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Feasibility of housefly larvae-mediated vermicomposting for recycling food waste added digestate as additive

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ABSTRACT

The development of methods for the efficient treatment and application of food waste digestate is an important research goal. Vermicomposting via housefly larvae is an efficient way to reduce food waste and achieve its valorization, however, studies on the application and performance of digestate in vermicomposting are rarely. The present study aimed to investigate the feasibility of the co-treatment of food waste and digestate as an additive via larvae. Restaurant food waste (RFW) and household food waste (HFW) were selected to assess the effects of waste type on vermicomposting performance and larval quality. Waste reduction rates of 50.9%–57.8% were observed in the vermicomposting of food waste mixed with digestate at a ratio of 25%, which were slightly lower than those for treatments without the addition of digestate (62.8%–65.9%). The addition of digestate increased the germination index, with a maximum value of 82% in the RFW treatments with 25% digestate, and decreased the respiration activity, with a minimum value of 30 mg-O_2/g-TS. The larval productivity of 13.9% in the RFW treatment system with a digestate rate of 25% was lower that without digestate (19.5%). Materials balance shows that larval biomass and metabolic equivalent had decreasing trends as the amount of digestate increased and HFW vermicomposting exhibited lower bioconversion efficiency than that of RFW treatment system regardless of the addition of digestate. These results suggest that mixing digestate at a low ratio (25%) during vermicomposting of food waste especially RFW could lead to considerable larval biomass and generate relatively stable residues.

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Introduction

Municipal solid waste sorting is widely implemented in China and generates large amounts of putrescible waste, such as restaurant food waste (RFW) and household food waste (HFW). According to Zhang et al. (2021), this type of waste was produced at 0.15 million ton/day in 2020 and is expected to increase rapidly over the next 5 years. Efficient and economical methods to handle such waste are needed to reduce environmental risks and achieve waste resource recycling. Traditional approaches for the treatment/disposal of putrescible waste, including anaerobic digestion, composting, and incineration, have various issues in terms of greenhouse gas emissions and resource recovery, despite their wide use (Lin et al., 2021; Wang et al., 2021). Hence, there is an urgent need to explore novel approaches for waste treatment.

Biotreatment via insects (also termed vermicomposting), including housefly larvae and black soldier flies, is considered an efficient means of reducing putrescible wastes, such as livestock and poultry manure, sewage sludge and food waste (Zhang et al., 2012; Zheng and Zhou, 2013; Čičková et al., 2015; Zhu et al., 2015; Negi et al., 2020; Shi et al., 2020; Cheng et al., 2021; Guo et al., 2021; Miranda et al., 2021). Vermicomposting with housefly larvae generates various useful products, including high-value materials for making organic fertilizer (Zhang et al., 2020) and high-quality insect protein for use in the fishing industry (Shi et al., 2020; Jiang et al., 2017). Furthermore, the whole vermicomposting process has advantages in terms of environmental impacts (i.e., carbon emissions) over conventional methods, like thermophilic composting, anaerobic digestion, and incineration (Guo et al., 2021; van Zanten et al., 2015; Mertenat et al., 2019; Pang et al., 2020; Song et al., 2021).

Digestate, an inevitable by-product of the anaerobic digestion of biowaste, is characterized by high water content and abundant organic matter (Lu and Xu, 2021) and may exert negative effects on the environment if improperly managed. Although some approaches (e.g., composting, incineration, and landfilling) have been applied to the treatment and/or disposal of digestate, they are not regarded as sustainable approaches owing to the low level of resource utilization (Peng et al., 2020; Xiao et al., 2022). To our best knowledge, few studies have evaluated treatment of digestate by vermicomposting with housefly larvae. Fu et al. (2022) found that digestate from the hydrogen/methane fermentation of food waste could be regarded as feed for the production of black soldier fly larvae with similar features to those of housefly larvae in terms of organic waste reduction. Nevertheless, the use of freeze-drying for the pretreatment of digestate in this previous study may result in an underestimate of bioconversion by black soldier flies. Additionally, understanding the characteristics of larva-handled food waste (i.e., vermicompost) is essential because thermal composting and other approaches are usually applied to improve the biostability and maturity of residues before use in agriculture (Song et al., 2021). Additionally, in comparison of black soldier fly larvae (around 2 weeks), housefly larvae could digest food waste more rapidly (typically in 3–4 days), indicating that it has a higher bioconversion efficiency. Hence, it is necessary to investigate the feasibility of digestate bioconversion via housefly larvae and to clarify the properties of the residues in order to explore a novel method for recycling digestate. In actual scenarios, bulking agents, like wheat bran, are added to food waste to adjust the water content of feedstock suitable for the survival and growth of housefly larvae (Cheng et al., 2017; Fu et al., 2022). Therefore, digestate, especially from dry anaerobic digestion with a relative loose structure, is expected to be a feasible alternative to wheat bran in the vermicomposting process of food waste via housefly larvae.

The present study aims to verify the feasibility of the cotreatment of food waste and dry anaerobic digestate via housefly larvae to explore a new pathway for recycling digestate. For this purpose, vermicomposting systems for food waste and digestate at different ratios were established. Given that the properties of food waste depend on its source, RFW and HFW were selected to investigate the effect of feedstock on the performance of vermicomposting. The properties of larvae-treated waste were evaluated, including waste reduction, typical physicochemical parameters (i.e., nutrients and heavy metals [HMs]), and maturity/biostability as well as larva-mediated valorization, productivity, and composition.

1. Materials and methods

1.1. *Putrescible waste and housefly larvae*

RFW and HFW that were in situ dewatered and crushed were collected from an anaerobic digestion facility in Nanjing, China. To provide palatable feedstock for housefly larvae, the digestate from the dry anaerobic digester receiving HFW with a solid retention time of 25 days was used for vermicomposting owing to its relatively loose structure (Appendix A Fig. S1), which has a weaker effect on larval activity compared to that of digestate from the wet anaerobic digester receiving RFW. Inert debris (i.e., metals and plastics) in waste was removed before the experiment. Physicochemical properties of different waste types are listed in the supporting materials (Appendix A Table S1). Initial wastes with a water content of approximately 80% were used for vermicomposting, following a previous study by Wang et al. (2015) of swine manure treatment via housefly larvae. This water content can reduce the cost of food waste dewatering, even though many studies adopt water content of approximately 70%.

