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Research Article

Integrated use of *Eisenia fetida* for bioconversion of water hyacinth and as alternative fish meal substitute in aquaculture

Gebeyehu Jemberu[1, 2], Tadillo Bela[3], Dagnachew Ayanew[3], Nega Tassie[3], Dessalegn Ejigu[3], Getachew Beneberu[3], Solomon Girmay[4], Ali Seid[3], Workiyie Worie Assefa[3], Ayalew Wondie[1], Yezbie Kassa[5], Asegedech Kebede[1], Anteneh Mulugeta[6], Belstie Fetene[7], Daniel Mengistu[8], Gashaw Tilahun*[1]

[1]Department of Fisheries and Aquatic Sciences, School of Fisheries and Wildlife, College of Agriculture and Environmental Sciences, Bahir Dar University, P.O. Box 5501, Bahir Dar, Ethiopia
[2]Department of Fisheries and Aquatic Sciences, College of Agriculture and Natural Resources, Gambela University, Gambela, Ethiopia
[3]Department of Biology, College of Science, Bahir Dar University, P.O. Box 79, Bahir Dar, Ethiopia
[4]Department of Wildlife and Ecotourism, College of Agriculture and Environmental Sciences, Bahir Dar University, P.O. Box 5501, Bahir Dar, Ethiopia
[5]Department of Biology, Faculty of Natural and Computational Science, University of Gondar, Gondar, Ethiopia
[6]Department of Agricultural Economics, College of Agriculture and Environmental Sciences, Bahir Dar University, P.O. Box 5501, Bahir Dar, Ethiopia
[7]Amhara National Regional State Environmental and Forest Protection Authority, Bahir Dar, Ethiopia
[8]Department of Geography and Environmental Studies, Faculty of Social Sciences and Geospatial Data and Technology Center, Bahir Dar University, P.O. Box 79, Bahir Dar, Ethiopia
***Corresponding author: gashindian@gmail.com**

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Abstract: *Invasive aquatic plants like Water Hyacinth (Pontederia crassipes) pose serious environmental threats, while conventional fishmeal use in aquaculture remains ecologically and economically unsustainable. This study investigates the potential to integrate the use of red worms (Eisenia fetida) for managing water hyacinth biomass through vermicomposting, and producing red earthworm meal as an alternative protein source for fish. Water Hyacinth, combined with cow manure in four treatment groups, was subjected to a process of vermicomposting trial using a CRD. Simultaneously, red earth worm meal obtained from the composting process was evaluated as Nile tilapia diets, replacing fishmeal at 0%, 25%, 50%, 75%, and 100% levels (T1–T5). A total of 225 fish fingerlings (7.39 ± 0.34 g; 6.13 ± 0.55 cm) were fed diets with 30% crude protein for 60- day feeding trials. The growth performance of fish was monitored biweekly, and feed was adjusted accordingly. Results showed a statistically significant difference ($p < 0.05$) in compost yield and quality among treatments. The highest vermi-compost productivity was observed in the mixture of water hyacinth sludge and cow manure. Vermi-compost from water hyacinth root + shoot and cow manure exhibited the highest electrical conductivity, while the water hyacinth root + cow dung treatment yielded the highest pH. Statistical analysis using SPSS v20 and Tukey's HSD revealed that partial replacement of fishmeal with red earth worm meal supported optimal growth without adverse effects. This study highlights a circular, eco-friendly approach to aquatic weed management and sustainable aquaculture by converting invasive weed biomass into nutrient-rich compost and protein-rich fish feed.*

Keywords: Fish farming, Invasive species, Lake Tana, Nile Tilapia, *Pontederia crassipes*
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1. Introduction

Invasive aquatic plant species such as water hyacinth (*Pontederia crassipes*) have become major ecological concerns worldwide. Native to South America, WH is now prevalent in most tropical and subtropical water bodies, including Lake Tana in Ethiopia. The rapid vegetative reproduction of *P. crassipes* can allow populations to double in just days and results in dense floating mats that shade out submerged vegetation, reduce dissolved oxygen, and alter ecosystem functioning (Lahon *et al*. 2023; Addis and Desta 2024; Zeleke *et al*. 2024). Therefore, managing and mitigating the impact of water hyacinth is critical for maintaining ecosystem services and local economies dependent on aquatic resources.

Conventional methods for water hyacinth control include mechanical removal, chemical herbicides, and biological control agents (Patel, 2012, Abebe and Kassahun, 2023; Getahun and Tsegaye, 2023). However, these methods have limitations such as high operational costs, environmental pollution, and potential negative effects on non-target species (Khan *et al*., 2013) with the exception of some biological controls. An alternative and sustainable approach involves the utilization of WH biomass as raw material for vermicomposting, a bio-oxidative process driven by earthworms, especially red worms. Vermicomposting converts organic waste into stabilized, nutrient-rich compost, enhancing soil fertility and structure (Edwards *et al*., 2011).

Earthworms are particularly efficient decomposers, capable of processing large quantities of water hyacinth into high-quality vermicompost within weeks (Eibne *et al*., 2020). The resulting vermi-compost is rich in macro and micronutrients such as nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), and essential trace elements, along with beneficial microbial populations that improve nutrient availability and plant growth (Arancon *et al*., 2004). Studies have demonstrated that vermin-compost derived from water hyacinth significantly improves soil physicochemical properties and crop yields, making it a valuable resource in organic farming and land reclamation (Masto *et al*., 2011).

