Evaluation of nutrition of Black Soldier Flies Larvae (BSFL) using Van Soest and fatty acid profile reared at different media

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Abstract. The current study aimed to evaluate nutrition of black soldier fly larvae (BSFL) using Van Soest and fatty acid profile reared at different media. A total of five different treatments were used in this study as follows: fruit waste, food waste, tofu by-product, 50% fruit waste + 50% food waste, and 50% fruit waste + 50% tofu by-product. The parameters observed were fatty acid profile (saturated and non-saturated fatty acid) and fiber composition (Neutral Detergent Fiber, Acid Detergent Fiber, Cellulose, hemicellulose, and lignin). Fatty acid profile analysis was done using gas chromatography and fiber composition using Van Soest method. The result presented the composition of fatty acid in BSFL dominated by saturated fatty acid, namely lauric acid (34.28 – 38.39%), palmitic acid (17.45 – 19.25%), arachidic acid (8.8 – 10.41%), myristic acid (7.35 – 9.36%), heptadecanoic acid (2.03 – 2.91%), and decanoic acid (1.23 – 1.42%). Non-saturated fatty acid; linolelaidic acid (13.51 – 16.86%), oleic acid (1.64 – 1.91%), nervonic acid (1.34 – 1.46%), dan linolenic acid (0.65 – 1.03%) measured higher as compared to the rest. Fiber components measured were NDF (20.19 – 29.11%), ADF (10.43 – 17.55%), hemicellulose (8.86 – 11.56%), cellulose (2.51 – 10.49%), and Lignin (0.14 – 0.75%). Respectively, in conclusion composition of growth media of BSFL influence the profile of fatty acid and fiber in BSFL. In addition, the media composed of 50% food wastes and 50% tofu by-product gave highest and most complete fatty acid profile as well as fiber composition.

1 Introduction

Indonesia is a developing country, populated by 270.20 million people [1] and still growing. This population growth is accompanied by the increasing production of waste [2]. The amount of organic waste produced by Indonesia reaches up to 60% of the nation’s total waste. A large portion of organic waste comes from food waste. The amount of food waste of Indonesia is around 300 kg per person annually, placing the nation second in global food waste production in large category [3]. Aside from food waste, organic waste types that are

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abundant and still minimally utilised are market waste, for instance, fruit waste, vegetable waste, and industrial waste such as tofu by-product [4]. Each of the mentioned waste are still containing large amount of nutrition. For example, tofu byproduct still contains around CP 21%, fat 3.79%, DM 48.37%, and ash 1.21% [5].

The use of insects has been one of the references to be a solution to utilisation of organic waste. It is also mentioned to be beneficial to the livestock industry. Insect is a natural organic bio converter that consumes organic wastes to produce beneficial products. [6] stated that bio-conversion by insects is more efficient, environment- friendly, as well as requiring minimum space. Out of some of the insects that have been the subject of research such as Black Soldier Fly Larvae (BSFL), Mealworm larvae (Tenebrio Molitor L.) and crickets (Orthoptera: Gryllidae). BSFL possesses a level of conversion and resilience that are superior to those of mealworms and crickets, which allow less difficult cultivation [7]. BSFL is also known to have the ability to consume higher diversity of organic matter as compared to other larvae [8] therefore, BSFL could be a solution for solving organic waste problem. In the adult phase, BSF is not considered a vector of pathogens to humans, animals, and plants [9]. According to [10] BSFL contains excellent nutrients, it possesses high protein, complete fatty acid, high feed conversion, short reproduction cycle, mineral, as well as high in vitamin. This implies that BSFL has the potential for a poultry feed alternative.

The composing nutrients of BSFL is influenced by the applied media on which it was reared. Good growth media results in BSFL with higher quality of nutrition. BSFL contains over 28% fat [8]. According to [11] the fat of BFSFL could be utilised for biodiesel, oleo chemical, soap, and consumption. A number of countries have started using BSFL fat as condiments for bread and margarine. According to a study of [12] the nutritional content in form of crude fat and crude fibre of BSFL reared on food waste respectively around 31.95% and 17.85%. Nutritional content of maggot reared in tofu by-product showed crude fibre 8.72% and crude fat 39.41% [2]. In comparison to other insects, BSFL contains high fat and fibre with the fat mainly in the form of saturated fatty acids (SFA) [13]. Since the imposed of antibiotic growth promoters (AGPs) in Europe 2006, it has continued with developing countries as well. There is an alternative to seeking replacement of AGPs [14].

