Hindawi
International Journal of Agronomy
Volume 2018, Article ID 6419467, 6 pages
https://doi.org/10.1155/2018/6419467

Research Article

Amino Acids, Mineral Nutrients, and Efficacy of Vermicompost and Seafood and Municipal Solid Wastes Composts

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Received 30 December 2017; Accepted 18 March 2018; Published 26 April 2018

Academic Editor: Manuel Tejada

Growing medium amino acids and mineral nutrients stimulate rhizosphere activities and plant growth. A greenhouse experiment was performed to compare amino acid and mineral nutrient profiles of seafood waste compost (SFWC) and municipal solid waste compost (MSWC) and vermicompost (VC). Their efficacies were also tested on onion (*Allium cepa* L. "Sweet Utah"). The control was Promix-BX™ alone. The MSWC, SFWC, and VC were composed of total of 36.4, 48.3, and 67.5 mg amino acids/100 g dry weight, respectively. Glutamic acid, aspartic acid, and glycine were the highest while methionine, histamine, and cysteine were the least in all the amendments. The VC had the highest Ca content but the least P and K contents. SFWC had the highest content of P and K and most of the determined micronutrients. The MSWC had significantly ($P < 0.05$) the highest N and leaf chlorophyll contents followed by the VC. The MSWC significantly ($P < 0.05$) increased anthocyanin content while the control recorded the least. The maximum quantum yield of photosystem II (Fv/Fm) and the potential photosynthetic capacity (Fv/Fo) were least in the VC treated plants. Dry matter was not affected by the type of amendment. Overall, plant growth was improved by the MSWC. Future research should investigate effect on secondary metabolites.

1. Introduction

Proteins are derived from amino acids and are essentially the basic component of all living cells [1, 2]. Amino acids and proteins play vital roles in the genetic and the physiological processes of organisms such as syntheses of nucleic acids, proteins, enzymes alkaloids, vitamins, terpenoids and chlorophyll, and formation of vegetative tissues [3–5]. These require specific and adequate quantities of plant tissue amino acids with each or combination of the amino acids performing specific function. Root uptake of amino acids can vary with the concentration, plant genotype, and rhizosphere microbial activities while microorganisms and plant roots compete for the available amino acids [6, 7]. For instance, onion *(Allium cepa)* is one of the few plants that use the amino acid, arginine, as a pool for nitrogen (N) during bulb formation and development [8]. This leads to an increase in chlorophyll concentration and the greening of onion leaf tissues because of the availability of free amino acids in the growing medium [9]. However, Kandil et al. [10] reported after a 2-year field trial that onion plant growth did not respond to foliar application of amino acids relative to the control treatment, but increased growth response was recorded for those onion plants that were sprayed with humic acid extracts.

Seafood such as fish, mollusk, and crustacean contains 10.3–23.3% proteins, which is composed of all the essential amino acids [11]. These proteins can be degraded during composting processes and humified into amino acids and sugars. The concept of composting seafood waste is relatively new in many parts of the world. Moreover, there are few studies that provided some information on chemical composition of seafood waste compost. On the other hand, vermicompost made by the process of earthworm (family Annelida) ingestion, digestion, and excretion, is well studied and widely used in agriculture. Many studies have confirmed the efficacy of vermicompost and its ability to enhance chemical constituents of plants such as kale (*Brassica oleracea* var. *sabellica* [12]) and aromatic and medicinal plants (*Plectranthus amboinicus* [13]) as compared to other natural amendments but excluded seafood waste compost. It was found that vermicompost increased the amino acids, flower yield, and essential oil content of *Matricaria chamomilla* [14]. Also, a study by Shaheen et al. [2] suggested that high rate of application of municipal solid waste compost increased the contents of onion bulb protein and mineral nutrients.

Onion is the second most consumed vegetable in the world with high content of antioxidants, vitamin C, folic acid, calcium (Ca), iron (Fe), and protein [15]. The content of these secondary metabolites and nutrients may be influenced by the composition of growing medium free amino acids as previously explained by Kopriva and Rennenberg [16] and Jin et al. [9]. Protein-rich seafood offal has relatively low content of connective tissue, which makes it easy to compost. During composting, proteins are broken down by microbial and enzymatic activities and humified into amino acids and sugars. The feedstock for vermicompost and municipal solid waste compost can vary, and the composition of the aggregate may not be richer in protein than seafood compost feedstock. Therefore, the objective of this study was to compare the amino acid and mineral nutrient profiles of seafood waste compost, municipal solid waste compost, and vermicompost and their agronomic impact on "Sweet Utah" onion.