Beyond their composting role, red worms themselves represent a sustainable and nutritious source of protein for aquaculture. Fishmeal, traditionally used as the primary protein source in fish feeds, is expensive and associated with overfishing concerns, prompting the search for alternative protein ingredients (Tacon and Metian, 2008). Red worms contain approximately 50-65% crude protein (Musyoka *et al*., 2019; Gunya and Masika, 2021), with a favourable amino acid profile, essential fatty acids, vitamins, and minerals suitable for fish nutrition (Henry *et al*., 2015). Incorporation of red worm meal in fish diets has been shown to enhance growth performance, feed conversion efficiency and immune responses in various cultured fish species such as tilapia, catfish, and carp (Sogbesan and Ugwumba, 2006; Kumar *et al*., 2018; Belay *et al*, 2025).

Utilizing red worms harvested from vermicomposting water hyacinth not only closes nutrient loops but also promotes circular economy principles in aquaculture and agriculture. This integrated approach addresses two environmental challenges: invasive aquatic weed management and sustainable fish feed production, while providing socio-economic benefits for small-scale farmers and fishers (Lalander *et al*., 2015). The dual utility of red worms thus represents a promising strategy for enhancing resource efficiency, reducing environmental impacts, and promoting sustainable livelihoods. However, limited studies have assessed the effectiveness of red earthworm meal as a direct replacement for fishmeal in tilapia diets under practical farming conditions. This study aimed to assess the dual function of red worms in transforming water hyacinth into nutrient-enriched compost and serving as a sustainable alternative to conventional fishmeal. The research particularly focused on their potential to enhance agricultural productivity and support Nile Tilapia farming around Lake Tana, where efficient management of water hyacinth is urgently needed.

2. Materials and Methods

2.1. Description of the study area

The process of vermicomposting was carried out under the constructed plastic shade at the shore of Lake Tana. The fish trial was carried out in concrete ponds covered with a greenhouse made of polyethene plastic in research site of Fisheries and Aquatic at the College of Agriculture and Environmental Sciences, Bahir Dar University, Ethiopia. The experimental fish were stocked in a hapa of $1m^3$ total volume (1m length x 1 m width x 1 m depth) installed inside the fish ponds in a triplicate and fed twice a day for 60 days.

2.2. Vermicomposting experiment

2.2.1. Water hyacinth preparation

Water hyacinth was collected in the early morning from infested areas of Lake Tana at the designated research site, following appropriate safety and environmental precautions. The harvested plants were transported immediately to the experimental field for processing. Different parts of the plant like leaves, petioles, and roots (Fig 1) were separated, air-dried under shade, and subsequently ground into smaller pieces.

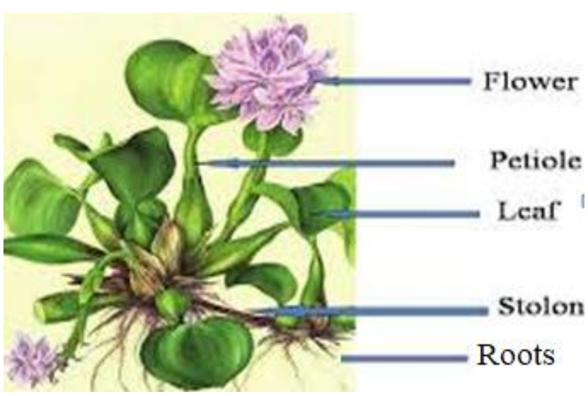

Figure 1: Diagrammatic illustration of different parts of water hyacinth

Source: (https://www.google.com/images20stolen%20production)

The dried material was then sieved using a mesh with whole dimensions of 1 cm × 2 mm to ensure uniform particle size. Cow manure, which was dried for 15 days at ambient temperatures (Wani and Rao, 2013) was added to the dried water hyacinth parts at 1:5 ratios as a co-composting material to reduce the emission of harmful gases and excess heat that could negatively affect earthworm activity.

2.2.2. Experimental treatment and design

The vermicomposting experiment was conducted using a Completely Randomized Design (CRD) with three replications. The size of the beds was 1 m × 1 m where the space between the beds was 0.5 m and between the blocks was one meter following the methods described by Gajalakshmi *et al*. (2002) and Gajalakshmi and Abbasi (2003) for high worm density production and to reduce worm mortality.

The total experimental area was 27 m² (6 m × 4.5 m). The treatments consisted of the dried and sieved products of the four water hyacinth parts each 17.5 kg in weight. These treatments were also supplemented with equal amount of cow manure (3.5kg). The total quantity of the input for vermicompost was thus 21 ± 00 kg (3.5 kg + 17.5 kg). The vermicompost inputs were turned manually every 15 days in order to provide proper aeration to earthworms (Singh and Kalamdhad, 2013). Most favorable conditions, including temperature (25–28°C) and moisture (65–75%), were maintained throughout the study period (Sharma and Garg, 2020).

Table 1: Experimental design and treatment arrangement used for vermi-composting

Treatments	Experimental beds 1	Road	Experimental beds 2	Road	Experimental beds 3
TC1	Root		Root		Root
	Inter-bed space		Inter-bed space		Inter-bed space
TC2	Root + Shoot		Root + Shoot + CM		Root + Shoot + CM
	Inter-bed space		Inter-bed space		Inter-bed space W=1m
TC3	Shoot		Shoot		Shoot + CM
	Inter-bed space		Inter-bed space		Inter-bed space
TC4	Sludge		Sludge + CM		Sludge + CM
	Inter-bed space W = 0.5m		Inter-bed space		Inter-bed space

2.2.3. Vermicompost harvesting and analysis

The matured vermi-compost was harvested after 90 days by removing the worms manually using light exposure techniques (Domínguez and Edwards 2011). The total quantity of vermi-compost produced was collected and dried to calculate the vermicompost productivity. The productivity of vermicompost was calculated based on the procedures described by (Ndegwa and Thompson, 2001) as indicated below.

$$\text{VP} = \left(\frac{Total\ vermicompost\ weight}{Total\ Prefeedstoch\ weight}\right) * 1 \qquad [1]$$

Moreover, samples of the dried pre-composting feedstock and composted vermicompost were taken and analyzed for their pH and EC.

