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Early report of effect of diet on physical properties of black soldier fly larvae meal

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Abstract

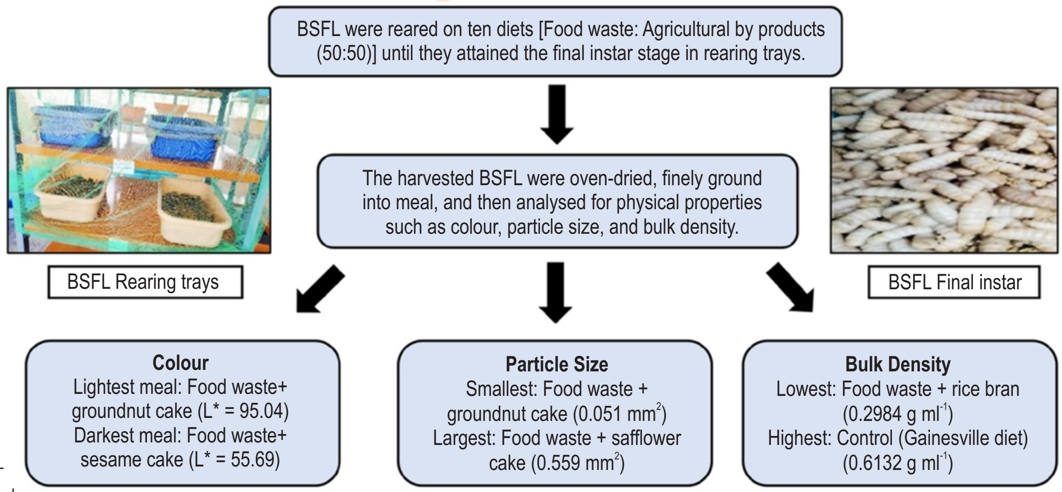
Aim: This study was undertaken to document the influence of a 50:50 combination of food waste and various agricultural byproducts on the particle size, bulk density and colour of Black Soldier Fly Larvae (BSFL) meal, addressing how dietary composition influences its potential as animal feed.

Methodology: Ten treatments, each combining equal proportions of food waste with specific agricultural byproducts, were prepared and replicated three times. BSF larvae were fed these diets, and the resulting meal was analysed for colour (using the CIELAB system) (Schneider *et al.*, 2012), particle size (measured with Image J software) (Schneider *et al.*, 2012), and bulk density following Okezie and Bello (1998).

Results: Significant variations were observed across treatments. The lightest meal was produced from food waste combined with groundnut cake (L* = 95.04), while the darkest meal came from food waste with sesame cake (L* = 55.69). Particle size was smallest in the groundnut cake treatment (0.051 mm^2) and largest in safflower cake (0.559 mm^2). The control group (Gainesville diet) exhibited the highest bulk density (0.6132 g ml^{-1}), while the lowest was found in food waste with rice bran (0.2984 g ml^{-1}).

Interpretation: The study indicates that dietary composition significantly influences BSFL meal's physical properties, providing insights for optimizing formulations in animal feed production.

Key words: Animal nutrition, Black soldier fly larvae, Feed formulation, *Hermetia illucens,* Waste management



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Introduction

The pressing challenge of supporting a rapidly increasing global population, as foreseen in "Limits to Growth" (Meadows *et al.,* 1972), has become a reality. The global population, current 8.1 billion, is projected to rise to 10.2 billion by 2100 (UN DESA, 2024), placing greater pressure on food systems. This growing demand, driven by increasing daily caloric intake and greater meat consumption linked to higher incomes, intensifies the strain on limited land and water resources, particularly for cereals and protein rich foods (Alexandratos and Bruinsma, 2012). Additionally, modern food systems must balance high productivity with environmental sustainability, animal welfare and product quality (Makkar, 2016). Given that feed accounts for up to 70% of total production costs in animal production, identifying cost-effective alternative feed sources is crucial for improving efficiency and ensuring economic viability (Makkar and Beever, 2013). One promising alternative is insect farming, which requires minimal land and water resources (Huis *et al.,* 2013) and can utilize pre- or post-consumer food waste as feed (Nguyen *et al.,* 2013; Leong *et al.,* 2016). Managing food waste - 931 million tonnes globally (UNEP, 2021) - is a major environmental challenge, as much of it ends up in landfills, leading to leachate, greenhouse gas emissions and pest attraction (Cameron *et al.,* 2005).

