I) and irrigated (I)) on the a) grain yield (± s.e) and b) protein (± s.e) of wheat. Different lower-case letters denote significant effects of frass application rate and different upper-case letters denote a significant effect of irrigation. n.s = not significant

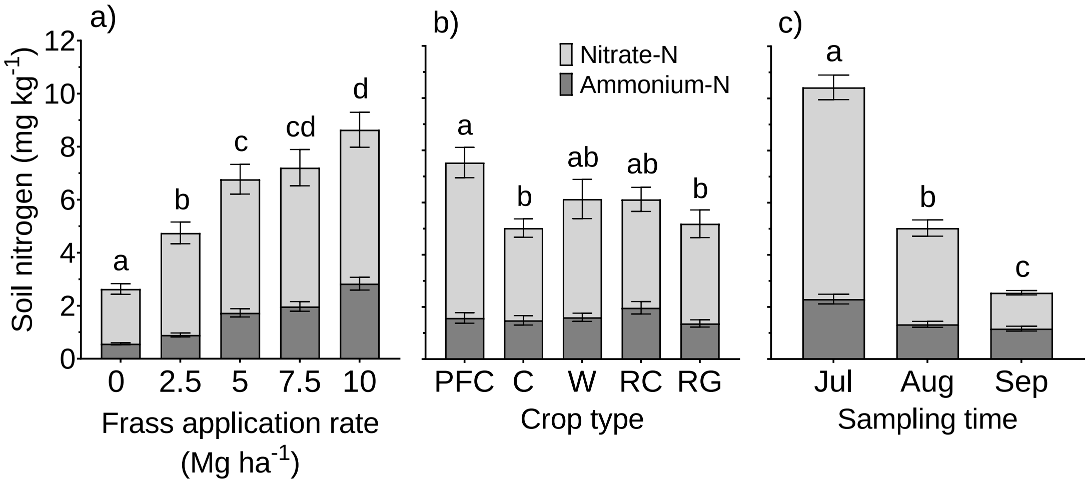


Figure 28. The effect of a) BSF frass application rate; b) crop type (plant free control (PFC), canola (C), wheat (W), red clover (RC), rye grass (RG)) and; c) soil sampling time (5 July (Jul), 17 August (Aug), 29 September (Sep)) on soil nitrogen (NO_3-N and NH_4-N). Different letters within each graph denote significant treatment effects.

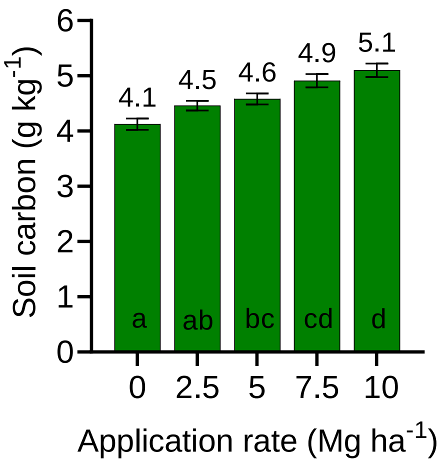


Figure 29. The effect of BSF frass application rate on total soil carbon (± s.e.). Values represent the treatment mean (n=20). Letters that differ indicate a significant effect of application rate on soil carbon according to Tukey's post-hoc test.

<u>Full economic and environmental cost benefit analysis</u>

Summary

The effectiveness of using Black Soldier Fly frass as a soil ameliorant was tested in field trials for wheat, and two pasture crops, rye grass and red clover. Experiments were conducted for both vegetable frass and manure frass to examine the effects of adding BSF frass on the yield and soil organic carbon and whether there are differences from the use of vegetable frass and manure frass (Figure 30).

Figure 30: Field experimental trials of BSF frass in wheat ryegrass and red clover

Methods

Partial budget analysis was conducted to examine the net gain of using black soldier fly as a soil ameliorant. Partial budget analysis involves comparing the changes in the costs and benefits with the use of a new technology (i.e., BSF frass). In this case, two scenarios were considered - the net benefits (1) between the control (no frass) and with the soil ameliorant using BSF vegetable frass, and (2) between the control and with BSF manure frass.

The main benefits included in the analysis are the additional yield resulting from the use of the frass and the increase in soil organic carbon. Values from the field experiment conducted by Dr Dan Kidd for yield and soil organic carbon were used, while price and cost data were based from market research data we conducted for the prices of inputs (frass) and outputs (commodity prices). The BSF frass prices ranged between $3.60 to $18+/kg. Several analytical scenarios reflecting different BSF frass price levels were performed (including lowest, mid and highest prices) but only the lowest price is presented in this report to reflect the minimum cost. The soil organic carbon price was based on ACCU market price. The price used in the model was the price on the day which had been fairly stable in September/October 2022 (during the final modelling period). In addition, model simulations, were done for various price levels up to $100 to examine the impact of BSF frass fertiliser. The sources of information for various input and output prices and costs are shown in the below tables.

The changes in GHG emissions were not included in the economic analysis as these were not measured in the field experiments, and the literature has varying results, so could not be used reliably in the modelling.

Results

For wheat, the addition of vegetable BSF frass increased the yield and soil organic carbon (SOC) by 1.62 t/ha and 0.97 g/kg of soil, respectively. The total gain from using vegetable frass was estimated to be about $579/ha while the additional cost was about -$36,000, thus giving a net loss of about $35, 421/ha (Table 27). BSF manure frass yielded lower total net loss at approximately $26,536/ha, with additional benefits estimated to be about $464/ha while the additional cost of the frass was about $27,000/ha (Table 28). Currently, the cost of BSF frass at approximately $3.60/kg means that adding BSF frass as a soil ameliorant is not economically viable for wheat production.

For rye grass, the use of BSF frass also increased the yield (3.6t/ha for vegetable frass and 0.94 t/ha for manure frass) and soil organic carbon. However, the additional gains from the higher yield and increase in soil organic carbon were not enough to offset the cost of the BSF frass. The additional gains from the higher yield and increase in soil organic carbon were valued at $1,485/ha while the additional cost was $36,000/ha, with extra net loss equalling $34,515/ha for vegetable frass. The additional net loss for ryegrass using manure frass was higher at about $35,598/ha (Tables 29 and 30).

For red clover, the experiment was only conducted with vegetable frass. As with wheat and rye grass, using BSF frass improved the yield of red clover and increased soil organic carbon which resulted in an economic benefit of $464/ha. However, the additional cost of the frass amendment was $36,000 giving a net loss of about $35,536/ha (Table 31).

Table 27: Partial budget analysis of using BSF vegetable frass in wheat production.

Losses				Gains			
Extra expenses	**Unit (t/ha)**	**Per unit cost ($/kg)**	**Total ($/ha)**	**Extra income**	**Unit (t/ha)**	**Per unit cost ($/t)**	**Total ($/ha)**
Cost of frass	10.00	3.60	36,000	Higher yield	1.62	330.00	535
			-	Increase in soil carbon (g/kg)	0.97	30.00	44
			-	Reduction in CO2 emission		5.38	-
Sub-total			*36,000*	*Sub-total*			*579*
Income forgone				**Expenses saved**			
Nil			-	Reduction in cost of fertiliser			-
Sub-total			-	*Sub-total*			-
Total losses (A)			***36,000***	***Total gains (B)***			***579***
Extra profit (B-A) =	**-$ 35,421.06**			**Value Cost Ratio (VCR) =B/A**	**0.02**		

Table 28: Partial budget analysis of using BSF manure frass in wheat production.

Losses				Gains			
Extra expenses	**Unit (t/ha)**	**Per unit cost ($/kg)**	**Total ($/ha)**	**Extra income**	**Unit (t/ha)**	**Per unit cost ($/t)**	**Total ($/ha)**
Cost of frass	7.5	3.6	27,000	Higher yield	1.33	330.00	439
			-	Increase in soil carbon (g/kg)	0.82	30.00	25
			-	Decrease in CO2 emission		5.38	-
Sub-total			*27,000*	*Sub-total*			*464*
Income forgone				**Expenses saved**			
Nil			-	Reduction in cost of fertiliser			-
Sub-total			-	*Sub-total*			-
Total losses (A)			***27,000***	***Total gains (B)***			***464***
Extra profit (B-A) =	**-$ 26,536.27**		**- 26,536**	**Value Cost Ratio (VCR) =B/A**	**0.02**		

Table 29: Partial budget analysis of using BSF vegetable frass in ryegrass production.

Losses				Gains			
Extra expenses	**Unit (t/ha)**	**Per unit cost ($/kg)**	**Total ($/ha)**	**Extra income**	**Unit (t/ha)**	**Per unit cost ($/t)**	**Total ($/ha)**
Cost of frass	10.00	3.60	36,000	Higher yield	3.60	400.00	1,441
			-	Increase in soil carbon (g/kg)	0.97	30.00	44
			-	Reduction in CO2 emission		5.38	-
Sub-total			*36,000*	*Sub-total*			*1,485*
Income forgone				**Expenses saved**			
Nil			-	Reduction in cost of fertiliser			-
			-				-
			-				-
Sub-total			-	*Sub-total*			-
Total losses (A)			***36,000***	***Total gains (B)***			***1,485***
Extra profit (B-A) =	**-$ 34,515.12**			**Value Cost Ratio (VCR) =B/A**	**0.04**		

Table 30: Partial budget analysis of using BSF manure frass in ryegrass production.

Losses				Gains			
Extra expenses	**Unit (t/ha)**	**Per unit cost ($/kg)**	**Total ($/ha)**	**Extra income**	**Unit (t/ha)**	**Per unit cost ($/t)**	**Total ($/ha)**
Cost of frass	10.00	3.60	36,000	Higher yield	0.94	400.00	377
			-	Increase in soil carbon (g/kg)	0.82	30.00	25
			-	Reduction in CO2 emission		5.38	-
Sub-total			*36,000*	*Sub-total*			*402*
Income forgone				**Expenses saved**			
Nil			-	Reduction in cost of fertiliser			-
Sub-total			-	*Sub-total*			-
Total losses (A)			***36,000***	***Total gains (B)***			***402***
Extra profit (B-A) =	**-$ 35,598.19**			**Value Cost Ratio (VCR) =B/A**	**0.01**		

Table 31: Partial budget analysis of using BSF vegetable frass in red clover production.

Losses				Gains			
Extra expenses	**Unit (t/ha)**	**Per unit cos**	**Total ($/ha)**	**Extra income**	**Unit (t/ha)**	**Per unit cos**	**Total ($/ha)**
Cost of frass	10.00	3.60	36,000	Higher yield	1.05	400.00	421
			-	Increase in soil carbon (g/kg)	0.97	30.00	44
			-	Reduction in CO2 emission		5.38	-
Sub-total			*36,000*	*Sub-total*			*464*
Income forgone				**Expenses saved**			
Nil			-	Reduction in cost of fertiliser			-
Sub-total			-	*Sub-total*			-
Total losses (A)			***36,000***	***Total gains (B)***			***464***
Extra profit (B-A) =	**-$ 35,535.59**			**Value Cost Ratio (VCR) =B/A**	**0.01**		

Discussion

The economic analysis shows that the use of BSF frass (either manure or vegetable frass) improves the yield of wheat, ryegrass and red clover. There are also additional environmental benefits in the form of improvements in soil organic carbon. However, due to the high cost of BSF frass, it is not economically viable to add BSF frass, as the additional gains from applying the frass is not sufficient to justify the additional costs. The current market cost of BSF frass is about $36/kg or $3,600/t. If a rate of 10 t/ha of BSF frass is used as soil ameliorant to get the maximum additional crop yield, the cost of applying BSF frass equates to about $36,000/ha, which is much higher than the combined agronomic and environmental benefits. Applying BSF frass will only become economically feasible if the market cost of frass decreases to make it more affordable to crop producers.

A simulation analysis was conducted to determine the break-even point for the use of BSF frass in the production of wheat, ryegrass and red clover (Table 32, 33). Based on the analysis, the price of BSF vegetable frass has to go down to about $57.89/t for profit to break-even in wheat, while for manure frass, the price has to go down to $8.60/t to meet break-even point. For ryegrass, the break-even price is about $148.49/t and $40.18/t for vegetable and manure frass, respectively; and for red clover, the price of BSF frass has to be about $46.42/t for a producer to break-even.

Table 32: Break-even point for using BSF frass as soil ameliorant for wheat, ryegrass and soil clover ($/ha)

	Price of BSF frass ($/t)			
	Vegetable frass	% change	Manure frass	% change
Wheat	57.89	-98.39	8.60	-99.76
Ryegrass	148.49	-95.88	40.18	-98.88
Red clover	46.42	-98.71	-	

A similar simulation analysis was conducted to examine the impact of increasing the price of soil organic carbon (currently $30/t). The analysis showed that even if the buying price of carbon is increased to about $100/t, the additional benefit would not be sufficient to cover the additional cost of BSF frass as soil ameliorant for these three crops.

Finally, the recommended rates of BSF frass application used in this model was 10t/ha, which was most often the highest yielding. However, where the yield did not differ significantly, lower rates of frass application would be recommended. We have also ran model simulations for these scenarios, but the results remain negative, although the net losses were slightly lower. The full analyses showing these various scenarios is currently being written for submission to a journal for publication.

Limitations and areas for future research

In this study, the same level of fertiliser was applied to all plots. Hence, the experimental treatments include inorganic fertiliser only (control) vs inorganic fertiliser at various levels of BSF frass application. Thus, the yield benefits from frass are in addition to fertilizer application. In future, a comparative analysis between treatments with no soil ameliorant (i.e., no inorganic fertiliser or BSF frass fertiliser) vs inorganic fertiliser only vs BSF frass fertiliser only will provide additional insights into the comparative economic performance between with and without soil ameliorants (inorganic fertiliser and BSF frass fertiliser). Similar analyses with alternative fertilisers (e.g., chicken manure) could also be conducted.

A second limitation of the study is that the field experiment was conducted in sandy soils. In such a sandy soil type, fertiliser application only gets the production (control), whereas frass application appeared to slow down the movement of urea or frass derived nitrogen. Moreover, as the experiment has only covered one year, we are unable to examine any continued benefit of frass beyond the year it is applied. It is possible that nitrogen inputs can be reduced when frass is also applied. This can benefit regenerative or conservation agriculture. Hence, it will be beneficial to extend the static analysis to multi-year experiments to enable analysis of dynamic changes in the system. This is a potential area for future research.

Finally, the effects measured in this study only included crop yield and soil organic carbon. It is likely that the use of BSF frass would also affect GHG emissions. Future studies could include measurement of GHG emissions to have a more comprehensive assessment of the environmental effects of the use of BSF frass as a soil ameliorant.

