

Use of Aquaculture Pond Sludge and Fish Waste to Produce Manure Worm, *Eisenia fetida* (Lumbricidae), Vermiwash

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ABSTRACT

Aquaculture pond sludge and fish waste are key environmental pollutants in the aquaculture and fish processing sectors, and they must be managed to reduce their negative environmental impacts. Aquaculture pond sludge and fish waste were used as bedding materials in this study to make *Eisenia fetida* vermiwash. The vermiwash was prepared using three substrate combinations: cow dung and *Gliricidia* leaves (control), cow dung and aquaculture sludge (ASV), and cow dung and fish waste (FWV) at a 2: 1 ratio. *E. fetida* (35–40 worms·kg⁻¹ of bedding material) were introduced to each vermiwash preparation unit, and vermiwash samples were collected weekly for four weeks after one month of worm introduction. Chemical oxygen demand (COD), pH, conductivity, potassium (K), total nitrogen (TN), and orthophosphate of vermiwash samples were measured. ASV contained significantly higher total nitrogen and less potassium than FWV and the control ($p<0.05$). While orthophosphate, pH, and COD among experimental vermiwash were not significantly different ($p>0.05$), the conductivity was significantly higher in FWV. Except for the low K and high TN levels, ASV was chemically comparable with the control vermiwash. *Phaseolus vulgaris* seeds exposed to ASV had a comparatively higher germination (63%) than those exposed to control (47%) and FWV (37%). The earthworm survival decreased to 9% in the FWV unit and increased by 49% and 33% in the control and ASV units, respectively. The findings demonstrated the feasibility of producing vermiwash from aquaculture sludge for integrated aquaculture and agriculture.

Keywords: Earthworm, Fish, Organic, Pond, Sludge, Waste

INTRODUCTION

The sludge generated by the aquaculture includes semi-solid and solid forms (Latt, 2002; Wudtisin *et al.*, 2015; Klein *et al.*, 2023) and contains uneaten feed particles, phytoplankton, detritus, animal wastes, mineral sediment, airborne particles, protozoa, bacteria, fungi and residues of fertilizer and therapeutic inputs (Rahman *et al.*, 2004; Mirzoyan *et al.*, 2010; Tan *et al.*, 2023). Accumulating excessive sludge in ponds leads to a reduction of space available for the cultured organism and may become a reason for the depletion of dissolved oxygen in pond water and the generation

of H₂S and NO₂ toxic gases (Hargreaves, 1998; Drózdz *et al.*, 2020; De Ungria *et al.*, 2023). Due to its deleterious effects, sludge must frequently be removed from the pond bottom and appropriately treated before being discharged into the environment (Wudtisin *et al.*, 2015; Belmeskine *et al.*, 2023).

Fish processing in the aquaculture industry, where unconsumable body parts are discarded, also generates solid waste, including fins, scales, gut, internal organs, gills, and other nonedible parts of the fish body (Laos *et al.*, 2002; Tan *et al.*, 2023). Fish waste in many countries (De Ungria *et al.*, 2023), including Sri Lanka, is dumped in landing

sites, landfills and local fish markets. These disposal activities cause to attract insects and flies, toxic gases emission, the dispersal of lousy odour in dumping sites and soil and groundwater contamination due to decaying organic matter leakage. Improper dumping of fish waste contributes to methane gas and CO₂ emission, major contributors to global warming (Saravanan *et al.*, 2023).

Vermitechnology is an application that includes vermiwash and vermicompost production, which benefit earthworms' capacity to decompose organic materials (Aira *et al.*, 2007; Jaybhaye and Bhalerao, 2015; Jasmin *et al.*, 2020). Production processes of vermiwash and vermicompost are more similar, but the final output varies. Vermicompost is a fine peat-like or humus like material produced using the active involvement of the earthworms (Zarei *et al.*, 2018; Belmeskine *et al.*, 2023; Klein *et al.*, 2023). Vermiwash is the liquid gathered by running water through the soil or organic substrate with active worms. The liquid contains water from earthworm bodies and is rich in nutrients such as nitrogen, potassium, magnesium, zinc, calcium, iron, and copper (Jaybhaye and Bhalerao, 2015; Sharif *et al.*, 2016; Zarei *et al.*, 2018). Vermiwash is a cost-effective and environmentally friendly approach to break down organic waste for sustainable agriculture (Aira *et al.*, 2007).