2.3. Red earthworm meal preparation

After the composting cycle, adult red worms were harvested and separated from vermi-compost and vermi-liquid manually following the method described by Jameson and Venkataramanujam (2002). The harvested worms were placed in a clean container for three hours to allow for gut evacuation of undigested material (Akpodiete and Okagbere, 1999). They were then thoroughly washed and blanched in boiling water for one minute to neutralize heat-sensitive toxins such as lysenin and haemolytic factors present in the coelomic fluid (Cayot *et al*., 2009). Subsequently, the worms were oven-dried at 60 °C for 72 hours (Musyoka *et al*., 2019), grounded into a fine powder, and sieved through a 2 mm mesh. The processed worm-meal was then sealed in airtight polyethylene bags and stored at 4°C until they are used for feed formulation (Gunya *et al*., 2016).

2.4. Fish feed formulation and feeding trial

2.4.1. Diet preparation and its proximate composition

The proximate composition of the feed ingredients of the red earthworm meal, fishmeal, and corn flour was analyzed to determine the crude protein, crude fibre, crude lipid, ash, and moisture contents and the gross energy and nitrogen-free extract (NFE) following the procedures described by AOAC (2012) and summarized in Table 2. For the adult earth worms were collected from the vermicomposting beds after the composting process was completed. The worms were manually separated from the compost using hand-sorting and gentle washing with clean water to remove adhering organic matter and debris. The cleaned worms were then placed on moist filter paper for 24 hours to allow purging of gut contents. During this time, they naturally expel (excrete) all the material in their digestive tract.

Experimental diets were formulated using two primary protein sources: the oven-dried red earthworm meal and fishmeal. Vegetable oil (5%) and a mineral-vitamin premix (2%) were included to provide essential fatty acids and micronutrients (Chapman, 1992). Cornmeal served as a binding agent. Ingredient proportions were calculated using a Microsoft Excel-based trial formulation method to meet the 30% crude protein requirement for Nile tilapia fingerlings (El-Sayed, 2018).

Table 2: Proximate composition of earthworm meal, fishmeal, and corn flour used as experimental feed ingredients

Compositions	Feed ingredients		
	Earthworm meal	Fishmeal	Corn flour
Crude protein [%]	43.70	59.40	6.75
Crude fat [%]	9.00	7.00	3.52
Crude fibre [%]	8.79	5.60	1.63
Moisture [%]	8.00	6.00	11.70
Dry matter [%]	92.00	94.00	88.30
Ash [%]	11.85	22.00	0.90
NFE	18.66	1.00	75.50
Gross energy [KJ/100g^{-1}]	470.28	464.11	316.23

Ingredients were mixed, homogenized, and moistened with 30% hot water for 5 minutes to ensure uniformity, then pelleted using a GEEPRS model pelletizer. Pellets were oven-dried at 40°C for 24 hours, crushed into crumbles, and sieved through a 1 mm mesh. The final pellet size was 2 mm, suitable for tilapia fingerlings, and stored in water-impermeable polyethylene bags at 4°C to prevent microbial growth (Kumari, 2009; Vodounnou and Tossavi, 2016).

The five iso-nitrogenous diets (30% CP), which were calculated in micro Excel were formulated with graded replacement levels of fishmeal by earthworm meal: T_{00}% (100% fishmeal), T_{25}%, T50%, T_{75}%, and T_{100}% (100% earthworm meal) (Table 3).

The replaced quantity of the treatment was adjusted using corn flow with the proximate analysis results indicated in Table 2.

Table 3: Percentage composition of experimental (diet kg^{-1} fed) for Nile tilapia fingerlings

Feed items	Treatment`s replacement level in %				
	T_{00}	T_{25}	T_{50}	T_{75}	T_{100}
Earthworm meal	00.00	16.25	32.50	48.75	65.00
Fishmeal	45.00	33.75	22.50	11.25	00.00
Corn flour	48.00	43.00	38.00	33.00	28.00
Minerals and Vitamins premix	2.00	2.00	2.00	2.00	2.00
Vegetable oil	5.00	5.00	5.00	5.00	5.00

2.4.2. Experimental procedures

A total of 225 healthy fingerlings of Nile tilapia (*Oreochromis niloticus*) with a mean weight of 7.39 ± 0.34 g and a mean length of 6.13 ± 0.55 cm were used for the experiment. Fingerlings were fed fish meal of Alema Koudijs for a week to acclimate them to the new environment (Aly *et al.*, 2020). The dead and weak fishes were removed daily during the time of acclimatization. At the end of acclimatization, the standard length and weight of each fingerling were measured and then the fingerlings were randomly assigned to 15 cages at a stocking density of 15 fingerlings per cage, which were fed with the respective experimental diets (Table 3).