Although over 2,000 insect species are consumed worldwide, only a few have been studied for livestock or aquaculture feed. Among them, Black Soldier Fly (BSF) (*Hermetia illucens* L.) stands out for its ability to convert organic waste into nutrient-rich biomass, feeding on a wide range of materials, including fruits, vegetables and animal remains (James, 1935; May, 1961). For over 30 years, research has highlighted BSF's potential in waste management, with larvae capable of reducing organic waste by 50% (Sheppard *et al.,* 1994). BSF larvae are nutritionally rich, containing 45.2% crude protein and 31.4% fat, making them a valuable feed ingredient, estimated at US$ 330 per ton (Hale, 1973; Newton *et al.,* 2005; Nguyen *et al.,* 2015). Furthermore, BSF larvae do not carry pathogens and can inactivate harmful bacteria like *Escherichia coli* and *Salmonella* (Erickson *et al.,* 2004). This species is widely distributed across tropical and temperate regions (Leclerq, 1969), with a life cycle well adapted to such environments. Eggs hatch within four days, and larvae feed for approximately two weeks, accumulating substantial fat reserves before pupating (Booth and Sheppard, 1984; Tomberlin and Sheppard, 2002).

Studies have demonstrated that BSF larvae meal is a viable feed replacement for livestock, including cockerels, pigs, catfish, and tilapia (Newton *et al.,* 1977; Bondari and Sheppard, 1981). Its balanced amino acid profile and high mineral content enhance growth performance, feed efficiency, and overall livestock health (Schiavone *et al.,* 2017; Kawasaki *et al.,* 2019; Zotte *et al.,* 2019; Rawski *et al.,* 2020). With growing interest in BSF-based feeds, regulatory frameworks are being developed to ensure safety, quality, and sustainability, guided by organizations such as the European Food Safety Authority (EFSA) and the U.S. Food and Drug Administration (FDA) (Raman *et al.,* 2022; Shah *et al.,* 2022). To improve the nutritional and physical properties of Black Soldier Fly Larvae (BSFL) meal, various processing techniques, like size reduction, mixing and pelleting are employed. These methods influence physical properties such as particle size, bulk density, and colour, which are critical for feed handling, storage, and processing efficiency (Syarif and Irawati, 1988; Chung and Lee, 1995; Jaelani and Firahmi, 2007; Kushwaha *et al.,* 2015; Syamsu *et al.,* 2015; Tanpong *et al.,* 2019). Additionally, fat content, drying methods, and the presence of phenolic compounds in the insect cuticle can affect meal characteristics, particularly colour variation, which influences its visual appeal in feed applications (Akpossan *et al.,* 2015; Janssen *et al.,* 2019; Pornsuwan *et al.,* 2023). Understanding the physical characteristics of BSFL meal is essential for its effective use in animal nutrition. Parameters such as bulk density, particle size, and friction coefficients play a crucial role in optimizing feed formulation, processing, and storage logistics (Tanpong *et al.,* 2019). In this context, the present study was carried out with the aim to assess the effects of diet on the physical properties of BSFL meal.

Materials and Methods

The experiment was conducted at the Black soldier fly breeding Unit, Agricultural Research Institute, Rajendranagar, Hyderabad, to evaluate the effects of different diets on the physical properties of BSFL meal, specifically colour, particle size and bulk density. The study consisted of 10 treatments, each replicated three times. Each treatment included a 50:50 proportion of food waste and one of the following amendments: T_1: Food waste + Sesame cake; T_2: Food waste + Safflower cake; T_3: Food waste + Cotton seed cake; T_4: Food waste + Groundnut cake; T_5: Food waste + Rice bran; T_6: Food waste + Red gram husk; T_7: Food waste + Green gram husk; T_8: Food waste + Unhatched eggs; T_9: Food waste + Wheat bran and T_{10}: Control (Gainesville diet: Corn: Alfalfa: Wheat bran at 20:30:50) (Hogsette, 1992). These byproducts were selected due to their abundance, nutritional value, cost-effectiveness, and potential to influence the physical properties of BSFL meal. Oilseed cakes (sesame, safflower, cottonseed, and groundnut) are protein-rich feed ingredients, while rice bran and wheat bran are commonly used as energy sources. Pulses byproducts (red gram husk and green gram husk) provide fibre, and unhatched eggs were included to assess their potential as an organic protein source. The Gainesville diet was used as a standard control (Hogsette, 1992). The BSF colony was maintained following the protocol of Sheppard and Tomberlin (2002).Upon reaching the final instar stage, larvae were harvested, oven-dried, and finely grounded into meal for further analysis.

