Cleaner and Circular Bioeconomy 15 (2026) 100229

Contents lists available at ScienceDirect

Cleaner and Circular Bioeconomy

journal homepage: www.elsevier.com/locate/clcb

# Biochar prepared from desalinated seaweed (*Gracilariopsis funicularis*) has a positive potential on cattle manure-waste paper vermidegradation and plant nutrient value

Elia N.M. Ruben [a,b,*], Simeon S. Hamukoshi [c,d], Bethold Handura [a], Hupenyu A. Mupambwa [a]

[a] Sam Nujoma Marine and Coastal Resources Research Centre, Sam Nujoma Campus, University of Namibia, P. Bag, 462, Henties Bay, Namibia
[b] Department of Crop Production and Agricultural Technologies, Ogongo Campus, University of Namibia, P. Bag 5507, Oshakati, Namibia
[c] Namibia Green Hydrogen Research Institute, University of Namibia, P. Bag 13301, Windhoek, Namibia
[d] National Commission on Research, Science and Technology, P. Bag 13253, Olympia, Windhoek, Namibia

ARTICLE INFO

*Keywords:*
Seaweed biochar
Nutrient retention
Vermicompost maturity
Phytotoxicity
Extractable nutrients

ABSTRACT

Seaweed biochar-amended vermicompost (VC) has the potential to improve the physicochemical properties of growing media for horticultural crop production. This study evaluated the agronomic suitability of cattle-manure-waste paper vermicompost amended with varying proportions of desalinated seaweed (*Gracilariopsis funicularis*) biomass-derived biochar (BC) at 0, 3, 6, and 9% (w/w). BC was prepared from desalinated seaweed biomass by soaking in deionised water at 25 °C for 6 h and incorporated into cattle manure prior to vermicomposting for 12 weeks, with sampling at 0, 4, 8, and 12 weeks. BC incorporation rate, vermicomposting time, and/or their interaction significantly ($P < 0.05$) influenced EC, nutrient dynamics, and heavy metal concentrations, whereas pH and Ca were not affected. Mature BC-amended vermicompost extracts (week 12) were evaluated for phytotoxicity using seed germination bioassays on tomato, cabbage, onion, Swiss chard, and carrot, with deionised water as the control. After 12 weeks carbon to nitrogen ratio (C/N) ranged from 11.43 to 17.60, EC (2.41–2.77 mS/cm), Olsen P (262.14–281.00 mg/kg), nitrate–nitrite nitrogen ($NO_3/NO_2$—N) (10.04–54.34 mg/kg), across treatments. All cation values increased over time, except for Ca at 9% seaweed biochar. All measured heavy metals (Cd, Cr, Cu, Ni, and Zn) were below the maximum permissible limits for biosolids soil applications after 12 weeks. For phytotoxicity, the Germination Index (GI) was improved in all selected vegetables under BC addition, except for cabbage which was reduced to 31.64 at 9% BC. Seaweed biochar at 9% rate showed most favourable results, slightly alkaline pH (7.85) and moderate EC (2.70 mS/cm), lowest C/N ratio (11.43), highest macronutrient (Olsen P; 281.00 mg/kg, exchangeable K; 8.90 g/kg, moderate N-forms), and the lowest Na concentration (3.44 g/kg). These findings suggest that this seaweed biochar-amended vermicompost may be suitable for ameliorating acidic growing media. Further evaluation of seaweed biochar-amended vermicompost in soilless media is recommended.

## 1. Introduction

Global population is projected to reach 9.8 billion by 2050 and 11.2 billion by the end of the 21st century, increasing the demand for food (United Nations Department of Economic and Social Affairs UN DESA, 2017). Meeting this demand requires intensifying crop production while maintaining environmental sustainability (Sands, 2023; Ajibade et al., 2023). Crop productivity is strongly influenced by nutrient availability, particularly fertilizer inputs and water management; therefore, strategies that improve nutrient-use efficiency are essential for sustainable agricultural systems (Xing and Wang, 2024). Among the potential alternatives, soil amendments such as manure, fly ash, rock phosphates, biochar, and compost are promising strategies for enhancing nutrient management, improving crop yields, and reducing dependence on chemical fertilizers (Ajibade et al., 2022; Mupambwa et al., 2024).

In contrast, manure use is limited by the slow release of macro-elements due to its slow degradation (Priya et al., 2024). Similarly, fly ash and rock phosphate do not supply complete range of essential nutrients, while biochar, although beneficial for improving nutrient and

* Corresponding author.
*E-mail address:* rubenelia6@gmail.com (E.N.M. Ruben).

https://doi.org/10.1016/j.clcb.2026.100229
Received 2 December 2025; Received in revised form 4 June 2026; Accepted 14 August 2026
Available online 15 August 2026
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water retention, generally contains lower nutrient concentrations (Ajibade et al., 2022; Mupambwa et al., 2024). Among these amendments, vermicompost, a product of the degradation of organic matter by earthworms, has gained attention due to its elevated micro- and macronutrient composition and ability to improve the physicochemical properties of growing media (Saha et al., 2022). Nevertheless, the application of this organic amendment is constrained by its susceptibility to nutrient leaching, particularly the nitrates and nitrites, which are available in water-soluble forms (Messiga et al., 2020).

There is a need for a technology that can improve the nutrient dynamics of vermicompost and water retention to optimize its agricultural suitability. Biochar, a black carbon-rich product resulting from the pyrolysis of organic biomass, is a promising amendment to improve the quality of vermicompost by addressing nutrient leaching issues and enhancing its properties (Antileo-Mellado et al., 2024). While biochar is often produced from agricultural and other organic waste residues (Manikandan et al., 2023), in arid countries such as Namibia, the availability of terrestrial residues is limited and may not support large-scale production without competing with other uses (Mupambwa et al., 2024). This highlights the importance of exploring alternative, locally abundant feedstocks, such as marine macroalgae, to ensure sustainable biochar production and avoid over-reliance on terrestrial biomass.

Interestingly, Namibia is blessed with the Atlantic Ocean, which is powered by a strong Benguela Current, making it productive and full of resources such as seaweeds, including *G. funicularis*, L. *pallida*, and *Macrocystis pyrifera* (Harb and Chow, 2022; Moloto et al., 2023). These seaweed species are abundant on the Namibian coast, which is influenced by the high nutrient levels, making seaweeds fast-replenishable resources (Katakula et al., 2020). Therefore, owing to their beneficial properties, seaweeds can serve as sustainable biochar feedstocks in countries like Namibia, helping to improve vermicompost quality and reduce the problem of seaweed accumulation on beaches (Dang et al., 2023). In addition to their abundance, seaweed-derived biochar is rich in nutrients, and when integrated into vermicompost, it is likely to address the nutritional management issues by generating an amendment with improved physical and chemical qualities (Cárdenas-Aguiar et al., 2024; Jia et al., 2022).

Various studies have investigated the co-application of biochar and vermicompost, many of which have focused on terrestrial biomass-based biochar, highlighting its impact on nutrient dynamics and crop performance (Antileo-Mellado et al., 2024; Zulfiqar et al., 2021). Others have investigated the standalone impacts of individual amendments, highlighting their benefits and drawbacks (Huang et al., 2024), while limited studies have explored the integration of biochar, particularly that from seaweed, in vermicomposting technologies. According to Katakula et al., 2020, seaweed-biomass-derived biochar has excessively high salt concentrations, making the pretreatment of biomass prior to biochar production a crucial step. Elevated salinity reduces the suitability of seaweed biochar for use as an amendment in vermicompost because it can negatively affect earthworm activity and compromise the compost quality (Katakula et al., 2022). In addition, research has shown that desalinating *G. funicularis*-derived seaweed biochar through soaking its biomass in cold water for 6 h can effectively reduce the salt concentration of resulting biochar (Ruben et al., 2025). In contrast, despite the increasing interest in seaweed biochar-vermicompost combinations (Katakula et al., 2022; Mupambwa et al., 2024), the potential of desalinated seaweed-derived biochar, particularly from *Gracilariopsis funicularis*, remains largely unexplored. This knowledge gap limits understanding of how seaweed-based biochar can enhance the nutrient retention and agronomic properties of cattle manure-waste paper vermicompost.

Therefore, this study sought to fill this gap by integrating desalinated *G. funicularis*-derived biochar with cattle manure-waste paper vermicompost and assessing its effects on nutrient dynamics and application optimization for improved agronomic use. The objectives of this study were as follows: (i) to evaluate the effects of different inclusion levels of desalinated *G. funicularis* biomass-derived biochar (BC) in cattle manure-waste paper vermicompost on the physicochemical characteristics and nutrient dynamics of the resulting vermicompost (VC); and (ii) to determine the phytotoxicity of the seaweed biochar-amended vermicompost using selected vegetable species (tomato, onion, carrot, Swiss chard, and cabbage).

## 2. Materials and methods

### 2.1. Study site and source of materials

The study was conducted at the University of Namibia, Sam Nujoma Campus, in the Erongo region of Namibia (14°30′19″E, 22°55′27″S). *G. funicularis* seaweed biomass was collected from the Henties Bay Coast in Namibia. Seaweed biomass sampling was conducted randomly along accessible intertidal zones within the coastal stretch. Deionized water used in this study was collected from a water-deionizing machine at the University of Namibia, Sam Nujoma Campus. The seaweed biomass was soaked in deionized water for 6 h to reduce salinity, and then oven-dried at 70 °C for 48 h as indicated by Ruben et al. (2025). The biomass was then converted to seaweed biochar through pyrolysis using a muffle furnace at 400 °C for 1 h, as recommended by Katakula et al., 2020. Sterile metal cylindrical cans (5 L) were used for seaweed biomass confinement during BC preparation. After pyrolysis, the seaweed biochar was ground into a fine powder using a mortar and pestle to increase surface area and ensure uniform mixing with the vermicompost. Cattle manure was sourced from local agri-shop in Swakopmund, and waste paper was collected from the Sam Nujoma Campus and shredded using an office paper shredder. The physicochemical characteristics of raw cattle manure and seaweed biochar used in this study are presented in Table 1. The earthworm species used in this study was *Eisenia fetida* and these were purchased from a local earthworm supplier. The vermicomposting process was carried out in rectangular vermi-bins (34 cm width × 60 cm length × 18.5 cm height). The overall workflow of seaweed biochar production and seaweed biochar-amended vermicompost preparation is summarized in a schematic diagram (Fig. 1). The seaweed biochar and BC-amended vermicompost used in this study, were prepared by the authors as described above. The vegetable seeds and Petri dishes used in the phytotoxicity test were purchased from a local hardware shop in Swakopmund, Namibia.