2.5. Data collection

The fishes were sampled every two weeks to monitor growth parameters including weight gain (WG), feed conversion ratio (FCR), specific growth rate (SGR) and survival rate using the equations described below. Sixty percent of the fish in each cage were sampled using a scoop net prior to feeding. Water quality parameters (temperature, pH, DO, and ammonia) were also monitored throughout the trial to ensure optimal conditions.

$$\text{Mean weight} = \frac{Total\ harvest\ weight}{Number\ of\ fishes\ harvested} \quad [2]$$

$$\text{Percent weight gain} = \left(\frac{FW-IW}{IW}\right)*100 \quad [3]$$

$$SGR = \left(\frac{FW - IW}{culture\ period\ in\ days}\right)*100 \quad [4]$$

$$SR = \left(\frac{Number\ of\ fishes\ at\ the\ end\ of\ experiment}{Number\ of\ fishes\ stocked}\right)*100 \quad [5]$$

Where

FW = final weight; IW = initial weight; SGR = specific growth rate and SR = survival rate

In addition key physicochemical water quality parameters (water temperature (°C), pH, and dissolved oxygen (DO, mg L^{-1}) were recorded biweekly

3. Results and Discussion

3.1. Vermi-compost maturity

A stable, humus-like product, which is an indicative for matured vermicompost was obtained after the composting process of 110 days. The final product exhibited a dark black color, which is a key indicator of compost maturity (**Fig** 2). Mature vermicompost typically resembles moist, loose soil with a homogeneous texture and aesthetically pleasing appearance (Majlessi *et al*, (2012). These observations are consistent with previous findings. Edwards *et al.* (2011) reported that vermicompost derived from various organic feedstocks exhibits a dark black color, mull-like soil odour, and a homogeneous, soil-like texture. Similarly, Sami *et al.* (2020) described matured vermicompost as dark brown in color, porous, granulated, and devoid of any foul smell supporting the maturity indicators observed in the current study.

Figure 2: Dark brown matured vermicompost obtained from the root part

The decomposition period in the present study lasted in 110 days, which is relatively shorter than that reported in other studies. For instance, Gajalakshmi *et al*. (2001a) and Gupta *et al*. (2007) documented composting durations of 147 days and 180 days, respectively, to reach a similar stage of vermicompost maturity. This may be attributed to the application of an adequate number of healthy earthworms—specifically, with the number of adult individuals of worms introduced per windrow pile—with a survival rate exceeding 90%. This high survival and activity level may have enhanced the decomposition efficiency. Garg *et al*. (2008) suggested that a stocking density of 27–53 earthworms per kilogram of feedstock provides optimal conditions for vermicomposting, which aligns with the density employed in the present study.

Pre-composting is essential to reduce earthworm mortality by eliminating volatile compounds such as ammonia and salts, which are toxic to *Eisenia fetida*. To ensure proper aeration and microbial activity during the composting process, the feedstock mixtures were manually turned every two weeks (Singh and Kalamdhad, 2013). Optimal environmental conditions were maintained throughout the study; temperature (25–28°C) and moisture content (65–75%) as indicated by Baghel, (2018);.Sharma and Garg, (2020). Moreover, plastic sheet was used to cover the compost, regular water sprinkling to sustain moisture levels, and the construction of a shade structure to protect earthworms from direct sunlight.

3.2. Vermicomposting productivity and biomass reduction

The productivity of vermicomposting of different parts of water hyacinth biomass with cow manure mixture in equal amounts of original feedstock weight (21 ± 00 kg) is summarized in Table 4. Among the substrate treatments tested, the Sludge combination yielded the highest vermicompost productivity of 15.18 kg (72.2%). This high performance may likely be attributed to the rich nitrogen and moisture content of sludge, along with its diverse microbial population, which enhances decomposition when combined with cow manure (Yadav and Garg, 2011). CM improves the carbon-to-nitrogen (C:N) ratio and introduces beneficial microbes to accelerate the composting process.

Water hyacinth roots as input for vermicomposting recorded the second-highest vermicompost yield of 13.47kg (64.3%)**,** suggesting that root biomass, though rich in lignin and cellulose can serve as an effective carbon source if supplemented nitrogen-rich cow manure (Li, Zhang, and Wang, 2023). This synergy enhances microbial decomposition and earthworm activity (Ndegwa and Thompson, 2001). Inputs sourced from root + shoot resulted in moderate productivity (53.2%)**.** The addition of shoot biomass may have altered the nutrient composition or produced excessive heat (Zhang, 2020) or acidity, especially if the shoots were immature, thereby temporarily inhibiting worm activity and reducing conversion efficiency.

The Water hyacinth shoots produced the lowest vermicompost yield of 11.5 (45.6%)**,** likely due to an imbalanced substrate composition. Shoots alone may lack sufficient fiber or may have high moisture content, leading to anaerobic conditions unfavorable for earthworm development and compost stabilization (Kale, 1998).

Overall, the average productivity across all four treatments was approximately 58%, indicating a substantial reduction in mass relative to the initial feedstock input. This aligns with findings by Lalander *et al*. (2015), who reported a 46% mass reduction during vermicomposting of CM and food waste. The decrease in mass is largely due to the biodegradation of organic material by both earthworms and microbial activity.

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Table 4: Yield of vermicompost obtained from different part of water hyacinth

Treatment	Pre-composting feedstock (kg)	Weight of vermi-compost yield (kg)	Vermi-compost productivity (%)
T1 (Root)	21 ± .00	13.47 ± .17	64.3
T2 (Root + shoot)	21 ± .00	11.17 ± .15	53.2
T3 (Shoot)	21 ± .00	9.56 ± .23	45.6
T4 (Sludge)	21 ± .00	15.18 ± .08	72.2
Total	21 ± .00	12.35 ± 2.25	58.8

A Univariate Analysis of Variance (ANOVA) was conducted to assess differences in vermicompost productivity across the various pre-feedstock treatments (Table 5) indicated a highly significant effect of treatment type on compost yield ($p < 0.01$), confirming that the composition of the initial pre-feedstock mixture had a strong influence on the amount of vermicompost produced. These align with observed differences in the degradation rates and nutrient compositions of the pre-feedstocks used, which likely contributed to the variation in productivity.