Earthworms grind organic residues into considerably finer particles by passing them via a grinding gizzard, a highly muscular thick-walled organ present in the eighth and seventh segments (Scheu *et al.*, 2002). While doing so, they enhance carbon and nitrogen mineralization through direct or indirect impact on the microorganism composition of the substrate (Aira *et al.*, 2007; Sharif *et al.*, 2016; Klein *et al.*, 2023). A wide variety of microorganisms, together with bacteria, algae, fungi, protozoa, actinomycetes and even nematodes, are usually found throughout the length of the earthworm gut. Suksomphap *et al.* (2022) indicated the positive effect of the coelomic fluid of earthworm, *Perionyx excavatus* on the seedling growth of *Aspergillus flavus*. It is generally known that the epigaeic earthworm species, *Eisenia fetida*,

E. eugeniae and *P. excavatus* have the potential as waste decomposers in soil (Kale *et al.*, 1982). *E. fetida*, commonly known as red wiggler or manure worm, can decompose several types of substrates, including solid industrial waste, agricultural residues, livestock excreta, woodchip, sewage sludge, sugar mill sludge, fly ash, solid waste of leather industry and bio-solids of the textile industry (Sharma *et al.*, 2009; Jaybhaye and Bhalerao, 2015; Suksomphap *et al.*, 2022; Belmeskine *et al.*, 2023). Due to the high carbon and nitrogen content (Rahman *et al.*, 2004), aquaculture pond sediment and fish discards may also be used to produce organic fertilizers like compost (Muendo *et al.*, 2014; Zhang and Sun, 2017; Klein *et al.*, 2023) and vermiwash. Though aquaculture pond sludge (Rahman *et al.*, 2004; Zhang and Sun, 2017; Belmeskine *et al.*, 2023; Tan *et al.*, 2023) and fish waste (Laos *et al.*, 2002; De Ungria *et al.*, 2023) have been used in compost production, their suitability in the production of vermiwash has yet to be investigated. The present study aimed to produce vermiwash using *E. fetida* grown in aquaculture pond bottom sludge and fish waste and assess the impacts of experimental vermiwash on the germination of common bean, *Phaseolus vulgaris*.

MATERIALS AND METHODS

Multiplication of Eisenia fetida

Approximately 300 *Eisenia fetida* adult worms were burrowed from a stock culture maintained in the vermicomposting unit of the Field Crop Research and Development Institute, Mahailuppallama, Sri Lanka. Earthworms were allowed to multiply in a 3:1 ratio of cow dung and *Gliricidia* leaves medium in a 50 L plastic container (Nayak *et al.*, 2019). The container was moisturized daily using 500 mL of well water. After two months, half of the initial decaying organic matter with earthworms was transferred into another container to multiply the earthworms. The study was conducted from June to November 2021 at the animal house of the Department of Zoology and Environmental Management, University of Kelaniya, Sri Lanka.

Preparation of vermiwash

Aquaculture pond sludge was collected from a semi-intensively managed tilapia culture pond (900 m^2) at the National Inland Fisheries and Aquaculture Training Institute, Kalawewa, Sri Lanka. During the culture period of six months, fish were fed commercially prepared pelleted feed with 32% protein twice daily and the fish stocking density was 2 m^2 . The ponds were not fertilized and the water was aerated using two paddlewheel aerators at night. After the culture period, the ponds were drained harvest and pond bottom sludge was collected up to 5–10 cm from the four corners, 3 m away from the dyke and the middle. The sludge collected from the pond bottom surface was mixed to make a composite mixture and was air dried at room temperature for five days before being sieved at a mesh size of 2 mm and used in the experiment. Fish waste, including gills, guts, fins, skin and small parts of flesh, was collected

from a local freshwater fish market. The collected fish waste was chopped (2–4 cm) and was then placed in a sieve for 15 min to drain any liquid contained in the fish waste. Then they were steamed using a kitchen steamer before being used in the experiment.

