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Adding value to Marlborough waste streams using insect bioconversion

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1 Background

A variety of waste streams are generated by Marlborough-based industries, including grape marc from the wine-making process and mussel shells from the aquaculture industry. These waste streams present an ongoing challenge in terms of appropriate means of disposal or re-use. This is our third year of research into the ability of insects to bioconvert grape marc and other waste streams from the Marlborough region into higher-value products. Our previous studies (Agnew et al. 2021, Page-Weir et al. 2022) identified mealworms (larvae of the beetle *Tenebrio molitor*) as a species capable of digesting both grape marc and ground mussel shells. In combination with a Plant & Food Research (PFR) programme investigating insects as potential components of aquafeeds, we have also started to characterise the fatty-acid and amino-acid profile of mealworms fed with these diets. The results of these previous studies are summarized below.

1.1 Grape marc

- Of nine insect species tested, only mealworms (larvae of the beetle *Tenebrio molitor*) and black soldier fly (*Hermetia illucens*) larvae were able to digest grape marc.
- When a small amount of wholemeal flour was added to the marc, mealworms were able to develop through to adult beetles and produce offspring, suggesting this diet is adequate for mealworm development.
- In small, vented click-clack boxes (15.1 cm l x 10 cm w x 4.8 cm h), approximately 100 mealworms were found to consume their body weight in grape marc over a 1-week period.
- When scaled up to larger boxes (57 cm l x 38 cm w x 32 cm d), 1 kg of mealworms consumed 2 kg of grape marc every 7–10 days.
- Mealworms were not able to consume the grape seeds, even when the seeds were milled into a flour.
- An analysis of the frass produced by the mealworms from feeding on grape marc by
 Hills Laboratories suggested that the frass may be useful as a soil amendment/fertiliser.

1.2 Wine lees

 Mealworms were unable to successfully feed and develop on wine lees. We have recommended further research to identify insect species that are able to bioconvert this byproduct.

1.3 Ground mussel shells

 Mealworms were able to feed and survive on ground mussel shells and developed normally if some flour or grape marc was added to the diet.

1.4 Mealworms as aquafeed components

- Mealworms fed with wholemeal flour and some vegetables were high in iron and zinc, had almost 50% crude protein and 40% fat.
- Regardless of food source, mealworms were found to be high in palmitic (C16:0) and oleic (C18:1n9c) acids, these two making up about 30% each of the total fatty acid content.
- The omega-3, alpha-linolenic acid (C18:3c9, 12, 15), and the omega-6, linoleic acid (C18:2n6c), represented 0.25% and 15% of the total fatty acids, respectively. Both of these are essential fatty acids that cannot be produced by the bodies of most animals, including fishes, and so need to be provided within their diet.

2 2022 research

Over the last 12 months we have conducted the following two pieces of work to determine if mealworms fed on Marlborough-sourced by-products could be used as a viable aquafeed components.

2.1 Scaling-up and assessment of environmental conditions

In our previous studies, our assessments of grape marc consumption by mealworms were conducted inside 23–25°C controlled-environment rooms, and mostly using small containers (Page-Weir et al. 2022). We have now scaled up to larger containers (35 cm x 50 cm x 100 cm), and placed the mealworms and grape marc in these containers in a variety of locations, to assess the effects of the environment (i.e. variable diurnal temperature and humidity patterns) on grape marc consumption. We assessed grape marc consumption at ambient indoor temperatures in Auckland, at outside-ambient conditions in both Marlborough and Auckland, as well as inside a tunnel house in Marlborough.

In addition, we assessed the consumption of grape marc by mealworms in rotatable compost bins to determine whether rotating the grape marc increased consumption: we proposed this because mealworms produce heat at high densities, and too much heat can reduce consumption rates (Deruytter et al. 2022).

2.2 Nutritional analysis of mealworms fed different diets

In combination with the PFR Aquafeeds project, we assessed the nutritional profiles of mealworms fed different foods, including ground mussel shells and ground grape marc. We simultaneously assessed mealworm survival on these different diets.