The earlier finding reveals the possibility of modification to be done in the composition of maggot nutrients such as the fatty acid and fibre fragments, which is due to the absence of information of which nutrition the maggot itself produce, or to what extent the nutrition of the media influences the fatty acid and fibre composition. Understanding of this matter is highly important if the maggots are to be reared in different media and to be applied to animal feed or other industries. This study aimed to evaluate the effect of different organic wastes as media of maggot growth in the fatty acid and crude fibre composition profile.

2 Materials and Methods

2.1 Materials

2.1.1 Material

The maggot was reared in Malang. Fatty acid profile test was done in Laboratorium Penelitian dan Pengujian Terpadu (LPPT) Gajah Mada University, Yogyakarta. Fiber content analysis was conducted in Laboratorium Nutrisi dan Makanan Ternak Universitas Brawijaya, Malang.

Black Soldier Fly Larvae (BSFL) was used priori reared for 28 days on organic waste media (fruit, food waste, and tofu byproduct). Fruit waste was obtained from markets (from apple, pear, dragon fruit, and starfruit) that were no longer in any quality for sale, food waste
was obtained from restaurants and households, Tofu byproduct was obtained from tofu factories, dried and ground shortly after, before being given to BSFL.

2.1.2 Media
Media of growth applied are fruit waste, food waste, and tofu by-product. Feed was provided at 5 kg per day. Nutrition content of every media is displayed on table 1.

Table 1. Nutritional content of BSFL growth media.

<table>
<thead>
<tr>
<th>Media</th>
<th>DM (%)</th>
<th>Ash (%DM)</th>
<th>CP (%DM)</th>
<th>Fat (%DM)</th>
<th>CF (%DM)</th>
<th>NFE (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fruit Waste</td>
<td>93.07</td>
<td>2.68</td>
<td>7.76</td>
<td>1.09</td>
<td>6.03</td>
<td>82.44</td>
</tr>
<tr>
<td>Food Waste</td>
<td>40.44</td>
<td>1.67</td>
<td>15.03</td>
<td>12.45</td>
<td>19.77</td>
<td>51.08</td>
</tr>
<tr>
<td>Tofu Byproduct</td>
<td>11.00</td>
<td>4.27</td>
<td>20.03</td>
<td>2.89</td>
<td>2.48</td>
<td>70.33</td>
</tr>
</tbody>
</table>

Source: Central Laboratory, Muhammadiyah University, Malang (2023)

DM – Dry Matter; CP – crude protein; CF – Crude Fibre; NFE – Nitrogen-Free Extract

2.2 Research method
Analysis of fatty acid profile by gas chromatography and fibre fragments analysis by van-soest method. Research method is descriptive with 5 treatments. The treatments tested include fruit waste (T0), food waste (T1), Tofu byproduct (T2), 50% fruit waste + 50% tofu byproduct (T3), and 50% fruit waste + 50% food waste (T4).

3 Results and discussion
3.1 Fatty Acid Profile
Table 2. shows the effect of different media of BSFL growth being determinant to fatty acid profile of BSFL. The dominant fatty acid in all treatments of BSFL media exhibited lauric fatty acid (34.28-38.39%), followed by palmitic acid (17.45-19.25%) and linoleic acid (13.52-16.86%). The larger fraction of BSFL contains a high saturated fatty acid (SFA) percentage, followed by poly-non-saturated fatty acid (PUFA; up to 18.57%), and mono-non- non-saturated fatty acid (MUFA: up to 4.2%). The high level of SFA in BSFL is a way to store energy for the next phase in the life cycle [16]. Storage of energy in the form of SFA is backgrounded by the lack of its vulnerability against oxidation as compared to unsaturated fatty acid (NSFA) [17]. Lauric Acid from SFA is the main element (up to 38.39%) in BSFL. In this study, the highest lauric fatty acid was obtained from T4 and followed by P3.