Vermiwash units were prepared according to Nayak *et al.* (2019). In the bottom of the tap-fitted plastic container (25 L), a bricks layer (5 cm) was spread, and a dry sand layer (5 cm) was applied. An insect proof net was kept on the sand layer to avoid the movement of earthworms toward the sand and bricks layer (Figure 1). A mixture of bedding material prepared according to Table 1 was layered on the insect-proof net. The cow dung used in preparation of the bedding material was collected fresh and sun dried two weeks. Then bedding materials of the control and each treatment were mixed thoroughly before they were placed in the experimental worm preparation units.

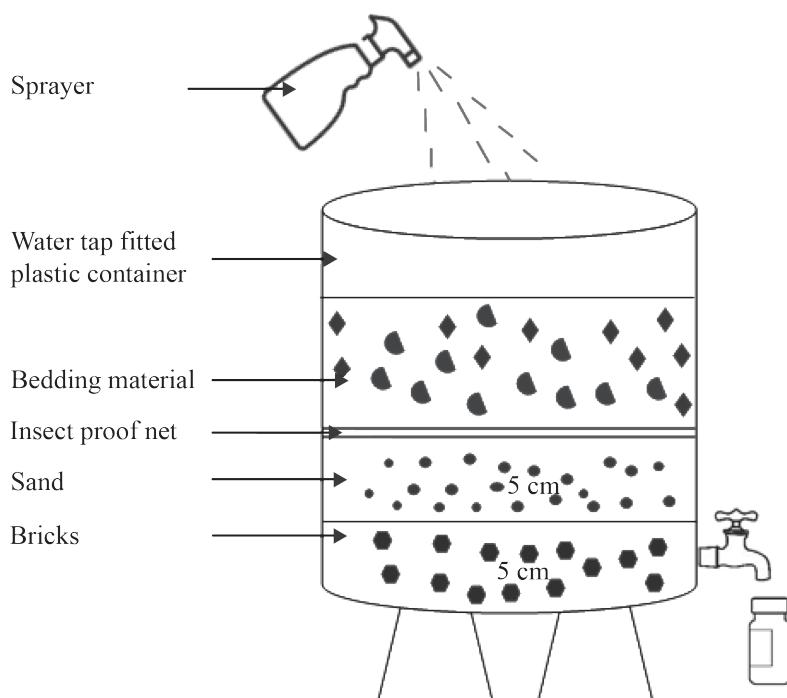


Figure 1. Vermiwash preparation unit.

Table 1. Bedding materials used in the experimental vermiwash preparation units.

Experiment	Bedding materials (2:1)
Treatment 1-Pond sludge	Aquaculture pond sludge and cow dung
Treatment 2-Fish waste	Fish waste and cow dung
Control	<i>Gliricidia</i> leaves and cow dung

Then 250–280 *E. fetida* with 4–9 cm body length were introduced with 1 kg of the initial decomposed mixture onto the experimental bedding material (6 kg). Water (250 mL) was sprayed on the surface of each vermiwash unit once a day using a handheld sprayer for three weeks to maintain moisture within containers. The bottom tap was kept open throughout this period and removed excess elute via the plastic tap following the industrial practice. Then the taps were closed for a week and 500 mL water was sprayed daily on all experimental units. The control and treatment vermiwash preparation units were triplicated and randomly arranged. Then vermiwash was collected from each vermiwash preparation unit to conduct chemical analysis. The procedure was repeated weekly to collect another three samples from treatment and control vermiwash preparation units.

Analysis of chemical parameters of aquaculture sludge and vermiwash

The sludge and vermiwash samples were filtered using Whatman 41 filter papers before the chemical analysis. The pH and conductivity of samples were measured in situ using a multiparameter water quality checker (HACH-HQ 40d) and COD was measured using the open reflux method (Burns and Marshall, 1965). Total nitrogen (TN) and orthophosphate (ortho-P) contents were determined by the Kjeldahl method (FAO, 2021) and the Ascorbic acid method (APHA, 2012), respectively. The total potassium was determined by flame photometer using a 50 ppm K standard solution (Jackson, 1967).