3 Scaling up: Auckland experiments

Scaling-up the process and transferring the mealworms and grape marc to ambient outdoor conditions at PFR in Auckland proved to be more difficult than expected. In September 2022 we purchased large bins (35 cm x 50 cm x 100 cm: Figure 1) and placed two bins in a shady position outside our laboratory building in Mt Albert. We made holes in the sides of the bin for ventilation and covered the holes in mesh to prevent mealworms escaping and other insects from entering the bins. Grape marc and mealworms were placed inside the bins, and a temperature and humidity datalogger was placed inside the bin just above the marc. The contents were monitored daily. This experiment was ended after seven days because the mealworms were found to have died. Investigations showed the temperature inside the boxes rose above 35°C and the odour within the box suggested that bacterial fermentation and decomposition had occurred within the box, producing a great deal of condensation and high temperatures inside the bin, and this had killed the mealworms.



Figure 1. Black bins with ventilation holes were used to see if the bioconversion of grape marc by mealworms could be scaled up and conducted in ambient conditions. The bins were elevated and placed in trays containing water to prevent ants and other crawling insects from accessing the grape marc and mealworms.

To reduce the temperature and prevent condensation forming in the boxes, new, raised lids were constructed (Figure 2). To prevent entry by other organisms, we covered the box with a fine-mesh cloth and put the lids on top. We first tested these boxes with grape marc but no mealworms, to ensure the temperatures and condensation levels inside the boxes were not too high for the mealworms. This worked well, with no condensation forming on the sides of the bins and the temperatures did not exceed 35°C.



Figure 2. Raised lids were constructed and put on the bins to increase air flow within the bins (intended for bioconversion of grape marc by mealworms). A fine-mesh cloth was placed over the top of the bin to prevent access by organisms other than mealworms. Bungee cords were used to secure the lid against the wind.

At the same time we tested whether stirring the grape marc on a daily basis would reduce temperature or condensation, but the stirring seemed to result in greater mould growth on the marc, so we did not continue to stir any of the marc from here on.

In November 2022 we set up two new bins with the raised lids. Each bin contained 12–13 L of grape marc, 500 g of meal worms and a temperature and humidity datalogger. Our plan was to maintain these mealworms on the grape marc over the summer to ensure this process would proceed throughout the warmer months of the year. All proceeded well, with the mealworms consuming grape marc and surviving well through to mid-January 2023 (Figure 3). Unfortunately new grape marc was added in late January that we then discovered was contaminated with mites. We needed to discard the contents of both bins to remove the mites.



Figure 3. Mealworms feeding on grape marc inside one of the black bins.

Similar testing of mealworms feeding on grape marc within the Auckland laboratory at room temperature has proceeded similarly to previous tests in controlled-temperature rooms.

4 Scaling up: Marlborough experiments

In January 2023 we set up four bins of grape marc and mealworms at PFR in Marlborough. Two of the bins were located inside a tunnel house (Figure 4), where higher temperatures could be maintained year-round, and two bins were located outside the tunnel house (Figure 5). Each bin contained grape marc, some flour, and mealworms. One of the inside bins and one of the outside bins each contained a temperature and humidity datalogger so that we could monitor conditions inside the bins. These showed that although the temperature inside the bins exceeded 35°C for up to 9 hours on several days, and the relative humidity (rH) ranged from 25% to 86% rH, the mealworms survived and continued to feed. This suggests that the raised lids are working well to maintain conditions that are suitable for mealworm survival and development. However, although the mealworms in these bins were seen to be alive when inspected in mid-February 2023, they had made little impact on the grape marc. This may have been a result of too few mealworms added at the beginning, and so a second batch of mealworms was added in March.



Figure 4. Two bins containing grape marc and mealworms in the tunnel house in Marlborough.



Figure 5. Two bins containing grape marc and mealworms set up outside the tunnel house in Marlborough.

In June 2023, the mealworms in these bins were still alive and feeding, however we have moved the two bins from outside to inside the tunnel house for the winter months: it's unlikely that mealworms, which are a Mediterranean species, will cope with very low temperatures. We have also started a regime of tilting two of the bins through 20° to 30° on a regular basis to determine if this will increase the speed of grape marc bioconversion.

5 Rotating compost bin experiments

In May 2023 we conducted a comparison of using a rotating compost bin versus our standard black bin on mealworm survival and bioconversion of grape marc by the mealworms (Figure 6). The bins were again located outside the PFR laboratory in Mt Albert. Each bin contained 3.4 kg of Sauvignon blanc marc, 1.7 kg of mealworms, and a datalogger. We rotated the composter one notch (approximately 30°) on each work day, Monday to Friday. After rotating through 90° in one direction we rotated it back through 180° in the other direction (30° change per day): we did not want to rotate the bin through 360° in case the door on the composter could not hold the weight of the marc and the mealworms. The contents of the bins and the health of the mealworms were assessed at the time of rotation.