Colour estimation: A 5 g sample of BSFL meal was collected from each treatment for colour analysis using the CIELAB system (Schneider *et al.,* 2012). The parameters measured included: L*[lightness on a scale from 0 (black) to 100 (white)], a* axis [redness (positive) to greenness (negative)], and b* axis [yellowness (positive) to blueness (negative)]. The colour values were analysed using Image J software.

Estimation of particle size: To assess particle size, BSFL meal was sieved through a 500 µm sieve. Images of the sieved sample were captured under a stereo zoom binocular microscope (10X magnification) and analysed with ImageJ software (Schneider *et al.,* 2012). Retention and passage percentages were calculated using formulas from Tanpong *et al.* (2019).

Bulk density measurement: Bulk density was measured as the mass-to-volume ratio using a pre-tared graduated cylinder. BSFL flour was filled upto 10 ml mark, gently compacted by tapping until no further settling occurred, and the final volume was recorded. The bulk density was calculated by dividing the sample's weight by its volume (gm l^{-1}) (Okezie and Bello, 1998).

Statistical analysis: Data analysis was carried out using Microsoft Excel, where mean and standard deviation (SD) for particle size and bulk density were calculated.

Results and Discussion

The dietary composition significantly influenced the colour properties of BSFL meal, as indicated by the variations in L* (lightness), a* (redness-greenness) and b* (yellowness-blueness) values across the treatments (Table 1; Fig. 1). Among the treatments, T_1 (Food waste + Sesame cake) exhibited the lowest L* value (55.69), indicating a darker meal, likely due to the pigmentation associated with sesame cake. This was accompanied by moderate a* (17.61) and b* (18.48) values, reflecting a balanced reddish-yellow tone. In contrast, T_2 (Food waste + Safflower cake) showed a lighter colour (L* = 66.14) with slightly reduced a* (16.29) and b* (18.15) values, indicating lower pigmentation. T_3 (Food waste + Cotton seed cake) produced a significantly lighter meal (L* = 81.31), with moderate a* (18.65) and b* (19.91) values imparting a subtle reddish-yellow tone. The lightest colour was observed in T_4 (Food waste + Groundnut cake), which had the highest L* value (95.04), along with the lowest a* (13.69) and higher b* (23.44), resulting in a pale-yellow tone characteristic of groundnut feed. T_5 (Food waste + Rice bran) maintained a moderately high L* (83.20) and balanced a* (18.44) and b* (19.20) values, contributing to a subtle yellowish hue. Similarly, T_6 (Food waste + Red gram husk) showed a high L* (92.85) and a distinct yellow tone with moderate a* (17.04) and elevated b* (23.22) values. T_7 (Food waste + Green gram husk) resulted in a moderately light colour (L* = 86.33) with vibrant reddish-yellow tones, as indicated by higher a* (18.85) and b* (21.19) values. A bright and distinctly yellow colour was observed in T_8 (Food waste + Unhatched eggs), which achieved a high L* (90.62) and the highest b* (26.68) value, while maintaining a moderate a* (14.87). T_9 (Food waste + Wheat bran) produced a darker shade with an L* of 71.11, paired with balanced a* (15.26) and b* (20.55) values, indicating a reddish-yellow tone. The control treatment, T_{10} (Gainesville diet), exhibited a lighter meal (L* = 88.29) with the highest a* (19.86) and moderately high b* (22.77), contributing to a vibrant reddish-yellow colour.

The results underscore the significant impact of dietary components on the physical appearance of BSFL meal. Ingredients such as sesame and safflower cakes contributed to darker tones due to their pigmentation, while groundnut cake and unhatched eggs enhanced lightness and yellowness, respectively. These findings align with earlier studies (Pornsuwan *et al.,* 2023; Adekunte *et al.,* 2010), which highlighted the role of feed composition and drying techniques in colour development. Phenolic compounds released during larval development likely contributed to the observed browning and pigmentation differences. Colour is a critical parameter in animal feed applications, influencing buyers' preferences and serving as an indicator of nutritional composition. Further research focusing on the specific pigments and bioactive compounds responsible for these variations will be crucial in improving the quality and commercial appeal of BSFL meal.