Conclusion

In conclusion, while the addition of BSF frass (either vegetable or manure frass) has agronomic and environmental benefits in terms of increased yield and increase in soil organic carbon, at the current market price of BSF frass, it is not economically viable to apply frass as soil ameliorant for wheat, rye grass and soil clover. The market price of BSF frass has to decrease considerably, before BSF frass becomes a viable option as a soil ameliorant for crop production of wheat, ryegrass and red clover.

Table 33: Sources of price and costs

	Price	Source
Wheat	330/t	https://www.awe.gov.au/abares/research-topics/agricultural-outlook/agriculture-overview
Rye and clover hay	400/t	https://www.farmtender.com.au/listing/hay-fodder/clover-hay/clover-rye-hay-330-bales
BSF frass fertiliser	$3.6/kg	https://wallingtons.com.au/black-soldier-fly-larvae-frass-fertiliser-5kg
Soil organic carbon	$30/t	https://www.renewableenergyhub.com.au/market-prices/

3.1.10 Research Activity 5.6 - Develop a granulated and/or pelletised fertiliser product

5.6(a) Optimisation trials for granulation to determine if the process requires other inputs such as heat/water/organics

5.6(b) Optimisation trials for pelletisation based on market analysis undertaken in 5.2(d)

5.6(c) Evaluate the performance of granules and pellets including dissolve rate – slow release and longevity effect on engineering properties and recipes

5.6(d) Ballistic modelling and optimisation of fertilisation products (granules and pellets) for mechanised delivery under different agricultural systems

Research activities 5.6(a-d) are written up as one research report, since these four activities are part of the same body of work. A technical report on the optimisation trials for granulation has been produced (Appendix M). This work was undertaken by the UWA agricultural engineering team (Isobel Sewell, Wesley Moss, Andrew Young, Andrew Guzzomi), in collaboration with Haver, Boecker, and Future Green Solutions.

Summary

The granulation/pelletisation of frass may be beneficial to crop agriculture industries by creating efficient handling, storage, transportation and mechanised dispersal of the product. Such products should reduce financial and energy costs of the operation. Activity 5.6 thus focused on the development of a granulated/pelletised fertiliser product produced using frass resulting from black soldier fly (BSF) reared on pork industry waste stream material. The primary aim was to understand the product's engineering properties to optimise handling, storage, transport and application for commercial use.

The effect of four binding options (no binder, potato starch, lignosulphonate, molasses), at varying concentrations and drying times, on the engineering properties of frass granules were investigated (Table 34, Figure 31). Specifically, compressive strength, sphericity, bulk density, terminal velocity and associated ballistic modelling of granules were determined.

This study has shown that it is possible to produce granules from pork industry waste products. Granules bound with molasses at 4% concentration, irrespective of a 24 or 120 hr drying time, had higher bulk density and significantly ($P < 0.05$) higher sphericity and compressive strength compared to the other binder options and concentrations investigated.

These results suggest that inclusion of a binder and drying is important for granule stability, with molasses at 4% concentration (24 or 120 hr drying time) being the preferred binder compared to those investigated in this study (Table 35).

Aerodynamics testing of leading binder candidates using a purpose-built vertical wind tunnel enabled granule terminal velocity to be measured and drag coefficients inferred. Drag coefficients were found to be approximately 0.69, projected areas approximately 11.6 mm^2 and approximate mass 0.021 g. Based on each binder type granule's average properties, granules exiting a disc spreader horizontally at 21 m/s and with no external wind, Molasses bound particles at 4% could be expected to travel within ~2% of the distance of 8% Molasses bound granule and ~10% further than the other candidates.

It was found that BSF, when reared on pork industry frass, whilst efficiently able to process the bulk of the waste stream, they did not digest the animal hairs. Hairs present in the frass also remained essentially intact post-ball milling whilst frass particle size was rendered finer. A large proportion of hairs then passed through vibrating sieving phases. The presence of hairs interfered with structural integrity of granule production and also may detract costumer adoption. It is therefore recommended that future research investigates the removal of animal hair which may be present in pork industry waste frass, without comprising frass quality, to improve the visual and structural stability of the product. It is also recommended to investigate additional binder concentrations, to optimise inclusion ratio against cost.

Table 34 Binder, concentration (%), form and drying time (hours) used in granule production trial.

Binder	Concentration (%)	Form	Drying Time (hr)
No binder	0	-	22
No binder	0	-	24
Potato starch	10	Powder	27.5
Lignosulphonate	10	Powder	24
Molasses	4	Liquid	24
Molasses	4	Liquid	120
Molasses	8	Liquid	96

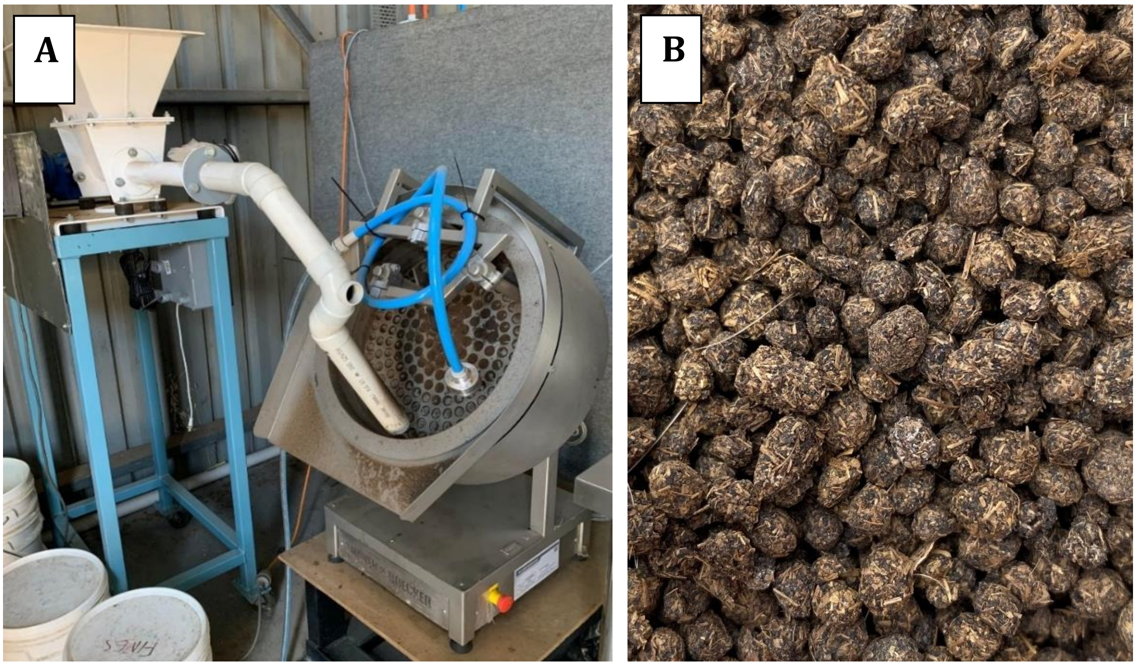


Figure 31 (A) Disc pelletiser (Haver and Boecker) purchased by project partner Future Green Solutions (FGS), and (B) resulting manure-based frass granules.

Table 35 Engineering properties of frass granules treated with two binders (molasses, potato starch), at varying concentrations and drying times

Binder	Molasses 4%	Molasses 4%	Molasses 8%	Potato Starch 10%	***P***
Drying Time (hr)	24.0	120.0	96.0	27.5	-

Moisture (%)	8.02	4.17	5.27	6.00	-
Sphericity	0.94 ± 0.01[a]	0.91 ± 0.01[a]	0.86 ± 0.18[b]	0.82 ± 0.01[b]	< 0.001*
Compressive Strength (MPa)	0.21 ± 0.02[a]	0.11 ± 0.01[a]	0.10 ± 0.03[b]	0.12 ± 0.01[b]	< 0.001*
Bulk Density (g/cm^3)	0.38	0.37	0.33	0.33	-
Drag Coefficient	0.69 ± 0.01	0.71 ± 0.03	0.64 ± 0.03	0.70 ±0.03	0.4238

* Indicates significant differences $P \leq 0.05$. Differing subscript indicates significant differences between pairs.

3.1.11 Research Activity 5.7 - Develop slow release encapsulated fertiliser product

5.7(a) Growth accelerator trials to develop tailored-release nitrogen fertiliser formulations with BSF frass or piggery waste stream materials.

5.7(b) Trials assessing effectiveness of the nitrogen inhibitors, sorbents and/or enhanced efficiency in these formulations

5.7(c) Field trial assessing effectiveness of the highest potential controlled delivery nitrogen fertilisers/sorbent/ enhanced efficiency and/or granulated products.

Research activities 5.7(a-c) are written up as one research report, since these activities are part of the same body of work. A technical report on the development of slow release encapsulated fertiliser products has been produced (Appendix N). This work was undertaken by our research partner organisation Department of Agriculture and Fisheries QLD. The team consisted of Dr Matt Redding, Ms Bri Smith, Ms Tara Rogan, Dr Sheik Rabbi, and Dr Ian Phillips.

Summary
Historically, industrial and agricultural production has followed a largely linear path: consume natural resources, produce a product, generate and discard waste materials, and dispose of the product at the end of its life cycle. In an increasingly resource-constrained world with threatened planetary boundaries (nutrient contamination, greenhouse gas emissions, species extinctions), global citizens have much to gain from replacing these approaches with more efficient, circular economy approaches where wastes become the efficient, valued inputs for other production.

This activity is predicated on the idea that some black soldier fly materials produced from wastes have characteristics that are superior for the formulation of fertilisers. In particular, a key concept for this study is that fertilisers formulated from these materials may allow nutrient cycles to be re-coupled (e.g. carbon, nitrogen, phosphorus, sulphur, and potassium), capitalising on the soil microbiome as part of the nutrient supply process, to replace leaky fertiliser practice. A brief explanation of the prospects for this hypothesis follows.

Conventional fertilisers such as urea deliver N cheaply in a convenient, low-bulk, and high-density form, with many advantages for the user. However, the nutrient use efficiency of these products can be relatively low, with common indications that 40 to 60% of applied N fertiliser

being taken up by the target crop (Figure 1). Recent sugar cane field trial production data from a wet tropics site in Australia found apparent crop uptake of 15 to 30% of urea-N applied. This inefficiency is a consequence of the uncoupling of C, N, and P cycles via the use of conventional highly soluble fertiliser forms. A range of additional mechanisms of increased nitrogen use efficiency (NUE) via organic N assimilation have been proposed. These include shaping the microbial communities that cycle N, recapturing exuded N compounds, and matching the N uptake capacity of the root system to the soil N supply in highly fertilized systems.

Materials that contain exactly the desired target ratios of the C:N and slight excesses of the other nutrients are uncommon, as most waste materials have C:N ratios higher than those of the average soil microorganism. Alternatively, organisms can be used to accumulate nutrients. Techniques include prokaryotic and algal accumulation, and plant uptake in wetlands or ponds. The challenge with extraction via algae and plants is to ensure that the final nutrient content of the accumulating organisms is higher than source waste materials. Black soldier fly (*Hermetia illucens L.*) provides another opportunity for recapturing nutrients in manures and turning by-products (larvae, frass) into animal feed and a residue-based fertiliser. Both defatted protein and larvae of the Black Soldier Fly have nutrient ratios close to the average for soil microorganisms, providing a very good raw substrate for the formulation of this type of prototype fertiliser.

Carbon containing nitrogen sources applied to soils can exacerbate nitrous oxide emissions (N_2O). As the heart of this study was to formulate carbon-containing nitrogen sources and apply them to soil, the study also investigated the impact of the nitrification inhibitor, 3,4-dimethylpyrazole-phosphate (DMPP) (Zerulla et al., 2001), on both N_2O emission and nutrient supply processes.

This research activity sought to develop fertiliser formulations from Black Soldier Fly materials that were:

1. Capable of delivering nutrient over an extended growing period relative to conventional fertilisers,
2. Resist leaching or water-borne losses, and
3. Better synchronise nutrient supply with plant uptake requirements (Figure 1).

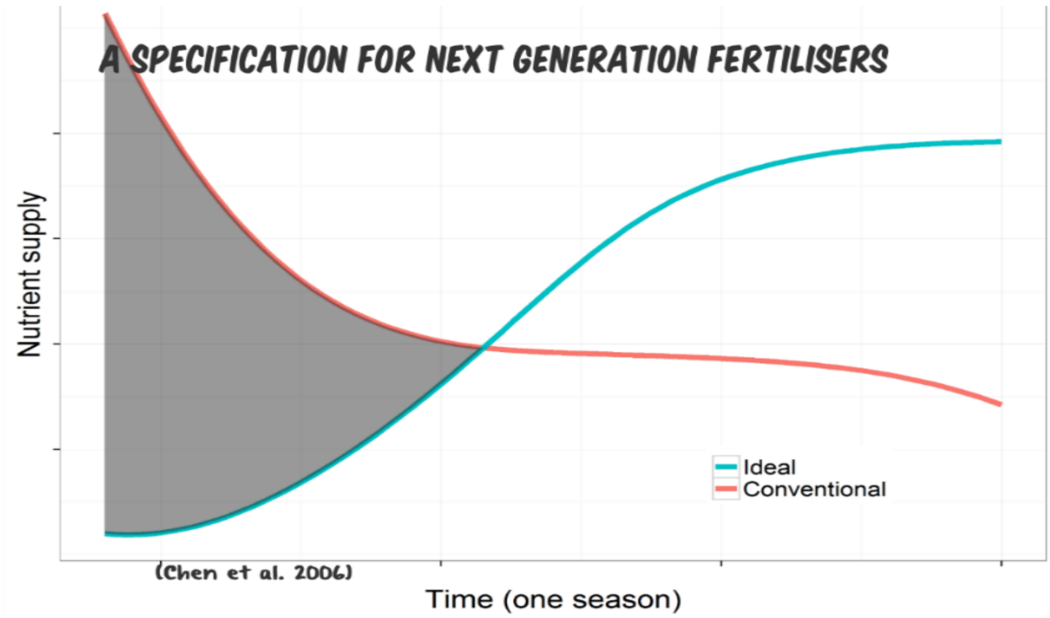


Figure 32. Nutrient release from conventional fertilisers is very poorly synchronised with plant requirements.

Main findings

(a) Growth accelerator trials to develop tailored-release nitrogen fertiliser formulations with BSF frassor piggery waste stream materials.

To produce a superior fertiliser product, the soldier fly larvae and protein were selected as candidate nutrient sources over the frass because the C:N:P:K:S ratios were more favourable (and comparable to soil microorganisms) and total N content was significantly higher than that of the frass.

- The results of the ferrosol growth accelerator trial identified two novel BSF derived fertiliser formulations that are superior to Urea and Urea+DMPP under the experimental conditions.
- Two novel BSF derived fertiliser products produced greater productivity than the urea reference (Figure 33) and in terms of nutrient use efficiency (in the Ferrosol experiment, urea: 18; urea+soldierflydefatprotein+DMPP: 55; urea+soldierflydefatprotein+DMPP: 49 %), greatly improved resistance to water borne losses (Table 36), retention of residual N close to the point of placement and reduced leaching potential (Figure 34).