Germination test for Phaseolus vulgaris

Aquaculture pond sludge, fish waste, distilled water and the control vermiwash were

used to test the germination of *P. vulgaris* seeds. The seeds were soaked in deionised water overnight before being used in the germination test. Twelve petri dishes were layered from Whatman number 1 filter paper and 20 seeds of *P. vulgaris* (Black Cora) were placed on them (Chattopadhyay, 2015). The experiment was conducted in a completely randomised design with three replications under environmental conditions of the laboratory and test solutions and distilled water was sprayed on seeds twice daily using a handheld spray gun. An injection cylinder was used to remove the residual solution from the previous day. The experiment was conducted continuously for three days and the number of germinated seeds was counted.

Determination of population variation of Eisenia fetida in different bedding materials

After 90 days of vermiwash preparation, the number of earthworms longer than 4 cm in decomposed bedding materials in control and two treatments were counted. The number of earthworms recorded in each unit was compared with the initial number of earthworms stocked in the culture units and population variation was calculated.

Statistical analysis

Chemical parameters (conductivity, pH, COD, TN, ortho-P and K) of vermiwash samples and the percentage germination of *P. vulgaris* seeds were analyzed by one-way ANOVA followed by Tukey's pair-wise comparison at 95% confidence. The number of earthworm population variations was compared by the Kruskal-Wallis test at a 95% confidence level. Statistical tests were conducted using MINITAB (version 14) software.

RESULTS AND DISCUSSION

Chemical parameters of aquaculture sludge and vermiwash

The mean pH, conductivity, COD, total nitrogen, ortho-phosphate and potassium in aquaculture sludge used in the study were 7.50 ± 0.01 , $293.33 \pm 3.51 \mu\text{S}\cdot\text{cm}^{-1}$, $9,690.67 \pm 4,924.65 \text{ g O}_2\cdot\text{L}^{-1}$, $0.78 \pm 0.07 \text{ mg}\cdot\text{L}^{-1}$, $0.204 \pm 0.005 \text{ mg}\cdot\text{g}^{-1}$, $10.67 \pm 2.08 \text{ ppm}$, respectively.

The pH of all the experimental vermiwash preparations gradually increased (Figure 2a) with time and showed a significant variation ($F = 13.78$, $p < 0.05$, One-way ANOVA) during the experimental period. However, there were no significant differences in pH among control, ASV and FWV samples during the experimental period ($F = 1.21$, $p > 0.05$; One-way ANOVA) (Table 2). The simultaneous impact of ammonium secretion, calciferous gland activity in earthworms (Lee, 1985), and the thermophilic phase of organic matter decomposition (Karak *et al.*, 2013) in the bedding material could be attributed to a gradual increase in the pH of experimental vermiwash solutions. Increased pH causes an increase in dissolved organic matter and consequently increases the mineralizable C and N (Curtin *et al.*, 1998). An increment of pH means decreased H^+ ions, and vermiwash contains basic properties that make them suitable for crop production. The variation in the pH of vermiwash solutions in the present study (7.15–7.57) is comparable with the pH values of vermiwash produced using cow dung and neem leaves (Chattopadhyay, 2015) and cow dung and hay (Sharif *et al.*, 2016) and well within the

recommended pH range for crop production (6.5–7.5) (Karak *et al.*, 2013).

Electrical conductivity, an indication of salt concentration and an indicative of solution electrolyte concentration (Lock and Janssen, 2002; Ding *et al.*, 2018), decreased with time in all vermiwash samples (Figure 2b). Conductivity in the ASV and the FWV significantly varied between the first and fourth weeks ($F = 15.44$, $p < 0.05$, One-way ANOVA). However, such a variation in conductivity was not observed in the control vermiwash ($F = 14.42$, $p > 0.05$, One-way ANOVA). The mean conductivity of FWV ($7.31 \pm 13.77 \text{ mS}\cdot\text{cm}^{-1}$) was significantly higher than the control and ASV ($F = 19.51$, $p < 0.05$, one-way ANOVA) (Table 2).