**Table 1**
Selected physicochemical characteristics of cattle manure and seaweed (G. funicularis) biochar used in this study before vermidegradation.

| Parameter | Seaweed biochar (BC) | cattle manure (CM) |
|---|---|---|
| pH | 8.75 ± 0.15 | 7.72 ± 0.06 |
| EC (mS/cm) | 5.40 ± 0.08 | 4.72 ± 0.03 |
| Total C (%) | 43.74 ± 2.00 | 30.18 |
| Total N (%) | 5.17 ± 0.05 | 1.65 |
| $NH_4$–N (mg/kg) | 14.17 ± 4.58 | 146.66 ± 19.93 |
| $NO_3/NO_2$–N (mg/kg) | 13.72 ± 0.72 | 88.63 ± 17.54 |
| Extractable P (mg/kg) | 244.74 ± 17.00 | 850.63 ± 7.59 |
| Ca (g/kg) | 1.95 ± 0.21 | 6.04 ± 0.27 |
| Mg (g/kg) | 0.17 ± 0.02 | 1.26 ± 0.10 |
| K (g/kg) | 21.29 ± 1.61 | 8.27 ± 0.28 |
| Na (g/kg) | 2.14 ± 0.16 | 3.80 ± 0.32 |
| Cr (mg/kg) | < 0.01 | 1.59 ± 0.04 |
| Cd (mg/kg) | < 0.01 | < 0.01 |
| Zn (mg/kg) | 3.49 ± 0.33 | 1.75 ± 0.11 |
| Cu (mg/kg) | < 0.01 | 1.92 ± 0.10 |
| Ni (mg/kg) | < 0.01 | < 0.01 |

**Note:** Values are presented as mean ± SD (n = 3), except for cattle manure total C and N (n = 1).

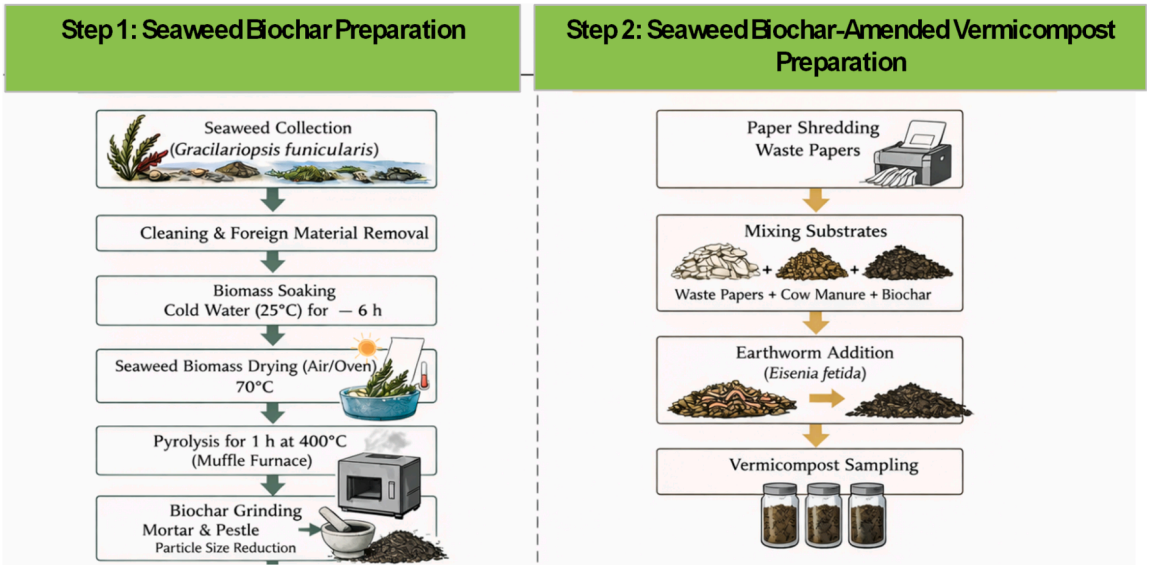


**Fig. 1.** Schematic diagram of the experimental methodology from seaweed (Gracilariopsis funicularis) collection to seaweed biochar-amended vermicompost sample collection prior to physicochemical analysis.

### 2.2. Treatments and experimental design

The vermicomposting experiment was conducted over a period of 12 weeks. The experiment used a Completely Randomized Design (CRD) with four treatments, namely, 0%, 3%, 6%, and 9% BC incorporation level on a weight-to-weight basis in cattle manure-waste paper vermicompost. The waste paper was mixed with crushed cattle manure to achieve a C:N ratio of 30:1. Incorporation levels were chosen to test a gradient of seaweed biochar addition around the levels previously recommended by Katakula et al., 2022 for nutrient enhancement, while assessing their effect on cattle manure-waste paper vermicompost quality. The seaweed biochar was added to the mixture at previously stated levels, and each treatment was replicated three times. The moistened seaweed biochar-amended vermicompost mixtures were allowed to stand for 1 week to allow volatile gases to evaporate before earthworms (*Esenia fetida*) were added at a stocking density of 25 g/kg of substrate as recommended by Mupambwa & Mnkeni (2016).

Sampling was then done at four-week interval starting from week 0 until week 12. At each time point, approximately consistent portions of seaweed biochar-amended vermicompost were collected randomly from each treatment to ensure adequate material for all planned analyses while minimizing disturbance to the remaining VC. The collected samples were oven-dried at 65 °C for 24 h, to remove excess moisture while preserving sensitive compounds in the seaweed biochar-amended vermicompost. The seaweed biochar-amended vermicompost samples were crushed using a mechanical grinder equipped with a 2 mm sieve, before analysis. Samples were handled independently to avoid cross-contamination at time of sample analysis and experimental setup.

### 2.3. Seaweed biochar-amended vermicompost sample analysis

#### 2.3.1. pH and electrical conductivity (EC)

Each seaweed biochar-amended vermicompost sample (5 g) was thoroughly mixed with deionized water at a ratio of 1:10 (w/v) and shaken on a reciprocating shaker at 120 rpm for 30 min. The pH and EC were measured using a multimeter with a glass electrode (HQ40d, Aqua Services & Engineering, Germany). Prior to measurement, the pH probe was calibrated using standard buffer solutions of pH 4.0 and 7.0, while the EC probe was calibrated with a sodium chloride (NaCl) standard solution of 1000 ± 10 μS/cm. The pH and EC were measured by placing multimeter probes in the solution (Agri Laboratory Association of Southern Africa AgriLASA, 2004).

#### 2.3.2. Total carbon and nitrogen

The total carbon and nitrogen contents were quantified using the dry combustion method with a Vario EL Cube Elemental Analyser (Elementar, Hanau, Germany). The analyser was calibrated using a certified reference material, specifically sulfamethazine, which contains 20.13% total nitrogen and 51.78% total carbon. The detection limits for the analyser were determined to be 0.01% for nitrogen and 0.25% for carbon. To conduct the analysis, a 0.1 g seaweed biochar-amended vermicompost sample was precisely weighed into an aluminum foil boat, along with approximately 5 mg of Tungsten Oxide ($WO_3$) powder additive, using an analytical balance. The aluminum foil boat containing the weighed seaweed biochar-amended vermicompost sample was then placed inside an operational Vario EL Cube Elemental Analyser, which automatically determined the total carbon and nitrogen contents in the sample.

#### 2.3.3. Extractable ammonium, nitrate and nitrite

Extractable ammonium nitrate and nitrite were extracted using 0.5 M potassium sulfate and analysed calorimetrically, as described by Okalebo et al., 2002. To extract ammonium nitrate and nitrite, a 5 g seaweed biochar-amended vermicompost sample was mixed with 50 mL of 0.5 M potassium sulfate and shaken for 1 h using a mechanical reciprocal shaker at 180 rpm. The extract was filtered through the same grade of filter paper (Whatman No 2). Colorimetric determination of $NH_4^+$-N and $NO_2/NO_3$-N in the extracts was performed following the procedures described by Okalebo et al., 2002. Briefly, for $NH_4^+$-N, the filtered extract was reacted accordingly with N1 and N2 reagents. N1 reagent was prepared by mixing sodium salicylate, sodium citrate, sodium tartrate, and sodium nitroprusside, whereas N2 reagent was prepared by mixing sodium hydroxide and sodium hypochlorite. The resulting blue color intensity was measured at 655 nm using a UV–Vis spectrometer (Genesys 10S, Thermo Fisher Scientific, USA), calibrated with standards ranging from 0 to 25 μg $NH_4^+$-N $mL^{-1}$. For $NO_2/NO_3$-N determination, the extract was reacted with salicylic acid and NaOH solutions according to Okalebo et al., 2002, and absorbance was read at 419 nm using the same spectrometer (Genesys 10S, Thermo Fisher Scientific, USA calibrated with standards ranging from 0 to 10 μg $NO_3$-N $mL^{-1}$.

#### 2.3.4. Extractable P

Extractable phosphorus was extracted using the Olsen method, and phosphorus was analysed using the ascorbic acid method (Kuo, 1996). All reagents used in the study were analytical reagent grade supplied by a local supplier in Namibia. To extract P, 5 g of seaweed biochar-amended vermicompost was shaken with 50 mL of 0.5 M sodium hydrogen carbonate in an extraction bottle on a reciprocal shaker at 180 rpm for 30 min (Schoenau and O' Halloran, 2008). Whatman No 2 filter paper was used to filter the extract, and the ascorbic acid method described by Kuo (1996) was used to analyse the P in the extracts. The ascorbic acid method involved the development of a blue color and the measurement of absorbance at a wavelength of 880 nm using a UV–Vis spectrometer (Genesys 10S UV–Vis, Thermo Fisher Scientific, USA)(Kuo, 1996).

#### 2.3.5. Extractable cations

Cations (Ca, Mg, Na, and K) were extracted using the ammonium acetate method (Agri Laboratory Association of Southern Africa AgriLASA, 2004). A 5 g seaweed biochar-amended vermicompost sample was extracted with 1 M ammonium acetate that had been adjusted to pH 7.0 using 1 M NaOH. The mixture was shaken on a reciprocating shaker at 180 rpm for 30 min, and the same grade of filter paper was used to out filter the extracts, which were analysed using an Atomic Absorption Spectrometer (AAS) (PinAAcle™ 900F, PerkinElmer Inc., USA). For quality assurance, certified standard solutions were used for calibration, and analytical blanks were included to detect possible contamination. Quality control (QC) standards were analysed after every ten samples to verify analytical precision and accuracy.

#### 2.3.6. Total concentrations of heavy metals

Heavy metals were analysed in the seaweed biochar-amended vermicompost to assess its suitability for application in soil and other growing media. To measure the total concentrations of heavy metals (Cd, Pb, Cr, As, Zn, and Ni), the seaweed biochar-amended vermicompost samples were digested using the wet digestion method described by Agri Laboratory Association of Southern Africa AgriLASA, 2004. A 0.5 g sample of seaweed biochar-amended vermicompost was digested with 10 mL of nitric acid (60%), heated at 150 °C for 30 min, before adding 4 mL of perchloric acid (65%), and heated again for 30 min at the same temperature. After the digestion process, the mixture was allowed to cool before being diluted in a 100 mL volumetric flask. The Whatman No 2 filter paper was used to filter the diluted solution, which was then analysed using a calibrated AAS (PinAAcle™ 900F, PerkinElmer Inc., USA). Quality assurance followed the same protocol used for cations analysis.

### 2.4. Phytotoxicity test

Seed germination bioassays were conducted to assess phytotoxicity, as they are a sensitive and rapid screening tool to detect potential inhibitory effects of seaweed biochar-amended vermicompost on plant growth (Ravindran et al., 2017; Duraisamy et al., 2021). However, these tests only assess early seedling growth and may not capture long-term or cumulative effects on plant development, highlighting their limitations. The phytotoxicity test was performed using matured seaweed biochar-amended vermicompost extract (at week 12), using the same treatments and deionized water used as a control, tested on five different vegetables, namely, tomatoes (*Solanum lycopersicum*), onions (*Allium cepa*), cabbage (*Brassica oleracea* var. *capitata)*, carrots (*Daucus carota*), and Swiss chard (*Beta vulgaris* subsp. *Cicla*). The extract was prepared by mixing seaweed biochar-amended vermicompost with deionized water at a ratio of 10:1 (solid-to-water ratio of 10:1) (Ravindran et al., 2017). The mixture was then shaken at 200 rpm for 1 h using a mechanical shaker, and the extract was filtered through Whatman No 2 filter paper. Seed germination was assessed following the bioassay protocol originally established by Tiquia and Tam (1998). In brief, two pieces of Whatman no.2 filter paper were placed inside a sterilized Petri dish, and 10 seeds for each vegetable were placed in each Petri dish. The seeds were moistened with the seaweed biochar-amended vermicompost extract, while the control treatment received deionized water only. Following seven days incubation period in a dark environment, the number of seeds that germinated and their root length were recorded. Relative Seed Germination (RSG), Relative Root Elongation (RRE), and Germination Index (GI) were calculated using Eq. (1), Eq. (2) and Eq. (3), respectively.

$$RSG\ (\%) = \frac{\text{Number of seeds germinated in the sample extract}}{\text{Number of seeds germinated in the control}} \times\ 100 \quad (1)$$

$$RRE\ (\%) = \frac{\text{Mean root elongation in the sample extract}}{\text{Mean root elongation in the control}} \times\ 100 \quad (2)$$

$$GI\ (\%) = \frac{\%\ \text{Seed germination}\ \times\ \%\ \text{Root elongation}}{100} \quad (3)$$

### 2.5. Data analysis

Sampling was not destructive in this study, and data on the chemical properties of seaweed biochar-amended vermicompost were analysed using repeated measures Analysis of Variance (ANOVA), using JMP statistical software student version 18.2.2 (SAS Institute, Inc.). Where sphericity assumptions could not be met, the Greenhouse-Geisser correction of *P* was used. For the phytotoxicity parameters, statistical analyses were conducted separately for each crop species using JMP statistical software version (JMP student edition version 18.2.2). For each species, a one-way ANOVA was applied with seaweed biochar inclusion level as the sole treatment factor. No combined or factorial model including crop species was used. Any comparisons between crops mentioned in the results are qualitative descriptions of differences in response, not outcomes of a joint statistical analysis. Mean comparisons were performed using the Least Squares Means Differences Student's *t*-test in JMP Statistical Software Student Edition, version 18.2.2. Where ANOVA indicated significant differences, post hoc tests were conducted using a student's T- test. All statistical tests were conducted at a significance level of $\alpha = 0.05$. Microsoft Excel was used to draw tables, while GraphPad Prism version 10.2.2 for Windows (GraphPad Software, www.graphpad.com) were used to plot graphs.