Table 36. Nitrogen recovered in various fractions for the Ferrol experiment. Note that soil N outside the treatment layer is no included in the "sum of Total N'.

Treat	Leached N	N Uptake	N Uptake > 57 days	Treatment Layer Residual N	Sum of Total N
	%	%	%	%	%
Urea+DefatSoldierflyProt+CarbonRatioAdjusted	54	8	0	27	88
Urea+CaneTrash	62	12	2	14	90
Urea+DefatSoldierflyProt	57	17	1	7	81
Urea	75	18	0	0	92
Urea+DMPP+CaneTrash	26	46	8	21	99
Urea+DefatSoldierflyProt+DMPP+ CarbonRatioAdjusted	10	49	0	14	73
Urea+DefatSoldierflyProt+DMPP	12	55	0	18	85
Urea+DMPP	35	56	1	1	93

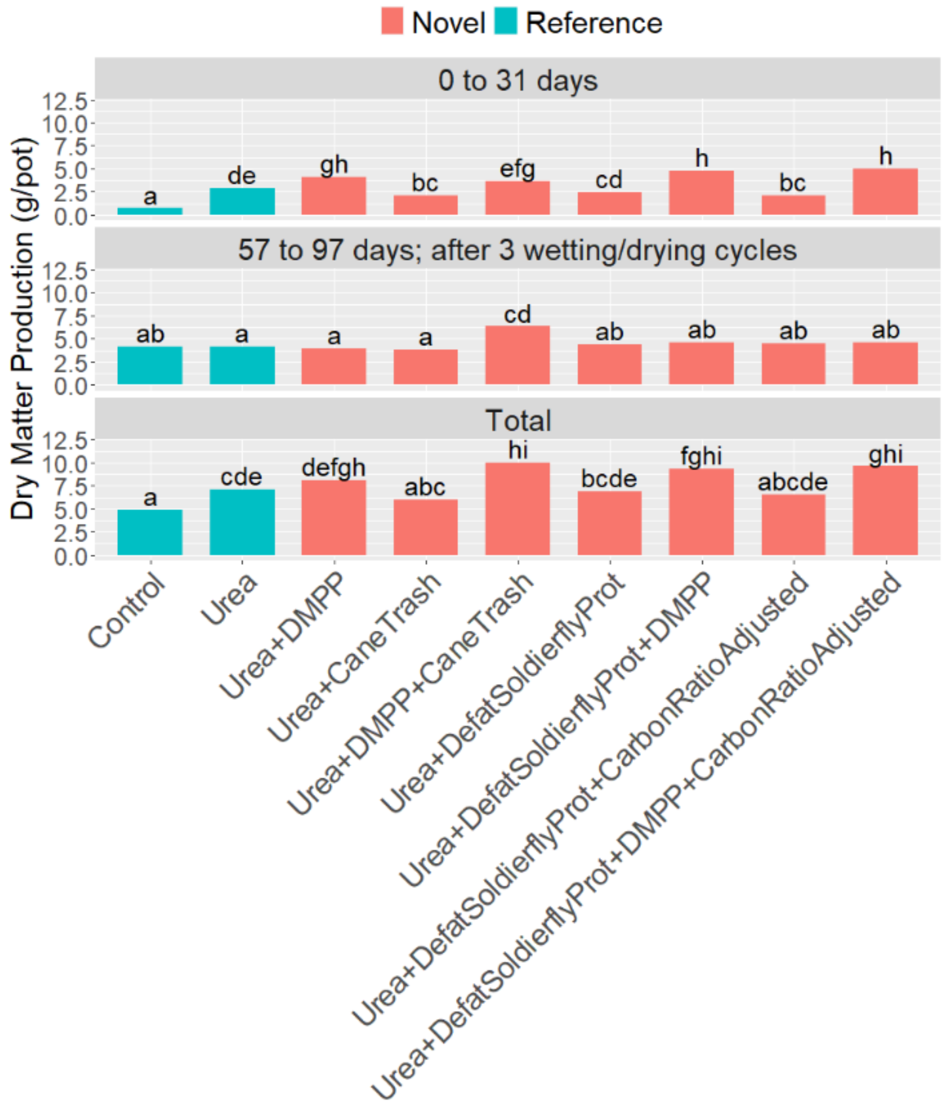


Figure 33 Dry matter production at first, second and total harvest for different formulations applied to Ferrosol soil. Columns appended with the same letter are not significantly different (*P< 0.05*)

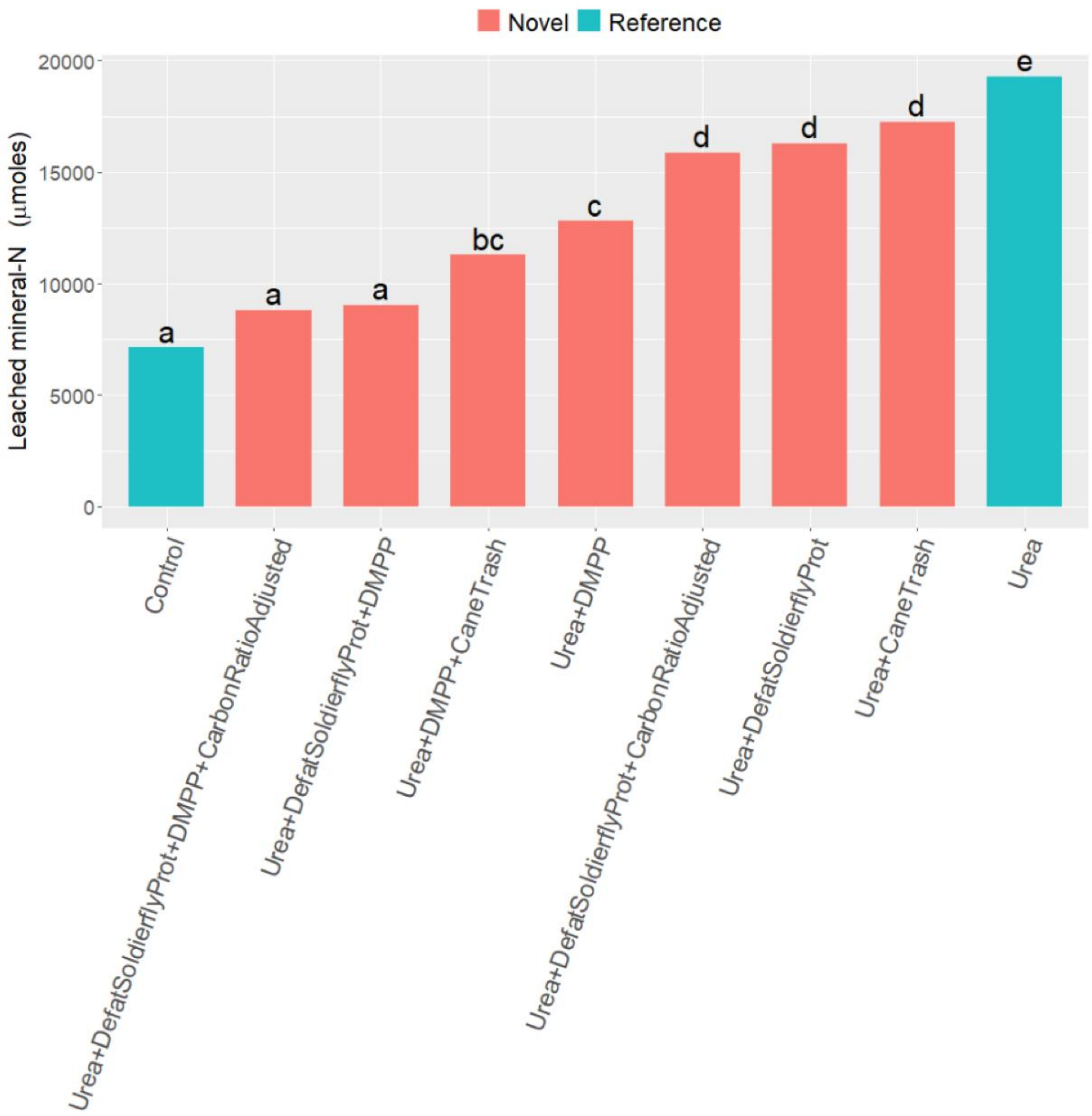


Figure 34. Leached mineral-N for the Ferrosol trial suggests that he novel treatments performed better than urea reference. Columns appended with the same letter are not significantly different (*P< 0.05*)

(b) Trials assessing effectiveness of the nitrogen inhibitors, sorbents and/or enhanced efficiency in these formulations

In addition to the growth accelerator trials that identified several successful inhibitor/enhanced efficiency formulations from soldier fly materials, a detailed reaction vessel trial was also conducted to identify nitrogen mineralisation and inhibitor efficacy. Inhibitor addition to both the defatted soldier fly protein and urea significantly decreased emission peaks and cumulative nitrous oxide emissions (Table 37), while the encapsulated form of the inhibitor had a lesser impact presumably due to the more gradual release of DMPP into the soil system early in the experiment –where emissions were at their highest.

Treatment	Nitrous Oxide nanomoles	LSD
Control	199	a
Urea DMPP	1183	a
Defatted protein DMPP	2552	ab
Urea encapsulated DMPP	4247	b
Urea	7205	c
Defatted protein encapsulated DMPP	7872	c
Defatted protein	11210	d

Table 37. Cumulative nitrous oxide emission when different defatted BSF protein formulations and encapsulation products are applied to the Ferrosol soil reaction vessel trial relative to Urea control. Where the mean is appended with the same letter, the two means are not significantly different (*P<0.05*)

Likewise, the formation of nitrate+nitrite in the soil is significantly slowed by the DMPP forms for both the protein and urea nitrogen additions, while greater nitrogen is retained as ammonium. Retention of nitrogen as ammonia via the use of inhibitors, however, increases the risk of ammonia volatilisation (Table 38; 85 days).

Nitrogen Source	Inhibitor Addition	Ammonia volatilisation (µmoles)	LSD
None	None	3942	a
Protein	None	6749	ab
Urea	None	7353	abc
Urea	Encapsulated DMPP	8177	abc
Protein	Encapsulated DMPP	9135	bc
Urea	Solution DMPP	11092	c
Protein	Solution DMPP	16417	d

Table 38. Ammonia volatilisation when different defatted BSF protein formulations and encapsulation products are applied to the Ferrosol soil reaction vessel trial relative to Urea control. Where the mean is appended with the same letter, the two means are not significantly different (*P<0.05*)

For a detailed account of the methods and project outcomes of the three key activities, refer to the full report in Appendix N.

3.1.12 Research Activity 5.8 - Develop a high quality animal feed product from approved waste materials (horticulture and meeting processing)

Activity 5.1 (KPI 3.4) and 5.8 (KPI 4.28) have been integrated into a desktop study on Nutritional analysis of Black Soldier Fly for intensive livestock (Appendix O) and a factsheet on Nutrition of animal feed products produced from Black Solider Fly larvae was produced.

Summary of report

BSF has been shown to be suitable as feed for ruminants, poultry pigs and fish. Meta-analyses have been conducted for salmonids and for all fish species.

Animal feed and other valued products can be created from Black Soldier Fly (BSF) larvae reared on a wide range of substrates, allowing the possibility of using alternative organic waste streams. However, there are regulatory challenges that have yet to be resolved in using certain types of substrates as such as animal manures if the intended end use of the BSH meals is to feed other animal species. For example, in Europe insect derived meals fed on waste streams are treated as animal by-products and, as such, are not allowed to be fed to pigs or poultry (Commission Regulation (EU) No 14/2011; Regulation (EC) No 1069/2009).

Although BSF Larvae can be fed fresh on a small scale, their greatest potential as an animal feed ingredient is when dried. BSF larvae meals are generally high in protein (36-66%), fats (5-39%), and contain useful levels minerals and vitamins. A component of all insect meals is the indigestible structural polysaccharide chitin. BSF larvae proteins are nutritionally similar to other commonly used protein sources such as soybean meal and fishmeal (Table 1). Variations in BSF ingredient nutrition quality and price are very important considerations for commercial production animals that require optimised nutrition.

Table 39 Comparison of Amino Acid levels in Black Soldier Fly meal with common protein feeds

Amino Acid (%) in Crude Protein

	Fish Meal, CP>60%[1]	Argentinian Soybean Meal, CP 46.2%[1]	Canola Meal[3]	Defatted Black Soldier Fly Meal[2]
Amino Acid	Mean(%)	Mean(%)	Mean(%)	Mean(%)
LYS	7.30	6.10	5.66	5.01
MET	2.68	1.34	1.93	1.94
CYS	0.84	1.41	1.76	0.48
M+C	3.52	2.75	3.69	2.44
THR	4.04	3.92	3.97	3.80
TRP	1.05	1.34	1.33	0.71
ARG	5.70	7.26	6.08	4.44
ILE	4.04	4.54	3.84	4.16
LEU	7.05	7.62	6.60	6.50
VAL	4.73	4.77	4.46	7.36
HIS	2.58	2.65	3.00	4.42
PHE	3.83	5.04	3.71	3.82
TYR	2.87	3.45	2.51	5.85

3.1.13 Research Activity 5.9 - Public perception

5.9(a) Conduct consumer surveys (end-users identified in Activities 5.2) to estimate willingness to pay for BSF fertiliser product for different purposes (e.g. home garden market, landscaping, agricultural production)

5.9(b) Conduct consumer surveys to determine public perceptions, attitudes, and emotional response towards BSF fertilisers/feed and existing alternative organic fertiliser products, and how these perceptions and emotions correlate to willingness to buy products

5.9(c) Identify what factors influence consumer perceptions, emotions, and willingness to pay e.g. price of product, type of product use, level of environmental benefits, product granulation, product certification, and individual consumer characteristics.

5.9(d) Evaluate how consumers' emotional response towards BSF fertiliser products could be influenced to increase willingness to buy these products.

Activities 5.9(a) to 5.9(d) were completed as one project activity by a research associate.

Commercial farmers were identified as the most likely consumers for the BSF products developed, given the scale of the BSF facility and potential to produce high quantities of converted BSF products. Target consumers were identified using the Australia's Agricultural Industries 2017 map, produced by ABARES and Department of Agriculture and Water Resources (dairy, horticulture, broadacre, cotton, sugar). Initial focus interviews were conducted with potential end-users mid-2020 (UWA Human ethics approval RA-4-20-5923; KPI 1.27).

The interview results (Figure 40) informed the development of a national survey of potential end-users. This 'end-users' survey' was developed to determine potential end-users' perceptions and willingness to pay for different BSF fertiliser products (KPI 2.29). In addition to interview results, results from the pot-trials and preliminary characteristics of the BSF products developed at the time were used to inform the survey - to ensure alignment between the end-users' survey and the products developed and tested in our project.