Table 2. Fatty acid content of BSFL

<table>
<thead>
<tr>
<th>Fatty acid type</th>
<th>T0</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
</tr>
</thead>
<tbody>
<tr>
<td>SFA</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Butyrate acid</td>
<td>0.35</td>
<td>7.84</td>
<td>&lt;0.1</td>
<td>0.16</td>
<td>0.16</td>
</tr>
<tr>
<td>Lauric acid</td>
<td>34.28</td>
<td>35.14</td>
<td>35.99</td>
<td>37.56</td>
<td>38.39</td>
</tr>
<tr>
<td>Miristic acid</td>
<td>8.58</td>
<td>7.35</td>
<td>8.9</td>
<td>9.36</td>
<td>9.31</td>
</tr>
<tr>
<td>Palmitic acid</td>
<td>19.25</td>
<td>17.74</td>
<td>18.34</td>
<td>17.45</td>
<td>18.43</td>
</tr>
<tr>
<td>Heptadecanoic acid</td>
<td>2.57</td>
<td>2.03</td>
<td>2.84</td>
<td>2.82</td>
<td>2.91</td>
</tr>
<tr>
<td>Decanoic acid</td>
<td>1.37</td>
<td>1.42</td>
<td>1.31</td>
<td>1.23</td>
<td>1.31</td>
</tr>
<tr>
<td>Arachidate acid</td>
<td>10.41</td>
<td>8.98</td>
<td>10.11</td>
<td>10.22</td>
<td>8.8</td>
</tr>
<tr>
<td>Heneicosanoic acid</td>
<td>0.44</td>
<td>0.24</td>
<td>0.51</td>
<td>0.49</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>Docosanoate acid</td>
<td>&lt;0.1</td>
<td>0.51</td>
<td>&lt;0.1</td>
<td>0.58</td>
<td>&lt;0.1</td>
</tr>
</tbody>
</table>
Lauric acid (C:12) is a type of fatty acid commonly found in BSFL that functions for antimicrobial, antivirus, and antifungal. That makes BSFL a potential feed component because in addition to the high protein content, the use for antibiotic is also applicable. The high lauric acid in BSFL is suspected to be because the mentioned fatty acid is synthesized by the BSFL itself through carbohydrate synthesis in a substrate and could increase as per decomposition of the organic substrate [17] The claim is supported by [18] that carbohydrate from substrate forms a number of fatty acids through glycolysis, oxidative decarboxylation, and later becomes fatty acid. The process exhibits the ability of BSFL to synthesize lauric acid coming from non-fat from its body. The NFE and lauric acid is very closely related due to NFE being group of fractions of digestible carbohydrates. However, in the current study, any evidences of high NFE being able to increase lauric acid content in BSFL are yet to be found. The provision of fruit waste mixed with food waste or tofu by-product (T3 and T4) could increase lauric acid in BSFL as compared to each as their own media (T0, T1, and T2). In comparison, the result of a study by [17] showed higher lauric acid in similar media of food waste (57.56%) and fruit waste (60.89%). the result is far higher in comparison to that of the current study. The content of lauric acid could vary depending on the nutrition of the media and environment of rearing. In a number of studies, lauric acid in food waste media showed 13.44%, 39.9%, and 7.4% [18,19,20] In the current study, the proceeding high SFA content is arachidate acid (8.8 – 10.41%), palmitic acid (17.45-19.25%) and myristic (7.35%-9.36%). This aligned with [23] that SFA content showed highest after lauric is myristic (5,71-8.89%) and palmitic (5.84-11.51%). According to [24] SFA composed of C12:0, C14:0, C16:0 could be synthesized by BSFL. Other SFA fatty acid such as butyrate (<0.1 – 7.84%), Heptadecanoic acid (2.03 – 2.91%), decanoic acid (1.23 – 1.42%), heneicosanoic acid (<0.1 – 0.51), and docosanoate acid (<0.1 – 0.58) was also detected in BSFL, however in very low quantity.

Unsaturated fatty acid (USFA) is split into Monounsaturated fatty acid (MUFA) and Polyunsaturated Fatty Acid (PUFA). The division is based on the bond, MUFA has a single bond, while PUFA has more than two. In the current study, the volume of MUFA is smaller as compared to PUFA. MUFA was dominated by oleic acid (1.64-1.91%) and nervonic acid (1,34-1,46%). Oleic acid could represent essential fatty acids that are important to consider in formulation of drugs and could potentially benefit wound recovery. Consumption of MUFA could lift production of high-density lipoprotein (HDL) in blood, suppressing low-density lipoprotein (LDL) in blood and supporting heart health. The highest amount of MUFA in the current study is obtained from T0, despite generally fruit does not produce MUFA. This implies that BSFL could synthesize MUFA on its own. This aligns with [24]
that MUFA such as oleic acid (C18:1) could be produced by BSFL on its own without interference of nutritional content from substrate.