The high conductivity in fish waste vermiwash may be due to its high protein content (Khoddami, 2012) and rich amino acids and inorganic ions, including Na^+ , Cl^- , and HCO_3^- (Chan *et al.*, 2007). It is known that the final product's conductivity significantly varies on the raw material used for vermicompost production (Atiyeh *et al.*, 2000). Laos *et al.* (2002) also reported a higher conductivity in vermicompost produced using fish offal. Electrical conductivity of $2.37 \text{ mS}\cdot\text{cm}^{-1}$ was recorded previously in vermicompost produced by a combination of aquaculture sludge, rice straw and water hyacinth (Birch *et al.*, 2010). Sharif *et al.* (2016) prepared a vermiwash using hay and cattle manure with a $2.82 \text{ mS}\cdot\text{cm}^{-1}$ conductivity. The electrical conductivity of aquaculture sludge vermiwash ($2.96 \text{ mS}\cdot\text{cm}^{-1}$) prepared in the present study is highly comparable with the values recorded by those two studies.

Table 2. Chemical parameters (mean \pm SD) of aquaculture sludge, fish waste and the control vermiwash after four weeks of the experiment.

Vermiwash type	pH	Conductivity ($\text{mS}\cdot\text{cm}^{-1}$)	COD ($\text{g O}_2\cdot\text{L}^{-1}$)	Nitrogen (%)	Orthophosphate (ppm)	Potassium (ppm)
Control	7.15 ± 0.59^a	6.20 ± 0.78^a	10.32 ± 1.89^a	0.10 ± 0.15^a	2.94 ± 1.34^a	888.30 ± 37.86^a
Aquaculture sludge	7.15 ± 0.23^a	2.96 ± 0.49^a	7.56 ± 1.44^a	0.22 ± 0.20^b	1.28 ± 0.99^a	230.50 ± 9.57^b
Fish waste	7.57 ± 0.41^a	10.50 ± 2.82^b	9.06 ± 3.58^a	0.09 ± 0.04^a	2.24 ± 0.88^a	592.50 ± 60.21^c

Note: Mean \pm SD in each column superscripted with different lowercase letters are significantly ($p < 0.05$) different.