Housefly larvae were selected for the bioconversion of food waste because they have a higher fecundity and shorter developmental duration compared with those of black soldier flies (Čičková et al., 2015). Larvae of housefly (*Musca domestica* L.) at 24 hr were obtained from the Organic Waste Recycling Center of Nanjing, China. The larval density was approximately 1000 individuals/g (wet weight).

1.2. *Experimental setup*

Vermicomposting of putrescible waste with housefly larvae was carried out in transparent plastic boxes with a size of 20 cm (length) × 12 (width) × 7 (height). Feedstock was prepared by mixing the dewatered dry digestate and RFW or HFW at five different ratios (dry weight), 1:0, 3:1, 1:1, 1:3, and 0:1 (Appendix A Table S2). The inoculation density was 1 g fresh larva/kg feedstock (dry weight), as described in a previous study by Shi et al. (2020). Each treatment was repeated in triplicate. The vermicomposting experiment was set up inside a mosquito net to prevent external flying insects from entering the reactors. The moisture content and temperature during the experiment were controlled at around 70% and 30°C, respectively, which are suitable for housefly larval growth. Vermicomposting was continued for 3 days. To increase contact between larvae and food waste, all reactors were turned over manually once a day. After vermicomposting, the prepupated housefly larvae were separated from the residues by using a metal mesh with a size of 2.5 mm under light conditions, and further manual separation was applied to ensure the full recovery of larvae. The obtained larvae were rinsed, weighed, and stored at -20°C until the determination of larval composition. The final solid residues were also weighed and stored at 4°C for physicochemical analyses.

1.3. *Sample measurement*

Food waste after larval composting was evaluated by various parameters classified into three groups: (1) waste reduction, including the waste reduction rate (WRR) and water content; (2) parameters related to organic fertilizer quality, including total and available nutrients (total nitrogen [TN], total phosphorus [TP], total potassium [TK], available nitrogen [AN], available phosphorus [AP], and available potassium [AK]), chloride, and HMs; (3) parameters related to maturity and biostability, including $\mathrm{SUVA}_{254\,(280)}$, dissolved organic carbon (DOC), germination index (GI), and respiration activity (RA_4). Briefly, the WRR was determined by weighing waste before and after vermicomposting and calculated following Eq. (1). The water content was measured by drying each sample in an electric thermostatic drying oven (GZX-9240MBE, BOXUN, China) at 105°C for 12 hr. TN, TP, and TK were determined following the Chinese agricultural standard NY/T525-2021. AN, AP, and AK were measured referring to the Chinese national standard GB/T 33891-2017. Chloride was analyzed by auto-potentiometric titration based on the Chinese national standard GB/T15063-2020. HMs (As, Cr, Cd, and Pb) were measured by inductively coupled plasma-mass spectrometry (Agilent 7700, Santa Clara, CA, USA) after sample digestion with HNO_3, HCl, HF, and $HClO_4$ at a volume ratio of 10:10:5:3. $\mathrm{SUVA}_{254\,(280)}$ was analyzed following the methods of Liu et al. (2019) and Eq. (2). Seed germination rates were evaluated according to the standard (NY/T 525-2021) and the GI was calculated following Eq. (3). RA_4 was determined by measuring oxygen consumption with a respirometer (BSBdigi-O_2, Selutec, Germany) referring to the Shanghai provincial standard DB31/T 1208-2020 and Eq. (4).

$$\mathrm{WRR} = \frac{W_i - W_f}{W_i} \times 100\% \tag{1}$$

where, W_i (g) represents the wet weight of the substrate before vermicomposting and W_f (g) is the wet weight of the substrate after vermicomposting.

$$\mathrm{SUVA}_{254(280)} = \frac{A_{254(280)}}{\mathrm{DOC}} \tag{2}$$

where, $\mathrm{SUVA}_{254(280)}$ is the relative molecular weight in DOC; $A_{254(280)}$ is UV absorbance at 254 nm (280 nm), and DOC (mg/L) is dissolved organic carbon.

$$\mathrm{GI} = \frac{\mathrm{GR_s} \times \mathrm{RL_s}}{\mathrm{GR_b} \times \mathrm{RL_b}} \times 100\% \tag{3}$$

where, $\mathrm{GR_s}$ (%) is the seed germination rate of the target sample; $\mathrm{GR_b}$ (%) is the seed germination rate of the blank, $\mathrm{RL_s}$ (mm) is the seed root length of the target sample, and $\mathrm{RL_b}$ (mm) is the seed root length of the blank.

$$\mathrm{RA}_4 = \frac{\sum_{i=1}^{96} m_{O_{2i}}}{40 \times \frac{300}{300+(300-m_{w1})-m_{w2}} \times (1 - C_w)} \tag{4}$$

where, RA_4 (mg-O_2/ g-(DM)) is respiration activity for 4 days; $\sum_{i=1}^{96} m_{O_{2i}}$ (mg) is accumulated oxygen consumption with 96 hr, m_{w1}, loss after sucti (on filtration, g, m_{w2} (g) is water loss during sample air drying and activation, and C_w (%) is water content of the measured sample.

Housefly larval productivity (LP) was calculated following Eq. (5), and crude protein, crude fatty acid, and amino acid contents in larvae were measured by referring to the methods of Jiang et al. (2017).

$$\mathrm{LP} = \frac{W}{M} \times 100\% \tag{5}$$

where, W (g) is the net weight of larvae after the bioconversion and M (g) is the initial weight of the waste in the reactor.

1.4. *Data analysis*

Differences in parameters among conditions were evaluated by one-way analysis of variance (ANOVA) using SPSS Statistics (Ver. 19.0, Chicago, IL, USA). Correlations between $\mathrm{SUVA}_{254(280)}$ and GI as well as between WRR and RA_4 or LP were also evaluated using SPSS. Significance was defined as $p < 0.05$.