A survey was conducted with potential end-users of BSF fertiliser products (KPI 2.30; Appendix Q). The survey targeted dairy farmers, broadacre crop growers, vegetable, and fruit growers. Questions were aimed at understanding growers' current fertiliser use, their preferences for different fertiliser characteristics, and their perceptions of BSF fertiliser products. We used a state-of-the art discrete choice experiment to enable estimation of respondents' willingness-to-pay for different fertiliser characteristics (Figure 41).

Figure 40 End-users interview results (result from 16 semi-structured interviews)

Interviews were conducted with potential end users in the Western Australian grains, pasture and horticulture industries and with a fertiliser agent/advisor. All interviewees were located in the Northern wheatbelt of Western Australia due to proximity to the researchers and COVID travel restrictions.

Information was gathered on the location and type of farming system, the fertiliser products currently used, current soil health management activities, what characteristics of fertiliser products are most important to the interviewee, what are desirable characteristics of new products, and what would be the appetite for sustainably badged products.

Grain growers apply fertiliser in-crop and prior to crops being sown, using different machinery. Pasture growers apply fertiliser using a spreader machine before sowing. Horticulture was solely an irrigated system with the fertiliser dissolved in water before being put through the irrigation line.

The types of products used by grain growers contain mainly nitrogen and potash, although other trace elements are valued in the blend. The pasture growers varied in their fertiliser use, from annual application of a wide range of nutrients to no application of nutrients (reliant on nutrition from previous year's crop). The main nutrients applied by pasture growers were phosphate, nitrogen and sulphur. The horticulture grower used products that contained a large variety of nutrients.

The key factors when choosing a fertiliser product were:

- Price (all interviewees).
- Handling, which includes storage, freight, application (volume density), hard granular (i.e. not dusty) (grains and pasture).
- Evenness of nutrient blend (grains only).
- Assurance that what is listed on the bag is what is in the granule (grains only).

The grain and horticulture farmers suggested that they would like to have information about the bacteria/beneficial microbes in the product. The grain and pasture farmers valued a slow release fertiliser (note that both farm on sandy soil types).

Figure 41 Example of the choice questions that were administered in the end-users' survey to elicit preferences for different BSF product characteristics.

Choice question 2 of 6: Consider the 3 options given below. Assume that these are the only options available, which **ONE** would you purchase? Please choose based on the characteristics of organic fertilisers / soil improvers you prefer and also your financial situation. Consider this question independent of the others.

	Option 1	Option 2	Option 3	Option 4
Form	Fine particles	Granules	Fine particles	None of these
Organic carbon content (w/w%)	60% (600 kg per tonne)	40% (400 kg per tonne)	60% (600 kg per tonne)	
NPK analysis (w/w%)	Low (1–5 N, 1–5 P, 1–5 K)	Medium (6–10 N, 6–10 P, 6–10 K)	Very Low (0.5–0.9 N, 0.5–0.9 P, 0.5–0.9 K)	
Price ($/tonne)	$1,800/tonne	$800/tonne	$150/tonne	

The survey was distributed to stakeholders through industry associations such as Dairy Australia, Hort Innovation, and The Australian Banana Growers' Council. The survey was also advertised on the project website and various social media. A total of 112 responses were obtained. After removing incomplete responses (47), respondents who were not farmers (4), and respondents from outside Australia (13), there were 48 usable responses.

A scientific paper has been completed to report on the estimated willingness-to-pay for BSF fertiliser products (Appendix P; KPI 4.29) and the consumer perceptions towards insect farming (KPI 4.30). Consumer perceptions were reported in Table 23. There were no association between a respondent's perceptions of BSF products and personal characteristics (such as age, gender identity, type of farmer). Of all respondent characteristics collected in the survey, only the 'alternative' farmer variable was significant in explaining preferences. Alternative' farmers were respondents who self-identified as organic, 'regenerative', or bio-dynamic producers.

Willingness-to-pay results were estimated based on mixed logit and latent class logit models (see Table 41 and Appendix P). While the mixed logit model results capture the unobserved heterogeneity in the end-user population, the latent class model results show the market segmentation amongst potential end-users. Farmers in 'Class 1' are more likely to be 'alternative' farmers. The class probability for Class 1 was 57%, indicating that there may be a suitable market for a BSF-based organic fertiliser or soil improver product.

The farmers in Class 1 are willing to pay significantly more for all of the product attributes, which means that the product is best marketed at potential buyers in this segment ('alternative' farmers). The WTP results (Table 42) show that farmers in Class 1 are particularly interested in a high carbon content product, being willing to pay upwards of $900/tonne for a product with a 40%–60% organic carbon content. These farmers are also willing to pay a lot more for product with low or medium nutrient concentrations compared to the 'conventional' farmers in Class 2. These results are not out of line with existing commercially available frass fertiliser products, which retail anywhere between $1,500/tonne to $3,000/tonne depending on the specific product customisation requirements. There are also commercially available blended frass fertiliser products such as blends of BSF frass with biochar (https://www.esenergy.com.au/biochar) that retail in the high volume agriculture market for around AU $2,200/tonne (Earth Systems, pers comm.).

Table 42: Willingness to pay for the attributes of BSF fertiliser/soil improver products. WTP values are relative to the base level. Only estimates significant at p<0.1 are reported.

	Mixed logit model results		Latent class model results			
			Class 1		Class 2	
Attribute	WTP-estimate	(95% Conf. Interval)	WTP-estimate	(95% Conf. Interval)	WTP-estimate	(95% Conf. Interval)
Form (Baseline: Granular)						
Superfine particles	-348	(-612 – -85)	ns		-389	(-644 – -134)
Liquid	ns		ns		-248	(-451 – -46)
Organic carbon content (Baseline: No organic carbon present)						
10% carbon	ns		1,132	(266 – 1,999)	ns	
20% carbon	241	(7 – 474)	585	(-34 – 1,205)	ns	
40% carbon	431	(182 – 680)	934	(305 – 1,563)	203	(-26 – 432)
60% carbon	463	(70 – 856)	1,299	(606 – 1,992)	446	(20 – 873)
NPK Analysis (Baseline: Very low NPK analysis)						
Low NPK analysis	ns		695	(153 – 1,237)	ns	
Medium NPK analysis	678	(383 – 972)	1,385	(761 – 2,009)	520	(214 – 826)

ns = not significant

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3.2 Contribution to program objectives

3.2.1 Generating knowledge, technologies, products that benefit primary producers.

3.2.1.1 BSF farming as a novel wastewater treatment technologies

Livestock industries in Australia produce significant amounts of manure that needs to be managed in a sustainable manner from an environmental and economic perspective. Manure management on Australian farms is a significant cost (estimated at AUD$100-200 million annually) impacting on the productivity, profitability and sustainability of businesses. Offsetting the high cost of synthetic fertilisers with organic manure derived sources would also benefit the productivity and profitability of Australian agricultural production systems.

Major constraints and barriers to solid manure use is the cost of transport and management of the materials (labour, storage, handling and reuse). Poor management of manures can also cause significant impact on the amenity and environment of the surrounding community.

This research project has shown Black Soldier Fly (BSF) farming is a potential waste management solution for livestock and agricultural waste. BSF farming can convert agricultural manures and waste into high quality fertiliser whilst significantly reducing the volumes of manure to be handled and transported. This project shows that treating livestock manures with BSF larvae can reduce manure volumes by over 80%, potentially reducing GHG emissions, odour nuisance, and storage costs. BSF larvae can be reared on livestock manures but can also process spent bedding and carcasses, although waste conversion efficiency is lower in such cases. The BSF larvae rearing process produces a product called ‘frass’ which is a mix of insect excrement, food residue and exoskeletons. Our project investigates the potential use of this frass as an alternative source of fertiliser in the agricultural sector.

Our research conducted in WA and QLD on poultry, dairy, piggery and abattoir wastes has produced the following benefits:

- Reduction in manure volumes of 70-80%
- Demonstrated low risk in pathogens compared to raw manures
- Reduced stable fly emergence compared to raw manures
- Reduced greenhouse gases (GHG) and nutrient leaching compared to raw manures
- Produces a stable product with no odour
- The frass-based fertiliser/soil conditioner product can outperform synthetic fertilisers
- The frass-based fertiliser/soil conditioner product can be granulated to reduce transportation cost and facilitate application

Using BSF technology to turn manures and waste products into highly valuable products that can be recycled back into the production on the farm provides the potential for a closed loop approach that could increase productivity and profitability, reduce waste, labour and transport costs, improve environmental impacts, and contribute to enhancing the sustainability of agriculture in Australia.

3.2.1.2 Development of novel fertilisers from insect meal

This project demonstrated that insect meal from BSF larvae could be converted into a high quality synthetic fertiliser (ammonium nitrate) via the process of protein and amino acid denaturation. The results show that BSF insect meal can be substituted for synthetic fertiliser (urea, DAP) leading to increase crop yield and reduced leaching potential. Inclusion of BSF insect meal derived from manure in fertiliser formulation increases plant growth and N/P utilisation efficiency.

Literature has suggested for several decades that elemental stoichiometry controls the turnover of nutrients in aquatic (Olsen et al., 1986, p. 1), forest, and other terrestrial soil systems (Cleveland and Liptzin, 2007; Spohn, 2016; Zechmeister-Boltenstern et al., 2015). More recent literature has made the connection to nutrient cycling in soil systems clearer (e.g.: Ma et al., 2022; Mehnaz et al., 2019). Incremental to this developing science, the work conducted in this project under Activity 5.7 demonstrated that these phenomena can be manipulated via fertiliser formulations to deliver nitrogen to plants more efficiently and resist losses to the environment.

Two BSF-derived prototype products were screened and evaluated in two growth accelerator trials, proving superior to urea in terms of nutrient use efficiency (in the Ferrosol experiment, urea: 18; urea + BSF protein + DMPP: 55; urea + BSF de-fatted protein + DMPP: 49 %), greatly improved resistance to water borne losses (Activity 5.7), and retention of residual N close to the point of placement. A third simple formulation proved a surprise performer, with many of the advantages of the two soldier fly formulations. This was the addition of a waste carbon source to Urea treated with DMPP (urea + DMPP + cane trash; Activity 5.7)).

3.2.1.3 Novel soil improver (frass) with reduced GHG and leaching potential that enhances soil fertility by overcoming soil constraints

Australia's organically grown produce is growing market with the domestic consumer market predicted to be worth A$2 billion by the end of 2018. This project developed a number of novel frass soil improver products have been tested. Applying manure derived frass to pasture, grain and horticultural crops in WA raised the soil pH leading to better soil function and stability while reducing greenhouse gas emissions and nutrient losses via leaching. Furthermore, pot and field trials found that microbes associated with frass supported shoot production and behaved like a biostimulant of plant growth. Consequently, they outperformed current organic products on the market and therefore have a strong competitive advantage in this growing high value, niche market.

Further modification of the frass via coating, pelletisation or granulation improved the nutrient retention and release to crops as well as making them easy to handle, store, transport and apply. In the granulated forms, CBH may consider back-loading freight trains thereby greatly reducing the transport cost and opening up the broadacre agriculture market.

Currently, soil amelioration practices is about $124/ha/year (annualised value). The average cost of soil amendment across grain growing regions in WA is about $41/ha. This has a benefit:cost ratio of 3:1 and a net benefit of about $83/ha/year. Agricultural semi-arid soils of Western Australia (WA) are characterised by poor soil structure coupled with low soil fertility, organic matter and carbon content making them susceptible to soil water repellence, soil acidity, soil compaction and herbicide resistance. In particularly, subsoil acidity has been identified as having the most significant economic impact a major yield-limiting constraint, affecting 8.5 million hectares of cropping soils in the south-west with an estimated to cost of at least $1.6 million/year in lost production potential.

We found that the application of frass to horticulture could partially ameliorate soil acidity by raising the pH improving N cycling, a major soil constraints across the Western Australia's cropping region. By raising the soil pH, frass could reduce the amount of liming required especially in acidic, semiarid areas of WA. Additionally, the application of frass increased the abundance of beneficial plant growth promoting rhizobacteria that improved root growth by increasing and extending soil nutrient supply.

However, while the addition of BSF frass (either vegetable or manure frass) has agronomic and environmental benefits in terms of increased yield and increase in soil organic carbon, at the current market price of BSF frass, it is not economically viable to apply frass as soil ameliorant for wheat, rye grass and soil clover. The market price of BSF frass has to decrease considerably, before BSF frass becomes a viable option as a soil ameliorant for crop production of wheat, ryegrass and red clover.

3.2.2 Strengthening pathways to extend the results of rural R&D, including understanding the barriers to adoption

While the financial costs of Black Soldier Fly production were not disclosed, the project's industry collaboration enables a conduit to commercialisation should the economic conditions be favourable - particularly given the positive results in terms of waste reduction potential, GHG mitigation, and the development of potential fertiliser products from BSF frass or protein.

3.2.2.1 Overcoming barriers to adoption by reducing the risk of pathogen and pests

The growing Western Australian Broiler industry produces in excess of 225,000m^3 of spent broiler litter per annum. By-products of intensive animal production have long been recognized as an important soil amendment, providing nutritive value for crops and pastures as well as soil health attributes through increased carbon storage and the development of robust soil microbial populations. Unfortunately, poor management practices in the past have led to stringent application restrictions being imposed through Health (Poultry Manure) Regulations 2001 and more recently through the Biosecurity and Agriculture Management Act 2007 (BAM Act). The regulation of manure disposal options have led to loss of important manure marketing options causing significant cost increases (>4 million) to the WA broiler growing industry.

Currently the Western Australian compost industry does not have sufficient scale, capacity or end market to process the entire allotment of broiler litter and therefore large quantities of litter are transported to horticulture and broadacre agricultural zones for pasture and crop fertilisation. The broiler industry recognises the importance of responsible manure management and therefore proposes to further develop application framework to ensure that the remaining markets are not closed to manure application, causing significant escalation of manure disposal costs and threatening the viability of the local poultry industry.

Recently, opportunities have arisen that could open up new markets for the use of spent litter in broadacre. To exploit these opportunities, guidelines on the best management practices for its land application are needed to avoid stable-fly outbreaks whilst improving crop productivity and soil quality.

Overall, the field trials showed that horticulture soils amended with manure frass exhibit very low emergence of stable fly and other nuisance. Additionally, using BSF waste technology to process manures and wastes led to a significant reduction in pathogen loading for majority of species.

Based on these research findings we will seek an amendment to the Biosecurity & Agricultural Management Act 2007 permitting manure derived frass to be applied to horticulture in previously banned Shires.

3.2.2.2 Understanding farmers perceptions toward Black Soldier Fly technology

The socio-economic work conducted in this project assessed farmers' attitudes towards using Black Soldier Fly as a waste management technology and using BSF-based fertiliser or soil improver products. The research overwhelmingly showed that farmer participants had no inherent disgust towards Black Soldier Fly-based products (Table 43) and that BSF technology could improve the sustainability of the agricultural industry. This means that farmers' feelings of disgust do not present a barrier to adoption.