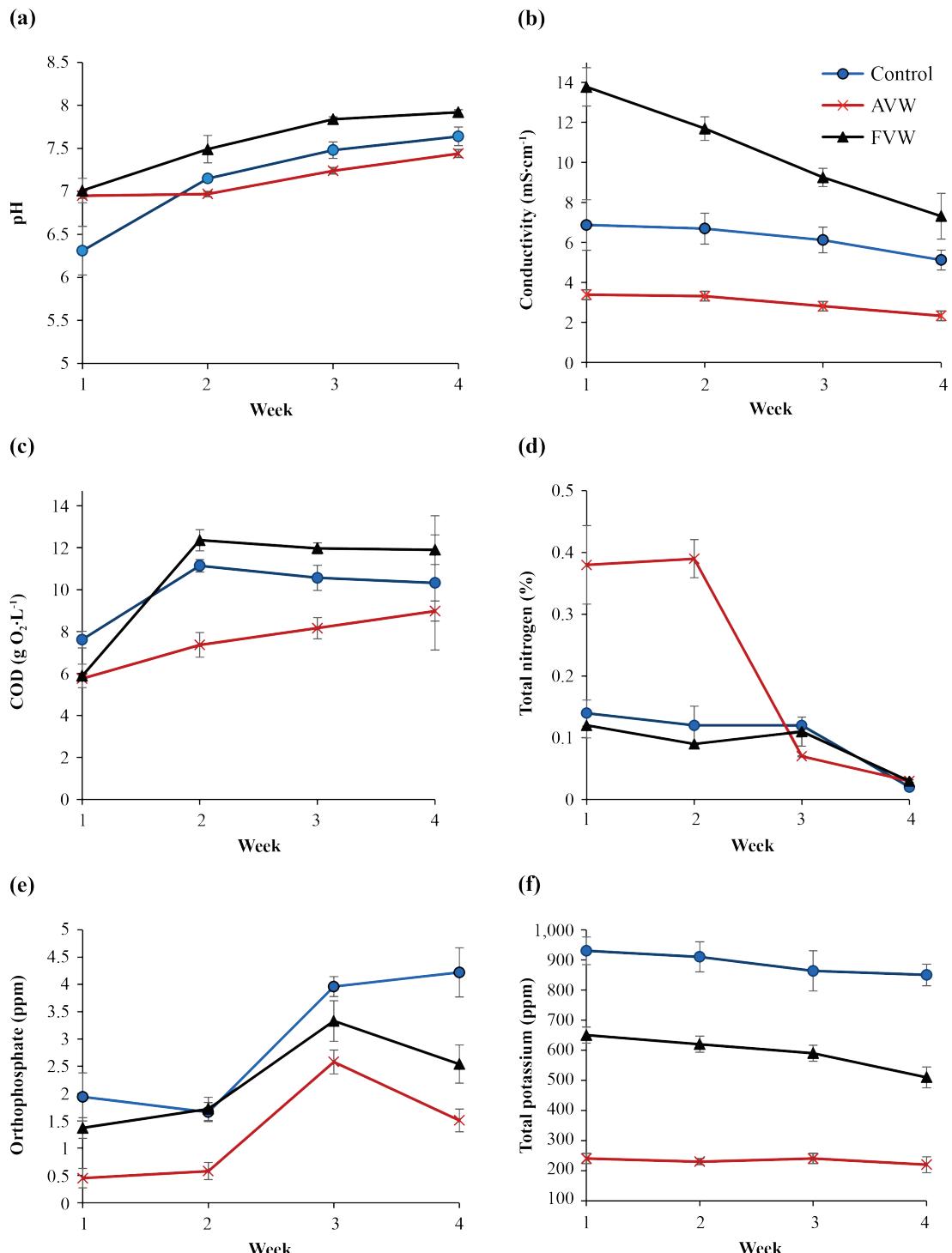


Figure 2. Weekly variation of pH (a), conductivity (b), COD (c), nitrogen (d), ortho-phosphate (e) and potassium (f) in vermiwash obtained from aquaculture pond sludge (AVW), fish waste sludge (FVW) and the control (*Gliricidia* leaves+cow dung).

The conductivity values of FWV and control vermiwash exceeded the upper limit of Indian standards ($4.0 \text{ mS}\cdot\text{cm}^{-1}$) for crop production (Karak *et al.*, 2013); hence their usage in agriculture is questionable.

The COD of ASV gradually increased with time (Figure 2c), having a comparatively lower average than that of FWV and the control (Table 2). COD in the control vermiwash was significantly higher in the fourth week than in the first week ($F = 16.31$, $p < 0.05$, one-way ANOVA). Such a variation was not observed in the other two vermiwash preparations. COD in control, aquaculture sludge and fish waste vermiwash varied in a narrow range ($7.96\text{--}10.32 \text{ g}\cdot\text{L}^{-1}$), showing the similarity of the organic matter content of bedding materials used in the study. As fish waste comprises flesh and blood with higher molecular weight proteins and nucleic acids, it had slightly increased COD than the other two vermiwash preparations.

Initial TN levels in all the vermiwash samples were significantly reduced ($F = 12.47$, $p < 0.05$, one-way ANOVA) by the fourth week of the experiment (Figure 2d). The highest initial total nitrogen level was recorded in the ASV ($0.38\pm0.11\%$) compared to FWV (0.12 ± 0.04) and the control (0.14 ± 0.04) ($F = 13.66$, $p < 0.05$, one-way ANOVA). Mean TN in the ASV was significantly higher than in the FWV and the control ($F = 18.74$, $p < 0.05$, one-way ANOVA) during the experimental period. Aquaculture pond sediment is well recognized to be a source of ammonia and a sink for nitrite and nitrate (Birch *et al.*, 2010; Jasmin *et al.*, 2020). Muendo *et al.* (2014) observed a gradual and continuous increase of nitrogen in the top 5 cm in Egypt's semi-intensively managed tilapia culture ponds. On average, the target organism recovers about 25% (range 11 to 36%) of N added as feed or other nutrient input (Hargreaves, 1998). This was reflected in the results of the present study having the highest N level in aquaculture sludge vermiwash (0.22%), while the fish waste vermiwash had the lowest (0.09%). According to the literature, total N in vermiwash was somewhat lower and

ranged from 0.01% (Nayak *et al.*, 2019) to 0.02% (Ansari and Sukhraj, 2010) and compared poorly with the higher N levels recorded in the present study (0.09 to 0.22%). As observed in the vermicompost production (Karak *et al.*, 2013) and vermicomposting (Klein *et al.*, 2023), the total N in all the experimental vermiwash preparations gradually decreased with time. This might be attributed to the volatilization of ammonia due to high microbial activity, which is more prominent in fish waste vermiwash than in the other two experiment vermiwash preparations.