2. Results and discussion

2.1. *Vermicomposting performance of food waste with digestate at various ratios*

As displayed in Fig. 1a, the highest WRRs for RFW and HFW after vermicomposting via the larvae were 65.93% and 62.80%, respectively, which were higher than the estimate (55%) obtained by Jiang et al. (2017) in a study of the recycling performance of housefly larvae for mixtures of food waste and rice chaff. The relatively high WRR in the current study may be explained by the pretreatment steps (i.e., dewatering and deoiling) because high levels of oil and salt in food waste have negative effects on larval growth (Man et al., 2021). Of note, the WRR decreased gradually as the proportion of digestate increased, regardless of the type of food waste. On the one hand, these results indicated that the larvae could digest large amounts of RFW and HFW in a short time (3 days), probably due to the strong ability of intestinal microorganisms in larvae to degrade organic matter (Xue et al., 2019; Liu et al., 2021) and significant water loss (Fig. 1b) caused by the rapid rise in pile temperature (Jiang et al., 2017). On the other hand, the

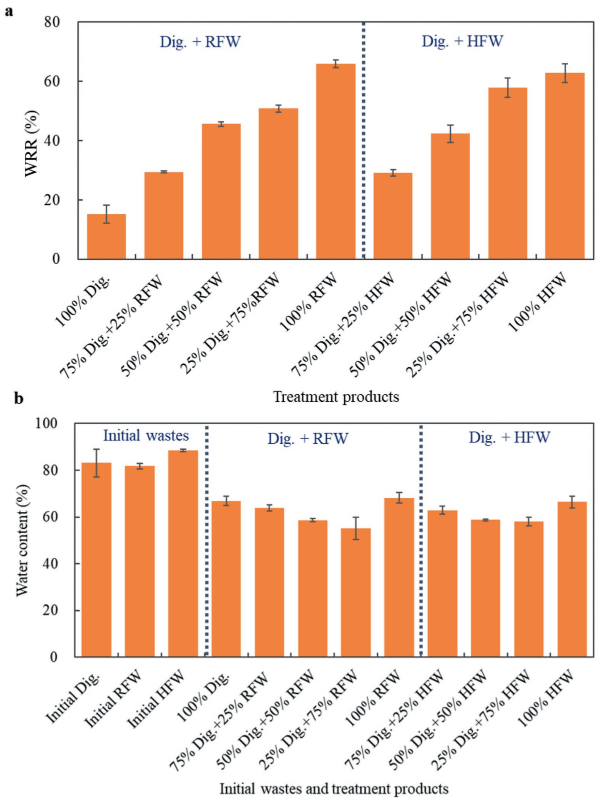

Fig. 1 – Waste reduction rate (WRR, a) and water content (b) for the initial wastes and the treatment products. WRR, waste reduction rate; Dig., digestate; RFW, restaurant food waste; HFW, household food waste.

digestate may serve as a bulking material for the vermicomposting of food waste, even though its palatability is slightly worse than that of the RFW and HFW. A recent study has confirmed that it is feasible to utilize earthworms to recycle digestate (Ning et al., 2021); however, the digestate needs to be composted to reduce the detrimental effects of foul-smelling gases (i.e., ammonia) on earthworms, suggesting that housefly larvae have a high tolerance to odor emitted by the digestate.

2.2. *Basic properties related to residue quality*

It is important to evaluate properties of final residues after the vermicomposting of food waste to ascertain whether additional measures, such as thermophilic composting, are needed. Based on the Chinese national standard for NY/T525-2021, several indicators, including nutrient, chloride, and HM contents, were analyzed.

2.2.1. *Nutrients*

Nutrient levels (TN, TP, and TK) in the final residues after waste bioconversion by housefly larvae are closely associated were their value as an organic fertilizer or soil amendment. As shown in Table 1, total nutrient contents of the food wastes increased after vermicomposting to varying degrees. Total NPK increased significantly from 4.5% (RFW) and 8.0% (HFW) to 6.5% and 12.3% ($p < 0.01$), respectively, indicating that the nutrient values meet the requirements ($\geq$ 4% and 1.5%) based on the standards NY/T 525-2021 and GBT33891-2017. Jiang et al. (2017) reported similar results, even though the treatment duration (6 days) using housefly larvae was longer than that (3 days) in the current study. It is worth noting that the P and K contents after vermicomposting did not change substantially; however, the N content was reduced, which may be ascribed to the loss of ammonia by volatilization (Zhu et al., 2020).

The changes of AN, AP, and AK were consistent with those of TN, TP, and TK. The inoculation of larvae increased the contents of AP and AK, especially for the RFW treatment, however, AN content decreased after bioconversion regardless of the type of food waste. similar results were observed in the studies by Jiang et al. (2017) and Zhu et al. (2020). AP and AK contents exhibited increasing trends as the proportion of digestate increased, while AN content had a decreasing trend for the HFW treatment. Therefore, adding digestate was beneficial to improve the level of available nutrients (AP and AK) in

Table 1 – Concentration of nutrients in the initial wastes and the treatment products.

		TN (%)	AN (%)	TP (%)	AP (%)	TK (%)	AK (%)
Initial wastes	Initial Dig.	1.60±0.19	0.37±0.03	3.51±0.28	0.13±0.01	2.70±0.16	0.076±0.011
	Initial RFW	4.43±0.35	0.56±0.04	2.07±0.17	0.27±0.02	0.89±0.07	0.124±0.020
	Initial HFW	3.41±0.28	0.33±0.03	3.66±0.24	0.19±0.02	2.28±0.19	0.081±0.008
Final residues	100% Dig.	1.21±0.05	0.30±0.02	0.86±0.07	0.15±0.01	0.55±0.05	0.063±0.005
	75% Dig.+25% RFW	2.78±0.09	0.35±0.02	4.99±0.35	0.14±0.10	2.97±0.24	0.075±0.005
	50% Dig.+50% RFW	2.23±0.11	0.49±0.04	3.61±0.23	0.18±0.02	2.54±0.22	0.118±0.016
	25% Dig.+75% RFW	1.76±0.12	0.45±0.04	4.66±0.38	0.23±0.01	2.23±0.18	0.140±0.012
	100% RFW	4.02±0.34	0.30±0.02	3.40±0.28	0.31±0.02	1.12±0.12	0.164±0.011
	75% Dig.+25% HFW	1.54±0.13	0.32±0.03	4.53±0.36	0.16±0.01	3.30±0.31	0.085±0.007
	50% Dig.+50% HFW	1.94±0.15	0.28±0.03	5.85±0.44	0.17±0.01	3.19±0.30	0.096±0.005
	25% Dig.+75% HFW	3.08±0.29	0.25±0.02	6.05±0.45	0.22±0.02	3.30±0.29	0.102±0.008
	100% HFW	2.48±0.17	0.14±0.1	6.43±0.51	0.26±0.02	3.70±0.33	0.121±0.009

Note: Value is shown as mean±standard deviation, $n = 3$. TN, total nitrogen; AN, available nitrogen; TP, total phosphorous; AP, available phosphorous; TK, total potassium; AK, available potassium. N.D., not detected. Dig., digestate; RFW, restaurant food waste; HFW, household food waste.