Table 43 Research participants agreement/disagreement with statements about Black Soldier Fly products and technology (% of total responses)

Statement	Disagree / somewhat disagree	Neutral	Somewhat agree / Agree
Producing organic fertilisers or soil improvers from livestock wastes using Black Soldier Flies is disgusting	94%	2%	4%
Organic fertilisers or soil improvers produced from livestock wastes using Black Soldier Flies will have an unpleasant odour	53%	36%	11%
Producing organic fertilisers or soil improvers from livestock wastes using Black Soldier Flies is a better alternative to handle agricultural waste compared to landfilling	8%	6%	85%
Organic fertilisers or soil improvers produced from livestock wastes using Black Soldier Flies will contribute to improved environmental sustainability of the agricultural industry	2%	21%	77%

The market research conducted in this project showed that farmers who self-identified as organic, bio-dynamic, or 'regenerative' farmers were more likely to purchase BSF-based fertiliser or soil improve products (Activity 5.9). This segment of the population is the most likely potential target market for a BSF based fertiliser product. Nevertheless, 'conventional' farmers showed a willingness to test the product, provided that the product:

- … can be applied using current machinery
- … has a clear benefit in terms of organic carbon or macro-nutrients content

- … has a price comparable to currently use fertiliser products (which will vary per product and over time)
- … macronutrient content meets maximum variability constraints
- … has been demonstrated through extended field trials

Given these findings, extending the results of this project would require (a) scale-up of BSF waste management technology and demonstrated on-farm application; (b) fine-tuning of BSF substrate to achieve a desired and consistent fertiliser product; and (c) on-farm field trials at trusted grower sites (e.g. through local grower groups) to demonstrate potential yield benefits of using BSF-based fertiliser or soil improver products.

3.2.3 Establishing and fostering industry and research collaborations that form the basis for ongoing innovation and growth of Australian agriculture

This research has cemented the collaboration between industry partner Future Green Solutions and the research providers. Further collaboration can grow this foundational project into a commercial fertiliser or soil improver product.

The knowledge gained from this project can be extended to other insect industries, with the potential to expand production around Australia. The UWA research team is currently working on a Cooperative Research Centre (CRC) proposal on Net Zero Emissions from Agriculture (with the University of Queensland and industry partners). The proposed CRC will further the knowledge gained in this project, but developing low-emissions, circular solutions to agricultural waste management. Such low-emissions, circular solutions are needed to enhance the sustainability of Australian agriculture, to meet local consumer demands for sustainable products, and ensure continued access to overseas export markets.

4 Collaboration

This project involved a collaboration between multiple research disciplines, as well as collaboration with industry, primary producers, and policy makers. The project contributed to the innovation and growth of insect farming as a new emerging industry in Australia that will benefit Australia's agricultural industries by reducing the cost of waste treatment and disposal, and producing sustainable alternatives to (currently costly) fertiliser products.

From the commencement of this project, a collaborative team was set up between Australian Pork limited and its industry partners Dairy Australia, Fisheries Research and Development Corporation, AgriFutures, Australian Egg, Australian Meat Processor Corporation, Future Green Solutions, The University of Western Australia, and the Queensland Department of Agriculture and Fisheries. These relationships have been strengthened through regular teleconferences throughout the lifetime of the project. This has enabled the project partners to contribute to the direction of the research and engagement through their representation on the project's formal Steering Committee. The collaborative project team meetings helped to bring together people working across the intensive industries. As recorded in the mid-project evaluation, the people involved in these collaborations might not have interacted if it had not been for this project.

Over the course of the project, the University of Western Australia and partners have built a network of relevant organisations, including government bodies (DPIRD and DWER), consulting firms (Rob Wilson Consulting and Scolexia consulting), grower groups (Mingenew Irwin Group), industry representatives and industry bodies (GRDC, Western Australian Primary Principals' Association, Western Dairies, and Western Australian Broiler Growers). Grower groups and industry bodies were directly consulted in the socio-economic work that aimed to assess farmers interest and preferences for BSF waste-management technology and BSF-based products. Through these collaborations we have kept industry informed on the emerging scientific evidence that BSF technology provides solutions for reducing biosecurity and environmental risks associated with waste management.

The project team members have also built collaborations between different industry sectors including pork, meat processing, poultry, and dairy, which has been fruitful on the research front as a blended mix of waste is proving the best substrate for the growth of the BSF larvae. Creating stronger ties between the industries has also had side benefits such as increasing discussions between partners due to greater understanding of industry priorities and willingness to engage on discussions around further collaborative projects. Some relationships have been slow to develop due to several changes in partner representatives, including a new project manager appointed during the COVID-19 lockdown. Some of these new collaborations include other academics and researchers within the University of Western Australia through sharing information and linkages to broader networks.

As a result of effective collaboration between the industry partners, the project has benefited from in-kind contributions of equipment to the project. For example, the use of Future Green Solution's disc pelletiser was instrumental to conduct some of the research work under Activity 5.6. The use of the Indian Ocean Marine Research Centre Molecular Lab allowed us access to the latest technology for conducting DNA extractions and quantification for microbial community analysis to conduct research work under Activities 5.3e, 5.4e and 5.5a. Information sharing among researchers in the lab also greatly assisted in improving the quality of our molecular results.

The formal collaborative links between research partners contributed significantly to the work conducted for Activity 5.7. In particular, all BSF-derived materials were provided by Future Green Solutions Pty Ltd, and significant information regarding caveats on the use of Black Soldier Fly derived materials in soils were provided by the UWA agronomic team.

Under Activity 5.2 and 5.9, the project explicitly involved stakeholders through interviews and survey activities, covering both waste producers and potential end-users of the BSF products. The involvement of the stakeholders subsequently increased the awareness and acceptance of BSF farming and helped to ensure that the BSF products being developed met the requirements of industry. We have also received some ad hoc enquiries from the public about Black Soldier Fly farming.

Informal collaborations between our researchers have contributed towards a series of outcomes for the project, including co-supervision and co-publications. For example, a manuscript section “A means to no ends – Frass as Fertilizer” for the review “Food for thought: valuable insect production pathways emerging from the transition to a circular food production model” has been submitted to the academic journal Nature and was funded by the Fisheries Research & Development Corporation (‘Insect Protein for Aquaculture Feed’).

Further afield, the in-kind collaboration with researchers at international universities has been productive when designing experiments and extending research results, including Wageningen University and University of Newcastle (UK). Another example has been working with Dr Hannes Schmidt from the Division of Microbial Ecology at the University of Vienna, who allowed us to conduct ddPCR on samples from Activity 5.3e. This extra data added value by allowing us to draw stronger conclusions about pathogen persistence in BSF products.

Importantly, the collaborations are leading to potential funding for emerging projects to support the ongoing research and development in this area. In particular, collaborations with Australian Meat Processor Corporation through Future Green Solutions, and research proposals with a number of Cooperative Research Centre (CRCs), including CRC for Plastics, CRC for protein invitations, CRC for Zero Net Emissions in Agriculture and Critical Supply Chain CRC.

5 Extension and adoption activities

Overview of activities

A Communications and Extension Plan was developed for this project, and later updated to include COVID-safe extension and outreach activities, which was approved by the Department for implementation.

The below table lists all communication and extension activities undertaken over the life of the project to date. The table also lists collaborations and engagement with stakeholders, which is part of the Communication and Extension Plan. The plethora of activities aimed to support adoption of the research outcomes. Note that we can not yet report on all outreach and extension activities, as we are actively growing post-final results over the next 6 months. There are some items in the list that are near completion or pending approval before they can be distributed.

The project has conducted a full range of extension activities including presentations and poster presentations at conferences (eg Future of Food Conference and Expo, TerraEnvision 2022), partner meetings, industry events, and field days (eg. UWA Farm Field Day) to increase awareness about insect farming and black soldier fly as a new waste treatment technology.

Media releases, an ABC primetime news segment, radio interviews, social media posts and newsletter articles have been published to keep stakeholders informed and anticipating results. The project has also developed a series of project factsheets and a professional video, which are published on the project website and circulated widely to stakeholders and partners upon approval.

An external-facing project website was developed in the second year of the project and has been kept up to date providing a central online location for all project information, acknowledgement of funding bodies, research profiles of the research team, news, contact details, and a location to provide accessible links of all types of publications developed by the project team. Where possible, data sets have also been made available via this project website.

Further, the project team attended the Extension Aus Bootcamp Program in December 2021 to learn more about the Community of Practice program and tips and tools to maximise the program. A dedicated webpage hosted by AgriFutures Australia on their Extension Australia portal was set up and has been kept updated.

The extension activities with growers was mostly done via participation in the research surveys. The two nation-wide online surveys were accompanied by a media strategy to ensure stakeholders were aware of the project. For example, relevant industry networks were accessed and introduced to the project before requesting assistance with information collection via industry newsletters and social media. These new connections are now established and will allow us to send invitations to the final webinars and distribute factsheets (and other promotional materials such as the final video).

Extension discussions to identify the thinking of the regulators as to the potential classification and likely requirements to meet certain classifications (compare composting, fertiliser, and manure standards) were undertaking in all states. These discussions were extremely hard to engage the right person.

A major barrier to some of the extension and adoption activities was generated by the outbreak of COVID-19 and the ongoing restrictions. The inability to travel to Western Australia during the COVID 19 pandemic prevented face to face extension activities going ahead as planned with regulators and policy makers. Further, border closures limited access to researchers and policy makers, which prohibited stakeholder communication and engagement during the project timeline. Inability to access potential demonstration sites and lack of face-to-face meetings created further delays in implementing the Communication and Extension Plan. However, video conferencing was used successfully between the project teams to progress extension activities, where possible. Utilising state-based staff for specific individual state-based activities assisted in progressing extension and adoption activities during COVID-19 restrictions.

Recommendations to facilitate adoption

Open communication and transparency need to be provided to the regulators and decision makers in order to facilitate and expedite the adoption of BSF products in Australia. Confidentiality can still be maintained in the public arena, however maintaining confidentiality and providing partial information will only risk creating uncertainty, confusion and the likely mis classification or application of policy, guidelines and standards that are not commensurate with the level of risk of the product. Extension of the process, product and benefits to the Agricultural departments and industry will also likely create interest in the adoption of the product.

To facilitate a greater understanding of the process and the product it is recommended that the BSF producers in Australia collaborate to produce a report that outlines in detail for regulators:

- The potential process of BSF farming
- The potential feedstocks likely to be used
- End product specifications (and likely classification)
- Australian and international research data- environmental, human and animal benefits

For both producers and regulators (to demonstrate general environmental duty), the following need to be prepared:

- Best Practice recommendations for reuse
- Duty of care statement
- Economic, agronomic and environmental analysis of BSF Frass compared to conventional fertilisers and raw manures.

Once this project has validated a proof-of-concept for using Black Soldier Fly larvae for livestock waste management, we expect the next step to involve demonstration sites where the use of BSF can be shown in real-life farming settings.

The following table lists all collaborations, stakeholder engagements and extension activities, including communication activities aimed at supporting extension and adoption.

Nature of activity	Number	Description
Articles for rural press, media releases, blogs and communications networks.		
Press Releases	3	1. Australian Pork Limited press release 2. University of Western Australia press release Link 3. Future Green Solutions press release Link
Newsletter articles	7	1. The UWA Institute of Agriculture Newsletter, December 2019 (page 4). Link. 2. Australian Eggs' "Eggstra" September 2020. Link. 3. National Poultry Newsletter, July 2021 Link 4. The UWA Institute of Agriculture Newsletter, August 2021 (page 4). Link.

		5. pig333.com Link 6. Queensland Country Life Link 7. The UWA Institute of Agriculture Newsletter, April 2022 (page 7). Link.
Rural Press	1	1. ABC News Link
Blog Posts	7	1. AgriFutures' Extension Australia website: a. What is black soldier fly, Reasoning behind the project Link b. Introduction to the Project – Turning Animal Waste into Fertiliser using BSF Link c. Understanding Current Waste Management Practices – Interviews Link d. Drivers and barriers to adoption of black soldier flies Link e. BSF Technology in short; what we know so far Link 2. UWA's Centre for Agricultural Economics and Policy a. Where is most livestock waste produced? Link b. ABC news informs the agricultural industry on Black Solider Fly research at UWA Link
Informal updates & briefing	6	1. Australian Pork Limited Update (4th September 2020, available in the project's Mid-Term Review Report) 2. WA DWER (2020) 3. 5x Project Updates a. Project Update #1: January - December, 2020 Link b. Project Update #2: January - June, 2021 Link c. Project Update #3: July - December, 2021 Link d. Project Update #4: January - June, 2022 Link
Media		
Twitter	14	Hashtags #BSFwastetoprofit #blacksoldierflyuwa 1. @IOA_UWA on 3rd December 2020 Link 2. @SAgE_UWA on 4th December 2020 Link 3. @Daniel_Kidd01 on 12th November 2021 Link 4. @RegenWa on 29th June, 2021 Link 5. @maritkragt on 7th November 2021 Link 6. @Daniel_Kidd01 on 25th August 2021 Link 7. @IOA_UWA on 13th August, 2021 Link 8. @Daniel_Kidd01 on 8th November 2021 Link 9. @maritkragt on 6th March 2022 Link 10. @uwaceep on 17th January 2022 Link 11. @IOA_UWA on 24th August, 2022 Link 12. @IOA_UWA on 1st July, 2021 Link 13. @FutureGreenSol on 22nd July 2022 Link 14. @AREatUWA on 1st April 2021 Link
Radio Programs	1	ABC's Great Southern and South West Rural Report (8th November, 2021). Link
TV Programs	1	ABC 7pm News (6th November, 2021). Link
Websites		