The ortho-P levels of the FWV, ASV and the control vermiwash slightly fluctuated over four weeks (Figure 2e), while the lowest initial orthophosphate level was recorded in the ASV ($1.28\pm0.99 \text{ ppm}$). No significant differences were detected in ortho-P concentrations among the control, ASV and FWV ($F = 2.26$, $p > 0.05$, One-way ANOVA). In the present study, aquaculture sludge vermiwash had the lowest mean ortho-P concentration ($1.28\pm0.99 \text{ ppm}$), followed by fish waste vermiwash ($2.24\pm1.34 \text{ ppm}$) and control vermiwash ($2.94\pm0.88 \text{ ppm}$). The reduction of orthophosphate in aquaculture sludge vermiwash may be attributed to the lower initial total phosphorous content in the bedding material. As Aye (2012) and Birch *et al.* (2010) indicated, *Gliricidia* leaves and aquaculture sludge contains 5.58% and 0.44% orthophosphate, respectively.

Total K in the FWV and the control gradually decreased with time, but such a variation was not observed in the ASV (Figure 2f). Total potassium among ASV, FWV and the control were significantly different ($F = 251.35$, $p < 0.05$, one-way ANOVA) during the experimental period. The K concentration of aquaculture vermiwash (232.5 ppm) is comparatively more similar to vermiwash prepared using dry grass and cattle dung (245.67 ppm) (Ansari and Sukhraj, 2010), and coconut leaf and cattle dung (205 ppm) (Gopal *et al.*, 2010). The highest K level detected in control vermiwash ($888.30\pm37.86 \text{ ppm}$) might be attributed to the high K content in *Gliricidia* leaves (6.03%), as mentioned by Aye (2012).

Germination of *Phaseolus vulgaris*

The germination rate of *P. vulgaris* exposed to ASV, FWV, control and distilled water is given in Table 3. The germination ratio of *P. vulgaris* was significantly lower when seeds were exposed to FWV ($F = 4.23$, $p < 0.05$, One-way ANOVA).

P. vulgaris seeds exposed to aquaculture sludge vermiwash had a comparatively higher germination ($63.33 \pm 3.28\%$) than those exposed to distilled water ($62.51 \pm 10.63\%$) and control ($47.14 \pm 18.09\%$) though the differences were not statistically significant. Vermiwash contains various extracellular enzymes, plant growth promoters like cytokinin, indole acetic acid, gibberellin-33, and micronutrients (Fe, Cu, Zn, Mn) and mucus secretion of earthworms, humic acid, volatile solids, total solids etc. (Sharif *et al.*, 2016; Nayak *et al.*, 2019). Green gram, *Vigna radiata*, when treated with vermiwash prepared from kitchen waste, had 70% germination rate (Jaybhaye and Bhalerao, 2015). Gopal *et al.* (2010) indicated the presence of plant growth-promoting substances and nutrients in coconut leaf vermiwash. Torri and Puelles (2010) have shown the presence of phytohormones in vermicompost and their

positive effects on germination and seedling growth. The higher mean germination ratio and lower standard deviation of aquaculture waste vermiwash treated seeds demonstrate the applicability of ASW in vermiwash preparation. The significantly lower seed germination in fish waste vermiwash treated seeds may be attributed to high ammonia, trimethylamine, dimethylamine and others volatile basic nitrogenous compounds and biogenic amines in decomposing fish muscles and viscera (Bulushi *et al.*, 2009; Altissimi *et al.*, 2017). Ammonia is found in fish muscle at an average concentration of $10 \text{ mg} \cdot 100 \text{ g}^{-1}$ wet weight and is increased by deamination activities of endogenous and bacterial enzymes (Altissimi *et al.*, 2017). As such, FWV may not suitable to be used in germination of crop seeds.