Table 2 – Contents of HMs detected in the initial wastes and final residues.

		As	Cd	Cr	Pb
Initial wastes	Initial Dig.	N.D.	N.D.	29.5±0.7	N.D.
	Initial RFW	N.D.	N.D.	14.2±6.0	N.D.
	Initial HFW	11.2±2.5	N.D.	16.4±3.0	N.D.
Final residues	100% Dig.	N.D.	N.D.	26.7±2.8	N.D.
	75% Dig.+25% RFW	N.D.	N.D.	42.8±0.7	N.D.
	50% Dig.+50% RFW	N.D.	N.D.	51.4±13.0	N.D.
	25% Dig.+75% RFW	N.D.	N.D.	40.4±10.7	N.D.
	100% RFW	N.D.	N.D.	38.4±20.3	N.D.
	75% Dig.+25% HFW	4.5±1.0	N.D.	52.4±21.3	N.D.
	50% Dig.+50% HFW	9.4±0.7	N.D.	42.8±12.4	N.D.
	25% Dig.+75% HFW	9.0±1.6	N.D.	26.4±5.7	N.D.
	100% HFW	13.3±2.2	N.D.	33.9±4.1	N.D.
Standards	GB/T 33891-2017[a]	≤10, 20, 35	≤1.5, 3.0, 5.0	≤70, 200, 300	≤120, 300, 400
	NY/T 525-2021[b]	≤15	≤3	≤150	≤50

Note: Value is shown as mean±standard deviation, n=3; unit, mg/kg; N.D., not detected. Dig., digestate; RFW, restaurant food waste; HFW, household food waste. Letters ‘a’ and ‘b’ are the Chinese national and industry standards, respectively and the value in the same is the corresponding threshold for each HM.

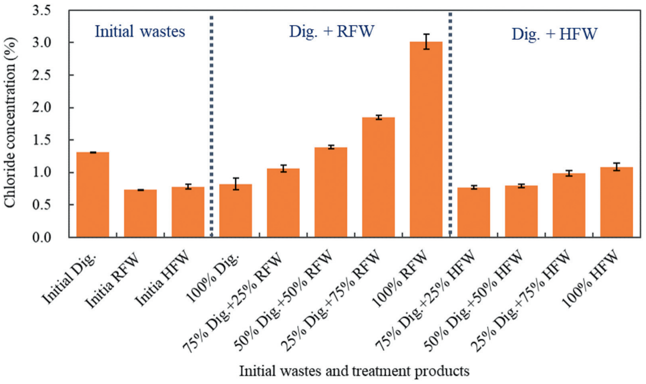

Fig. 2 – Content of chloride in the initial wastes and different treatment products. Dig., digestate; RFW, restaurant food waste; HFW, household food waste.

the larva-mediated food waste especially for the RFW, which is possibly related to the enhanced larval biomass owning to the addition of digestate.

2.2.2. *Chloride*

Fig. 2b shows that vermicomposting increased chloride contents to varying degrees with the increment of digestate ratios. However, the highest content (3.0%) of chloride detected in the residue from the RFW vermicomposting system did not exceed the threshold established by the agricultural standard NY/T 525-2021, indicating that excessive chloride was not an issue, regardless of the addition of digestate. The high content of chloride in the RFW treatment system without the addition of digestate may be explained by the high degree of degradation of organic matter (Appendix A Fig. S2). The chloride contents in both initial RFW and HFW were approximately 1.7%, and dehydration in the pretreatment stage could further reduce the chloride content due to its water solubility. Ding (2017) reported that chloride in commercial organic fertilizers should be controlled below 1.0%, since the risk of crop damage would increase when the chloride content exceeds this threshold. In other words, the chloride content in organic fertilizer needs to be comprehensively evaluated in combination with plant tolerance and soil characteristics.

2.2.3. *HMs*

As presented in Table 2, Cd and Pb were not detected in all samples and As was detected only detected in the initial HFW, with a content of 11.2 mg/kg (dry weight), similar to that in residue after vermicomposting. Further, the addition of digestate reduced the content (4.5–9.4 mg/kg) of As in food waste, probably due to the “dilution effect” of the digestate. Cr was detected in the initial digestate and food waste, with a relatively constant content of 14–29 mg/kg (dry weight); however, its content after vermicomposting increased up to 52.4 mg/kg (dry weight). Overall, the content of each HM in all residues did not exceed the limit based on the agricultural standard (NY/T525-2021) and national standard (GBT 33891-2017), indicating that HM pollution in the vermicomposting of food waste and digestate is negligible. The HM content was mainly determined by the source of food waste, consistent with the results of previous studies (Jiang et al., 2017; Lu and Xu, 2021).