Main website	1	1. Project website https://www.bsfwastetoprofit.com/
Project webpages on associated websites	4	1. A Community of Practice website hosted by AgriFutures Australia on their Extension Australia portal: https://extensionaus.com.au/blacksoldierfly/introduction-to-the-project/ 2. Research Project under the UWA Centre for Engineering Innovation: Agriculture and Ecological Restoration: https://www.uwa-cei.org/blacksoldierfly 3. Research Project under the UWA Centre for Agricultural Economics and Development: https://www.uwacaed.org/research 4. Future Green Solutions: https://www.futuregreensolutions.com.au/
UWA webpages for student projects (now expired)	2	1. PhD project https://www.uwa.edu.au/projects/consumer-perceptions-towards-insect-farming-products 2. PhD Project https://www.uwa.edu.au/projects/developing-and-assessing-a-high-quality-fertiliser-product-based-on-black-soldier-fly-frass
Videos		
Project video	1	Closing the loop: Black Soldier Fly technology to convert agriculture waste (2022). YouTube (Forthcoming, 2022). Available from the project website: https://www.bsfwastetoprofit.com/blog
Short videos for social media content	2+	@Degleeson on 24th March 2021 Link @jp_wisdom on 19th May 2021 Link More are under development, coming soon, available from project website: https://www.bsfwastetoprofit.com/blog
Formal and informal reports, publications, posters, factsheets, tools and other project material		
Peer reviewed journal articles	8	Submitted: 1. Kragt, M.E., Dempster, F. & Subroy, V. (2022) *Black Soldier Fly can enhance circularity in agricultural production: Farmers' perceptions and willingness-to-pay for insect-based fertilisers*. Journal of Cleaner Production. 2. Jenkins, S.N., Middleton, J.A., Huang, Z., Mickan, B.S., Andersen, M.O., Wheat, L., Waite, I.S. and Abbott, L.K. (2023). *Combining frass and fatty acid by-products of Black Soldier Fly larvae farming has potential as slow release fertiliser*. Science of the Total Environment In Preparation: 3. Gurung, S.K., Mickan, B.S., Jenkins, S.N., Middleton, J.A., Wheat, L., Rengal, Z., Siddique, K.H.M. and Solaiman, Z.M. (2023). *De-bottlenecking the black soldier fly manure biorefinery: utilising the frass for fertiliser and*

		impacts on soil biological processes. Journal of Cleaner Production. 4. Langa, S.P., Middleton, J. A., Gleeson, D.B. and Jenkins, S.N. (2023). *Processing poultry manure with Black Soldier Fly technology lowers greenhouse gas emissions from soil*. Science of the Total Environment 5. Middleton, J. A., Martin, B. and Jenkins, S.N. (2023). *The fate and persistence of pathogen in frass amended soil*. Science of the Total Environment 6. Gurung, S.K., Jenkins, S.N., Wheat, L., Rengal, Z., Siddique, K.H.M. and Solaiman, Z.M. (2023). *Waste-to-value: utilising black solider fly frass for nutrient recovery from industrial waste for fertiliser.* Insects. 7. Enkhbat. G., Kidd, D., Wisdom, J., Middleton, J. A., Jenkins, S.N., Wheat, L., and Ryan, M. (2023). *The effect of Black Soldier fly frass and irrigation on the productivity and yield of agronomically important crop and pasture species.* Journal of Cleaner Production 8. Cook, D. Waite, I.S., Middleton, J. A., Wheat, L., and Jenkins, S.N.(2023). *Applying Black soldier fly frass to horticulture reduces stable fly emergence.* Insects
Technical Reports	9	1. Tascon, A., Dempster, F. & Kragt, M. (2021). *Spatial Analysis of Farm Animal Waste in Australia*. The University of Western Australia. 2. Langa, S., Middleton, J., Gleeson, D. & Jenkins, S. (2022). *Processing poultry manure with Black Soldier Fly technology lowers greenhouse gas emissions 2 from soil*. The University of Western Australia. 3. Dempster, F. Harold, T., Subroy, V. & Kragt, M. (2022). *Market potential for Black Soldier Fly fertiliser products.* The University of Western Australia. 4. Dempster, F. Harold, T., Subroy, V. & Kragt, M. (2022). *End-users' perceived benefits and perceptions of insect farming*. The University of Western Australia. 5. Pearson, L. and Wilson, T.B. (2022). A Review of the Biosecurity Risks of Using Black Soldier Fly (Hermetia illucens) in the Australian Agriculture Sector. Scolexia. 6. Quinteros, J.A., Wilson, T., Pearson, L., Price, J., Kragt, M. and Scott, P. (2022). Potential environmental risk of Black Soldier Fly (Hermetia illucens) By -Products: A review of the current literature. Scolexia and The University of Western Australia. 7. Bruerton, K., Pearson, L. and Scott, P. (2022). Nutritional analysis of Black Soldier Fly for intensive livestock. Scolexia. 8. Pearson, L., Wilson, T.B. and Scott, P. (2022). Pathogen and Heavy Metal Loading at the Manure Input, Larval and Frass Stages of Black Soldier Fly Production. Scolexia. 9. Price, J. (2022). Potential Environmental Regulation Classification and Regulatory Requirements for Manure Derived Black Soldier Fly Frass. Scolexia.

Performance Reports	5	1. Milestone Report 2 2. Milestone Report 3 3. Mid-Term Review 4. Milestone Report 4 5. Final Report
Datasets	1	1. Tascon, A. (Creator) (4 Aug 2021). Spatial Analysis of Farm Animal Wastes in Australia. The University of Western Australia. Excel file available at: 10.26182/r3zp-4j91
Fact Sheets for industry	6	1. Biosecurity Risks of Black Soldier Fly (BSF) Larvae as an Animal Feed (Appendix C) 2. Fact of Fiction: Black Solider Flies Link 3. Nutrition of BSF products used for animal feed Link 4. Creating fertilisers from agricultural waste using Black Solider Fly farming (Appendix I) 5. Stable Fly emergence Link 6. Potential environmental regulation classification and requirements for manure derived Black Soldier Fly frass Link
Economic tools	2	1. Benefit Costs Analysis Tool (available upon request, used in Research Activity 5.5d) 2. Benefit Transfer Tool (available upon request, used in Research Activity 5.5d)
Imagery	100+	1. Photographs of project work and BSF technology imagery. Files stored in Google Photos Drive Link 2. Black Solider Fly Technology – Research End Users (2021). *UWA researchers are developing innovative solutions for converting agricultural waste into valuable commodities*. Poster. University of Western Australia. 3. UWA Shenton Park Research Facility (2021). *Black Solider Fly Field Trials*. Poster. University of Western Australia.
Banners	2	UWA Soil Science Future Green Solutions
Non-academic (industry and government) engagement, including conferences, meetings and forums		
Presentations and formal discussions	7	1. Presentation on *Reducing greenhouse gas emissions with Black Soldier Fly frass* delivered by E/Prof Lyn Abbott and student Silvestre Lange at the Soil amendment industry forum in Brookton, 9th July 2019. 2. Dr Kragt discussed the project at Australian Farm Institute's Agriculture Round Table in Canberra, 15 Oct 2019. 3. Presentations on *Soil biological fertility and climate resilience* and *Building carbon, what are the options for dryland ag soils, what works, and where is the evidence?* delivered by Prof Lyn Abbott at Talkin' Soil Health hosted by Wheatbelt NRM in York on 12th March 2020. Link 4. Meeting with Minister MacTeirnan and Minister Amber Jade-Sanderson (as Environment Minister) at the Shenton Park Research Facility to present how BSF technology works. Delivered by Luke Wheat. 5. Presentation on *Manure as a Substrate for Black Soldier Fly Larvae Rearing: An Australian Analysis of Biosecurity Risks* delivered by Lachlan Pearson eon the Insect Industry

		Program at The Poultry Information Exchange (PIX) and Australasian Milling Conference (AMC) event on Swine and Poultry, Gold Coast, 15-17th May, 2022 6. Panel member for discussions at The Poultry Information Exchange (PIX) and Australasian Milling Conference (AMC) event on Swine and Poultry, Gold Coast, 15-17th May, 2022 7. Presentation on performance of BSF farming on agriculture waste streams and the manure frass v untreated manures for stable fly establishment in horticultural crops at the Annual General Meeting of the Stable Fly Action Group in Gingin, Western Australia, October 2022.
Meetings and connections made with industry and research organisations (degree of involvement varies)	57	1. Almonds Australia 2. AgriFutures Australia 3. Agrifutures 'Ignite Network' 4. Animal Health Australia 5. Association of Australia, Dairy NSW, 6. Australian Banana Growers' Council 7. Australian Eggs 8. Australian Farm Institute 9. Australian Meat Processor Corporation (AMPC) 10. Australian Pork limited (APL) 11. Birchip Cropping Group 12. CSBP (part of Wesfarmers Group) 13. Dairy Australia 14. Elders 15. Fisheries Research and Development Corporation (FRDC) 16. FlyFarm 17. Future Green Solutions (FGS) 18. Grain growers 19. Grains Research Development Corporation (GRDC) 20. Grassdale Fertilisers Pty Ltd 21. Grower Group alliance 22. Harvey Beef 23. Hort Innovation 24. Incitec Pivot 25. Indian Ocean Marine Research Centre 26. IAPA Association 27. Insect Association of Australian 28. Insect Protein Association of Australia 29. Liebe Group 30. Meat & Livestock Australia 31. Mingenew Irwin Group 32. Moora-Miling Pasture Improvement Group 33. National Farmers' Federation 34. North Central Catchment Management Authority (VIC) 35. Nuffield Australia 36. NSW Farmers' Association 37. Perth NRM (and Wheatbelt) 38. Rangeland 39. Regen WA

		40. Rob Wilson Consulting 41. SoilsWest 42. Southercross Agricultural Exports 43. Summit Fertiliser 44. South West Catchments Council (WA) 45. Stock Feed Manufacturers Council of Australia 46. The Australian Banana Growers' Council 47. University of Western Australia (UWA) 48. UWA Institute of Agriculture 49. UWA Smart Farms National Landcare Project 50. Vegetables WA 51. Western Australian Broiler Growers 52. WA Citrus and Horticulture Innovation Australia 53. West Australian Pork Producers' Association 54. Western Australian Primary Principals' Association 55. Western Dairies 56. Wine Australia 57. Worrolong Produce
Workshops with key industry stakeholders	1	Industry representatives from Margaret River and Augusta regions interested to trial BSF technology (3rd August 2020)
Industry networking events	6	1. *Extension Aus Bootcamp Program* at AgriFutures, December 2021. 2. *The Future of Food Conference and Expo*, Mandurah, 23-34th September 2021. 3. *The Asian Food and Feed Insect Association Annual Event*, Bangkok and Online, 16-18th of June 2021. 4. Insect Oil: A new tool driving sustainable Pig Production. *Pig Progres Webinar* Dr. Graziano Mantovani Cargill Animal Nutrition Chloé Phan Van Phi InnovaFeed Vincent ter Beek Pig Progress. 20th June 2021. 5. Redefining biowaste to insects to feed. Dr Moritz Gold. *Centre for Advanced Food Engineering Seminar Series*, University of Sydney 12th November 2021. 6. *Challenges and opportunities for insect meal in animal feed Webinar* Featured Liz Koutsos, president, Enviroflight, and Chloé Phan Van Phi, head of marketing and sales, InnovaFeed. Thu, July 22, 2021 1:00 AM - 2:00 AM AEST.
Government organisations engaged with	9	1. WA Department of Primary Industries and Regional Development (DPIRD) 2. WA Department of Water and Environmental Regulation (DWER) 3. QLD Department of Agriculture and Fisheries (DAF) 4. QLD Department of Environment and Science 5. VIC Environmental Protection Authority 6. NSW Department of Primary Industries (DPI) 7. NSW Environment Protection Authority 8. TAS Environment Protection Authority 9. SA Environment Protection Authority

Scientific conferences, meetings and forums		
Conferences Presentations	7	1. Presentation on *Black soldier fly technology to convert piggery manure into fertilizer* delivered by Denise Woods at the Australasian Pig Science Association (APSA) 17th Biennial Conference, 17-20th November 2019, Hilton Adelaide, Adelaide. 2. Presentation on *Black soldier fly technology to convert piggery manure into fertilizer* delivered by Dr Sasha Jenkins at the State of Soil Science WA, 4-6th December 2019, UWA Business School, Western Australia. 3. Presentation on *Valuing the costs of waste management in the pork industry* delivered by Dr Sasha Jenkins at the Australasian Pig Science Association (APSA) conference, 18th November 2021. 4. Presentation on *Assessing the potential of black soldier fly technology to convert piggery manure into slow-release fertiliser* delivered by Dr Sasha Jenkins at the Australasian Pig Science Association (APSA) conference, 18th November 2021. 5. Presentation on *Increasing the throughput of fertiliser product screening: machine vision and microdialysis* delivered by Matt Redding at the Soil Science Society of Australia, Cairns 2021. 6. Presentation on *Using BSF technology to turn livestock waste into profitable products* delivered by Dr Marit Kragt at the Terra Envision Conference, Utrecht, Netherlands. 28th June, 2022. 7. Gurung, S.K., Mickan, B.S., Middleton, J., Jenkins, S., Rengel, Z., Siddique, K. and Solaiman, Z.M (2022). *Waste to value: the use of black soldier fly frass for sustainable soil health and food production*. Poster, 22nd world congress of soil science in Glasgow, United Kingdom, 31 July - 5 August, 2022.
Poster presentations	3	1. Gurung, S.K., Mickan, B.S., Middleton, J., Jenkins, S., Rengel, Z., Siddique, K. and Solaiman, Z.M (2022). *Waste to value: the use of black soldier fly frass for sustainable soil health and food production*. Poster, 22nd world congress of soil science in Glasgow, United Kingdom, 31 July - 5 August, 2022.
Meetings with research organisations	4+	1. Wageningen University 2. University of Newcastle (UK) 3. University of Vienna 4. University of Queensland
Engagement on research proposals for ongoing collaborations	8	1. National Soil Science Challenge 2. National Landcare Program – Smart Farming Partnerships. 3. Plastics CRC 4. CRC for protein invitations 5. CRC Zero Net Emissions in Agriculture 6. Critical Supply Chain CRC 7. Grains Research Development Corporation

		8. Australian Meat Processor Corporation (discussions to progress investment decisions in further R&D activities).
Seminars, webinars, trainings and workshop		
Tutorials	1	1. Interactive tutorial on *'The Fly Show' – Use of Black Soldier Fly Technology in waste management* delivered by Dr Jen Middleton for ENVT1104 at UWA in 2022.
Research Seminars	3	1. Seminar on *Closing the loop: Black Soldier Fly technology to convert agricultural waste* delivered by Marit Kragt, Fiona Dempster & Vandana Subroy for the ARE/AARES Research Seminar Series on 21 May 2021 at UWA Link 2. Seminar on *Key research skills in the biological sciences. My research journey: from aquatic ecology to biotechnology* delivered by Dr Jen Middleton for BIOL1131 in 2022 at UWA. 3. Seminar on *Using black soldier fly technology to turn livestock waste into profitable products* delivered by Marit Kragt for the SAGE Seminar Series on 3 November 2022 at UWA.
Webinars	2	Two Webinars for industry partners and project stakeholders are currently being organised to transfer knowledge and research results. These are aimed to be conducted during January – March, 2023.
Postgraduate research supervision related to this grant	5	1. Sun Kumar Gurung is conducting a PhD on Utilising Black soldier frass as a soil improver in horticulture. Supervised by Sasha Jenkins & Kadambot Siddique 2. Maria Belen Castillo is conducting a Masters on Black soldier fly castings as a high quality fertilizer product. Supervised by Sasha Jenkins & Deirdre Gleeson 3. ZhouDa Huang conducted a Masters on coating BSF frass to reduce leaching potential. Supervised by Sasha Jenkins & Lyn Abbott. 4. Silvestre Langa conducted a Masters on assessing GHG potential of BSF frass. Supervised by Sasha Jenkins & Deirdre Gleeson 5. Jing Yi Lin is conducting a Masters on optimising BSF larval growth and conversion rates of manure based feedstocks. Supervised by Marit Kragt
Site visits and farming activities		
Site visits	4	1. Dairy Farm in Margaret River/Augusta region on 4th August 2020 2. Ridgefield Farm in collaboration with Wheatbelt NRM and Smart Farms National Landcare Project on 10th September 2020 3. UWA Students from unit ENVT1104 visited the Black Soldier Fly production facility and trial sites on 24 March 2021. 4. Australian Pork Limited site visit to UWA and Shenton Park represented by Gemma Wyburn, 4 – 6th May, 2022.
Field Days	7	1. Dowerin GWN7 Machinery Field Days, 25th August 2021