3.3. Selected quality parameters of the vermicompost

The results on selected quality parameters of the vermicompost are summarized in Table 6. Significant changes in pH and electrical conductivity (EC) were observed after vermicomposting. The pH of the raw feedstocks initially ranged from 7.6 to 8.1, indicating a slightly alkaline nature. After vermicomposting, the pH of the vermicompost decreased and stabilized within a nearly neutral range of 6.7 to 7.2. This shift suggests enhanced microbial activity and organic acid formation during decomposition, leading to improved compost quality. Moreover, electrical conductivity, which indicates the concentration of soluble salts, increased across all treatments following vermicomposting. Initial EC values of the feedstocks ranged from 2.93 to 3.33 mS/cm, whereas vermicompost EC values increased to a range of 3.31 to 3.85 mS/cm. The highest EC value (3.85 ± 0.01 mS/cm) was recorded in the treatment TC2 (root + shoot reflecting higher mineralization and nutrient availability in this mixture.

Table 5: Summary of statistical tests of vermicompost productivity among treatments (Univariate ANOVA)

Dependent Variable: VC weight		Multiple Comparisons		
(I) Treatments	(J)Treatments	Mean Difference (I-J)	Std. Error	Sig.
Root	Root + Shoot	2.2967	.13740	.000*
	Shoot	3.9100	.13740	.000*
	Sludge	-1.7133	.13740	.000*
Root + Shoot	Root	-2.2967	.13740	.000*
	Shoot	1.6133	.13740	.000*
	Sludge	-4.0100	.13740	.000*
Shoot	Root	-3.9100	.13740	.000*
	Root + Shoot	-1.6133	.13740	.000*
	Sludge	-5.6233	.13740	.000*
Sludge	Root	1.7133	.13740	.000*
	Root + Shoot	4.0100	.13740	.000*
	Shoot	5.6233	.13740	.000*

Table 6: pH and electrical conductivity of the pre-feedstocks and their vermicomposts obtained from them

	pH		Electrical conductivity (ms/cm)	
Parts of Water hyacinth	Pre-feedstock	Vermin-compost	Pre-feedstock	Vermin-compost
Root	8.10 ± .01	7.20 ± .01	2.93 ± .03	3.39 ± .02
Root + shoot	7.80 ± .01	7.10 ± .10	3.33 ± .01	3.85 ± .01
Shoot	7.60 ± .02	6.70 ± .01	3.21 ± .01	3.65 ± .01
Sludge	7.90 ± .01	6.80 ± .02	3.17 ± .02	3.31 ± .01
Average	7.85 ± .19	6.95 ± .22	3.16 ± 29	3.71 ± .22

Among all treatments, the compost derived from Water hyacinth roots had the highest pH (7.2 ± 0.01), while the vermin-compost sourced from the combination of roots and shoots of Water hyacinth had the highest EC value (3.85). These findings highlight that both the type and combination of organic pre-feedstocks influence the final physicochemical properties of the vermicompost

Reduction of pH was observed in all treatments. Several other authors have also reported pH reduction during vermicomposting (Sharma and Garg, 2019; Balachandar *et al.*, 2021). This near-neutral range of pH was also reported in an earlier study on a weed by Yadav and Garg (2016). The differences in pH of vermicomposts are probably related to the raw materials used for vermicomposting (Alves *et al.*, 2001). The pH reduction during vermicomposting may be attributed to the production of various compounds such as organic acids, ammonia, nitrates, orthophosphates and the release of CO_2 gas. The mineralization of nitrogen and phosphorus and the production of intermediate organic acids by the biodegradation of the substrate could be the significant reason for the lowering of pH during the process (Ndegwa *et al.*, 2000). This reduction in pH may further be augmented by the decomposition of organic materials and intestinal secretion of earthworms that play a central role in neutralizing the carboxylic and phenolic groups of humic acids (Pramanik *et al*., 2007). Composts with pH near neutral or slightly alkaline are preferred for many crops.

It is evident from the literature that usually EC of vermicompost is higher than that of pre-feedstocks (Sharma and Garg, 2020; Balachandar *et al*., 2021). The EC increment was recorded after the vermicomposting process and it was within the safe limits of phytotoxicity (Putranta *et al*., 2019). This increasing trend may be attributed to the formation of ions due to mineralization in the presence of earthworms as reported by Yadav and Garg (2011). The EC value is not only important for the assessment of vermicompost quality, but also determines the survival of the earthworm species (Ananthavalli *et al*., 2019).

The elevated electrical conductivity in our study was observed with root + shoots + manure treatments, which might reflect higher soluble salts or mineral release, consistent with rapid mineralization. However, high EC may risk salt stress if applied in certain soil types or in plant culture. We also observed the highest pH in the root + manure mixture, which may be due to the buffering capacity of manure, lower acidifying compounds, or faster nitrogen mineralization producing ammonia, which tends to raise pH. These dynamics are reported in similar vermicomposting studies (Adhikari *et al*., 2022; Gueye *et al*., 2022; López-González *et al*., 2016).

3.4. Water quality, growth and feed utilization of Nile Tilapia

3.4.1. Water quality parameters during the feeding trial

Key physicochemical water quality parameters (water temperature (°C), pH, and dissolved oxygen (DO, mg L^{-1}) were measured consistently across all the treatments in the experimental greenhouse concrete fish ponds inside the cage feeding trial (Table 7)

The mean water temperature ranged from 27.97 ± 0.48°C to 28.03 ± 0.48°C, remaining within the ideal range for tilapia growth (Vodounnou and Tossavi, 2016; Samuel *et al*., 2018). The pH values varied slightly across treatments, ranging from 7.28 ± 0.12 to 7.37 ± 0.22, indicating a neutral to slightly alkaline environment conducive to fish health and metabolic activity (El Feky and El-Sherif, 2019).

