Effect of Supplemental Organic Material on *Tenebrio molitor* L. (Coleoptera: Tenebrionidae) Larvae Growth

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Abstract: An increasing global population, environmental concerns, and the rising prices of conventional types of animal protein have generated a need for and interest in substitutive protein sources. Yellow mealworms, the larval form of *Tenebrio molitor* L. (Coleoptera: Tenebrionidae), are a promising new alternative protein source in the food industry. These larvae can be raised on agricultural by-products and supplemental organic material such as fruits and vegetables. This experiment evaluated the growth rate and survival of yellow mealworms when provided with a supplemental diet of different fruits and vegetables in a substrate of 50% brewer's yeast in wheat bran. Quantitatively, larvae provided carrots as the supplemental material has the highest survival rate at 95%, and larvae given potatoes had the greatest average weight gain (M=25.1 mg). While no statistically significant correlation was found between the type of organic supplemental material and weight gain (p=0.783) and larval survival rate (p=0.201), it was determined that organic supplements with a greater carbohydrate composition resulted in higher weight gains, and high moisture levels in the provided organic materials contributed to larval deaths during the experiment.

Keywords: *Tenebrio molitor,* yellow mealworm, larvae, growth rate, weight

Entomophagy, the consumption of insects, has drawn increasing attention as the world population grows and the demand for animal protein continues to rise. Increases in prices for standard animal proteins, such as beef, pork, and chicken, as well as greenhouse gas emissions from livestock production have produced a demand for alternative protein sources that can possibly be filled through the nutrients provided by various species of insects. Traditional animal protein prices are expected to rise by at least 30% between 2000 and 2050, increasing food scarcity and insecurity, but greater efficiencies in insect production could help combat these issues (Van Huis 2013).

Insects are much more environmentally friendly to produce than traditional livestock animals, have a much higher agricultural land use efficiency, and are able to convert agricultural by-products and food waste into nutritious food products (Alexander et al. 2017). In particular, the larval form of *Tenebrio molitor* L. (Coleoptera: Tenebrionidae), the yellow mealworm, has risen in popularity as a possible alternative source of protein due to various benefits that include, but are not limited to, their high

protein content, well-balanced amino acid profile, low greenhouse gas emissions, and efficient feed conversion rate (Liu et al. 2020). *T. molitor* protein and caloric yields are second only to high protein primary producers, such as soybeans, and all essential amino acids can be obtained from their consumption (Alexander et al. 2017). The global warming potential for the rearing of mealworms is lower than that of all other common animal protein sources. To obtain the same product weight, mealworms require only 43% of the land required for milk production and 10% of the land required for beef production (Oonincx et al. 2012). The feed conversion efficiency is increased by the fact that 100% of mealworm mass can be consumed, compared to only 40% in beef production (Alexander et al. 2017).

These omnivorous larvae can be raised on a variety of fruit and vegetable scraps, as well as the cereal grains that the adult beetles prefer. Despite preliminary research indicating higher growth rates and pupal weights when *T. molitor* larvae diets are supplemented with fresh plant material, conclusive data documenting growth rates after supplementation with fresh fruits and vegetables are limited (Liu et al. 2020).

Materials and Methods

T. molitor larvae were obtained through an online supplier (Rainbow Mealworms, Compton, CA) at an approximate size of ¼ inches. Twenty larvae were then separated and placed individually into uniform two-ounce plastic mini cups with lids (Meijer, Walker, MI). Each lid was punctured multiple times with a needle in order to provide proper air circulation. The initial weight of each larva, in milligrams, was obtained using a digital jewelry scale (Smart Weigh Scales, New York, NY) The mini cups were then filled with a prepared substrate mixture of 50% animal grade brewer's yeast (Josh's Frogs, Owosso, MI) and wheat bran (Bob's Red Mill, Milwaukie, OR).

The organic materials used in this experiment were Autumn Glory® apples (Domex Superfresh Growers, Inc., Yakima, WA), carrots, Sungold™ kiwifruits (Zespri International Limited, Mount Maunganui, New Zealand) and russet potatoes, with Fluker's Orange Cube Complete Cricket Diet (Fluker's Cricket Farm, Inc., Port Allen, LA) used as a control. Each material was provided *ad libitum* to 4 *T. molitor* larvae.

The larvae were sifted from the substrate and weighed every three days for 15 days. The organic material within each cup was also removed every three days and replaced in order to prevent mold growth within the mealworm cups. Upon observation, any deceased larvae, which could be identified through a darker black color and lack of movement, were removed and the number of deaths were recorded.

Statistical Analysis. The total average weight differences and larval survival rates were analyzed using a one-way ANOVA test in Excel for Windows 10 (Microsoft Corporation, Redmond, WA) to determine significance.

Results

The highest average survival percentage after 15 days was observed in the group that was supplemented with carrots at 95%, and the

lowest survival percentage was seen in the control group, treated with Fluker's Orange Cube Complete Cricket Diet at 75%. *T. molitor* larvae supplemented with apples or potatoes had the second highest survival percentage at 85%. Mealworms supplemented with kiwi were observed to have an 80% survival rate. However, the differences observed between the survival percentages were not statistically significant (p=0.201) (Table 1).

The total average weight gain was observed to be greatest in the mealworms that were supplemented with potatoes (M=25.1) and lowest in the mealworms supplemented with apples (M=12.7), but this effect was not significant (p=0.783) (Table 1). Mealworms in the control group (M=18.7) and those treated with carrots (M=22.0) and kiwi (M=19.8) had moderate weight gains.