## 3. Results

The results presented in this study reflect changes observed over a 12-week vermicomposting period, with measurements taken at four sampling intervals (weeks 0, 4, 8, and 12).

### 3.1. Effect of BC on pH and electrical conductivity (EC) of seaweed biochar-amended vermicompost

BC incorporation level had no significant effects ($P > 0.05$) on changes in seaweed biochar-amended vermicompost pH (Table 2). Furthermore, the pH of the seaweed biochar-amended vermicompost did not change significantly over the 12-week vermicomposting period (Table 2). The pH values remained within an alkaline range of 7.72 to 8.20 throughout the 12-week period (Fig. 2A). The control (0% BC) showed non-significant increasing trend over time, increasing from 7.87 at week 0, to 8.21 at week 12 (Fig. 2A). Although not statistically significant ($P > 0.05$), seaweed biochar-amended treatments showed a slight numerical decrease in pH over time (Fig. 2A). The 3% BC treatment exhibited a consistent decrease from a pH of 8.05 to 7.77, while the 6% and 9% BC treatments also followed a downward trend from a pH of 8.2 and 8.14, respectively, both reaching a pH of 7.85 at week 12 (Fig. 2A). After 12 weeks, pH values ranged between 7.77 and 8.21, with

**Table 2**
Repeated measures Analysis of Variance (ANOVA) of various chemical properties of Seaweed biochar-amended vermicompost.

| Source of variation | | | | | | |
|---|---|---|---|---|---|---|
| Parameters | Between Subjects | | Within subjects | | | |
| | Seaweed biochar level (BC) | | Time (T) | | BC × T | |
| | *F-value* | *P-value* | *F-value* | *P-value* | *F-value* | *P-value* |
| pH | 0.6372 | ns | 3.4747 | ns | 2.5548 | ns |
| EC (mS/cm) | 1.3548 | ns | 73.6818 | <.0001 | 2.3987 | ns |
| C/N ratio | 22.5147 | 0.0012 | 82.885 | <.0001 | 6.6006 | 0.0032 |
| $NH_4$—N (mg/kg) | 25.5538 | 0.0008 | 208.108 | <.0001 | 31.8464 | <.0001 |
| $NO_3$/$NO_2$—N (mg/kg) | 2.8135 | ns | 2.2788 | ns | 18.8015 | <.0001 |
| Extractable P (mg/kg) | 12.6419 | 0.0053 | 6.1753 | 0.0092 | 1.88 | ns |
| Ca (g/kg) | 2.1535 | ns | 3.2144 | ns | 1.6699 | ns |
| Mg (g/kg) | 3.2968 | ns | 30.5130 | 0.0003 | 1.588 | ns |
| K (g/kg) | 23.0628 | 0.0011 | 13.4141 | 0.0004 | 1.742 | ns |
| Na (g/kg) | 2.3352 | ns | 25.6562 | 0.0002 | 1.7471 | ns |
| Cr (mg/kg) | 5.5277 | 0.0367 | 23.3146 | 0.0002 | 3.5499 | 0.0381 |
| Cd (mg/kg) | 10.4016 | 0.0086 | 25.365 | 0.0011 | 1.6541 | ns |
| Zn (mg/kg) | 14.1953 | 0.0039 | 32.587 | 0.0002 | 1.1484 | ns |
| Cu (mg/kg) | 4.8868 | 0.0474 | 56.1668 | <.0001 | 4.9293 | 0.0097 |

**Note:** ns= not significant (*P-value>0.05*).

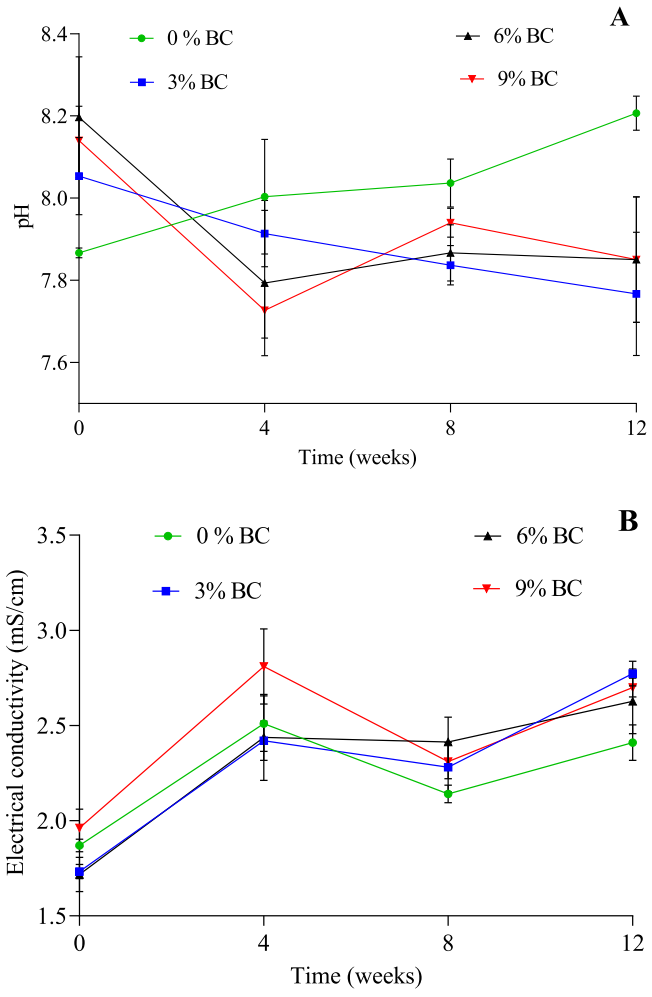


**Fig. 2.** Effect of different rates of *G. funicularis* seaweed-derived biochar amendment on pH (A) and EC (B) in cattle manure-waste paper vermicompost over 12-week vermicomposting period. Error bars indicate standard deviation.

the control maintaining the highest value and 3% BC the lowest (Fig. 2A).

There were no significant influences of seaweed biochar incorporation level on EC; however, EC increased significantly over time ($P < 0.05$), whereas seaweed biochar level had no significant effect (Table 2). Irrespective of the seaweed biochar level, the EC values increased over time, with moderate fluctuations between week 4 and week 8 (Fig. 2B). At 0% BC, EC rose from 1.87 mS/cm at week 0 to 2.41 mS/cm at week 12 (Fig. 2B). A similar trend was observed in the seaweed biochar-amended treatments, although the magnitude of change varied slightly (Fig. 2B). The inclusion of 3% BC increased the EC from 1.73 mS/cm at week 0 to 2.77 mS/cm at week 12. Similarly, the EC in 6% BC and 9% BC treatments increased to 2.63 mS/cm and 2.70 mS/cm after 12 weeks, from a value 1.73 mS/cm and 1.96 mS/cm, respectively (Fig. 2B). Across all treatments, the EC values after 12 weeks ranged from 2.41 mS/cm to 2.77 mS/cm with the highest value observed in 3% BC-amended vermicompost, followed by 9% BC > 6% BC > 0% BC (Fig. 2B).

### *3.2. Effect of BC on the C/N ratio of seaweed biochar-amended vermicompost*

There were significant effects ($P < 0.05$) of seaweed biochar inclusion level and time on the C/N ratio, with a significant interaction indicating that the effect of seaweed biochar depended on sampling time (Table 2). The C/N ratio decreased over time across all treatments, with lower C/N ratios observed at higher seaweed biochar incorporation rates at the same sampling times (Fig. 3). In the control (0% BC), the C/N ratio decreased from 47.66 at week 0 to 17.60 at week 12 (Fig. 3). Similar patterns were observed in the BC-amended treatments, whereby the 3% BC treatment showed a consistent decline from 38.40 at week 0 to 13.11 at week 12 (Fig. 3). In the 6% BC treatment, the C/N ratio decreased from 30.98 at week 0 to 12.33 at week 12 (Fig. 3). The lowest C/N values were recorded in the 9% BC treatment, which decreased from 15.04 at week 0 to 11.43 at 12 weeks (Fig. 3). After 12 weeks, C/N ratios ranged from 11.43 to 17.60, with the lowest values being recorded in 9% BC level and the highest in 0% BC level (Fig. 3).

### *3.3. Effect of BC on extractable N and P in seaweed biochar-amended vermicompost*

There were significant effects ($P < 0.05$) of seaweed biochar inclusion level on the extractable ammonium ($NH_4$—N) of seaweed biochar-amended vermicompost over the 12 weeks period (Table 2). Furthermore, there was a significant two-way interaction ($P < 0.05$) between

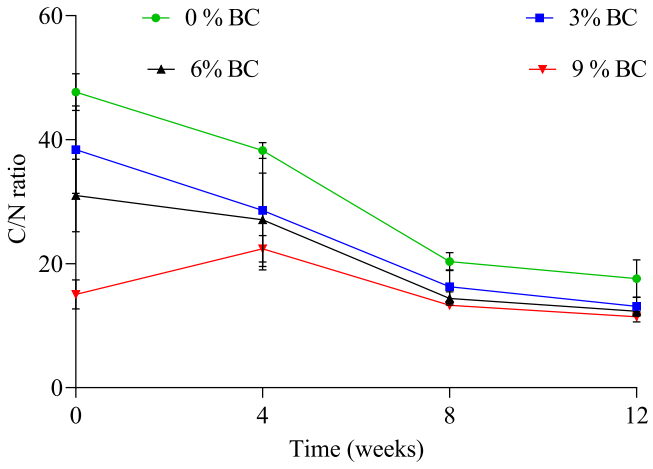


**Fig. 3.** Effect of different rates of *G. funicularis* seaweed-derived biochar amendment on C/N ratio in cattle manure-waste paper vermicompost over 12-week vermicomposting period. Error bars indicate standard deviation.

time and seaweed biochar level (Table 2). Generally, $NH_4$—N concentrations showed variable patterns across treatments and over time (Fig. 4A). In the 0% BC treatment, $NH_4$—N exhibited an increasing trend over time, starting from 29.38 mg/kg at week 0 to 151.32 mg/kg at week 12 (Fig. 4A). In the seaweed biochar-amended treatments, $NH_4$—N levels rose sharply during the early stages, reaching the highest concentrations at week 4 or week 8, before declining toward week 12 (Fig. 4A).The 3% BC treatment showed the highest $NH_4$—N concentration which was 195.81 mg/kg, followed by 9% BC with 174.23 mg/kg and 6% BC with 148.17 mg/kg at week 8 (Fig. 4A). After 12 weeks of vermicomposting, $NH_4$—N concentrations ranged from 88.69 mg/kg to 151.32 mg/kg, with the highest $NH_4$—N concentration recorded in the 0% BC treatment, followed by 6% BC > 9% BC >3% BC (Fig. 4A).

There was a significant two-way interaction ($P < 0.05$) between seaweed biochar level and sampling time on extractable nitrate and nitrite concentrations ($NO_3/NO_2$—N) in seaweed biochar-amended vermicompost (Table 2). This indicates that $NO_3/NO_2$-N dynamics depended on both seaweed biochar level and vermicomposting stage. $NO_3/NO_2$-N concentrations showed varied trends across treatments, with the control (0% BC) exhibiting a consistent increase over time, whereas the 9% BC treatment showed a decreasing trend (Fig. 4B). In contrast, the 3% and 6% BC treatments displayed no consistent pattern throughout the vermicomposting period (Fig. 4B). In the control, $NO_3/NO_2$-N concentration values increased from 3.55 mg/kg at week 0 to 54.34 mg/kg at week 12 (Fig. 4B). In contrast, in the 3% BC treatment, the highest $NO_3/NO_2$-N concentration was at week 8 with 29.83 mg/kg and the lowest was observed after 12 weeks with 10.04 mg/kg (Fig. 4B). In the 6% BC treatment, $NO_3/NO_2$-N concentrations rose from 17.60 mg/kg at week 0, with minor fluctuations between week 4 and week 8, before reaching a value of 32.37 mg/kg after 12 weeks (Fig. 4B). The $NO_3/NO_2$-N concentration in the 9% BC treatment ranged from 48.91 mg/kg at week 0 to 11.43 mg/kg at week 12 (Fig. 4B). By week 12, $NO_3/NO_2$-N concentration values ranged from 10.04 mg/kg to 54.34 mg/kg across treatments, with the highest value recorded in the control treatment, followed by 6% BC > 9% BC > 3% BC (Fig. 4B).