2. Materials and Methods

2.1. Composting and Compost Utilization. Composting of seafood waste obtained from a local grocery store started in September 2015 and finished in January 2016. The seafood waste compost was made by adding approximately 10 kg of shredded fish and crustacean waste including the offal to a 10 L rubber-maid container with perforations on the walls for aeration. Dry rye straw was cut into pieces and added to the seafood waste as source of carbon (C) with the aim of achieving a C : N ratio of 25–30 : 1. Estimation of the C : N ratio was based on the formula by Richard and Trautmann [17]. These materials were added in layers, that is, the seafood waste on top of the straw followed by 100 g of finished seafood waste compost as a booster, and then strawed before sprinkling 1 L of water to moisten the mass. The layers were repeated in that same order until the container was one-third full before placing on a lid. The mass was turned and mixed once a month. The compost reached a peak temperature of 58°C (thermophilic stage) before cooling down at the mesophilic and maturation stages to ≤20°C after four months and allowed to cure for another month. The finished seafood waste compost was then stored at 4°C. For comparative analysis, mature municipal solid waste compost (7 months old) from Fundy Compost (Brookfield, Nova Scotia) and vermicompost (6 months old) produced by red wiggler worms *(Eisenia fetida)* from Cathy's Crawly Composters (Bradford, Ontario) were the other treatments used in the study.

2.2. Growing Medium Preparation. Based on previous Manure compost study by Abbey and Appah [18], 25% (w/w) of the seafood waste compost, municipal solid waste compost, and the vermicompost were individually incorporated into 400 g of Promix-BX soilless potting mix (Premier Horticulture Inc., Quakertown, PA, USA) purchased from Halifax Seeds, Nova Scotia, using 25.4 cm diameter plastic pots. The control treatment was a 400 g of the Promix-BX potting mix alone without the addition of any amendment. Each of the potted medium was saturated with water and allowed to stand for 24 h to attain field water-holding capacity prior to transplanting "Sweet Utah" onion seedlings.

2.3. Plant Culture. Seeds of "Sweet Utah" onion were initially sown in a 72-plastic cell-tray in the greenhouse and nurtured for five weeks after germination. Seedlings were not fertilized until after transplanting into the individual organic amendments. Three seedlings were transplanted into each pot, which was later thinned out to two seedlings per pot two weeks after transplanting (WAT). Each amendment was applied in two splits, that is, at planting and at 5 WAT. The greenhouse set temperature was 24°C in the day and 16°C at night and 72% relative humidity. Supplementary lighting was provided by a 600 W HS2000 high pressure sodium lamp with NAH600.579 ballast (P.L. Light Systems, Beamsville, ON, Canada). Approximately, 200 ml of tap water was added to each potted-plant every two days until the time of harvest.

2.4. Amino Acid and Mineral Nutrient Analyses. Approximately 200 g samples of each organic growing medium amendment (i.e., seafood waste compost (SFWC), municipal solid waste compost (MSWC), and vermicompost (VC) was sent to Covance Laboratories (Madison, Wisconsin, USA) for analyses of 17 amino acids (i.e., aspartic acid, threonine, serine, glutamic acid, proline, glycine, alanine, valine, isoleucine, leucine, tyrosine, phenylalanine, lysine, histidine, arginine, cysteine, and methionine). The Covance Laboratory reference method used for the amino acids analyses was Amino Acids TAALC_S.17 as described by Henderson and Brooks [19]. Additionally, approximately 200 g samples of the different types of compost and 20 g of harvested plants per treatment were sent to the Nova Scotia Department of Agriculture Laboratory Services (Truro, Nova Scotia, Canada) for the analyses of macro- and micronutrients, crude protein, and crude fat using the dry ash method based on Association of Analytical Communities method 968.08 as described in AOAC [20].