Dissolved oxygen levels were also maintained within an acceptable range, fluctuating from 5.09 ± 0.26 mg L^{-1} to 5.22 ± 0.22 mg L^{-1}. These values reflect sufficient oxygenation to support the physiological needs of the fish throughout the experimental period. The DO values more or less agree with the findings of DoF (2009), who noted that the range of dissolved oxygen suitable for fish culture would be 5.0 to 8.0 mg L^{-1} Boyd (1998) also found the desired concentration of DO 5 to 15 mg L^{-1}, which are more or less similar findings with the present study.

The mean value of ammonia (NH_3) was 0.12±0.01 mg L^{-1}, which was within the range reported by Rahman (2005), who recorded ammonia values ranging from 0.01 to 0.82 mg $L^{-1.}$ Ammonia is a very important parameter for good fish production. Ammonia in water is found in two forms: ammonium ions ($NH4^+$), which are non-toxic, and un-ionized, toxic ammonia (NH_3). The appropriate range of ammonia for fish farming is <0.1 mg L^{-1} (Boyd, 1998).

Table 7: Physicochemical parameters of water during the feeding trial

Treatments	Temperature, ^{0}C	pH	DO (mgL^{-1})	Ammonia (mgL^{-1})	Nitrate (mgL^{-1})	Nitrite (mgL^{-1})
T_{00}	27.91±0.53	7.29±0.07	5.22±0.09	0.12±0.01	4.24±0.01	0.02±0.00
T_{25}	28.03±0.48	7.32±0.10	5.12±0.16	0.12±0.01	4.24±0.01	0.02±0.00
T_{50}	28.02±0.43	7.28±0.12	5.22±0.15	0.12±0.01	4.24±0.01	0.02±0.00
T_{75}	27.96±0.43	7.37±0.22	5.11±0.13	0.12±0.01	4.24±0.01	0.02±0.00
T_{100}	27.97±0.48	7.33±0.15	5.09±0.26	0.12±0.01	4.24±0.01	0.02±0.00

DO is dissolved oxygen and pH is the hydrogen ion concentration

3.4.2. Growth and survival of Nile Tilapia fingerlings

The growth performance and survival rate of *Oreochromis niloticus* fingerlings that fed diets with varying inclusion levels of red earthworm meal are summarized in Table 8.

Growth performance: The diets used to replace fish meal with red earthworm meal showed significant difference ($p<0.05$) in weight gain and final length across the treatments. Fishes fed on T25 and T50 showed improved growth with higher final weights, better specific growth rate and more efficient feed conversion ratio than the control with 100% fishmeal (T_{00}%). These results suggest moderate Red earth worm meal inclusion supports the optimal growth of fishes. In contrast, fishes fed on T75 and T100 showed reduced performance where fishes fed on 100 replacement of fishmeal with Red earth worm meal exhibited significantly lower weight gain and poorer FCR, indicating reduced nutrient utilization.

Nile tilapia fingerlings recorded higher weight gain at diet T50, which was formulated with an equal proportion of earthworm meal and fishmeal throughout the study period. The results were consistent with the findings of Olele and Okonkwo (2014); Ahmed *et al*. (2023); Belay *et al*. (2025), but did not agree with Aly *et al*. (2020), who recommended the replacement of red earth worm (T_{75}) by fishmeal. This may be due to the culture substrate used for this experiment was water hyacinth, and this aquatic weed may affect the nutrient composition of red earthworm. The growth of Nile tilapia fingerlings was very good and showed that each diet can completely replace the fishmeal with red earthworm meal due to the growth recorded at the control treatment, which was comparable with all diets, including fourth and fifth diets (T_{75} T_{100}). This growth response pattern implies the beneficial effect of earthworm meal in culturing Nile tilapia. According to Olele and Okonkwo (2014), the amino-acid profile of the earthworm diet in combination with the protein composition in the body tissue of the fish stimulates faster growth. This justifies the ability of red earthworm meal to provide productive nitrogen and digestible amino acids needed for the metabolic activity of the fish.

Furthermore, it could be explained by the advantages of combining two crude protein sources to create a single, superior diet (Djissou *et al*., 2016; Olele and Okonkwo, 2014); it was also suggested that red earthworm meal could be used in place of fishmeal to a large extent and that a combination of both promotes better growth (Olele and Okonkwo, 2014). Indeed, the nutritive value of a given protein diet does not depend merely on its amino acid profile but also on its level of digestibility.

The fiber content of the red earthworm diet is known for enhancing the digestibility of feeds and thus leading to better growth performance of fishes in aquaculture. Based on the growth pattern, the contents including the fiber of diet three in in equal combination with fishmeal surpassed the nutritive value of diet one which was red worm meal free (Table 8) This was the apparent reason for the higher weight gain of fingerlings fed an equal proportion than feeding fishmeal. For the sake of this study, the amino acid profile of the feed ingredients has not yet been conducted. However, based on researchers' findings, the lowest value in mean weight increment for the diet without earthworm meal diet one could be attributed to insufficient amino acid and/or the absence of easily digestible fiber in the fishmeal diet when compared to that of the other experimental diets (fishmeal and earthworm meal combined)

(Amerio, 1983; Hilton, 1983; Olele and Okonkwo 2014).