There were significant influences ($P < 0.05$) of seaweed biochar inclusion level on the Olsen extractable P content across the 12 weeks of vermicomposting (Table 2). Across all treatments, extractable P increased from week 0 to week 12 (Fig. 5). In the 0% BC treatment, extractable P increased from 188.71 mg/kg at week 0 to 274.91 mg/kg at week 12, with notable fluctuations between week 4 and week 8 (Fig. 5). Similarly, the 3% BC treatment showed an increase from 218.91 mg/kg to 262.14 mg/kg (Fig. 5). In the 6% BC treatment values ranged from 206.74 mg/kg at week 0 to 267.63 mg/kg after 12 weeks (Fig. 4). The 9% BC treatment maintained relatively higher extractable P concentration values throughout the vermicomposting period, increasing from 237.42 mg/kg at week 0 to 281.00 mg/kg at week 12 (Fig. 5). After 12 weeks, the extractable P concentration in the seaweed biochar-amended vermicompost followed the order 9% BC > 0% BC > 6% BC > 3%, with values ranging from 262.14 mg/kg to 281.00 mg/kg (Fig. 5).

### 3.4. *Effect of BC on exchangeable cations (Ca, Mg, K, and Na) in seaweed biochar-amended vermicompost*

There was no significant influence of seaweed biochar incorporation level and time on changes in calcium (Ca) of the seaweed biochar-amended vermicompost throughout the 12 weeks period (Table 2). Although not statistically significant, numerical fluctuations in Ca concentrations were observed across treatments over time. In the seaweed biochar-treated VC treatments, Ca values initially decreased gradually for the first 8 weeks, followed by an increase toward week 12 (Fig. 6A). In the control treatment, Ca concentrations showed an inconsistent trend across the vermicomposting period (Fig. 6A). In the 0% BC treatment, the Ca concentration ranged from 5.91 g/kg recorded at week

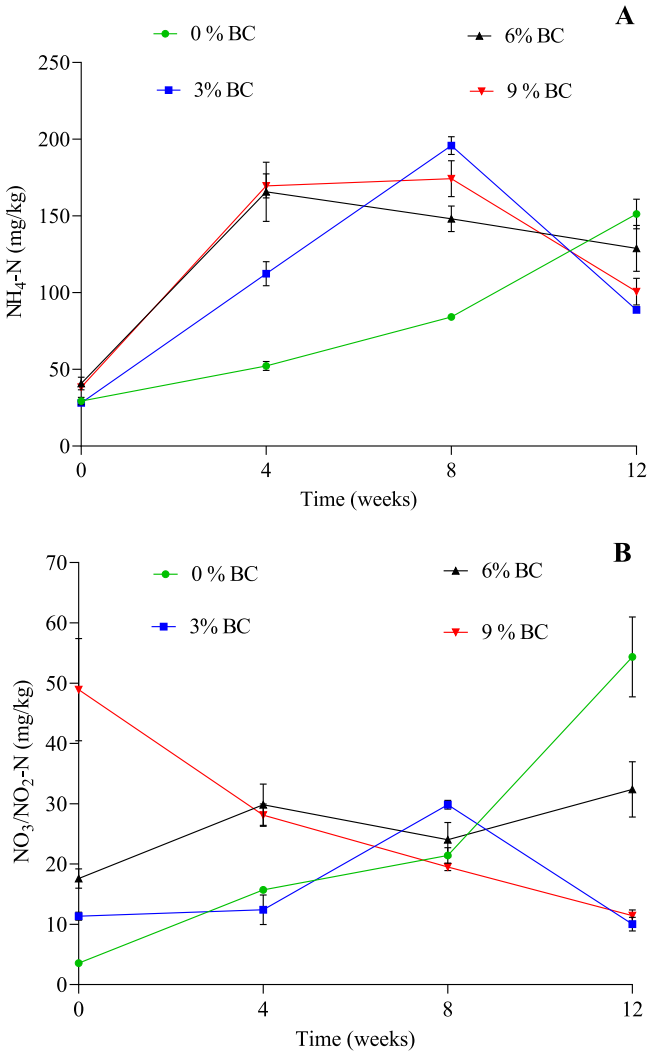


**Fig. 4.** Effect of different rates of G. funicularis seaweed-derived biochar amendment on $NH_4$—N (A) and $NO_3/NO_2$—N (B) in cattle manure-waste paper vermicompost over 12-week vermicomposting period. Error bars indicate standard deviation.

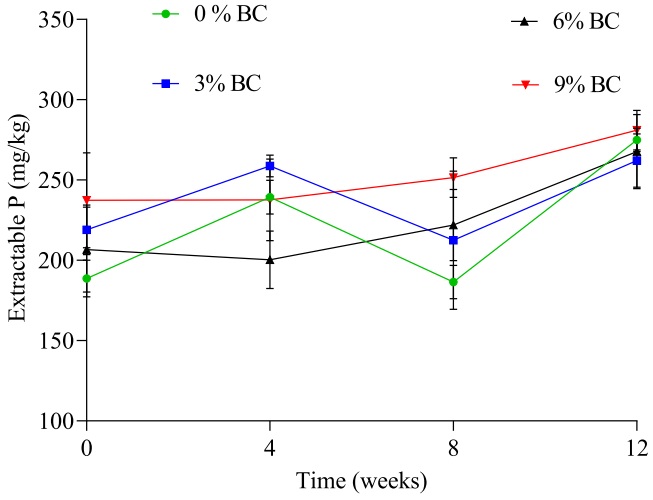


**Fig. 5.** Effect of different rates of *G. funicularis* seaweed-derived biochar amendment on extractable phosphorus in cattle manure–waste paper vermicompost over a 12-week vermicomposting period. Error bars indicate standard deviation.

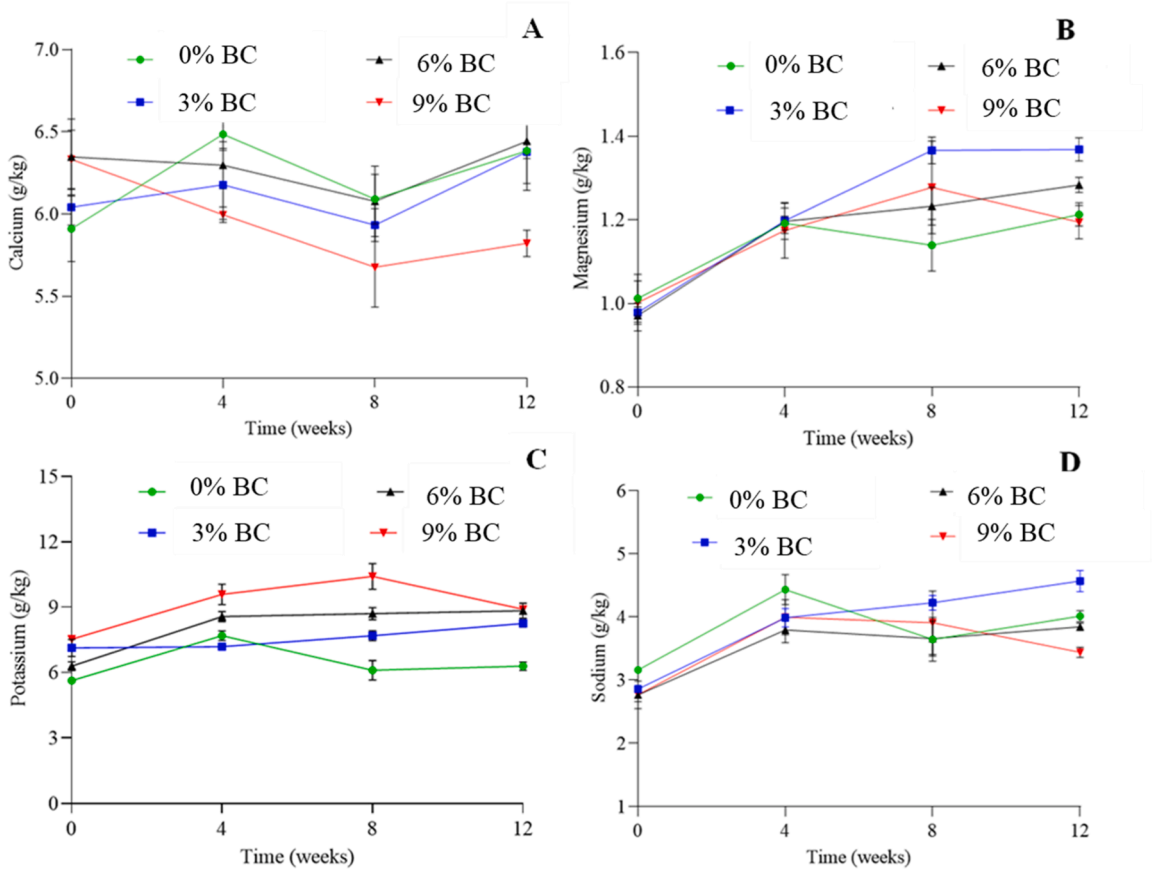


**Fig. 6.** Effect of different rates of *G. funicularis* seaweed-derived biochar amendment on Ca (A), Mg (B), K (C), Na (D) in cattle manure-waste paper vermicompost over 12-week vermicomposting period. Error bars indicate standard deviation.

0 to 6.48 g/kg recorded at week 4 (Fig. 6A). In the 3% BC inclusion treatment, Ca values decreased from 6.04 g/kg at week 0 to 5.93 g/kg at week 8, followed by a recovery to 6.38 g/kg at week 12 (Fig. 6A). Similarly, in the 6% BC treatment, values decreased from 6.33 g/kg at week 0 to 6.08 g/kg after 8 weeks, then rose to 6.44 g/kg after 12 weeks (Fig. 6A). At 9% BC incorporation rate, Ca content decreased gradually from 6.33 g/kg at week 0 to a 5.69 g/kg at week 8, followed by a slight recovery to 5.82 g/kg at week 12 (Fig. 6A). After 12 weeks of vermicomposting, the Ca concentration values ranged from 5.93 g/kg to 6.44 g/kg, with the lowest value recorded in the 9% BC treatment and the highest in the 6% BC treatment (Fig. 6A).

There were no significant influences of seaweed biochar inclusion on changes in magnesium (Mg), although there were significant differences ($P < 0.05$) in Mg concentrations over time (Table 2). In all treatments, the Mg concentrations increased over the vermicomposting period, although differences among seaweed biochar treatments were not statistically significant (Fig. 6B). At 0% BC incorporation rate, the Mg concentration in seaweed biochar-amended vermicompost increased from 1.00 g/kg at week 0 to reach 1.21 g/kg at week 12 (Fig. 6B). In the 3% BC inclusion, the lowest Mg concentration value was 0.97 g/kg recorded at week 0, while the highest value was 1.37 g/kg recorded after 12 weeks (Fig. 6B). In the 6% BC treatment the concentration values ranged from 0.97 g/kg at week 0 to 1.28 g/kg at week 12, in the 9% BC treatment values ranged from 1.00 g/kg at week 0 to 1.28 g/kg at week 8 (Fig. 6B). After 12 weeks of vermicomposting, the highest Mg concentration values was recorded at 3% BC with 1.37 g/kg, followed by 6% with 1.28 g/kg, 0% BC with 1.21 g/kg and 9% BC with lowest value of 1.19 g/kg (Fig. 6B).