Table1: Effect of different diets on physical properties of black soldier fly larvae meal

Treatments	L*	a*	b*	Particle size (mm^2)	Retain (%)	Passing (%)	Bulk density (g ml^{-1})
T_1: Food waste + Sesame cake (50:50)	55.69	17.61	18.48	0.213±0.1008	86.60	13.71	0.4290±0.000702
T_2: Food waste + Safflower cake (50:50)	66.14	16.29	18.15	0.559±0.1390	96.25	3.67	0.3855±0.001097
T_3: Food waste + Cotton seed cake (50:50)	81.31	18.65	19.91	0.267±0.0691	95.57	4.47	0.3811±0.000379
T_4: Food waste + Groundnut cake (50:50)	95.04	13.69	23.44	0.051±0.0226	29.56	70.30	0.6059±0.002163
T_5: Food waste + Rice bran (50:50)	83.20	18.44	19.20	0.249±0.0622	90.36	10.13	0.2984±0.000551
T_6: Food waste + Red gram husk (50:50)	92.85	17.04	23.22	0.230±0.0341	97.21	2.97	0.4057±0.000586
T_7: Food waste + Green gram husk (50:50)	86.33	18.85	21.19	0.321±0.0406	90.17	9.87	0.3209±0.000458
T_8: Food waste + Unhatched eggs (50:50)	90.62	14.87	26.68	0.213±0.0359	34.65	65.10	0.6061±0.004508
T_9: Food waste + Wheat bran (50:50)	71.11	15.26	20.55	0.220±0.0176	90.17	9.87	0.5135±0.003485
T_{10}: Control [Gainesville diet (Corn: Alfalfa: Wheat bran) (20: 30: 50)]	88.29	19.86	22.77	0.170±0.0532	90.56	9.37	0.6132±0.001539

Values in the columns for particle size and bulk density are presented as mean ± S.D., indicating variability among replicates for each treatment.

Fig. 1: Effect of different diets on BSFL meal colour.

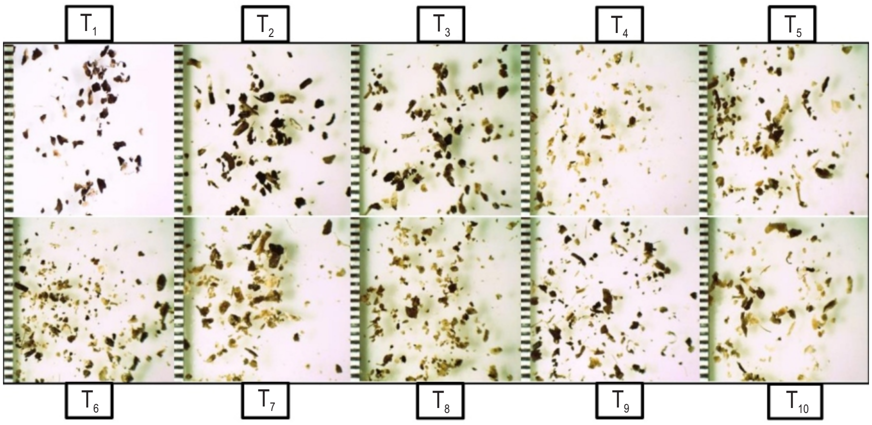

Fig. 2: Effect of different diets on BSFL meal particle size.

The study examined the particle size distribution in BSFL meal across ten treatments, each comprising food waste combined with different supplementary ingredients (Table 1; Fig. 2). Significant variations were observed among treatments, which were likely influenced by the physical properties of each additive. Treatment T_2 (Food waste + Safflower cake) resulted in the largest particle size, with a mean of 0.559 mm^2, and the highest retain percentage (96.25%), suggesting a more cohesive and dense particle structure. This may be attributed to the texture of safflower cake, enhancing compactness and particle uniformity. Similarly, T_7 (Food waste + Green gram husk) displayed a substantial particle size 0.321 mm^2 with a high retain percentage (90.17%), indicating a similar pattern of compact particle formation. In contrast, T_4 treatment yielded the smallest particles, averaging 0.051 mm^2, with the lowest retain percentage (29.56%), suggesting a loose and fine structure with increased sieve passage (70.30%). This result aligns with the granular nature of groundnut cake, promoting finely ground particles. T_8 treatments also exhibited smaller particle size, with a mean of 0.213 mm^2 and a passing percentage of 65.10%, implying that softer or easily fragmented materials contributed to finer, more dispersed particles. Treatments T_1, T_5 and T_9, which included sesame cake, rice bran, and wheat bran, displayed moderate particle sizes (0.213 mm^2, 0.249 mm^2 and 0.220 mm^2) and retain percentages around 86-90%, indicating an intermediate density and particle distribution. The compactness in these treatments may be attributed to bran and seed cake materials that provide consistent particle sizes upon grinding.