2.5. Plant Growth Determination. The onion plants were harvested as green onions and all the growth data were collected at final harvest at 8 WAT. The number of green leaves per plant was recorded for each treatment. Plant height was measured from the stem disc of the onion plant to the tip of the longest leaf. Total plant fresh weight was recorded for each treatment. Onion bulb development was estimated from the ratio of bulb-to-pseudostem diameter. The diameter of the swollen base of the pseudostem was measured as the bulb diameter while the pseudostem diameter was measured from the middle portion of the pseudostem. The root mass of each plant was washed under running tap water and the fresh weight was recorded. Leaf chlorophyll content was measured by leaf greenness using SPAD 502 Chlorophyll meter (Spectrum Technologies Inc., Illinois, USA). Leaf anthocyanin content was determined using portable ACM-200 Anthocyanin meter (OptiSciences Inc., New Hampshire, USA). Both chlorophyll and anthocyanin contents were estimated from the 3rd and 4th leaves of each plant. Photosynthetic activities of the tagged leaves were determined using Os30p+ Chlorophyll Fluorometer (Opti-Science Inc., NH, USA) based on the methods of Kitajima and Butler [21] and Kalaji et al. [22] with some modifications. The middle portion of the 3rd and 4th leaves excluding the midrib was clipped with a dark adaptation clip with the window closed for 25 mins prior to recording of the photosynthesis indices on the Chlorophyll Fluorometer. The data collected included variable fluorescence (Fv = Fm − Fo), where Fo and Fm are minimum and maximum fluorescence, respectively. Data on maximum quantum efficiency as determined by the ratio of variable florescence to maximum florescence (Fv/Fm) and the ratio of efficiency of electron donation to photosystem II estimated by the ratio of variable fluorescence to minimum fluorescence (Fv/Fo) were also recorded.

2.6. Experimental Design and Data Analysis. The single-factor experiment was laid out in a completely randomized design with three replications. Each of the experimental treatment was comprised of ten pots with each containing two plants. The pots were periodically rearranged on the greenhouse bench to offset any unforeseen variations in environmental condition. Data collected were subjected to one-way analysis of variance (ANOVA) using Minitab v. 18.1. Differences in experimental treatments were identified using the Fisher's least significance difference (LSD) method when the ANOVA suggested a significant difference at $P \leq 0.05$. Graphs with standard error bars were also plotted using Microsoft Excel.

3. Results and Discussion

The concentrations of the individual 17 determined amino acids were consistently and significantly ($P < 0.05$) highest in the VC and least in the MSWC, that is, VC > SFWC > MSWC (Figure 1). Moreover, the sum total of the 17 analyzed amino acids in the MSWC, SFWC, and VC was approximately 36.4, 48.3, and 67.5 mg/g dry weight, respectively. These amino acids were part of the humic substances in the MSWC, SFWC, and the VC. The differences in the total and individual amino acid concentrations can be attributed to variations in the starting feedstock, the biodegradation process employed, the structure of the microbial community, and overall activities of hydrolyzing enzymes [23, 24]. According to Fuentes et al. [23], the composition of amino acids in compost may range between 4% and 20%. For instance, the three amino acids with the highest concentrations in the different types of organic amendments were glutamic acid (5.55–6.90 mg/g), aspartic acid (4.56–9.14 mg/g), and glycine (3.37–6.59 mg/g). On the contrary, the three amino acids with the least concentrations were cysteine (0.44–0.81 mg/g), histamine (0.62–0.98 mg/g), and methionine (0.67–0.78 mg/g). All the other remaining 11 amino acids were intermediate in amounts (Figure 1). The significance in the differences in the amounts and the types of amino acids can have an impact on the chemical properties and efficacies of the three different organic amendments.

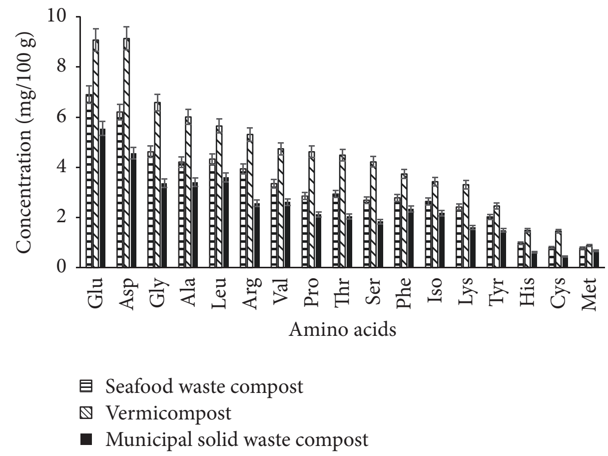

Figure 1: Amino acids composition in seafood waste compost, vermicompost, and municipal solid waste compost. Glu, glutamic acid; Asp, aspartic acid; Gly, glycine; Ala, alanine; Leu, leucine; Arg, arginine; Val, valine; Pro, proline; Thr, threonine; Ser, serine; Phe, phenylalanine; Iso, isoleucine; Lys, lysine; Tyr, tyrosine; His, histidine; Cys, cysteine; and Met, methionine.