Weight gain: Though the weight gain recorded at diets four and five was comparable with the control diet, the growth showed a significantly decreased weight gain of Nile tilapia fingerlings from diet four to diet five. This might be due to the oven-dried red earthworm, which was found to be unpalatable to Nile tilapia fingerlings due to haemolytic factors in the foul-smelling coelom fluid. Hilton (1983) stated that diets containing earthworm meal reduced feed intake and growth. Some ideas have been made by researchers to explain why earthworm meal-based diets reduced growth rates. The coelomic fluid, a yellow fluid in earthworms, could make feeds unpalatable when they contained bulky amounts of earthworms and could thus decrease feed intake (Tacon *et al*., 1983; Tuan et al., 2015).

Average daily mean weight gain: The average daily mean weight gain per day was significantly increased (*P<0.05*) from diet one T_{00} to diet three (T_{50}) and showed a little decline from diet four ($T_{75)}$ to diet five ($T_{100)}$. In this regard, the results of the present study are in line with El-Sherif and El-Feky (2009), who reported an average daily weight gain between 0.12 and 0.39 g/day. This study revealed a slightly greater value of daily weight gain with Vodounnou and Tossavi (2016), who reported a range between 0.14±0.00 to 0.25±0.02 g/day under an unconventional feed source.

Specific growth rate: The specific growth rate of Nile tilapia fingerlings was assessed. The effects of replacement red earthworms on specific growth rate (SGR) for Nile tilapia were statistically significant (*P<0.05*) within the five diets; the third diet (T_{50}) showed the best specific growth rate, followed by diets two and four, while diet one showed the least specific growth rate at the end of the experiment. In this growth parameter, results are more or less coincide with Medard *et al*. (2018), where they scored a specific growth rate values within a range of 2.017 ± 0.28 to 4.01 ± 0.09. This study also scored a better specific growth rate than Djissou *et al*. (2016), where they noted that a specific growth rate in the range of 1.17±0.10 to 1.83± 0.10 under an unconventional feed source for Nile Tilapia (Vodounnou and Tossavi, 2016).

Table 8: Growth performance and survival rate of Nile tilapia fed on different diets within twelve weeks

Parameter	T_{00}	T_{25}	T_{50}	T_{75}	T_{100}
IML (cm)	5.90±0.66^{a}	5.67±0.58^{a}	5.67±0.76^{a}	5.17±0.76^{a}	5.83±0.29^{a}
FML (cm)	14.43±0.51^{a}	14.97±0.38^{a}	16.90±0.17^{b}	14.53±0.50^{a}	14.47±0.50^{a}
MLG (cm)	8.53±0.55^{a}	9.30±0.46^{a}	11.23±0.75^{b}	9.37±0.23^{a}	8.63±0.32^{a}
IMW (g)	7.36±0.34^{a}	7.77±0.60^{a}	7.26±0.99^{a}	6.90±0.26^{a}	7.64±0.49^{a}
FMW (g)	31.65±1.86^{a}	35.25±1.52^{a}	39.32±1.98^{b}	31.65±2.13^{a}	31.96±2.10^{a}
MWG (g)	24.29±2.10^{a}	27.48±1.20^{a}	32.06±1.10^{b}	24.75±1.89^{a}	24.32±2.47^{a}
MDWG (g)	0.26±0.05^{a}	0.32±0.04^{a}	0.42±0.03^{b}	0.27±0.04^{a}	0.26±0.05^{a}
FCR	2.00±0.29^{a}	1.60±0.33^{a}	1.43±0.33^{a}	1.89±0.20^{a}	1.98±0.19^{a}
SGR	2.01±0.90^{a}	2.31±0.47^{a}	2.78±0.71^{b}	2.23±0.40^{a}	2.03±0.34^{a}
SR (%)	100±0.00^{a}	97.78±0.71^{a}	100±0.00^{a}	95.56±1.41^{a}	100±0.00^{a}
PER	3.40±0.90^{a}	3.60±0.70^{a}	4.24±0.53^{a}	3.65±0.79^{a}	3.40±0.87^{a}

IML [cm] * Initial mean length; FML [cm] * = Final mean length; MLG [cm]* = Mean length gain; IMW [g]* = Initial mean weight; FMW [g]* = Final mean weight; MWG [g]* = Mean weight gain; DMWG [g]* = Daily mean weight gain; SGR* = Specific growth rate and SR [%]* = Survival ratio; FCR = Feed conversion ratio; PER = Protein efficiency ration; * = significant difference at p< 0.05

Feed conversion ratio (FCR): The FCR values in this study indicated comparatively better feed utilization in diets containing equal proportions of earth worm meal and fishmeal (diet three), while diets without fishmeal (diet five) showed relatively less favorable FCR values as indicated in Table 8, however, these differences were not statistically significant ($P > 0.05$)

So, with the five treatments throughout the experimental periods, it suggested that replacement of FM by red earthworm meal of up to $_{100}$% is possible without any negative effect on the FCR. The mean FCR for diet three suggested comparatively better feed utilization and intake at this protein level, implying relatively improved efficiency and diet quality, although the differences among treatments were not statistically significant (Arora *et al*., 2019). This is also evidence that the fiber and higher methionine contents of earthworm meal are necessary for optimal growth and utilization of nutrients, addressing both earthworm meal and fishmeal. In this regard, our finding was consistent with other authors (Aly *et al*., 2020; Vodounnou and Tossavi, 2016), who noted feed conversion ratio between 1.43±0.01-2.59±0.29. Based on their findings, they asserted that since earthworm meal has higher methionine content than fishmeal, it would have a lower feed conversion ratio (FCR), and in combination with fishmeal would result in higher nutritive value for the fingerlings at a $_{50}$% inclusion level.