		2. UWA Farm Field Day, 10th September 2020 3. UWA Farm Field Day, 11th November 2021 4. UWA Farm Ridgefield Open Day, 3rd September 2021 5. UWA Shenton Park Field Station Open Day, 23rd Sept 2022 6. UWA Shenton Park Field Station Open Day, 12th Oct 2021 7. Dowerin GWN7 Machinery Field Days, 24th August 2022
Industry focus groups, committees, surveys and champions		
WA Industry Focus Group	1	1. Dr Fiona Dempster interviewed 16 end users, consisted of mostly current and past farmers, plus agronomists and senior farm management staff.
Surveys	3	1. Nation-wide survey on the socio-economic costs and benefits and perceptions of insect farming and waste treatments. Sent to dairy farmers, broadacre crop growers, vegetable, and fruit growers. 38 farmers successfully engaged. 2. Nation-wide survey on assessing the availability of livestock wastes and costs of current waste management practices. Sent to livestock producers and meat processers. 16 quality responses received. 3. Mid-Term Project Evaluation. 4 (out of 5) funding partners successfully engaged.
Steering Committee meetings	11	1. Gemma Wyburn (Australian Pork) 2. Sasha Jenkins (UWA) 3. Marit Kragt (UWA) 4. Luke Wheat (Future Green Solutions) 5. Matthew Redding (Qld DAF) 6. Janine Price (Scolexia) 7. Matthew Deegan (AMPC 8. Cath Lescun (Dairy Australia) 9. Mini Singh (Australian Eggs) 10. Lechelle Van Breda/ Georgina Toose (AgriFutures Australia) 11. Crispian Ashby (FRDC)

6 Lessons learnt

In this section, we focus on the key lessons learned to date from a project administration perspective.

Ensuring clear understanding of partner positions prior to contracting to avoid unnecessary delays due to communication issues. This includes securing background IP rights, contracts and any terms and conditions before progressing with the project application. Background IP from a previous project on encapsulation was to be utilised in the project, however this was unable to be realised due to restrictive conditions around the use of the IP and future ownership questions that came to light late in the contracting process between partners. This lead to some frustration and the alteration of the project plan, which could have been avoided if this information was secured in the project initiation/application phase.

The need to keep detailed notes and minutes for cross industry projects. This project has been well managed on this front and effort has been made to record as much discussion as possible. This was well planned as several project partners have had representatives change throughout the project to date (some several times) including the project manager. Having the detailed notes has allowed rapid onboarding of new representatives and the revisiting of the rational of the project and its progress has also been beneficial for those that have not changed out as it provides a reminder for why the project is important and where it is up to for those partners that may be spread thin with competing priorities.

The power of digital communication. As most of the experimental work had just commenced when COVID 19 triggered the lockdowns, digital communication has proven very effective as a means to keep partners updated and engaged, and as an extension platform. Having a working knowledge of these is crucial, especially going forward where the project aims to take a more digital approach to its extension to avoid any further impacts due to cancelled events. Efforts at extension have been altered as meetings and conferences were cancelled, with some able to be undertaken by local staff while others have been progressed digitally.

Planning of sequential research activities. The work conducted in this study relies on the generation of BSF frass and larvae for further testing and analysis. This poses a risk to the research activities if BSF products are not delivered. Given the experimental nature of rearing Black Soldier Fly, some delays have been experienced in this step of the process – with significant research effort going into designing the optimal substrate for BSF rearing (Activity 5.1). While it was anticipated that waste streams from individual industries could be used for the BSF larvae, in most cases larvae growth was improved on mixed waste streams (to ensure the right moisture content, consistency, and nutritional profile). While in this project, all research activities were able to continue using the mixed waste substrate, it is recommended that future projects consider contingency plans to formulate research schedules that can progress despite any delays in earlier research activities.

The importance of contingency plans. The project was slowed due to several of the key researchers becoming seriously unwell during 2020, including one main researcher being required to take extended sick leave. This was relatively well managed by the research partners who appointed / increased the FTE of other researchers to contribute to the BSF production process and testing, and another senior researcher on the team who was rapidly able to take on the majority of the administration of the project while the other researcher was convalescing. The importance of a core project manager (lead researcher) who is on top of all the project activities and involved in all project coordination is essential for this type of research grant. To this end, we recommend funding a dedicated research lead who is responsible for all area of research management - including experimental design and communicating across disciplines. This requires a researcher with strong interdisciplinary skills, which may not always be the primary disciplinary researcher.

7 Appendix - additional project information

3.1 Project, media and communications material and intellectual property

Nature of materials / activities	**Number**	**Details** (Please provide details if appropriate (eg links to publicly available documents)
Media appearances – press and TV	6	1. Australian Pork Limited press release 2. University of Western Australia press release Link 3. Future Green Solutions press release Link 4. ABC News feature on 7pm news, aired on 6th November 2021. Link 5. ABC News article published 6th November 2021 “Black soldier fly recruited to recycle livestock waste into fertiliser for food production” Link 6. ABC local radio program Great Southern and South West Rural Report, aired 8th November 2021. Link
Brochures, fact sheets, posters and newsletters	21	1. The UWA Institute of Agriculture Newsletter, December 2019 (page 4). Link 2. Australian Eggs’ “Eggstra” September 2020. Link 3. National Poultry Newsletter, July 2021 Link 4. The UWA Institute of Agriculture Newsletter, August 2021 (page 4). Link 5. pig333.com Link 6. Queensland Country Life Link 7. The UWA Institute of Agriculture Newsletter, April 2022 (page 7). Link 8. Australian Pork Limited Update, 4th September 2020 9. WA DWER, 2020 10. Project Updates 11. Project Update #1: January - December 2020 Link 12. Project Update #2: January - June 2021 Link 13. Project Update #3: July - December 2021 Link 14. Project Update #4: January - June 2022 Link 15. Project Update #5: July - December 2022 Link 16. Biosecurity Risks of Black Soldier Fly (BSF) Larvae as an Animal Feed (Appendix C) 17. Fact or Fiction: Black Solider Flies Link 18. Nutrition of BSF products used for animal feed Link 19. Creating fertilisers from agricultural waste using Black Solider Fly farming (Appendix I) 20. Stable fly emergence Link

		21. Potential environmental regulation classification and requirements for manure derived Black Soldier Fly frass Link
Websites	7	1. Project website: https://www.bsfwastetoprofit.com/ 2. A Community of Practice website hosted by AgriFutures Australia: https://extensionaus.com.au/blacksoldierfly/introduction-to-the-project/ 3. UWA Centre for Engineering Innovation: Agriculture and Ecological Restoration: https://www.uwa-cei.org/blacksoldierfly 4. UWA Centre for Agricultural Economics and Development: https://www.uwacaed.org/research 5. Future Green Solutions: https://www.futuregreensolutions.com.au/ 6. PhD projects https://www.uwa.edu.au/projects/consumer-perceptions-towards-insect-farming-products 7. https://www.uwa.edu.au/projects/developing-and-assessing-a-high-quality-fertiliser-product-based-on-black-soldier-fly-frass
Social media presence	24	Hashtags #BSFwastetoprofit #blacksoldierflyuwa **Twitter & Facebook** 1. @IOA_UWA on 3rd December 2020 Link 2. @SAgE_UWA on 4th December 2020 Link 3. @Daniel_Kidd01 on 12th November 2021 Link 4. @RegenWa on 29th June, 2021 Link 5. @maritkragt on 7th November 2021 Link 6. @Daniel_Kidd01 on 25th August 2021 Link 7. @IOA_UWA on 13th August, 2021 Link 8. @Daniel_Kidd01 on 8th November 2021 Link 9. @maritkragt on 6th March 2022 Link 10. @uwaceep on 17th January 2022 Link 11. @IOA_UWA on 24th August, 2022 Link 12. @IOA_UWA on 1st July, 2021 Link 13. @FutureGreenSol on 22nd July 2022 Link 14. @AREatUWA on 1st April 2021 Link **Blogs** 15. What is black soldier fly, Reasoning behind the project Link 16. Introduction to the Project – Turning Animal Waste into Fertiliser using BSF Link 17. Understanding Current Waste Management Practices – Interviews Link 18. Drivers and barriers to adoption of black soldier flies Link 19. BSF Technology in short; what we know so far Link

		20. ABC news informs the agricultural industry on Black Solider Fly research at UWA Link 21. Where is most livestock waste produced? Link **Videos** 22. Closing the loop: Black Soldier Fly technology to convert agriculture waste (2022). YouTube. Date forthcoming, available on project website: https://www.bsfwastetoprofit.com/blog 23. @Degleeson on 24th March 2021 Link 24. @jp_wisdom on 19th May 2021 Link
Scientific conference presentations and posters	7	1. Presentation on *Black soldier fly technology to convert piggery manure into fertilizer* delivered by Denise Woods at the Australasian Pig Science Association (APSA) 17th Biennial Conference, 17-20th November 2019, Hilton Adelaide, Adelaide. 2. Presentation on *Black soldier fly technology to convert piggery manure into fertilizer* delivered by Dr Sasha Jenkins at the State of Soil Science WA, 4-6th December 2019, UWA Business School, Western Australia. 3. Presentation on *Valuing the costs of waste management in the pork industry* delivered by Dr Sasha Jenkins at the Australasian Pig Science Association (APSA) conference, 18th November 2021. 4. Presentation on *Assessing the potential of black soldier fly technology to convert piggery manure into slow-release fertiliser* delivered by Dr Sasha Jenkins at the Australasian Pig Science Association (APSA) conference, 18th November 2021. 5. Presentation on *Increasing the throughput of fertiliser product screening: machine vision and microdialysis* delivered by Matt Redding at the Soil Science Society of Australia, Cairns 2021. 6. Presentation on *Using BSF technology to turn livestock waste into profitable products* delivered by Dr Marit Kragt at the Terra Envision Conference, Utrecht, Netherlands. 28th June, 2022. 7. Gurung, S.K., Mickan, B.S., Middleton, J., Jenkins, S., Rengel, Z., Siddique, K. and Solaiman, Z.M (2022). *Waste to value: the use of black soldier fly frass for sustainable soil health and food production*. Poster, 22nd world congress of soil science in Glasgow, United Kingdom, 31 July - 5 August, 2022.

Scientific seminars and tutorials	4	1. Seminar on *Closing the loop: Black Soldier Fly technology to convert agricultural waste* delivered by Marit Kragt, Fiona Dempster & Vandana Subroy for the ARE/AARES Research Seminar Series on 21 May 2021 at UWA Link 2. Seminar on *Key research skills in the biological sciences. My research journey: from aquatic ecology to biotechnology* delivered by Dr Jen Middleton for BIOL1131 in 2022 at UWA. 3. Interactive tutorial on 'The Fly Show' – Use of Black Soldier Fly Technology in waste management delivered by Dr Jen Middleton for ENVT1104 at UWA in 2022. 4. Seminar on *Using black soldier fly technology to turn livestock waste into profitable products* delivered by Marit Kragt for the SAGE Seminar Series on 3 November 2022 at UWA.
Intellectual Property	3	1. A novel approach using BSF proteins to develop a protein/amino based fertiliser product. Trade secret (difficult to work out independently). 2. Economic survey data (farmers' preferences survey). Copyright, data housed in excel database. 3. Economic survey data (waste producers survey). Copyright, data housed in excel database.
Technical Reports	9	1. Tascon, A., Dempster, F. & Kragt, M. (2021). *Spatial Analysis of Farm Animal Waste in Australia*. The University of Western Australia. 2. Langa, S., Middleton, J., Gleeson, D. & Jenkins, S. (2022). *Processing poultry manure with Black Soldier Fly technology lowers greenhouse gas emissions 2 from soil*. The University of Western Australia. 3. Dempster, F. Harold, T., Subroy, V. & Kragt, M. (2022). *Market potential for Black Soldier Fly fertiliser products.* The University of Western Australia. 4. Dempster, F. Harold, T., Subroy, V. & Kragt, M. (2022). *End-users' perceived benefits and perceptions of insect farming*. The University of Western Australia. 5. Pearson, L. and Wilson, T.B. (2022). A Review of the Biosecurity Risks of Using Black Soldier Fly (Hermetia illucens) in the Australian Agriculture Sector. Scolexia. 6. Quinteros, J.A., Wilson, T., Pearson, L., Price, J., Kragt, M. and Scott, P. (2022). Potential environmental risk of Black Soldier Fly (Hermetia illucens) By -Products: A review of the current literature. Scolexia and The University of Western Australia. 7. Bruerton, K., Pearson, L. and Scott, P. (2022). Nutritional analysis of Black Soldier Fly for intensive livestock. Scolexia.

		8. Pearson, L., Wilson, T.B. and Scott, P. (2022). Pathogen and Heavy Metal Loading at the Manure Input, Larval and Frass Stages of Black Soldier Fly Production. Scolexia. 9. Price, J. (2022). Potential Environmental Regulation Classification and Regulatory Requirements for Manure Derived Black Soldier Fly Frass. Scolexia.

7.2 Equipment and assets

No equipment and assets over $10,000 have been purchased under this contract.

The following table lists equipment and assets of value used to obtain research outcomes.

Item purchased	Date of purchase	Purchase price (GST exclusive)
Drying Oven (small) Model: WAS-FA.10.240 OVEN FORCED AIR Make: WA Scientific Instruments	28/01/2022	$4,265
Professional Moisture Analyser Model: MB90 Make: OHAUS (Instrument Choice)	3/02/2022	$3,895
A disc pelletiser Model: Haver SC 500 Make: Haver and Boecker	N/A	External grant

7.3 Monitoring and evaluation

The final monitoring and evaluation report (Appendix R) was outsourced to an independent consultant, conducted by Advanced Choice Economics Pty Ltd.

7.4 Budget

The project grant budget will be spent in full by the end of the grant. A final financial report approved by the Accountant will be submitted within 60 days of submitting the final milestone report.