Population variation of *Eisenia fetida* in different vermiwash preparations

The survival of earthworms in different experimental setups was significantly different ($p < 0.05$, Kruskal-Wallis test) and the highest number of earthworms survived in the control vermiwash preparation unit (Table 4) and the lowest number of earthworms survived in the fish waste vermiwash preparation unit. Many earthworm cocoons and

Table 3. The % germination (mean \pm SD) of *Phaseolus vulgaris* exposed to aquaculture sludge, fish waste and control vermiwash preparations, and distilled water.

Experiment	% Germination
Control vermiwash	47.14 ± 18.09^a
Distilled water	62.51 ± 10.63^a
Aquaculture sludge vermiwash	63.33 ± 3.28^a
Fish waste vermiwash	37.31 ± 3.51^b

Note: Mean \pm SD superscripted with different lowercase letters are significantly ($p < 0.05$) different.

Table 4. Mean (\pm SD) number, % multiplication/reduction of *Eisenia fetida* cultured in aquaculture sludge, fish waste and control vermiwash preparation units after four weeks.

Vermiwash preparation unit	Mean number of earthworms (>4cm)	% Multiplication/reduction
Control	373.21 ± 14.32^a	+49
Aquaculture pond sludge	333.41 ± 20.31^b	+33
Fish waste	23.12 ± 18.41^c	-91

Note: Different superscript letters in a column denote significant ($p < 0.05$) differences indicated by the Kruskal Wallis test.

hatchlings were observed in the ASV and control vermiwash preparation units. A noticeable increase of earthworms in control (49%) and aquaculture sludge vermiwash preparation (33%) units indicate their suitability as bedding materials in vermiwash production. Sharif *et al.* (2016) showed a similar increase in the earthworm population when vermiwash was prepared, using hay and cattle manure as bedding materials. A significant reduction of earthworms was detected in the fish waste vermiwash unit, where nearly 227 earthworms died (91% of initial inoculation) within three months. Due to the low survival rate of *E. fetida* in the fish waste vermiwash preparation unit, fish waste may not have been converted into vermiwash with desirable qualities.

In the fish waste vermiwash preparation unit, high conductivity, ammonia, and biogenic amines in decomposing fish waste may be hazardous to different life stages of earthworms reducing their population size (Bulushi *et al.*, 2009; Altissimi *et al.*, 2017). When the salt concentration is high in the culture medium, it imposes an arresting effect on neurosecretory activity and osmotic potential within the body, reducing survival, biomass, growth rates and reproduction of earthworms (Sharif *et al.*, 2016). An increase in the earthworm population in the FWV units cannot be expected as cocoon production is the most sensitive parameter of *E. fetida* for higher salinity (Guzyte *et al.*, 2011). The increased osmotic pressure due to the high conductivity of FWV may inhibit plant nutrient absorption (Samarakoon *et al.*, 2006) if used as a plant fertilizer.

As a rapidly growing industry, aquaculture generates a significant amount of pond sludge (Hargreaves, 1998; Wudtisin *et al.*, 2015; Dróżdż *et al.*, 2020) that needs to be disposed of in an environment-friendly manner to enhance the industry's sustainability. The current study results indicate the possibility of producing vermiwash using aquaculture pond sludge for agricultural purposes. Further, according to the findings, aquaculture pond sludge can be used as a suitable growing medium for earthworm production. The results of this study can be efficiently utilized in integrated aquaculture, which combines aquaculture with the rearing of livestock and the production of crops.

CONCLUSION

Aquaculture pond sludge vermiwash had high nitrogen, low conductivity, K, COD and *Phaseolus vulgaris* seed germination potential comparable to distilled water and control vermiwash. As a bedding material, aquaculture sludge supports the multiplication of *Eisenia fetida* compared to fish waste. Therefore, aquaculture sludge is more suitable compared to fish waste in producing vermiwash with desirable qualities for agricultural purposes. Using fish waste in vermiwash production was unsuccessful as it could not support a healthy earthworm population and *P. vulgaris* seed germination under experimental conditions. However, vermiwash prepared using cow dung and *Gliricidia* leaves supports the multiplication of earthworms than fish waste and aquaculture sludge bedding materials.

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