2.3. *Maturity and bio-stability of bio-converted residues*

2.3.1. *SUVA$_{254\ (280)}$*

SUVA$_{254(280)}$ is closely related to aromaticity and relative molecular mass (Wang et al., 2013; Zhao et al., 2015). Fig. 3a shows that the two indicators exhibited synchronous changes after vermicomposting. SUVA$_{254(280)}$ values for the initial RFW and HFW were lower in than those for the digestate, implying that the digestate contained a certain amount of humus substances, which can be partially attributed to the degradation of organic matter during the anaerobic digestion of food waste (Peng et al., 2020). Both indicators increased after the vermicomposting of food waste and increased with an increase in the proportion of digestate. Obviously, the presence of housefly larvae could accelerate the degradation of RFW and HFW, and the addition of digestate could enhance their

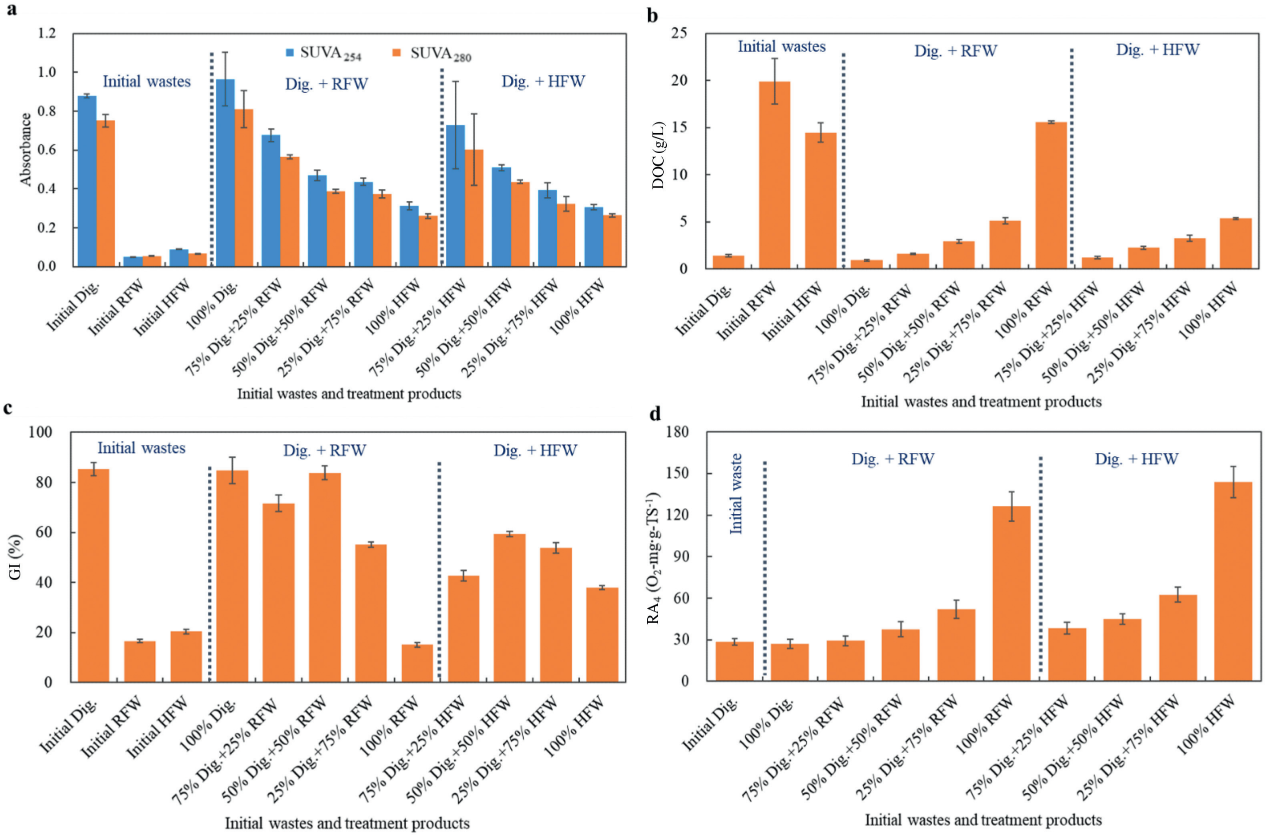

Fig. 3 – Biostability and maturity of initial wastes and different treatment products. (a), SUVA$_{254}$ and SUVA$_{280}$; (b), dissolved organic carbon (DOC); (c), respiration activity (RA$_4$); (d) germination index (GI). Dig., digestate; RFW, restaurant food waste; HFW, household food waste.

Table 3 – Different standards for evaluation of biostability and maturity of composted biowaste.

	GB/T 33891-2017 [a]	NY/T 525-2021 [b]	DB31/T 1208-2020 [c]	References
SUVA$_{254(280)}$	/	/	/	0.247(0.180) (Lou et al., 2020)
DOC (g/kg)	/	/	/	4 g/kg (Zmora-Nahum et al., 2005)
GI (%)	≥95, ≥80, ≥65	70≥	/	/
RA$_4$ (mg-O$_2$/g-DM)	/	/	≤30	10 (EC, 2001)

Note: Letters ‘a’, ‘b’, and ‘c’ are the Chinese national, industry, and local standards, respectively and the value in the same line is the corresponding threshold for each parameter.

maturation. Lou et al. (2020) claimed that manure-based compost with SUVA$_{245}$ and SUVA$_{280}$ values of 0.247 and 0.180, respectively, is completely mature (Table 3). This conclusion is inconsistent with the results of the current study, which may be explained by differences in the characteristics of treated putrescible waste (Lou et al., 2020). In the present study, these two indicators cannot be used to determine whether larvae-treated food waste have reached good stability; however, they can be used to characterize relative maturity.

2.3.2. DOC

DOC is one of the most sensitive indicators of microorganisms in the soil environment and is frequently applied to assess compost biostability (Cui et al., 2018). Zmora-Nahum et al. (2005) found that 4 g/kg of DOC could be a threshold for a mature compost. As summarized in Fig. 3b, changes in DOC were contrary to those of SUVA. The initial RFW and HFW exhibited higher DOC contents than those of the initial digestate, suggesting that the latter was characterized by a relatively high stability. After vermicomposting, significant decreases in DOC were observed for the two types of food waste. As the proportion of digestate increased, especially for 25%, DOC decreased significantly ($p < 0.01$), indicating that the biostability of the residue increased remarkably. However, this discrepancy was not obvious for the HFW vermicomposting system. It is possible that the addition of digestate could improve the palatability of larval feedstock, as supported by the results of the LP analysis (described below).