There were significant influences ($P < 0.05$) of seaweed biochar incorporation level on the potassium (K) content of the seaweed biochar-amended vermicompost throughout the 12 weeks period of vermicomposting (Table 2). The potassium content showed an increasing trend over time, with lower BC incorporation rates generally maintaining lower values than higher BC incorporation levels throughout the 12 weeks of vermicomposting (Fig. 6C). In the control, the lowest potassium concentration was recorded at week 0 with a value of 5.63 g/kg, while the highest was recorded at week 4 with 7.69 g/kg (Fig. 6C). In the 3% BC treatment the K concentration values ranged from 7.13 g/kg recorded at week 0 to 8.25 g/kg at week 12, while in the 6% BC treatment, values ranged from 6.29 g/kg at week 0 to 8.84 g/kg at week 12 (Fig. 6C). At the highest BC incorporation rate (9% BC), potassium concentration rose from 7.53 g/kg at week 0 to a peak at week 8 with 10.42 g/kg (Fig. 6C). At week 12, the highest concentration was recorded 9% BC with 8.90 g/kg, followed by 6% with 8.84 g/kg, 3% with 8.25 g/kg and control with the lowest value of 6.29 g/kg (Fig. 6C).

There were no significant effects of seaweed biochar level on changes in sodium (Na) concentrations in the seaweed biochar-amended vermicompost, although there were significant differences ($P < 0.05$) in Na content throughout the 12 weeks period (Table 2). The sodium concentrations in the seaweed biochar-amended vermicompost generally increased toward week 12, although differences among seaweed biochar treatments were insignificant (Fig. 6D). In the control, the lowest Na concentration value was recorded at week 0 with 3.16 g/kg, while the highest value was recorded after 12 weeks with 4.43 g/kg (Fig. 6D). In the 3% BC treatment the Na concentration values ranged from 2.86 g/kg at week to 4.57 g/kg after 12 weeks of vermicomposting, while at 6% BC incorporation rate values ranged from 2.17 g/kg at week 0 to 3.84 g/kg at week 12 (Fig. 6D). In the 9% BC treatment, the lowest Na concentration of 2.77 g/kg was recorded at week 0, while the highest value was recorded at 3.99 g/kg (Fig. 6D). After the 12 weeks period, the Na

concentration values ranged from 3.44 g/kg to 4.57 g/kg, with treatments following the order 3% BC > 0% BC > 6% BC > 9% BC (Fig. 6D).

### 3.5. Effect of BC on heavy metal concentrations (Cr, Cd, Zn, and Cu) in seaweed biochar-amended vermicompost

There were significant influences ($P < 0.05$) of seaweed biochar incorporation rate on the chromium (Cr) content of the seaweed biochar-amended vermicompost throughout the 12 weeks period of vermicomposting (Table 2). Moreover, there was a significant two-way interaction ($P < 0.05$) between the factors on the Cr content of the seaweed biochar-amended vermicompost (Table 2). Across all treatments, fluctuated over time without a consistent trend, indicating treatment- and time-dependent variation (Fig. 7A). To start with the 0% BC treatment, over time the Cr concentration values ranged from 1.38 mg/kg to 2.08 mg/kg, with lowest value recorded at week 4 and highest was recorded at week 8 (Fig. 7A). Similarly, in the 3% BC treatment, the lowest (1.34 mg/kg) was recorded at week 4 while the highest value of 2.03 mg/kg was observed at week 8 (Fig. 7A). In contrast, at higher BC rates, the highest Cr concentration values were recorded at week 0, which were 2.06 mg/kg in the 6% BC and 1.70 mg/kg in the 9% BC treatment; however, these values dropped toward week 12 (Fig. 7A). After 12 weeks, the lowest Cr content was observed in the 9% BC treatment with 1.51 mg/kg, followed by 6% BC with 1.61 mg/kg, the control with 1.78 mg/kg, and 3% BC, which recorded the highest value of 1.80 mg/kg (Fig. 7A).

There were significant effects ($P < 0.05$) of seaweed biochar inclusion level on the Cadmium (Cd) content of seaweed biochar-amended vermicompost, as well as the significant changes in Cd content across the vermicomposting period (Table 2). Overall, the Cd concentration increased over time across all treatments, with no detectable levels (< 0.01) from week 0 to week 4 (Fig. 7B). In the 0% BC treatment, Cd concentration was only detected at week 12 with 0.17 mg/kg (Fig. 7B). In the 3% BC, Cd concentration values ranged from a 0.10 mg/kg at week 8 to 0.25 mg/kg at week 12 (Fig. 7B). Similarly, in the 6% BC treatment, Cd increased from undetectable levels to 0.22 mg/kg at week 8 and 0.24 mg/kg at week 12 (Fig. 7B). In the 9% BC treatment, Cd concentration in seaweed biochar-amended vermicompost rose from 0 at week 4 to a value of 0.20 mg/kg at week 8 and 0.23 mg/kg at week 12 (Fig. 7B). After 12 weeks, the lowest Cd content was observed in the control with 0.17 mg/kg, followed by 9% BC with 0.23 mg/kg, 6% BC with 0.24 mg/kg, and 3% BC, which recorded the highest value of 0.25 mg/kg (Fig. 7B).

There were significant influences ($P < 0.05$) of seaweed biochar incorporation rate on the Zinc (Zn) content across the 12 weeks of the vermicomposting (Table 2). Across all treatments, the Zn concentration increased over time, with higher seaweed biochar rates maintaining higher values (Fig. 7C). In the control, Zn increased from 0.45 mg/kg at week 0, reaching a value of 1.42 mg/kg at week 12 (Fig. 7C). In 3% BC treatment, Zn concentration values increased from 0.66 mg/kg at week 0 to a peak of 1.78 mg/kg at week 8, then slightly decreased to 1.60 mg/kg after 12 weeks (Fig. 7C). Similarly, in the 6% BC, Zn increased from 0.93 mg/kg at week 0 to 2.16 mg/kg at week 8, followed by a slight decline to 1.76 mg/kg at week 12 (Fig. 7C). In the 9% BC, Zn concentrations increased from 1.37 mg/kg at week 0 to 2.15 mg/kg at week 8, before decreasing to 1.69 mg/kg after week 12 (Fig. 7C). After 12 weeks, the highest Zn content was recorded in the 6% BC treatment, followed by 9% BC > 3% BC > the control (Fig. 7C).

There were significant influences ($P < 0.05$) of BC inclusion level on the copper (Cu) content of the seaweed biochar-amended vermicompost over time (Table 2). There was also a significant two-way interaction ($P < 0.05$) between time and seaweed biochar level on the Cu concentration (Table 2). Cu concentrations increased over time in all treatments, although the pattern varied among the seaweed biochar levels at each

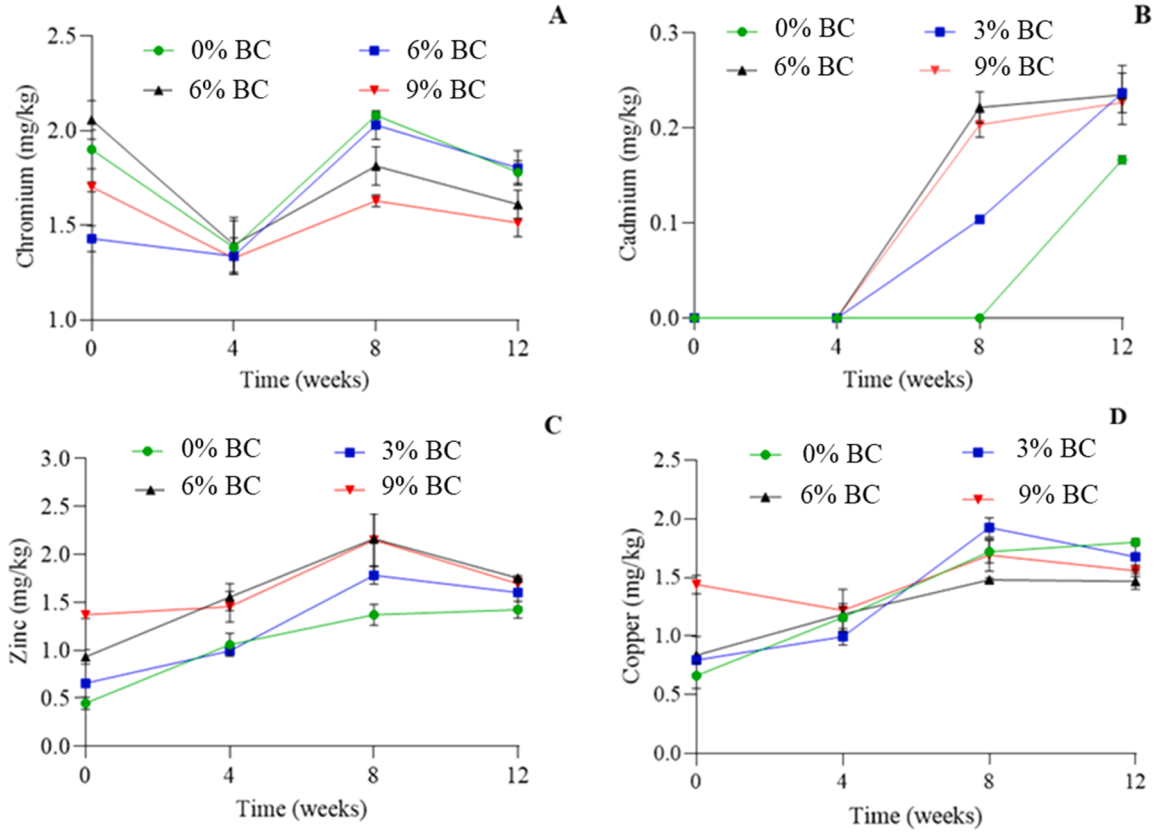


**Fig. 7.** Effect of different rates of *G. funicularis* seaweed-derived biochar amendment on Cr (A), Cd (B), Zn (C), Cu (D) in cattle manure-waste paper vermicompost over 12-week vermicomposting period. Error bars indicate standard deviation.

sampling time (Fig. 7D). In the 0% BC treatment, Cu increased from 0.66 mg/kg at week 0 to 1.80 mg/kg at week 12 (Fig. 7D). In the 3% BC, Cu concentration value rose from 0.79 mg/kg at week 0, reaching maximum of 1.93 mg/kg at week 8, followed by a slight decrease to 1.68 mg/kg at week 12 (Fig. 7D). The 6% BC and 9% BC treatments followed the same pattern, whereby in 6% BC, Cu content peaked with 1.48 mg/kg at week 8, followed by a slight decline to 1.47 mg/kg at week 12 (Fig. 7D). In 9% BC, it peaked with 1.69 mg/kg at week 8 before slightly decreasing to 1.56 mg/kg at week 12 (Fig. 7D). After 12 weeks, the highest Cu content was observed in the 0% BC treatment, followed by the 3%, 9%, and 6% BC treatments (Fig. 7D). Overall, heavy metal concentrations showed variable trends across treatments and time, reflecting complex interactions between seaweed biochar addition and vermicomposting processes.

### 3.6. Effects of seaweed biochar-amended vermicompost on crop phytotoxicity

There was a significant effect ($P < 0.05$) of seaweed biochar (BC) inclusion level on relative seed germination (RSG) in carrot and Swiss chard (Table 3). The RSG had no consistent pattern across the five crops, indicating species-specific responses to seaweed biochar addition. The highest RSG was observed in tomato, reaching 112.04% at 3% BC level, whereas Swiss chard recorded the lowest RSG, at 0% BC with 26.11% (Table 3). In tomato, the lowest RSG (100.00%) was observed in the 0% BC treatment, while the highest (112.04%) was observed at 3% BC level (Table 3). Cabbage RSG showed a decreasing trend with increasing BC levels, with the highest value of 100.37% observed in the 3% BC treatment and the lowest value of 75.56% recorded at 9% BC (Table 3). In contrast, in carrot, the RSG increased with higher BC inclusion, from 80.55% at 0% BC to 108.33% at 9% BC (Table 3). Similarly, Swiss chard showed a strong increasing RSG response to BC addition, with the lowest RSG of 26.11% at 0% BC and the highest value of 77.79% at 9% BC (Table 3). In onion, RSG remained relatively stable across treatments, with values ranging from 93.33% (at 0%,3% and 9% BC) to a value of 96.67% at 6% BC level, indicating no significant effect of seaweed biochar inclusion (Table 3).