Red earthworm meal replacement: The effects of replacing fishmeal with earthworm meal (Table 8) on the protein efficiency ratio (PER) were not statistically significant (*P>0.05*). The non-significant variation in protein efficiency ratio among treatments indicated that red earthworm meal can effectively replace fishmeal without compromising protein utilization efficiency. This finding is consistent with the reports of Adeniyi and Folorunsho (2015); Devic *et al.* (2018) and Samuel (2021), who observed PER values within a similar range (3.1–3.4) in related studies.

The present results suggest that up to 100% replacement of fishmeal with REWM is feasible without detrimental effects on feed performance indicators. According to Muin *et al.* (2017), the protein efficiency ratio serves as a reliable measure of dietary protein quality, with higher PER values reflecting improved protein utilization. The maintenance of acceptable PER values across all treatments in the current study implies that REWM provides a protein source of adequate nutritional quality for fish growth. These findings are in agreement with those of Susanto *et al*. (2020); Ahmed *et al*. (2023) and Belay *et al*. (2025), who similarly reported that replacing fishmeal with red earthworm meal did not adversely affect growth performance, feed conversion, or overall feed efficiency. Collectively, the evidence supports the potential of REWM as a sustainable and cost-effective alternative to fishmeal in aqua feed formulations.

The drop in performance at higher replacement levels (e.g. 75> up to 100%) in many such studies, including ours, can be tied to several factors. Fishmeal has a well-balanced amino acid profile and high digestibility. REWM may lack certain essential amino acids or have lower digestibility due to chitin content. Moreover, earthworm meal contains chitin in its exoskeleton. If inclusion is too high, the undigested chitin may reduce nutrient absorption or increase gut passage, leading to suboptimal feed conversion. In addition, as inclusion of REWM increases, feed texture, taste, binding, and sinking/swelling behavior might change, leading to lower feed intake or higher feed wastage.

Survival Rate: It was one of the growth parameters addressed in this study. Even though Nile tilapia fingerlings fed diets one ($_{00}$T), three ($_{50}$T), and five ($_{100}$T) had the highest survival value and no deaths were scored, and the fingerlings fed diet four ($_{75}$T) had the lowest survival followed by diet two ($_{25}$T), but there was no statistically significant difference in survival rate within each of the treatments (*P>0.05*). The numeric values scored here indicated the higher physicochemical quality in the growing environment and the good quality of different diets. This result is in agreement with Vodounnou and Tossavi (2016).

Overall, the survival in our study, including 100% replacement, remained within acceptable limits. These results indicate that replacing fishmeal with *Eisenia fetida* meal, even fully, did not harm the health or survival of Nile Tilapia fingerlings. The improved survival in 50% may reflect an optimal nutrient balance at this replacement level.

3.5. Limitations of the study

- Direct measurement of apparent digestibility coefficients (ADC) for REWM vs fishmeal, especially for amino acids and for crude protein, is needed to more precisely evaluate the limitations.

- If certain essential amino acids are limiting, feed formulation might require supplementation to get near equivalence to fishmeal in high replacement diets.
- Methods such as defatting earthworm biomass, partial de-chitinization, or processing to reduce chitin may allow higher replacement levels without negative effects.

Overall, our findings strongly support the concept that partial replacement of fishmeal with red earthworm meal, and vermicomposting of invasive aquatic biomass + manure is a viable component of a circular and sustainable aquaculture and environmental management system. The results support the feasibility of converting invasive water hyacinth biomass into value-added vermicompost while maintaining earthworm viability. With careful optimization, especially in substrate mixing, feed formulation, chitin management and scale, it is possible to increase the utility and impact of such systems.

3.6. Conclusion

The study highlights the potential of integrating invasive plant biomass into vermicomposting systems as a sustainable method for both organic waste management and invasive species control. Among the treatments evaluated Sludge showed the highest vermicompost productivity (72.2%), indicating the effectiveness of nutrient-rich and microbial active substrates. Similarly, water hyacinth root supplemented cow manure also demonstrated high productivity (64.3%), suggesting that such biomass can serve as a valuable carbon source when appropriately balanced with nitrogen-rich materials like cow manure.

Simultaneous use of earthworm derived from vermicomposting systems represents a promising and sustainable alternative to conventional fishmeal in aqua feed. Earthworms such as *Eisenia fetida* are rich in high-quality protein, essential amino acids, lipids, and micronutrients, making them nutritionally comparable to fishmeal. Additionally, rearing earthworms on organic waste such as cow manure, sludge, and invasive plant residues not only supports waste recycling but also reduces the environmental pressure on wild fish stocks for being used for fishmeal production. Empirical evidence indicates that a 50% replacement level of fishmeal with earthworm meal constitutes the optimal substitution threshold, balancing nutritional adequacy, growth performance, and feed conversion efficiency in aquaculture species such as tilapia. This level of replacement not only maintains or enhances fish survival and health but also amplifies ecological benefits by promoting resource circularity, reducing dependence on finite marine protein sources, and valorizing invasive plant biomass. Consequently, the incorporation of earthworm meal into aqua feeds supports both sustainable aquaculture development and the advancement of circular bioeconomy frameworks.

Based on the findings, two key recommendations are proposed for future research. First, further studies should investigate large-scale vermicomposting systems using water hyacinth and other invasive aquatic plants, focusing on optimizing decomposition efficiency, nutrient enrichment, and ensuring the safety of the resulting compost for agricultural use. Second, additional research is needed to evaluate the long-term effects of partially or fully replacing conventional fishmeal with red worm meal in aqua feeds, particularly regarding fish growth performance, reproduction, immune response, and disease resistance across different species and culture conditions.

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Conflict of interest

The author declared no conflict of interest associated with this work.

Data availability statement

Data will be made available upon request.

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