2.3.3. GI

GI is a widely used indicator for the maturity of compost (Luo et al., 2018). Generally, a higher GI after the biotreatment of putrescible waste indicates that the residues show good maturity. As shown in Fig. 3c, the residues after vermicomposting (except in the sole RFW system) showed substantial increments in GI ($p < 0.05$). In particular, the GI values for the residues from the co-treatment systems including RFW and digestate at ratios of 50% and 75% were 70% and 82%, respectively, indicating that the addition of digestate could enhance vermicompost maturity. The residues from the HFW treatment systems displayed increases in GI ($< 60\%$), and these values were below the threshold ($\geq 70\%$) based on the agricultural standard (NY/T525-2021, Table 3). These findings suggest that the larvae-mediated HFW still contains toxic substances for plant growth. Luo et al. (2018) and Chen et al. (2021) have reported that partially degraded compost is commonly rich in plant polyphenols and organic acid and is characterized by a low pH and high electrical conductivity, thus reducing rates of seed germination. In particular, plant polyphenols derived from the plant peel, root, fruit, and leaf may be abundant in the HFW, explaining the lower *GI* compared to that for the RFW.

2.3.4. RA$_4$

RA$_4$ is widely used to evaluate the biostability of biowaste and its treated residue. Generally, RA$_4$ values below 20 mg-O$_2$/g-TS indicate good biostability (Arias et al., 2012; Table 3). As shown in Fig. 3d, the residues from RFW vermicomposting had a significantly lower RA$_4$ concentration (96.2 mg-O$_2$/g-TS) than that of residues from HFW vermicomposting (150 mg-O$_2$/g-TS). Both types of vermicomposting resulted in similar decreasing trends in RA$_4$ as the amount of digestate increased. RA$_4$ of the residue from the vermicomposting system with 25% digestate was highly similar to that of the initial digestate (26.9 mg-O$_2$/g-TS). The inoculation of housefly larvae promoted the partial degradation of organic matter in the food waste (Appendix A Fig. S2). However, the final residues, especially those from HFW, were still biologically unstable with relatively high RA$_4$ values.

Further, the addition of digestate improved the biological stability of the product. For instance, RA$_4$ values for the RFW and HFW vermicomposting systems with 50% digestate were reduced to less than half of the RA$_4$ values of the initial food waste. These results suggest that in addition to the effect of intestinal microorganisms in housefly larvae, the addition of digestate with a low RA$_4$ (30 mg-O$_2$/g-TS) could further enhance residue stability. Overall, regardless of whether digestate is added or not, the food waste after larval composting requires further treatment (i.e., additional composting) before use as organic fertilizer in agricultural applications or as organic media for greening. Similarly, a previous study has reported that residues obtained by the treatment of food waste with black soldier fly larvae did not satisfy relevant agricultural standard (NY/T 525-2021), requiring further composting for at least 5 weeks (Song et al., 2021). Normally, the time to the bioconversion of organic waste is longer for black soldier fly larvae (14–21 days) than for housefly larvae (3–4 days) (Čičková et al., 2015; Palma et al., 2019). The two types of scavenging insects mainly digest and degrade easily degradable components of putrescible waste, including polysaccharides, fat, and protein. As expected, trends in RA$_4$ for the vermicomposting systems are roughly consistent with trends in GI, thus verifying that the degradation of food waste via housefly larvae is controlled by diverse microorganisms from the larval intestinal tract (Liu et al., 2021) and the properties of the substrate.

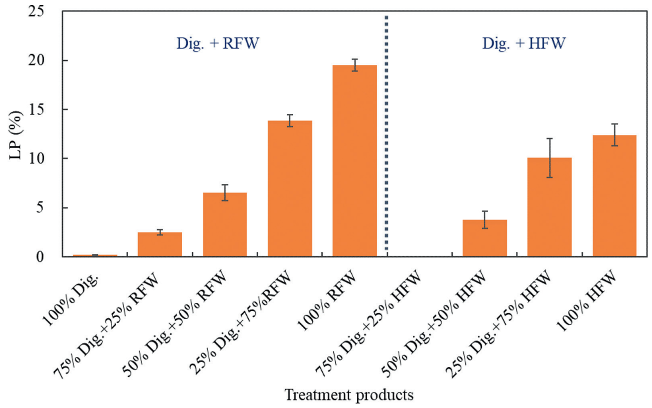

Fig. 4 – Housefly larvae productivity (LP) for different treatments. Dig., digestate; RFW, restaurant food waste; HFW, household food waste.

2.4. *Valorization of housefly larvae*

2.4.1. LP

Vermicomposting of food waste via housefly larvae is regarded as a complex process involving both waste degradation and larval growth. Understanding the growth state of housefly larvae is essential for evaluating the performance of food waste vermicomposting. Additionally, the composition of harvested larvae (i.e., protein, fat, and amino acids) is closely associated with the extent of valorization. As shown in Fig. 4, the RFW vermicomposting system without the addition of digestate had the highest LP of approximately 20%, which is in line with the maximum WRR (Fig. 1). Shi et al. (2020) have observed that larval composting for the treatment of food waste from a three-phase separation facility could lead to a LP of 18.9%, which is similar to the estimate in the current study. It should be noted that the LP for both vermicomposting systems declined significantly as the amount of digestate increased ($p < 0.05$). LP values for the vermicomposting of RFW were significantly higher than those of the HFW system for all digestate ratios ($p < 0.05$). These findings show that the properties of feedstock had an important impact on larval biomass. The digestate alone was not suitable as feedstock for housefly larvae, which is likely related to the low amount of DOC (Fig. 3b) available to their microorganisms. Accordingly, housefly larvae preferred RFW over HFW, since the former usually contained high amounts of protein and fat (Carmona-Cabello et al., 2020). Given the high cost of vermicomposting in terms of maintaining the conditions for larval growth, HFW should be mixed with RFW to feed housefly larvae. It is worth noting that the LP was as high as 15% when the proportion of digestate for the RFW treatment was 25%, and this was remarkably higher than that for the vermicomposting of HFW alone (10%). Based on comprehensive analyses of WRR and LP, 25% digestate added to food waste during vermicomposting would result in a higher larval biomass, providing a feasible option for resource utilization of digestate.