Table 1. Larval survival percentage and total average weight gain in relation to supplemental organic material provided.

Diet	Survival (%)	Total Average Weight Gain (mg)
Fluker's (Control	75	18.7
Carrots	95	22
Apples	85	12.7
Potatoes	85	25.1
Kiwi	80	19.8
p-Value	0.201	0.783

All of the supplemental organic materials had a higher larval survival rate than the commercially produced control. All supplements except apple produced a greater average weight gain than the control group.

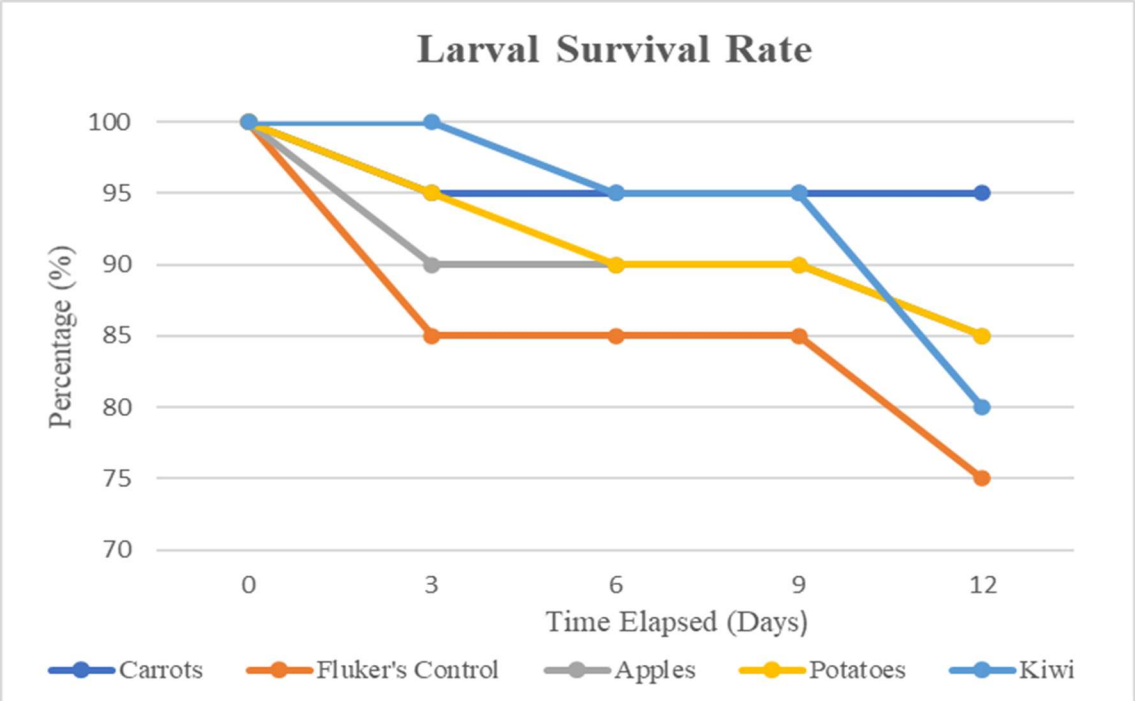

Fig. 1. Larval survival rates as a percentage of total sample size of larvae of *Tenebrio molitor* (n=20).

Discussion

Tenebrio molitor, has the potential to serve as a sustainable source of edible protein for the human population (Oonincx and de Boer, 2012). Insect growth rate and nutritional composition can be altered through changes in diet to increase production efficiency. Since this typically occurs in high protein diets, it was unexpected that the mealworms in the control group, given the supplement with the greatest protein content, did not have the greatest average weight gain (Van Broekhoven et al. 2015).

While the larval survival rate and total average weight gain values produced in this study were not significantly different, the values obtained can still be compared and conclusions drawn. In particular, it was found that the largest total weight gain was not found in the control but rather in the group that was provided with potatoes as a dietary supplement. While this opposes previous expectations, potatoes contain around 18 grams of carbohydrates per every 100 grams of potato. They also contain several nutrients and minerals and are relatively high in protein compared to the other fruits and vegetables tested (Duroy et al. 2009). This could be the reason behind this outcome. Carbohydrates are broken down and turned into ATP within cells, which are then used as energy. As such, a larger amount of carbohydrates results in increased energy production, which also increases growth and therefore weight gain. It also seems likely that the protein and mineral levels were high enough in potatoes to support development. This study suggests that potatoes may have a place in mealworm diet supplementation to increase larval weight gain for use as an alternative protein source.

It was also observed that Fluker's organic supplement produced the lowest larval

survival rate at 75% (Table 1). This lower survival rate could be attributed to the high moisture content of the supplement (92%) (Fluker Farms 2021). This moisture content resulted in a build-up of moisture within the enclosures used in the experiment, which encouraged mold growth towards the end of the 15 days of observation. Mold was also observed in the apple and carrot enclosures towards the end of observation, which can also be attributed to their higher moisture contents at 86% to 89% and 85.3% respectively (Sharma 2012, Ferretti 2014). Overall, even with the air circulation provided, mold growth still occurred, and this information could prove useful towards future studies regarding the steps that should be taken to ensure proper ventilation when using more moist supplements. Larger sample sizes and a longer observation window could aid future studies in obtaining more data and gaining a greater understanding of the effects of these supplements throughout development.

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