The high amino acids in the VC as compared to the MSWC and SFWC can be attributed to microbial and enzymatic activities in the gut of the red wiggler worms during the process of worm ingestion, digestion, and excretion [25]. Furthermore, Vialykh et al. [24] suggested that variations in the composition of amino acids in soils and humic substances were due to the presence of wide range of amino acids in microbial biomass. This can explain the high amino acids content in the VC, that is, 40% greater than the SFWC and 85% greater than the thermophilically produced MSWC (Figure 1). The amino acid concentrations of the SFWC were also 33% greater than those of the MSWC due to the protein-rich seafood feedstock [11]. Comparatively, glutamic acid and aspartic acid were highest in all the three organic amendments. However, methionine was the least in the VC while histamine and cysteine were the least in the SFWC and the MSWC. It was reported that free amino acids improve microflora growth, which in turn enhanced soil nutrients assimilation [5, 7]. Thus, the variations in amino acid composition in the three different organic growing amendments can influence nutrient uptake by the plants growing in them.

The mineral nutrient contents of the SFWC, MSWC, and VC were significantly ($P < 0.05$) different (Figures 2 and 3). Overall, Ca and N were highest in all the organic amendments while magnesium (Mg) and phosphorus (P) were the least (Figure 2). Comparatively, the VC had the highest Ca content but the least P and potassium (K) contents while N, P, and K were highest in the SFWC (Figure 3). This was expected because of the high protein and mineral nutrient contents of seafood [11].

Onion plant tissue content of K was the highest followed by Ca and N, and the least were Mn and Zn, irrespective of the amendment (Figure 4). However, it was found that the uptake of K and Zn were highest in plants grown in the SFWC whereas plants grown in the MSWC had the highest

Table 1: Onion "Sweet Utah" plant growth and development parameters as affected by different growing medium organic amendments.

Treatment	% DM[2]	CP	CF	SPAD value	Anthocyanin	Fv/Fm	Fv/Fo
SFWC[1]	10.7[a]	6.04[b]	2.11[ab]	46.3[b]	6.8[b]	0.791[a]	3.805[a]
VC	10.5[a]	6.77[ab]	2.15[ab]	47.1[ab][3]	7.0[b]	0.768[b]	3.241[b]
MSWC	10.7[a]	7.95[a]	2.22[a]	51.4[a]	8.6[a]	0.783[ab]	3.626[a]
Control	11.0[a]	5.83[c]	1.91[b]	43.2[b]	5.2[c]	0.780[ab]	3.566[a]
P value	ns	0.05	0.05	0.02	0.01	0.03	0.01

[1]SFWC, seafood waste compost; VC, vermicompost; MSWC, municipal solid waste compost; [2]DM, dry matter content; CP, crude protein; CF, crude fat. [3]Means in columns followed by the same letter are not significantly different at $\alpha = 0.05$, Least Significant Difference; ns, not significantly different at $\alpha = 0.05$.

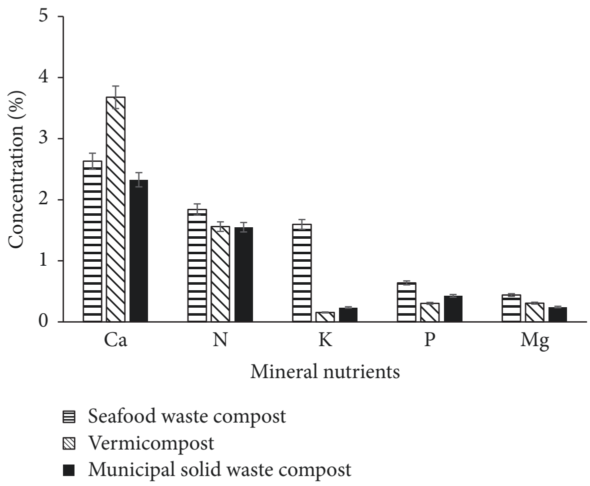

Figure 2: Macronutrients composition in seafood waste compost, vermicompost, and municipal solid waste compost. Calcium (Ca); nitrogen (N); potassium (K); phosphorus (P); and magnesium (Mg).