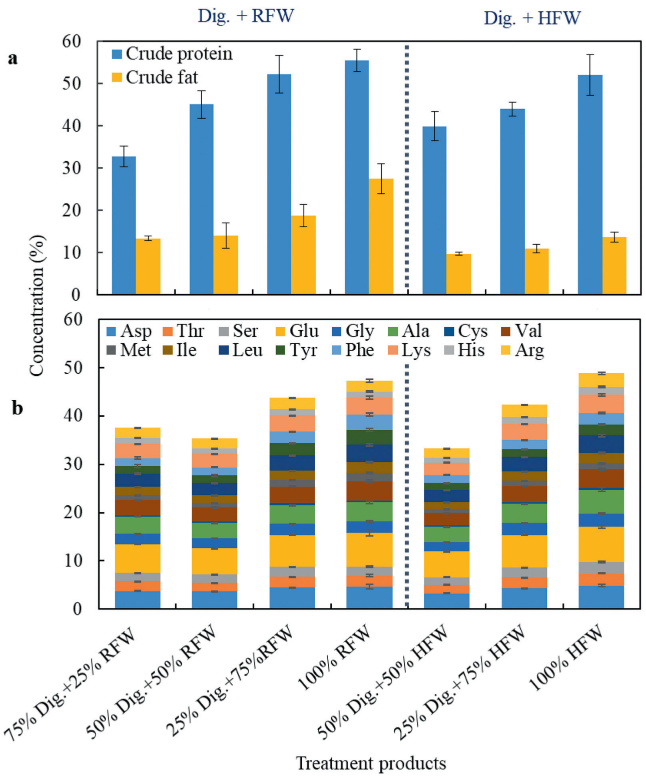

Fig. 5 – Contents of protein (a), fat (a) and amino acid (b) for different treatments products. Asp, aspartic acid; Thr, threonine; Ser, serine; Glu, glutamic acid; Gly, glycine; Ala, alanine, Cys, cystine; Val, valine, Met, methionine; Ile, isoleucine; Leu, leucine; Tyr, tyrosine; Phe, phenylalanine; Lys, lysine; His, histidine; Arg, arginine. Dig., digestate; RFW, restaurant food waste; HFW, household food waste.

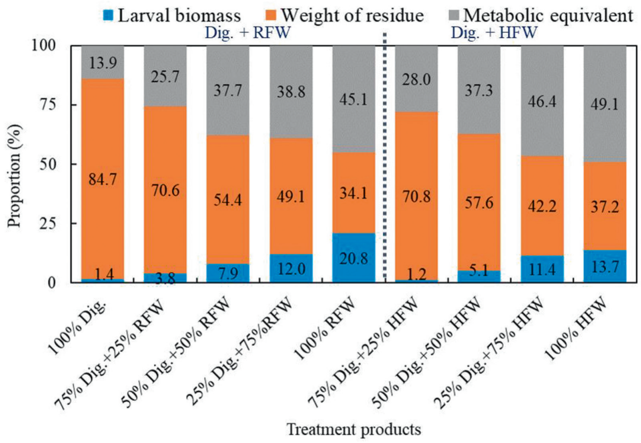

Fig. 6 – Materials balance between treatment substrate and housefly larvae under different vermicomposting conditions. Dig., digestate; RFW, restaurant food waste; HFW, household food waste.

2.4.2. *Crude protein, crude fat, and amino acids*

In addition to LP, the larval composition is an important parameter for assessing the performance of vermicomposting of food waste. Fig. 5a shows that the highest content (46%) of crude protein was observed in the larvae from the RFW, which was apparently higher than the content (35%) for the corresponding HFW treatment. The crude protein content decreased significantly as the amount of digestate increased, regardless of the type of food waste, consistent with the results for LP. The highest content (26%) of crude fat was also recorded in the larvae-aided RFW treatment and the content of crude fat was affected by the type of food waste and amount of digestate. Crude protein contents of 52.3% and 55.5% for the larvae harvested from the RFW treatments with 25% and 0% digestate were observed, respectively, while the protein content (52.0%) of the HFW treatment without the digestate was higher than the reference value (> 50%) of the national standard GB/T19164-2003, which could result in a relatively high quality of housefly larvae with abundant protein. The observed protein contents are consistent with those obtained from the vermicomposting of pig manure (Zhu et al., 2015) and food wastes (Gao et al., 2019) but exceed that reported by Jiang et al. (2017). In particular, the HFW treatment with digestate ratios of 75% and 100% had no detectable larvae, further confirming that the digestate is not palatable as feedstock for housefly larvae, as discussed above for the LP.

In total, 16 nonessential/essential amino acids were detected across all treatments (Fig. 5b). The highest amino acid contents (47.2%–48.8%) were observed in the vermicomposting systems without added digestate, followed by the treatment systems with 25% digestate (42.3%–43.8%) and 50% digestate (33.3%–35.3%). These results suggest that the addition of digestate had an obvious effect on the composition of amino acids in larvae, which may be related to the C-to-N ratio of substrates (Palma et al., 2019). Adjusting the C-to-N ratio could alter the life cycle, thus impacting the larval composition. In the present study, several amino acids, including aspartic acid, glutamic acid, valine, leucine, and lysine, declined as the amount of digestate increased, regardless of the type of food waste, implying that the addition of digestate in the vermicomposting of food waste reduces the quality of larvae. Higher contents of amino acids such as methionine, tyrosine, and phenylalanine were observed in the bioconversion of RFW without digestate than in the corresponding HFW treatment, whilst higher contents of serine, glycine, and alanine arginine were observed in the bioconversion of HFW without digestate; these results are partially in line with those of a previous study by Palma et al. (2019). However, studies of the mechanisms by which substrate properties, such as the C-to-N ratio, influence amino acid contents in larvae are needed.