After two weeks we noticed some moisture on the walls of the rotating bin and some of the marc appeared to be wet. It is not possible to know if the moisture was from condensation or if some rainwater had been able to get into the bin. However, the mealworms were alive and there was no obvious fermentation odour or mould visible.



Figure 6. Rotating compost bin and standard black bin, both containing grape marc and mealworms, set up in May 2023.

In early June 2023 we assessed that there was very little grape marc left in the compost bin so we removed and weighed the contents of both bins. In the black bin, the mealworms had maintained their weight at 1.7 kg and had reduced the grape marc from 3.4 kg to 1.8 kg: this included remaining seeds, stems and some grape skins (Figure 7a). In the rotating compost bin, the mealworms had increased their weight to 1.8 kg and only 1.3 kg of grape marc remained (Figure 7b).



Figure 7. Grape marc and mealworm contents of (a) the black bin and (b) the rotating compost bin after 26 days.

The dataloggers showed that the temperature inside the two bins was similar throughout, with daily fluctuations inside the black bin between 9°C and 20°C (average +/- SEM = 14.8° C +/- 0.1° C) and daily fluctuations inside the rotating bin between 9°C and 21°C (average +/- SEM = 15.6° C +/- 0.1° C). However relative humidity (rH) within the two bins was very different, with conditions in the black bin remaining reasonably stable between 75% and 85% rH throughout (average +/- SEM = 80.1 rH +/- 0.1), whereas humidity in the rotating bin rose slowly from 74% to 95% rH (average +/- SEM = 87.6 rH +/- 0.3) over the 26 days. The moisture in the compost bin may have contributed to the increased bioconversion of the grape marc, and certainly the contents looked different from those in the black bin (Figure 7a versus 7b).

Figure 8 shows the frass produced by the mealworms feeding on the grape marc in the black bin (Figure 8a) and the rotating compost bin (Figure 8b). We aim to have these two different samples analysed for their potential as soil amendments.



Figure 8. Frass (excrement) produced by mealworms feeding on grape marc in the black bin (a) and the rotating compost bin (b).

6 Nutritional analyses of mealworms fed on different diets

We have analysed the amino acid profile of mealworms that have been fed on a variety of diets, including grape- and mussel-derived diets, as well as their standard wheat-based diet. Interestingly, the mealworms were found to have different profiles based on diet. For instance, mealworms reared on diets containing mussel shells showed an increase in the concentration of taurine relative to larvae reared on purely plant-based diets (Figure 9). This is exciting because taurine is a non-protein forming amino acid that is important for fish nutrition.

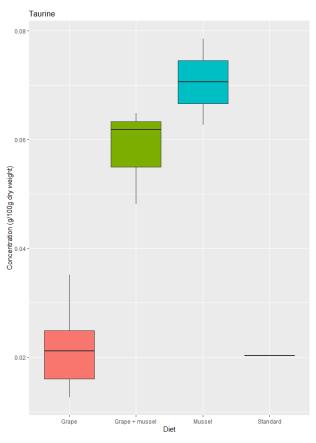


Figure 9. Concentration of the amino acid Taurine in mealworms fed with different diets: Grape = grape marc only; Grape + mussel = grape marc and ground mussel shells; Mussel = ground mussel shells only; Standard = mealworm standard diet (wholemeal flour).

7 Summary

Our results from this year show that the bioconversion of grape marc by mealworms can be conducted in large bins and at ambient temperatures in both Auckland and Marlborough. This suggests that a mealworm bioconversion facility could be built in Marlborough and may not need continuous heating or cooling. The facility could operate similarly to black soldier fly facilities where bioconversion occurs in tall stacks of large bins. Further research into the effects of tilting/rotating the bins, as well as the ideal humidity level within the bins, on bioconversion rates would be useful, as would determining the effects of these two factors on the quality of the frass produced by the mealworms.

The results from the PFR-funded aquafeeds programme indicate that insect meal produced from the mealworms may be a valuable aquafeed component. Further research is needed to determine the best diet for mealworms in order to produce the highest quality insect meal in terms of its value for aquafeeds.

8 References

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