uptake of Mn, sodium (Na), N, Ca, and Mn as compared to the other treatments. Magnesium uptake did not vary among the plants irrespective of the growing medium amendment. Also, plant uptake of N, Mn, and P were not different between the SFWC and VC. Previous report suggested that cysteine and methionine were involved directly in the assimilation of N, S, and C in plants [16]. This suggested that the high amino acid content of the VC can be associated with an increase in N and crude protein contents of plants grown in the VC as compared to onion plants grown in the other amendments. It was reported that N uptake may be inhibited by amino acids accumulation in the cytoplasm of plant cells [26], which was contrary to the present result. Onion plant tissue N content was reduced by the low N content of the control treatment, that is, Promix-BX soilless potting mix alone (Figure 4). Percentage dry matter and crude fat contents of onion "Sweet Utah" were similarly affected by the three different types of organic amendments with moderate increase by the MSWC and moderate reduction by the control (Table 1). There was no significant ($P > 0.05$) association between percentage dry matter, crude fat, and crude protein contents in the present study.

Leaf greenness (chlorophyll content) as determined by SPAD value and anthocyanin content were significantly ($P < 0.05$) increased by the MSWC followed by the VC although values for the VC were not significantly ($P > 0.05$) different from those for the SFWC. Similar observation was made in previous onion bulb studies by Jin et al. [9]. The low pH of the control Promix-BX soilless potting mix (5.8) and the SFWC (5.9) may be the cause of the reduced leaf greenness and leaf tissue anthocyanin content. The lack of correlation (not presented) between growth and yield responses versus the amino acid and mineral nutrient compositions of the three different organic amendments conformed to the report by Kandil et al. [10].

The maximum quantum yield of PSII photochemistry and the potential photosynthetic capacity as determined by Fv/Fm and Fv/Fo, respectively, were significantly ($P < 0.05$) reduced in onion plants grown in the VC as compared to the other amendments including the control plants (Table 1). Lower values of these physiological indices were indications of lower chloroplasts photochemical activities and reduced photosynthesis [27]. The ratio of Fv/Fo represented the structural alterations in PSII and also indicated the electron donation efficiency [28, 29]. Although there were significant ($P < 0.05$) differences in photosynthetic activities between treatments, the percentage dry matter content (Table 1), and plant growth and yield components such as the number of leaves, plant height, pseudostem and bulb diameters, bulbing ratio, and roots and foliage, fresh weights were not significantly ($P > 0.05$) affected (data not presented) as reported by Kandil et al. [10]. The reason for the relatively low photosynthetic efficiency recorded for VC despite the high amino acids and mineral nutrients contents is not clear and will be investigated in future studies. However, it is possible that acquired chemicals were used in secondary metabolism instead of primary metabolism as there is a tradeoff between the two.

In conclusion, the findings demonstrated distinct variations in amino acid and mineral nutrient profiles of the SFWC, MSWC, and VC. Plant growth was improved by the MSWC as compared to the others. Comparatively, the VC had the highest amount of amino acids and mineral nutrients followed by the SFWC. Although the VC did not improve plant growth, previous research confirms VC enhancement of plant biosynthesis of secondary metabolites. Results from earlier studies were contradictory. Some studies showed that amino acids application increased plant growth and yield [4, 16] while others such as the study by Kandil et al. [10] did not find any significant difference. Unlike previous investigations, the current study did not apply pure amino acids as experimental treatment and as such the effects were