2.5. *Comprehensive analysis*

2.5.1. *Materials balance*

Materials balance between substrate and housefly larvae can clearly elucidate efficiency of vermicomposting. As presented in Fig. 6, the addition of housefly larvae significantly promoted the transformation of RFW to larval body of 50.4 g along with the metabolic equivalent of 109.2 g. Comparatively, slightly lower bioconversion efficiency with the larval biomass of 33.2 g and metabolic equivalent of 118.8 g were ob-

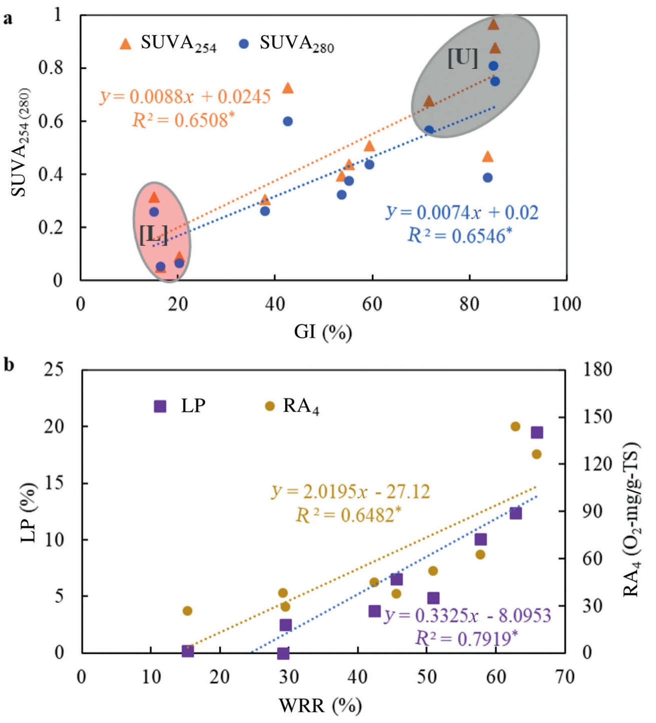

Fig. 7 – Correlation analysis between $SUVA_{254}$ ($SUVA_{280}$) and germination index (GI), as well as between waste reduction rate (WRR) and larvae productivity (LP), RA_4. The regions [L] and [U] represent samples with low and high maturity, respectively. RA_4, respiration activity for 4 days. Asterisk (*) represents significant difference between two parameters at level of $p < 0.05$.

served in the HFW vermicomposting system. Further, for the two different vermicomposting systems, larval biomass and metabolic equivalent had decreasing trends as the amount of digestate increased. These findings indicate that high digestate addition ratios such as 50% and 75% negatively affected the degradation of feed and the growth of larvae regardless of the type of food waste. In other words, there were large amounts of substrates in the vermicomposting system that was not able to be fully digested via housefly larvae, thus leading to lower WRR (Fig. 1a). Given a higher larvae biomass (Fig. 4, Fig. 5) and relatively mature residue (Fig. 3b, 3c, 3d) from composting of RFW added with 25% digestate, co-treatment of RFW and digestate with a low ratio via housefly larvae is recommended.

2.5.2. *Correlation analysis*

To gain an in-depth understanding of effects of digestate on the quality of residues after the vermicomposting of food waste via housefly larvae, multiple correlation analyses between $SUVA_{254(280)}$ and GI, WRR, and LP as well as RA_4 were performed. As shown in Fig. 7a, there was a significant positive correlation between $SUVA_{254(280)}$ and GI ($p < 0.05$). The RFW, HFW, and housefly larvae-treated residues contained harmful substances (corresponding to the lower area [L] in the plot). The upper area [U] consisted of the initial digestate and the residues from the RFW treatment with 75% and 50% digestate, which clearly suggests that these samples were rich in humus. The area between the upper and the lower regions represented the residues of the RFW treatment without digestate addition and the HFW treatment with digestate. Although the GI of residues from the vermicomposting of HFW increased due to the presence of digestate, the maturity was still lower than the threshold (70%) based on the agricultural standard NY/T 525-2021. This could be explained the fact that the HFW with a high lignin content had poor larval palatability compared with that of the RFW (Fig. 4) because the former usually contains abundant plant polyphenols (Aiello et al., 2020).

As shown in Fig. 7b, the WRR showed positive correlations with both LP and RA_4 ($p < 0.05$). This suggests that the extent of waste reduction mainly depended on the efficiency of digestion by larvae. In other words, the larger the weight gain of larvae, the higher the waste reduction. The RFW with high DOC (Fig. 3b) was a good feedstock for larvae, leading to a significant increase in LP. Similarly, a higher growth rate of black soldier fly larvae was observed after the thermal pretreatment of waste activated sludge with higher soluble chemical oxygen demand (Liew et al., 2022). Furthermore, the addition of digestate to the RFW at 25% resulted in higher LP, WRR, and maturity values compared to those for the HFW without digestate. Therefore, it is a feasible option to add digestate at an appropriate rate to RFW for vermicomposting; this strategy not only guarantees a substantial larval biomass but also contributes to relatively stable residues as a good source of organic fertilizer. The benefits of vermicomposting of RFW mixed with digestate may be explained by the importance of ‘balance meat and vegetable’. Rehman et al. (2017) revealed that vermicomposting of dairy manure with abundant cellulose and chicken manure via black soldier flies caused a remarkable waste reduction. Palma et al. (2019) also found that a decrease in the C-to-N ratio of feedstock could significantly increase the productivity of black soldier fly larvae (*Hermetia illucens* L.). A study of the evolution of microorganisms in the vermicomposting system of RFW and digestate revealed that microorganisms, especially bacteria, in the intestinal tract of larvae play an important role in food waste biodegradation (Gold et al., 2020; Liu et al., 2021).

3. Conclusion

Resource utilization of food waste digestate is a major concern in China. The cotreatment of food waste and added digestate by housefly larvae was investigated in the present study. Although the addition of digestate decreased the productivity and nutrient levels in housefly larvae, the RFW mixed with digestate at 25% was recommended from the viewpoints of waste reduction and larval valorization. Materials balance shows that HFW treatment system exhibited lower bioconversion efficiency than that of RFW treatment system regardless of the addition of digestate. These results reveal that the combination of food waste and digestate for treatment by housefly larvae not only provides a new option for digestate resource utilization but improves the maturity of food waste. In-depth investigations of the mechanism underlying “balance meat and vegetable" during vermicomposting of food waste and digestate via housefly larvae are required.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A Supplementary data

Supplementary material associated with this article can be found in the online version, at doi:10.1016/j.jes.2022.07.016.

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