There was a significant effect ($P < 0.05$) of seaweed biochar (BC) inclusion on relative root elongation (RRE) in most vegetable species, except for carrot (Table 3). Across the five vegetables, the highest RRE was observed in carrot with 96.63% at 9% BC inclusion, while the lowest RRE was observed in Swiss chard with 15.36% at 0% BC (Table 3). In tomato, RRE peaked at 0% BC with 81.55%, declined to 62.53% at 3% BC, and then showed a slight recovery to 71.66% at 9% BC (Table 3). Cabbage RRE exhibited a decreasing trend with increasing BC, with the highest value of 74.69% observed at 0% BC and the lowest value of 41.15% at 9% BC (Table 3). In the case of carrot, the observed RRE values were relatively high across all treatments, ranging from 69.66% at 0% BC to 96.63% at 9% BC, with no significant differences between the treatments (Table 3). Swiss chard exhibited an increasing trend in RRE with higher BC levels, from 15.36% at 0% BC to 85.17% at 9% (Table 3). For onion, the RRE fluctuated across treatments, with the lowest value of 44.91% observed at 3% BC and the highest of 94.91% at 0% BC (Table 3).

There was a significant effect ($P < 0.05$) of seaweed biochar (BC) inclusion level on the germination index (GI) for most vegetable species, except for tomato (Table 3). The observed GI values varied across the crop species; and the highest GI was observed in carrot with 105.16% at the 6% BC level, whereas the lowest GI was observed in Swiss chard with 3.99% at 0% BC (Table 3). In tomato, GI values ranged from 70.03% at 3% BC to 81.55% at 9% BC, showing no significant differences across treatments (Table 3). In cabbage, GI decreased with increasing seaweed biochar levels, with 72.32% at 0% BC and 31.64% at 9% BC (Table 3). Both carrot and Swiss chard showed an increasing trend in GI with higher BC inclusion, reaching 105.16% and 85.17%, respectively, at 9% BC (Table 3). In onion, GI fluctuated across treatments, with the lowest value of 41.99% observed at 3% BC level and the highest value of 90.48% in the 6% BC treatment (Table 3).

## 4. Discussion

This study addressed two main objectives: (i) to evaluate the effects of BC inclusion on the physicochemical properties and nutrient dynamics of seaweed biochar-amended vermicompost, and (ii) to assess its phytotoxicity and agronomic suitability. The discussion below is structured to address these objectives. In this discussion, emphasis is placed on statistically significant results, while non-significant findings are cautiously interpreted as trends to provide additional insight into vermicomposting processes

### 4.1. Physicochemical properties of seaweed biochar-amended vermicompost

To address the first objective, the effects of seaweed biochar addition on key physicochemical properties (pH, EC, and C/N ratio) were evaluated. Among the important physicochemical properties of organic amendments, pH is one of the critical properties that needs to be considered because it affects nutrient availability to crops. The consistent alkaline pH range in the current study aligns with Ajibade et al., 2022, who reported similar ranges in manure-based vermicompost. The alkaline pH recorded may be attributed to the initial pH of the manure and seaweed biochar used which was alkaline (Table 1). No statistically significant effect ($P > 0.05$) of seaweed biochar inclusion on pH was observed, therefore observed patterns are interpreted as trends rather than definitive effects. Unamended seaweed biochar-amended vermicompost pH increased over time due to enhanced vermidegradation of basic ammonia and continuous release of carbonates by microbes, corresponding to Katakula et al., 2022. In contrast, seaweed biochar inclusion showed a non-significant decreasing trend in pH over time, possibly due to the adsorption of basic ions, release of acidic functional groups by seaweed biochar in the vermicompost matrix, and organic acids by microbes. Similar trends were reported by Kumar et al., 2023, who reported that in rock phosphate-amended vermicompost, the pH decreases with time. Vermicompost with a pH within the alkaline range can be a suitable amendment, particularly for the remediation of acidic soils, because slightly alkaline amendments may help neutralize

**Table 3**
Influence of extracts of seaweed biochar-amended vermicompost on Relative Seed Germination (RSG, %), Relative Root Elongation (RRE, %), and Germination Index (GI) for selected vegetables.

| Seaweed biochar incorporation level | Bioassay crop | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Tomato | | | Cabbage | | | Carrot | | | Swiss chard | | | Onion | | |
| | GI | RRE | RSG | GI | RRE | RSG | GI | RRE | RSG | GI | RRE | RSG | GI | RRE | RSG |
| **0% BC** | 81.55 | $81.55^{a}$ | 100 | $72.32^{a}$ | $74.69^{a}$ | 96.67 | $56.29^{b}$ | 69.66 | $80.55^{b}$ | $3.99^{c}$ | $15.36^{c}$ | $26.11^{c}$ | $88.76^{a}$ | $94.91^{a}$ | 93.33 |
| **3% BC** | 70.03 | $62.53^{c}$ | 112.04 | $52.50^{b}$ | $52.29^{b}$ | 100.37 | $88.09^{ab}$ | 89.62 | $97.22^{ab}$ | $48.69^{ab}$ | $68.80^{b}$ | $69.44^{a}$ | $41.99^{b}$ | $44.91^{b}$ | 93.33 |
| **6% BC** | 70.17 | $67.15^{bc}$ | 104.17 | $57.84^{ab}$ | $59.64^{b}$ | 96.67 | $83.86^{ab}$ | 89.1 | $94.44^{ab}$ | $31.54^{b}$ | $65.59^{b}$ | $48.15^{b}$ | $90.48^{a}$ | $93.69^{a}$ | 96.67 |
| **9% BC** | 75.99 | $71.66^{b}$ | 105 | $31.64^{c}$ | $41.15^{c}$ | 75.56 | $105.16^{a}$ | 96.63 | $108.33^{a}$ | $67.13^{a}$ | $85.17^{a}$ | $77.79^{a}$ | $76.57^{a}$ | $82.06^{a}$ | 93.33 |

**Note**: Means within the same column followed by different superscript letters differ significantly ($P < 0.05$).

excess hydrogen ions in acidic soils and reduce the solubility of toxic elements such as aluminium and manganese (Cárdenas-Aguiar et al., 2024). In addition, slightly alkaline conditions may enhance the availability of essential nutrients, while also supporting microbial activity involved in nutrient cycling (Cárdenas-Aguiar et al., 2024). These mechanisms highlight the broader potential of this amendment for improving soil chemical conditions in acidic environments.

Similar to pH, the electrical conductivity (EC) of vermicompost is a crucial factor in determining its agronomic suitability, as it influences salinity dynamics (Gondek et al., 2020). Electrical conductivity increased significantly over time ($P < 0.05$), whereas seaweed biochar inclusion level had no significant effect, indicating that temporal changes were the primary driver of EC variation The increasing EC pattern with time in this study indicates that soluble saline ions increase with VC maturity, likely due to mineralisation of organic matter, releasing ions in the vermicompost matrix, as reported in other vermicomposting studies (Ananthavalli et al., 2019; Kumar et al., 2023). Raza et al., 2020 reported a decreasing trend in EC over time in unamended cow and pig manure vermicompost, which contradicts the EC pattern observed in this study. The current study exact EC values after 12 weeks ranging from 2.41 mS/cm to 2.77 mS/cm are consistent with Kumar et al., 2023 and Mupambwa at al. (2024), who reported also lower EC values (< 4 mS/cm) in rock phosphates and in fly ash amended vermicompost, respectively. In contrast to these findings, Katakula et al., 2022 and Mupambwa at al. (2024) reported that seaweed biochar-amended vermicompost exhibited EC values exceeding the permissible limit of 4 mS/cm, thereby compromising its agronomic suitability. Despite the observed increase in extractable cation concentrations during vermicomposting, EC values remained within moderate ranges. This may be attributed to the cation exchange capacity of biochar, which can retain nutrients on exchange sites while limiting the proportion of ions freely dissolved in the compost solution (Nemati et al., 2015). Consequently, nutrient enrichment may occur without a proportional increase in electrical conductivity. This deviation may be due to the water pre-treatment of seaweed biomass prior to seaweed biochar preparation employed in this study, which aids in salt reduction in seaweed biochar and therefore in seaweed biochar-amended vermicompost (Purnomo et al., 2021). In addition, variations in the chemical composition of different manures, such as goat and cow manure, can influence the characteristics of the resulting vermicompost (Nleya et al., 2025). The presence of seaweed biochar appeared to slightly elevate the EC, indicating a cation-enriched amendment (Gondek et al., 2020), while maintaining the EC value within a safe range of 2.63 mS/cm – 2.77 mS/cm our study. This highlights that pre-treated seaweed biochar can generate a safe vermicompost for soil application in a wide range of crops, without inducing salt stress when applied at optimised rates. Blending seaweed biochar-amended vermicompost with soilless media of lower EC and neutral pH may buffer pH and electrical conductivity because the low-salt media dilute the soluble ions from the vermicompost (Huang et al., 2019). In addition, the seaweed biochar's cation exchange sites can adsorb excess cations, together moderating salinity and alkalinity levels (Han et al., 2018). This approach reduces stress on sensitive crops and improves overall nutrient availability.

In organic amendments, the C/N ratio is a crucial and widely used parameter to determine maturity, with C/N ratios < 20 indicating a fully mature and safe vermicompost (Kong et al., 2024). The C/N ratio was significantly influenced ($P < 0.05$) by both seaweed biochar level and time, including a significant interaction, indicating that the effect of BC depended on vermicomposting duration. In this study, it was interesting to note that seaweed biochar-amended vermicompost in all treatments fell within the mature vermicompost range of 11.43 to 17.60, with 9% seaweed biochar showing the lowest C/N value, indicating more mature vermicompost (Mnkeni and Mupambwa, 2023). Across all treatments, the C/N ratio decreased over time likely due to active microbial degradation of carbon-rich substrates and relative enrichment of nitrogen as organic matter stabilised (Ananthavalli et al., 2019). However, this decline was more pronounced at higher seaweed biochar application rates, indicating a significant interaction between vermicomposting time and BC addition, where seaweed biochar accelerated the rate of vermicompost maturation, which is consistent with Ananthavalli et al., 2019. The seaweed biochar-amended vermicompost values of the BC treatments, particularly at higher rate (9% BC) with a C/N ratio <15 indicate more stable vermicompost, as reported by Katakula et al., 2022, which suggests that seaweed biochar can enhance the biodegradation process. The reduction in C/N ratio with increasing seaweed biochar addition may be due to the porous and adsorptive nature of biochar, which can provide additional surface area for microbial colonization and promote faster carbon mineralization (Gong et al., 2023). Biochar may also adsorb compounds that inhibit microbial activity, thereby helping to minimize nitrogen losses during vermicomposting (Gong et al., 2023). Consequently, higher seaweed biochar rates may enhance microbial activity and improve organic matter stabilization, leading to lower C/N ratios compared to the control. Moreover, the incorporation of desalinated seaweed biochar and waste paper not only enhanced nutrient retention and seaweed biochar-amended vermicompost maturity but also contributes to sustainable waste management and circular economy practices by repurposing materials that would otherwise be discarded. This pattern is however not consistent with Antileo-Mellado et al., 2024, who reported that biochar-amended treatment had higher C/N ratio (14.55) than unamended pig and poultry manure vermicompost (11.69–11.97). Variations in the C/N ratio between the present study and other studies may be attributable to the different raw materials and vermicomposting methods used, as reported by Ji et al., 2023. Overall, this seaweed biochar-amended vermicompost can be applied to enrich growing medias with organic carbon, while ensuring minimal nitrogen losses. This suggests that the effect of seaweed biochar on BC-amended vermicompost maturity is time-dependent, with higher application rates enhancing the rate of organic matter stabilisation as vermicomposting progresses.