In summary, the project grant funding was spent in accordance with the budget outlined in the Research Agreement. A majority of funds were spent on staff resources at UWA and sub-contractor invoices (Scolexia and Future Green Solutions) to contribute to the research activities. The remaining funds were spent on supplier invoices to pay for data analysis, laboratory testings, equipment, communication activities, travel and general consumables and charges.

7.5 List of appendices

Appendix A: Activity 5.1a-d: Optimising Black Soldier Fly frass and larvae production on single and blended waste streams

Appendix B: Activity 5.2: Waste Producers Survey

Appendix C: Activity 5.3a: A Review of the Biosecurity Risks of Using Black Soldier Fly (Hermetia illucens) in the Australian Agriculture Sector AND Factsheet

Appendix D: Activity 5.3c and 5.4c: Pathogen and Heavy Metal Loading at the Manure Input, Larval and Frass Stages of Black Soldier Fly Production

Appendix E: Activity 5.3f and 5.4g: Potential Environmental Regulation Classification and Regulatory Requirements for Manure Derived Black Soldier Fly Frass

Appendix F: Activity 5.4a: Potential environmental risk of Black Soldier Fly (Hermetia illucens) By -Products: A review of the current literature.

Appendix G: Activity 5.4d: Conduct pot trials to determine the nutrient and heavy metal leaching potential of different BSF frass products applied to a range of soil types.

Appendix H: Activity 5.4e: Processing poultry manure with Black Soldier Fly technology lowers greenhouse gas emissions from soil

Appendix I: Activity 5.5a: Combining frass and fatty acid by-products of Black Soldier Fly larvae farming has 1 potential as slow release fertiliser AND Factsheet

Appendix J: Activity 5.5b: De-bottlenecking the black soldier fly manure biorefinery: utilising the frass for fertiliser and impacts on soil biological processes

Appendix K: Activity 5.5c: Conduct pot trials on targeted pasture, broadacre or horticulture crops as determined from the market analysis and industry engagement

Appendix L: Activity 5.5d Conduct field trials to evaluate the effectiveness of using frass to ameliorate soil constraints (e.g. acidity, water repellency).

Appendix M: Activity 5.6: Develop a granulated fertiliser product

Appendix N: Activity 5.7: Develop slow release encapsulated fertiliser product

Appendix O: Activity 5.8**:** Nutritional analysis of Black Soldier Fly for intensive livestock

Appendix P: Activity 5.9d: Using Black Soldier Fly to enhance circularity in agricultural production: Farmers' perceptions and willingness-to-pay for insect-based fertilisers

Appendix Q: Activity 5.9d: End Users Survey

Appendix R: Section 7.3: Monitoring and Evaluation Report

Supplementary Data

Activity 5.4 Assess the environmental risks of Black Soldier Fly products.

Activity 5.4(d) Conduct pot trials to determine the nutrient and heavy metal leaching potential of different frass products applied to a range of soil types.

Table S1. Effect of soil amendments on the total leaching of ammonium nitrogen (NH_4-N) from two soil types (sand and sandy loam). Post-hoc pairwise comparison test results (Fisher's), least squares means and standard error for all interactions from three-way factorial analysis of variance. Letters denote significant difference between treatment means (alphabetised from highest to lowest mean).

NH_4	LS means	Standard error					
Soil type-Sand*Substrate-granulated frass (+binder)*Application rate (t ha-1)-High	1.144	0.165				D	E
Soil type-Sand*Substrate-granulated frass (+binder)*Application rate (t ha-1)-Low	0.498	0.165					
Soil type-Sand*Substrate-granulated frass (-binder)*Application rate (t ha-1)-High	9.187	0.165	A				
Soil type-Sand*Substrate-granulated frass (-binder)*Application rate (t ha-1)-Low	2.291	0.165			C		
Soil type-Sand*Substrate-No amendment*Application rate (t ha-1)-Nil	0.318	0.165					
Soil type-Sand*Substrate-NPK fertiliser*Application rate (t ha-1)-High	5.759	0.165		B			
Soil type-Sand*Substrate-NPK fertiliser*Application rate (t ha-1)-Low	1.496	0.165				D	
Soil type-Sand*Substrate-Pork frass*Application rate (t ha-1)-High	0.207	0.190					
Soil type-Sand*Substrate-Pork frass*Application rate (t ha-1)-Low	0.140	0.165					
Soil type-Sand*Substrate-Pork manures*Application rate (t ha-1)-High	0.094	0.165					
Soil type-Sand*Substrate-Pork manures*Application rate (t ha-1)-Low	0.220	0.190					
Soil type-Sandy loam*Substrate-granulated frass (+binder)*Application rate (t ha-1)-High	1.109	0.165				D	E
Soil type-Sandy loam*Substrate-granulated frass (+binder)*Application rate (t ha-1)-Low	1.094	0.165				D	E
Soil type-Sandy loam*Substrate-granulated frass (-binder)*Application rate (t ha-1)-High	1.545	0.165				D	
Soil type-Sandy loam*Substrate-granulated frass (-binder)*Application rate (t ha-1)-Low	0.801	0.165					
Soil type-Sandy loam*Substrate-No amendment*Application rate (t ha-1)-Nil	0.676	0.165					
Soil type-Sandy loam*Substrate-NPK fertiliser*Application rate (t ha-1)-High	1.331	0.165				D	E
Soil type-Sandy loam*Substrate-NPK fertiliser*Application rate (t ha-1)-Low	0.881	0.165					E
Soil type-Sandy loam*Substrate-Pork frass*Application rate (t ha-1)-High	0.739	0.233					
Soil type-Sandy loam*Substrate-Pork frass*Application rate (t ha-1)-Low	0.649	0.165					
Soil type-Sandy loam*Substrate-Pork manures*Application rate (t ha-1)-High	0.635	0.165					
Soil type-Sandy loam*Substrate-Pork manures*Application rate (t ha-1)-Low	0.679	0.165					

Table S2. Effect of soil amendments on the total leaching of nitrate nitrogen (NO_3-N) from two soil types (sand and sandy loam). Post-hoc pairwise comparison test results (Fisher's), least squares means and standard error for all interactions from three-way factorial analysis of variance. Letters denote significant difference between treatment means (alphabetised from highest to lowest mean).

NO_3	LS means	Standard error					Gro
Soil type-Sand*Substrate-granulated frass (+binder)*Application rate (t ha-1)-High	16.716	1.870					E
Soil type-Sand*Substrate-granulated frass (+binder)*Application rate (t ha-1)-Low	11.330	1.870					
Soil type-Sand*Substrate-granulated frass (-binder)*Application rate (t ha-1)-High	28.468	1.870	A				
Soil type-Sand*Substrate-granulated frass (-binder)*Application rate (t ha-1)-Low	19.193	1.870			C	D	E
Soil type-Sand*Substrate-No amendment*Application rate (t ha-1)-Nil	2.953	1.870					
Soil type-Sand*Substrate-NPK fertiliser*Application rate (t ha-1)-High	24.191	1.870	A	B	C		
Soil type-Sand*Substrate-NPK fertiliser*Application rate (t ha-1)-Low	18.537	1.870				D	E
Soil type-Sand*Substrate-Pork frass*Application rate (t ha-1)-High	25.903	2.159	A	B			
Soil type-Sand*Substrate-Pork frass*Application rate (t ha-1)-Low	13.712	1.870					
Soil type-Sand*Substrate-Pork manures*Application rate (t ha-1)-High	15.761	1.870					
Soil type-Sand*Substrate-Pork manures*Application rate (t ha-1)-Low	12.346	2.159					
Soil type-Sandy loam*Substrate-granulated frass (+binder)*Application rate (t ha-1)-High	18.123	1.870					E
Soil type-Sandy loam*Substrate-granulated frass (+binder)*Application rate (t ha-1)-Low	14.810	1.870					
Soil type-Sandy loam*Substrate-granulated frass (-binder)*Application rate (t ha-1)-High	18.812	1.870			C	D	E
Soil type-Sandy loam*Substrate-granulated frass (-binder)*Application rate (t ha-1)-Low	17.302	1.870					E
Soil type-Sandy loam*Substrate-No amendment*Application rate (t ha-1)-Nil	16.225	1.870					E
Soil type-Sandy loam*Substrate-NPK fertiliser*Application rate (t ha-1)-High	23.664	1.870	A	B	C	D	
Soil type-Sandy loam*Substrate-NPK fertiliser*Application rate (t ha-1)-Low	21.349	1.870		B	C	D	E
Soil type-Sandy loam*Substrate-Pork frass*Application rate (t ha-1)-High	15.302	2.644					
Soil type-Sandy loam*Substrate-Pork frass*Application rate (t ha-1)-Low	17.849	1.870					E
Soil type-Sandy loam*Substrate-Pork manures*Application rate (t ha-1)-High	15.407	1.870					
Soil type-Sandy loam*Substrate-Pork manures*Application rate (t ha-1)-Low	16.539	1.870					E

Table S3. Effect of soil amendments on the total leaching of phosphate phosphorus (PO_4-P) from two soil types (sand and sandy loam). Post-hoc pairwise comparison test results (Fisher's), least squares means and standard error for all interactions from three-way factorial analysis of variance. Letters denote significant difference between treatment means (alphabetised from highest to lowest mean).

PO_4	LS means	Standard error			Groups		
Soil type-Sand*Substrate-granulated frass (+binder)*Application rate (t ha-1)-High	10.609	0.235		B			
Soil type-Sand*Substrate-granulated frass (+binder)*Application rate (t ha-1)-Low	2.547	0.235				D	
Soil type-Sand*Substrate-granulated frass (-binder)*Application rate (t ha-1)-High	12.991	0.235	A				
Soil type-Sand*Substrate-granulated frass (-binder)*Application rate (t ha-1)-Low	0.895	0.235					E
Soil type-Sand*Substrate-No amendment*Application rate (t ha-1)-Nil	0.052	0.235					
Soil type-Sand*Substrate-NPK fertiliser*Application rate (t ha-1)-High	0.054	0.235					
Soil type-Sand*Substrate-NPK fertiliser*Application rate (t ha-1)-Low	0.149	0.235					
Soil type-Sand*Substrate-Pork frass*Application rate (t ha-1)-High	10.911	0.271		B			
Soil type-Sand*Substrate-Pork frass*Application rate (t ha-1)-Low	2.688	0.235				D	
Soil type-Sand*Substrate-Pork manures*Application rate (t ha-1)-High	6.111	0.235			C		
Soil type-Sand*Substrate-Pork manures*Application rate (t ha-1)-Low	1.089	0.271					E
Soil type-Sandy loam*Substrate-granulated frass (+binder)*Application rate (t ha-1)-High	0.024	0.235					
Soil type-Sandy loam*Substrate-granulated frass (+binder)*Application rate (t ha-1)-Low	0.061	0.235					
Soil type-Sandy loam*Substrate-granulated frass (-binder)*Application rate (t ha-1)-High	0.046	0.235					
Soil type-Sandy loam*Substrate-granulated frass (-binder)*Application rate (t ha-1)-Low	0.018	0.235					
Soil type-Sandy loam*Substrate-No amendment*Application rate (t ha-1)-Nil	0.015	0.235					
Soil type-Sandy loam*Substrate-NPK fertiliser*Application rate (t ha-1)-High	0.020	0.235					
Soil type-Sandy loam*Substrate-NPK fertiliser*Application rate (t ha-1)-Low	0.014	0.235					
Soil type-Sandy loam*Substrate-Pork frass*Application rate (t ha-1)-High	0.041	0.332					
Soil type-Sandy loam*Substrate-Pork frass*Application rate (t ha-1)-Low	0.027	0.235					
Soil type-Sandy loam*Substrate-Pork manures*Application rate (t ha-1)-High	0.047	0.235					
Soil type-Sandy loam*Substrate-Pork manures*Application rate (t ha-1)-Low	0.039	0.235					

Table S4. Effect of soil amendments on the total leachate volume from two soil types (sand and sandy loam). Post-hoc pairwise comparison test results (Fisher's), least squares means and standard error for all interactions from three-way factorial analysis of variance. Letters denote significant difference between treatment means (alphabetised from highest to lowest mean).

Leachate volume	LS means	Standard error					
Soil type-Sand*Substrate-granulated frass (+binder)*Application rate (t ha-1)-High	2.586	0.121	A				
Soil type-Sand*Substrate-granulated frass (+binder)*Application rate (t ha-1)-Low	2.206	0.121		B	C	D	
Soi type-Sand*Substrate-granulated frass (-binder)*Application rate (t ha-1)-High	2.642	0.121	A				
Soil type-Sand*Substrate-granulated frass (-binder)*Application rate (t ha-1)-Low	2.484	0.121	A	B			
Soil type-Sand*Substrate-No amendment*Application rate (t ha-1)-Nil	1.994	0.121			C	D	E
Soil type-Sand*Substrate-NPK fertiliser*Application rate (t ha-1)-High	2.514	0.121	A	B			
Soil type-Sand*Substrate-NPK fertiliser*Application rate (t ha-1)-Low	2.313	0.121	A	B	C		
Soil type-Sand*Substrate-Pork frass*Application rate (t ha-1)-High	1.832	0.14					E
Soil type-Sand*Substrate-Pork frass*Application rate (t ha-1)-Low	2.141	0.121			C	D	E
Soil type-Sand*Substrate-Pork manures*Application rate (t ha-1)-High	1.807	0.121					E
Soil type-Sand*Substrate-Pork manures*Application rate (t ha-1)-Low	1.883	0.14				D	E
Soil type-Sandy loam*Substrate-granulated frass (+binder)*Application rate (t ha-1)-High	1.236	0.121					
Soil type-Sandy loam*Substrate-granulated frass (+binder)*Application rate (t ha-1)-Low	1.301	0.121					
Soil type-Sandy loam*Substrate-granulated frass (-binder)*Application rate (t ha-1)-High	1.729	0.121					
Soil type-Sandy loam*Substrate-granulated frass (-binder)*Application rate (t ha-1)-Low	1.049	0.121					
Soil type-Sandy loam*Substrate-No amendment*Application rate (t ha-1)-Nil	0.652	0.121					
Soil type-Sandy loam*Substrate-NPK fertiliser*Application rate (t ha-1)-High	0.935	0.121					
Soil type-Sandy loam*Substrate-NPK fertiliser*Application rate (t ha-1)-Low	0.7	0.121					
Soil type-Sandy loam*Substrate-Pork frass*Application rate (t ha-1)-High	0.972	0.171					
Soil type-Sandy loam*Substrate-Pork frass*Application rate (t ha-1)-Low	1.151	0.121					
Soil type-Sandy loam*Substrate-Pork manures*Application rate (t ha-1)-High	1.429	0.121					
Soil type-Sandy loam*Substrate-Pork manures*Application rate (t ha-1)-Low	1.564	0.121					