Treatments with red gram husk (T_6) and cotton seed cake (T_3) demonstrated slightly smaller particles (0.230 mm^2 and 0.267 mm^2), with high retention rates (97.21% and 95.57%), supporting the retention of uniform particles, possibly due to the fibrous nature of the husks. In comparison, the control diet (T_{10}), produced particles of 0.170 mm^2 with a retention of 90.56%, further demonstrating how ingredient composition affects the physical characteristics of BSFL meal. Larger particle sizes observed in T_2 (0.559 mm^2) and T_7 (0.321 mm^2) treatments may be advantageous in feed applications requiring increased texture and density, while finer particle distributions, as seen in T_4 (0.051 mm^2) and T_8 (0.213 mm^2) treatments could enhance nutrient absorption and feed processing. These results align with the findings of Pornsuwan *et al.* (2023), who noted similar particle size variations based on drying techniques, with microwave drying producing larger particles compared to hot-air oven method. Particle sizes ranged between 180 and 850 μm (20-80 mesh size).

Particle size significantly impacts feed preparation and animal nutrition by ensuring uniformity during storage, transportation, and mixing, and preventing nutrient imbalances. Smaller particles improve nutrient accessibility, feed efficiency, and animal performance (Svihus, 2011). Common feed ingredients like corn and soybean meals typically have particle sizes between 300 and 800 μm (Gao *et al.,* 2020), where finely ground feed benefits young animals by enhancing intake and nutrient absorption. Conversely, larger particles can hinder enzymatic digestion and intake (Vu *et al.,* 2015). Finer particles also optimize nutrient utilization, improving animal performance due to increased surface area (Kiarie and Mills, 2019). Such observations are consistent with the studies of Vukmirović *et al.* (2016) and Gao *et al.* (2020), which emphasize the critical role of particle size in enhancing feed efficiency and maintaining nutritional balance.

The analysis of bulk density values for BSFL meal across various treatments revealed significant insights into the physical properties and potential applications of BSFL meal (Table 1). Among the treatments, T_{10} (Control: Gainesville diet) exhibited the highest bulk density (0.6132 g ml^{-1}), suggesting a compact structure conducive for storage and transport. Similarly, T_8 and T_4 treatments displayed high bulk densities of 0.6061 g ml^{-1} and 0.6059 g ml^{-1}, respectively, indicating that these ingredients contribute to a denser, compact particle structure, potentially enhancing the nutritional value and ease of handling for production.

In contrast, T_5 treatment yielded the lowest bulk density (0.2984 g ml^{-1}), likely due to the incorporation of rice bran, which may produce a looser particle structure, potentially impacting its suitability for compact storage. Treatments such as T_2 and T_3 yielded moderate bulk densities of 0.3855 g ml^{-1} and 0.3811 g ml^{-1}, suggesting a more porous structure that could affect flowability in feed formulations. Intermediate bulk density values were observed in T_9 and T_6 treatments with densities of 0.5135 g ml^{-1} and 0.4057 g ml^{-1}, respectively. These findings imply that lighter ingredients, such as wheat bran and red gram husk, contribute to a reduced bulk density compared to more compact treatments. Additionally, T_1 and T_7 treatments showed bulk densities of 0.4290 g ml^{-1} and 0.3209 g ml^{-1}, respectively, supporting the trend that lighter components generally result in lower density.

The composition of dietary components play a critical role in influencing bulk density, as evidenced by these results. Previous studies by Bondari and Sheppard (1987) and Spranghers *et al.* (2017) align with our findings, indicating that diet composition can significantly impact the physical characteristics of BSFL meal. Additionally, Tanpong *et al.* (2019) reported differences in the bulk density when using various ingredients, such as citric acid by-products and cassava root meal, underscoring how ingredient density influences BSFL meal properties. Factors such as particle size and moisture content also affect the bulk density, with higher moisture levels potentially enhancing packing efficiency. Bulk density plays a crucial role in determining packaging requirements, material handling, and applications in industries, including the food sector (Zayas, 1997). High bulk densities imply ease of handling and storage, making these formulations more practical for commercial production, while lower densities may be preferred for feed formulations requiring optimal mixing and flowability.

In conclusion, this study demonstrates that the dietary composition significantly impacts the physical properties of BSFL meal, influencing its colour, particle size and bulk density. Variations in colour profiles highlight how different feed components affect the appearance of meal, which can play a crucial role in buyers' preferences and nutritional evaluation. The observed differences in particle size and bulk density further suggest that ingredient choices can enhance the meal's physical characteristics, making it suitable for various applications in animal nutrition and feed formulations. Understanding these dietary impacts can guide future research and improve the quality and utilization of BSFL meals across different industries.

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