### 4.2. *Macronutrients in seaweed biochar-amended vermicompost*

In relation to nutrient dynamics, nitrogen bioavailable forms and extractable phosphorus transformations were assessed to determine how seaweed biochar influences nutrient availability in cattle manure-waste paper vermicompost. Nitrogen is one of the most essential nutrients for crop growth and is typically absorbed as ammonium, nitrate, or nitrite (Singh and Singh, 2024). The $NH_4$-N and $NO_3/NO_2$-N concentrations were significantly influenced by the interaction between vermicomposting time and seaweed biochar level, indicating that the effect of BC on nitrogen dynamics varied across different vermicomposting stages. For instance, in the control, $NH_4$-N concentrations in seaweed biochar-amended vermicompost increased with time, consistent with the findings of Zhou et al., 2021. This pattern reflects the continuous earthworm-facilitated mineralisation of organic N by mechanisms such as fragmentation and enzyme secretion, which in turn enriches the vermicompost matrix with N forms (Yakkou et al., 2024). Gong et al., 2023 reported the opposite trend; in chicken and green manure vermicompost, $NH_4$-N concentration initially increased before gradually decreasing toward week 12, possibly attributable to different substrates and experimental techniques used. In contrast, Gong et al., 2023 reported similar time-dependent patterns in biochar-amended treatments, supporting an interaction between vermicomposting duration and biochar addition. This declining pattern at a later stage could be attributed to the seaweed biochar's porous structure which provides a conducive environment for nitrifying bacteria to convert more ammonium into nitrate/nitrates, reducing $NH_4$-N levels after 12 weeks (Liao et al., 2021). In addition, $NH_4$-N may decrease with time in BC treatments because biochar can adsorb ammonium ions and reduce its concentration (Antileo-Mellado et al., 2024). After 12 weeks, unamended vermicompost exhibited the highest $NH_4$-N, possibly due to delayed nitrification in the absence of seaweed biochar addition, suggesting a

potential role of biochar in influencing nitrification processes. However, the $NH_4$-N concentration range (88.69 mg/kg-151 mg/kg) in the present study was lower than the values (290 mg/kg-350 mg/kg) in vermicompost reported by Jia et al., 2022, highlighting the contribution of vermicompost substrate variation toward final $NH_4$-N ranges. Overall, these findings suggest that seaweed biochar influences ammonium dynamics in a time-dependent manner, with its effects becoming more evident at later stages of vermicomposting.

Similar to ammonium, in the control, $NO_3/NO_2$—N also increased with time, likely due to biological oxidation of ammonium to nitrates/nitrites by autotrophic nitrifying bacteria (Saba et al., 2023). At higher seaweed biochar levels, $NO_3/NO_2$-N decreased gradually over time, indicating a significant interaction between BC addition and vermicomposting duration. This trend is likely due to the ability of seaweed biochar to immobilise ammonium ions on its surface and influence microbial activities, particularly as the vermicomposting process advances (Wu et al., 2023). This indicates that seaweed biochar does not enhance nitrate availability but rather regulates nitrogen transformation by reducing nitrification and promoting nitrogen retention within the vermicompost matrix. In addition, seaweed biochar may contain inhibitory compounds such as phenolic and polyaromatic hydrocarbons which can be disruptive to nitrifying bacterial activities (Han et al., 2022). Comparable findings were reported by Jia et al., 2022, who observed that the addition of biochar decreased the nitrification index in vermicompost, likely due to a decline in nitrate concentration, while $NH_4$-N remained constant or increased. Moreover, values observed in this study in amended treatments at week 12 ranging 10.04 mg/kg- to 32.37 mg/kg, are generally lower than nitrate value reported in unamended cattle manure-waste paper vermicompost (up to 97.7 ± 8.7 mg/kg) in the study by Ferraz et al., 2022. After 12 weeks, higher $NO_3/NO_2$—N in the unamended vermicompost reflects more complete nitrification, which could be favoured by abiotic factors such as pH, EC, temperature, and oxygen in a required range by microbes to be active (Saba et al., 2023). In contrast, biochar may inhibit nitrification by altering these factors by influencing the denitrifying community and through the adsorption of ammonium ions, leading to lower nitrate/-nitrite levels (Wu et al., 2023). These results further confirm that nitrogen transformation pathways in seaweed biochar-amended vermicompost are strongly influenced by the combined effects of time and seaweed biochar, rather than by either factor alone.

Regarding the usability of this seaweed biochar-amended vermicompost, elevated $NH_4$-N, such as that in unamended vermicompost, may be toxic to crops and microbial communities and can increase the risk of nitrogen losses; hence, $NH_4$-N is less stable than nitrates/nitrites (Zhou et al., 2021). In contrast, seaweed biochar-treated vermicompost may be more suitable for agronomic application because it is more likely to have reduced ammonia ($NH_3$) volatilisation (Canatoy et al., 2024). Unamended vermicompost may be more prone to nitrate leaching and nitrogen losses in the form of $N_2O$ and ammonia gas (Canatoy et al., 2024; Wu et al., 2023). The inclusion of seaweed biochar may address these issues by providing this amendment with good N retention (Priya et al., 2024).

Phosphorus is a critical element for plant growth, development, and yield, primarily assimilated by plants in the form of phosphate ions (Singh and Singh, 2024). The level of phosphorus in seaweed biochar-amended vermicompost was significantly influenced by seaweed biochar addition. Across treatments Extractable P concentrations at week 12 are significantly higher than initial stages, in line with findings by Dugassa & Worku (2025). The increase in extractable phosphorus in mature seaweed biochar-amended vermicompost could be attributed to the secretion of enzymes, such as phosphatases, and microorganisms by earthworms during decomposition (Oyege et al., 2023). These enzymes and microorganisms facilitate the conversion of organic phosphorus from organic compounds into forms that are available to plants (Oyege et al., 2023). The higher extractable P observed at greater seaweed biochar rates-treated vermicompost (up to 281.00 mg/kg) may be due to the relatively high extractable P content (244.7 mg/kg) in the seaweed biochar (Table 1). In addition, biochar may enhance phosphorus availability through its liming effect and its ability to modify pH, which can increase P solubility, as well as by reducing phosphorus fixation through adsorption of metal ions such as Fe and Al that typically bind phosphate (Zhang et al., 2025). Extractable P values (262.14 mg/kg-281.00 mg/kg) observed in seaweed biochar-treated vermicompost, are higher than values (24.65 mg/kg −27.75 mg/kg) reported in crop residue-cow manure vermicompost by Dube et al., 2024. However, the present study values are consistent with those of Mupambwa et al., 2024, who reported Olsen P values for seaweed biochar-amended vermicompost within this same range. The discrepancies between the present study and Dube et al., 2024 can be attributed to differences in the chemical composition of the feedstock used, for example, the higher extractable P (850.63 mg/kg) observed in cattle manure used in the current study (Table 1). According to Bai et al., 2013, most crops prefer soils containing a minimum of 20 mg/kg of phosphorus. Seaweed biochar-amended vermicompost exhibited adequate Olsen P content (> 20 mg/kg), highlighting that it can be a good source of phosphorus to crops. In addition, it can be used to enrich hydroponic nutrient-inert growing media, such as coco peat with phosphorus (Hernández-Rodríguez et al., 2017). It is important to note that seaweed biochar particle size and production conditions can affect its interaction with seaweed biochar-amended vermicompost, including nutrient retention and microbial activity; however, these factors were not assessed in the present study.

### 4.3. Cations in seaweed biochar-amended vermicompost

The evaluation of exchangeable cations further contributes to understanding nutrient dynamics under different seaweed biochar application levels. The presence of exchangeable cations (Ca, Mg, K, Na) in growing media is important because cations influence salinity and soil pH trends, which in turn impact nutrient availability in the media (Yang et al., 2024). In particular, Ca, Mg, and K are essential plant macronutrients (Singh and Singh, 2024), while sodium is categorised as a non-essential plant element because of its toxicity at elevated concentrations; however, in small quantities, it can be useful in some plants (Nunes et al., 2022). The cations in this study showed varying patterns over time and across treatments, indicating that the cation concentrations in seaweed biochar-amended vermicompost were influenced primarily by vermicomposting processes, with limited or non-significant effects of seaweed biochar for certain cations. Calcium (Ca) concentrations were not significantly affected ($P > 0.05$) by seaweed biochar inclusion or time; therefore, observed variations in Ca concentrations are interpreted as non-significant trends. Magnesium (Mg) was significantly influenced by time ($P < 0.05$) but not by seaweed biochar inclusion, indicating that changes were primarily driven by vermicomposting progression. Similarly, Na concentrations varied significantly over time but were not significantly affected by seaweed biochar inclusion level. For instance, Ca concentration decreased at the early stage of vermicomposting in BC-treated treatments, which may be attributed to processes such as adsorption of $Ca^{2+}$ions to negatively charged seaweed biochar surfaces which reduces the available exchangeable calcium (Guo et al., 2020). Moreover, calcium concentrations likely decreased due to microbial cell wall stabilisation which uses calcium (Hao et al., 2024), and through precipitation to water-insoluble salts such calcium carbonate and phosphates (Chen et al., 2022; Penn and Camberato, 2019). After eight weeks, Ca concentration then started increasing, likely due to mineralisation of previously immobilised Ca by microbial organisms (Kumar et al., 2023), and desorption of Ca ions from BC surfaces due to changes in pH towards seaweed biochar-amended vermicompost maturity (Han et al., 2018).

The increase observed in Na, Mg, and K over time can be attributed to the continuous degradation of manure by microbial organisms and earthworms, releasing ions of these cations, which previously existed in

organic forms (Kumar et al., 2023). In unamended vermicompost, for all cations, the concentrations initially increased, possibly due to fast mineralisation because unamended VC lacks the adsorption effect that biochar provides (Ananthavalli et al., 2019). In the control, after four weeks, cations then decreased gradually, likely due to leaching; hence, there was no seaweed biochar in this treatment (Alvarez et al., 2019).

The findings for the increase in cation concentrations over vermicomposting time are consistent with those of Dume et al., 2023, who reported that available Mg and K concentrations in sewage sludge vermicompost increased toward maturity due to biomass weight reduction. Comparable findings in biochar-amended vermicompost were also reported by Raza et al., 2020, who reported higher cation concentrations at maturity compared to initial values. However, the findings of Dume et al., 2023 and Raza et al., 2020 contradict the decreasing pattern observed for calcium at a 9% BC rate. After 12 weeks, Ca, Mg, and Na concentrations were lowest at the higher BC rate (9% BC), indicating that the greater adsorption capacity of seaweed biochar at higher rates may have immobilized these cations (Jia et al., 2022). These findings are inconsistent with those of Abban-Baidoo et al. (2024), who reported that biochar-treated compost recorded higher cation concentrations at maturity compared to unamended compost, indicating variations in trends between vermicompost and conventional compost. Potassium maintained higher concentrations at higher BC rates throughout the vermicomposting period, likely due to the addition of seaweed biochar which had an initial higher initial K content (Table 1). In addition, seaweed biochar may enhance potassium availability by reducing leaching losses through its high cation exchange capacity (CEC), which enables retention of $K^+$ ions on exchange sites (Antileo-Mellado et al., 2024). This retention may allow for a more gradual release of potassium during vermicomposting, thereby maintaining higher available K levels over time.

Our study reported lower final Mg (1.19 g/kg –1.37 g/kg), Ca (5.82 g/kg – 6.44 g/kg), and K (8.25 g/kg – 8.90 g/kg) values compared to the poultry litter BC-amended bagasse vermicompost of Antileo-Mellado et al., 2024, which recorded Mg (3.38 g/kg), Ca (74.07 g/kg), and K (18.01 g/kg). However, our results align with their eucalyptus-sawdust BC-amended bagasse VC, which recorded Mg (1.93 g/kg), Ca (6.51 g/kg), and K (18.43 g/kg), reflecting a similar mineral proportion trend of Mg < Ca < K at maturity. Discrepancies in the cation ranges and patterns between the present study and others can be due to variations in vermicomposting feedstocks and vermicomposting conditions, such as moisture, duration, and stock density (Ji et al., 2023).

These findings suggest that this BC-enriched vermicompost can enhance soil with essential cations, especially at a higher BC rate (9% BC), which exhibits the lowest Na level while maintaining other elements, particularly Mg and K, within acceptable ranges (Ishfaq et al., 2022; Santos et al., 2024). Other studies reported enhanced performance of horticultural crops in media with up to 17.1 g/kg of calcium (Zapałowska et al., 2025). In this study, Ca ranges (5.82 g/kg to 6.44 g/kg) were lower than these values, highlighting that this amendment can be safely applied to some crops. In contrast, other studies reported that Ca content > 400 mg/kg (equivalent to 0.4 g/kg) in growing media can be detrimental to horticultural tree species seedlings, suggesting that seaweed biochar-amended vermicompost in this study may not be suitable for such species (Li et al., 2023; Zhang et al., 2023). Variations in the literature regarding Ca thresholds suggest that the suitability of the media is driven by crop-specific nutrient requirements (Zapałowska et al., 2025; Zhang et al., 2023). However sodium level of 3.4 g/kg observed at 9% BC was the lowest recorded in this study, it may still be phytotoxic to salt sensitive crops, as it can cause osmotic stress, germination and growth inhibition (Benazzouk et al., 2019). Therefore, blending this amendment with other soilless growing media which have lower cation concentrations and leaching practices may be considered in the future to achieve maximum agronomic suitability of this amendment. It should be noted that in this study, seaweed biochar-amended vermicompost applications in soil were not evaluated; therefore, the long-term effects on soil salinity and nutrient balance remain unknown.