Table 2. displayed that PUFA content in all treatments are high (14.8 – 18.57%). The composing components of PUFA include essential fatty acids, namely omega-3 and omega-6. Linoleic acid is a type of omega-6 fatty acid. In this study, the amounts of linoleic acid obtained from each treatment are T0 16.86%, P1 13.51%, P2 16.68%, P3 14.42%, and P4 15.26%. The highest to contain are T0 and T2 due to presence of the fatty acid in the media. According to [24] linoleic acid is considered a type of fatty acid not being produced by BSFL itself. This fatty acid is highly influenced by the content contained in the substrate. The composition of PUFA include Linoleic acid, EPA, and DHA are essential fatty acid type omega-3. In table 2. It is visible that DHA (0.59 – 1%) and EPA (<0.01%) content are found in small amounts. T2 displayed the highest value of DHA as compared to the rest of the treatments. DHA content of BSFL in all treatments are smaller in comparison to [21] that obtained 5% DHA in ensiled muscles media. Such could occur due to the difference in fatty acid content in the substrate. [25] Suspected that there is a positive correlation between the percentage of fatty acid in the media and in the larvae’s body. According to [31] the low content of EPA and DHA occur due to the increasing quantity of SFA. It was proven that the highest SFA content is T1(81.25%) producing DHA content (0.59%) that is the lowest compared to the rest.

Fatty acid of BSL closely resembles the fatty acid content of palm oil and coconut oil. Therefore, it is possible to utilise fatty acid of BSFL in the related industry [26]. The presence of BSFL fatty acid makes plant based protein fatty acid also obtainable from animals. [27] found that fatty acid in BSFL is suitable for production of high quality biodiesel. From the mentioned matter. BSFL fat seemingly has good potential for being utilized in production of feed, food, and fuel

3.2 Fibre Composition of BSFL

Fibre composition of BSFL on each treatment is displayed in Table 3. On the table, it is displayed that the fibre compositions in BSFL differ according to the media of growth implemented. NDF content on this study is 20.19 – 29.11% where the content of NDF at the highest was held by T1 (29.11%) which was organic food waste. This takes place because in Table 1. it is displayed that the fibre composition in the food waste media was the highest compared to the rest. Food waste contains abundant plant and animal protein based, presence of vegetables in the food waste could increase the fibre content in the media and influence BSFL nutritional content. ADF in each treatment namely T0 11.66%, T1 17.55%, T2 10.80%, T3 10.43%, T4 11.75%. In a study of [26] content of NDF and ADF of BSFL each are respectively 35.53% and 29.27%. In this study it is also mentioned that the lower the content of crude fibre, NDF, and ADF in BSFL could indicate the declining chitin content. ADF content is a fraction of fibre that is soluble in acid detergent composed of cellulose, lignin, and silica that could indicate that the feed has low digestibility [27]. Therefore, the utilisation of BSFL for ruminant feed as well as non-ruminants should decide NDF and ADF content that are low. According to [28] crude fibre in insects represents chitin content due to chitin (linear polymer unit b-(1-4) N-Acetyl-D-glucosamine) has structure similar to that of cellulose (linear polymer unit b-(1-4) N-acetyl-D-glucosamine) and in fraction of ADF insects have proven to contain nitrogen.

<table>
<thead>
<tr>
<th>Code</th>
<th>NDF (%)</th>
<th>ADF (%)</th>
<th>HEMICELLULOSE (%)</th>
<th>CELLULOSA (%)</th>
<th>LIGNIN (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T0</td>
<td>20.52</td>
<td>11.66</td>
<td>8.86</td>
<td>3.27</td>
<td>0.16</td>
</tr>
</tbody>
</table>
In table 3. The hemicellulose content in BSFL is 8.86 – 11.56%. Hemicellulose is a fraction of crude fibre easiest to degrade, obtained from lowering of NDF with ADF. The high content of hemicellulose in the current study as in T1, 11.56% and lowest in T0, 8.86%. Cellulose and lignin content that are a fraction of fibre more challenging and difficult to digest into simpler carbohydrates. Lowest content of cellulose was on T2, 2.51%. Lowest in lignin content was T3, showing 0.14%. The decreasing content of lignin in feed, the higher the digestibility of the feed. That occurs because lignin is one of the fibre compulsions that is difficult to break down into simpler sugar. To maggots, lignin is fibre that is very difficult to turn into fatty acid [31]. In the digestive system of BSFL, there are lignin digesting microorganisms. These microorganisms will convert lignin into cellulose that will be used by maggots for energy [30]. Lignocellulose converts lignin into simple sugars in the form of enzymes that are produced by BSFL [2]. A number of studies showed that providing crude fibre could decrease the protein and fat in maggots. Maggot is suspected to not have enzymes for decomposing fibre. However, hosts fungi and microbes in the back of its middle digestive system with pH of 6-8 that could degrade crude fibre [31].

4 Conclusion

After analysis of fatty acid profile and fibre composition, it is discovered that the composition of fatty acid and fibre are influenced by the media of growth, although the exerted difference is relatively small. According to the potential of application, the content of fatty acid in BSFL could be utilised for alternative to protein based plant oil in food, feed, as well as biofuel production.

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