### 4.4. Heavy metals content in seaweed biochar-amended vermicompost

To further assess the quality and agronomic suitability of the seaweed biochar-amended vermicompost, heavy metal concentrations were analysed to evaluate potential environmental and health risks. The accumulation of heavy metals can be toxic to plants, and their accumulation in agricultural produce may lead to health issues (Kujawska, 2023). However, some trace metals, including Zn, Cu, and Ni, are characterised as micronutrients and are vital for promoting plant biological processes and preventing deficiency diseases (Singh and Singh, 2024).

Among the heavy metals assessed, Ni was not detected, possibly because the seaweed biochar and cattle manure used had no detectable Ni levels (Table 1). Cd was not detected at an earlier stage, likely because it existed in recalcitrant organic matter or mineral-bound forms, as stated by Goswami et al., 2016, and such forms of heavy metals may not be detected by the digestion analysis employed in this study. Previous studies have reported dual trends of heavy metals in vermicompost over time. Some studies have reported heavy metal accumulation in vermicompost over time (Hait and Tare, 2012; Paul et al., 2020), whereas others have reported heavy metal reduction in vermicompost over time (Ananthavalli et al., 2019; Mupondi et al., 2018). In the present study, an increasing trend was observed, but with a decreasing pattern at later stages. The increase in heavy metal content, as observed for Zn, Cu, and Cd, towards week 12 could be attributed to the degradation of organic matter, which reduced the seaweed biochar-amended vermicompost bulkiness, enabling such metal concentrations to rise, as reported by Hait & Tare (2012). In contrast, the reduction in heavy metal content over time, as observed for most metals, particularly from week 8, is likely due to the bioaccumulation of trace metals in earthworm tissues (Goswami et al., 2016). The observed lower Cr and Cu values in the seaweed biochar-amended treatments at later stages suggest a time-dependent effect of BC. This can be explained by the increasing ability of biochar to immobilise these heavy metals through adsorption as the vermicompost matures, reducing their concentration in the vermicompost matrix (Qiu et al., 2020). In addition, higher Cr and Cu in the unamended treatments could be due to the high content observed in the original manures used in this study (Table 1). In contrast, higher Zn values observed in seaweed biochar-amended treatments throughout the vermicomposting period can be explained by the higher initial Zn value (3.49 mg/kg) in seaweed biochar, as shown in Table 1; a similar trend was reported by Mupambwa et al., 2024.

The seaweed biochar-amended vermicompost in the present study had values of Cr < 600 mg/kg, Cu < 4300 mg/kg, Zn < 7500 mg/kg, Ni < 420 mg/kg, and Cd < 85 mg/kg, which are safe for soil applications according to the United States Environmental Protection Agency (U.S. Environmental Protection Agency US EPA, 2020). The heavy metal values in this study were consistent with other studies that reported values within the same ranges for seaweed biomass/biochar and rock phosphate-enriched cow manure vermicompost (Ananthavalli et al., 2019; Mupondi et al., 2018). Therefore, this seaweed biochar-amended vermicompost is suitable for agronomic applications without health hazards or toxicity to crops. In addition, seaweed biochar–based vermicompost is rich in Zn, indicating its potential use in zinc-deficient soils to enhance crop growth (Vuković et al., 2021). This could, in turn, help alleviate Zn deficiency in humans consuming crops grown in soils incorporated with this seaweed biochar-amended vermicompost (Vuković et al., 2021). Overall, the interaction between vermicomposting time and seaweed biochar addition played a critical role in influencing nutrient availability and transformation. Significant interaction effects observed for C/N ratio, $NH_4$-N, $NO_3^-$/$NO_2^-$-N, and Cr indicate that seaweed biochar not only affects nutrient levels but also modifies the rate and direction of biochemical processes over time.

Specifically, higher BC application rates enhanced seaweed biochar-amended vermicompost maturity, altered nitrogen transformation pathways, and promoted heavy metal immobilisation as vermicomposting progressed. These findings demonstrate that the impact of seaweed biochar is dynamic rather than static and highlight the importance of considering both seaweed biochar level and vermicomposting duration when evaluating seaweed biochar-amended vermicompost quality.

### 4.5. *Phytotoxicity and agronomic potential of seaweed biochar-amended vermicompost*

To address the second objective, phytotoxicity tests were conducted to evaluate the biological safety and maturity of the seaweed biochar-amended vermicompost. Phytotoxicity is characterised by delayed seed germination, inhibited plant growth, or any detrimental effects on plants induced by specific substances, such as phytotoxins or environmental conditions (Ravindran et al., 2017). Therefore, assessing the phytotoxicity of organic amendments such as biochar, manure, and vermicompost prior to their application to crops is crucial to ensure safety and optimise yield performance. In this study, the phytotoxicity of seaweed biochar-amended vermicompost was evaluated using Germination Index (GI), Relative Root Elongation (RRE), and Relative Seed Germination (RSG) in five vegetables. Phytotoxicity indicators (RSG, RRE, and GI) showed mostly significant responses ($P < 0.05$) to seaweed biochar inclusion, justifying detailed interpretation of treatment effects.

Starting with RSG, at low seaweed biochar rates, Swiss chard demonstrated significantly ($P < 0.05$) reduced germination, whereas other species such as tomato, onion, and cabbage attained higher RSG values of up to 108.33%. These findings are in line with those of Chawhan and Chintapalli (2025), who reported that germination percentage varies among vegetable species under the same conditions due to differences in their tolerance and physiological characteristics. Unlike other species, Swiss chard likely has a less permeable seed coat, which limits water and oxygen uptake and increases sensitivity to phytotoxic compounds such as soluble salts and ammonia in the seaweed biochar-amended vermicompost extract, reducing its RSG (Upretee et al., 2024). Interestingly, species with initially low RSG values showed higher germination under seaweed biochar-amended vermicompost, indicating that poor germination in unamended vermicompost was not solely due to seed physiology but also to the presence of toxins (Lončarić et al., 2024). These findings correspond with Duraisamy et al. (2021), who reported that vermicompost extracts may contain phytotoxic compounds that inhibit seed germination in certain vegetable species. The inclusion of seaweed biochar likely improved RSG through immobilisation of toxic organic compounds in unamended vermicompost (Huang et al., 2024), thereby creating a more favourable environment for seed germination. In addition, a lower C/N ratio (11.43) in the seaweed biochar-amended vermicompost indicates a more mature vermicompost, which tends to have reduced phytotoxicity and promotes seed germination, as indicated by (Lončarić et al., 2024). These findings highlight the importance of blending seaweed biochar with cattle manure-waste paper vermicompost to enhance its suitability, reduce phytotoxicity, improve seed germination, and broaden its applicability to diverse crop species.

Swiss chard still had the lowest RRE values in unamended vermicompost, highlighting its sensitivity to phytotoxic compounds such as phenolics and short-chain organic acids, which can inhibit cell division and elongation in the root apical meristem (Verdeguer et al., 2020). The significant ($P < 0.05$) improvement in RRE up to 85.17% with seaweed biochar addition may be attributed to the ability of biochar to adsorb these soluble toxic compounds (Huang et al., 2024). These results suggest that seaweed biochar not only mitigates toxicity and enhance germination but also promotes root development, particularly in sensitive species. Similar results were reported by Tasnim et al., 2021, who found that vermicomposting with biochar increased root elongation compared to vermicompost alone.

The germination index is widely used to assess the phytotoxicity of organic amendments, integrating both RSG and RRE (Jakubus and Bakinowska, 2018). According to Ravindran et al., 2017, GI values below 50% indicate phytotoxicity, values between 50% and 80% indicate moderate toxicity, and values above 80% indicate a lack of phytotoxicity. Similar criteria were adopted in this study to evaluate seaweed biochar-amended vermicompost toxicity. In this study, unamended vermicompost was within toxic to non-toxic ranges, reflecting species-specific responses, as also reported by Chawhan and Chintapalli (2025). In contrast, Duraisamy et al., 2021 reported that mineralised unamended vermicompost extracts are non-toxic and improve germination. This deviation can be explained by variations in the tested crops species and vermicompost feedstock. The lower GI observed in unamended vermicompost in this study may be attributed to higher levels of ammonia, salts, and organic acids present in manures (Ravindran et al., 2017).

Seaweed biochar inclusion improved GI in species such as Swiss chard and carrot, likely because of its ability to enhance nutrients and adsorb toxins (Huang et al., 2024). These findings contrast with those of Mupambwa et al., 2024, who reported inhibited GI in vegetables (< 50%), in the seaweed biochar-amended vermicompost treatment. These differences may have resulted from the use of pretreated seaweed biochar feedstock in the present study, which reduced salts and phytotoxic compounds (Stévant et al., 2018), thus improving germination. In cabbage, seaweed biochar inclusion at 3–9% induced phytotoxicity, likely due to elevated K and EC values (2.60–2.77 mS/cm) at these rates, which were higher than at lower BC levels (Chawhan and Chintapalli, 2025). Increased EC may create osmotic stress during germination, while elevated potassium concentrations can further contribute to ionic imbalance, both of which may inhibit seed germination in sensitive species such as cabbage (Chawhan and Chintapalli, 2025). Onion and tomato remain within the moderate to non-toxic range, suggesting greater tolerance to a wide range of conditions such as salinity and osmotic stress in certain species (Muhammad et al., 2024). From an agronomic perspective, vermicompost amended with pretreated seaweed biochar can enhance nutrient availability and is safe for application to certain species. In contrast, excessive application may induce stress to sensitive species, emphasizing the need to match crop type with amendment level to achieve maximum seed germination.

## 5. Conclusions and recommendations for future studies

The addition of 9% *G. funicularis* BC produced seaweed biochar-amended vermicompost with slightly alkaline pH (7.85), lower EC (2.70 mS/cm), the lowest C/N ratio (11.43), highest macronutrient levels (Olsen P 281 mg/kg, K 8.90 g/kg), moderate N-forms, and the lowest Na concentrations (3.44 g/kg). Seaweed biochar-amended vermicompost with up to 9% BC was not phytotoxic to most tested vegetables, except cabbage. It should be noted that the phytotoxicity results are based on seed germination assays, which assess early seedling growth and may not capture long-term effects; thus, these findings should be interpreted as an initial screening.

Based on these findings, the seaweed biochar-amended vermicompost with BC amendment at 9% may have potential for enriching acidic soils with macronutrients, although soil validation studies are still required. Future studies should focus on evaluating the amendment performance in horticultural soilless media as a nutrient source while simultaneously buffering its pH and EC through blending. In addition, future studies should evaluate the economic feasibility of producing seaweed biochar-amended vermicompost from *G. funicularis* and other abundant seaweed species. This would include assessing the costs associated with collection, processing, and application at scale, to ensure that the proposed amendment is both agronomically effective and economically sustainable. Lastly, the long-term impacts of repeated seaweed biochar-amended vermicompost applications on soil salinity

and nutrient balance may also be evaluated in future.

## Consent to participate, and consent to publish

Not applicable.

## Clinical trial number

Not applicable.

## Ethical approval and guidelines compliance

The experiments conducted in this study involving plant materials were performed in accordance with the ethical standards of the University of Namibia, under reference number: OGC0033.

## Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work, the author(s) used Open Paper Pal in order to improve the English readability of the manuscript. After using this tool/service, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the publication.

## Funding

This research was funded by the Daures Green Hydrogen Village Project, within the Namibia Green Hydrogen Research Institute under the University of Namibia (R404) with funding from Southern African Science Service Centre for Climate Change and Adaptive Land Management (SASSCAL). The study was also partly funded by the New Zealand Government to support the objectives of the Global Research Alliance on Agricultural Greenhouse Gases" under grant number 2023-EJPSOILS-CarouNd-UN under the University of Namibia (R411 project).

## CRediT authorship contribution statement

**Elia N.M. Ruben:** Writing – review & editing, Writing – original draft, Methodology, Formal analysis. **Simeon S. Hamukoshi:** Supervision, Funding acquisition. **Bethold Handura:** Visualization, Methodology. **Hupenyu A. Mupambwa:** Writing – review & editing, Validation, Supervision, Funding acquisition.

## 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.

## Acknowledgments

The authors acknowledge staff members and students from the University of Namibia , Sam Nujoma Campus, who assisted in experimental setup of this study.

## Data availability

Data will be made available on request.

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