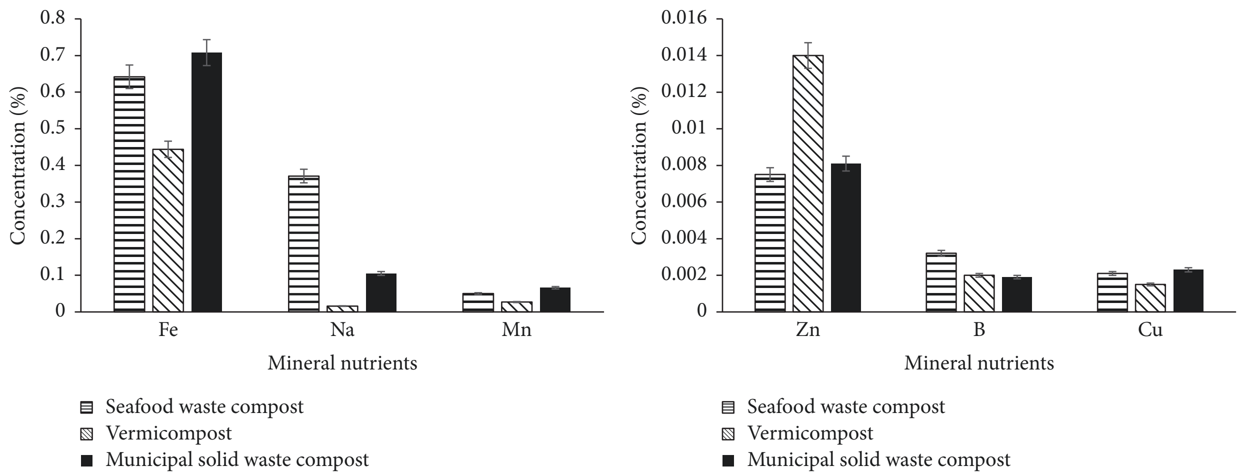

Figure 3: Micronutrients composition in seafood waste compost, vermicompost, and municipal solid waste compost. Iron (Fe); sodium (Na); manganese (Mn); zinc (Zn); boron (B); and copper (Cu).

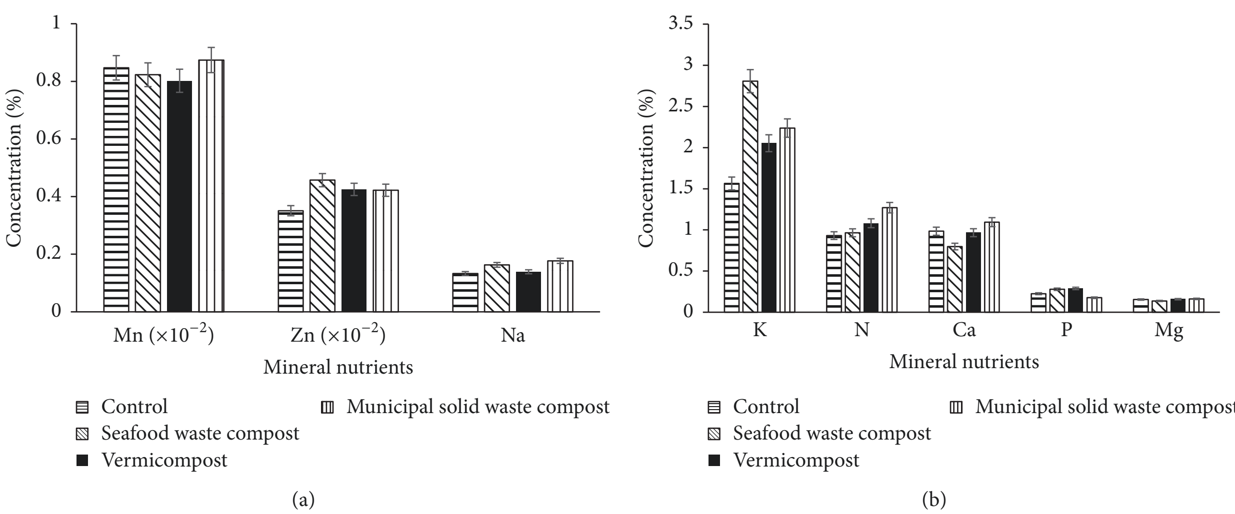

Figure 4: Micronutrients (a) and macronutrients (b) uptake by onion "Sweet Utah" plants grown in peat moss (control), seafood waste compost, vermicompost, and municipal solid waste compost. Manganese (Mn × 10^{-2}%); zinc (Zn × 10^{-2}%); sodium (Na); potassium (K); calcium (Ca); phosphorus (P); and magnesium (Mg).

not expected to be the same. Also, Hansen [8] reported reductions in amino acid content of onion bulbs as the plant ages. Therefore, the early harvest of the plants as immature green onions in the present study might be a contributory factor to the no-effect on plant growth and yield. Thus, future research should investigate and compare SFWC, MSWC, and VC effect on secondary metabolite biosynthesis in mature onion plants.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Acknowledgments

The authors wish to thank Ms. Lindsey Gillard for providing the compostable seafood waste for this project. This research benefitted from the start-up fund provided by Dalhousie University Faculty of Agriculture.

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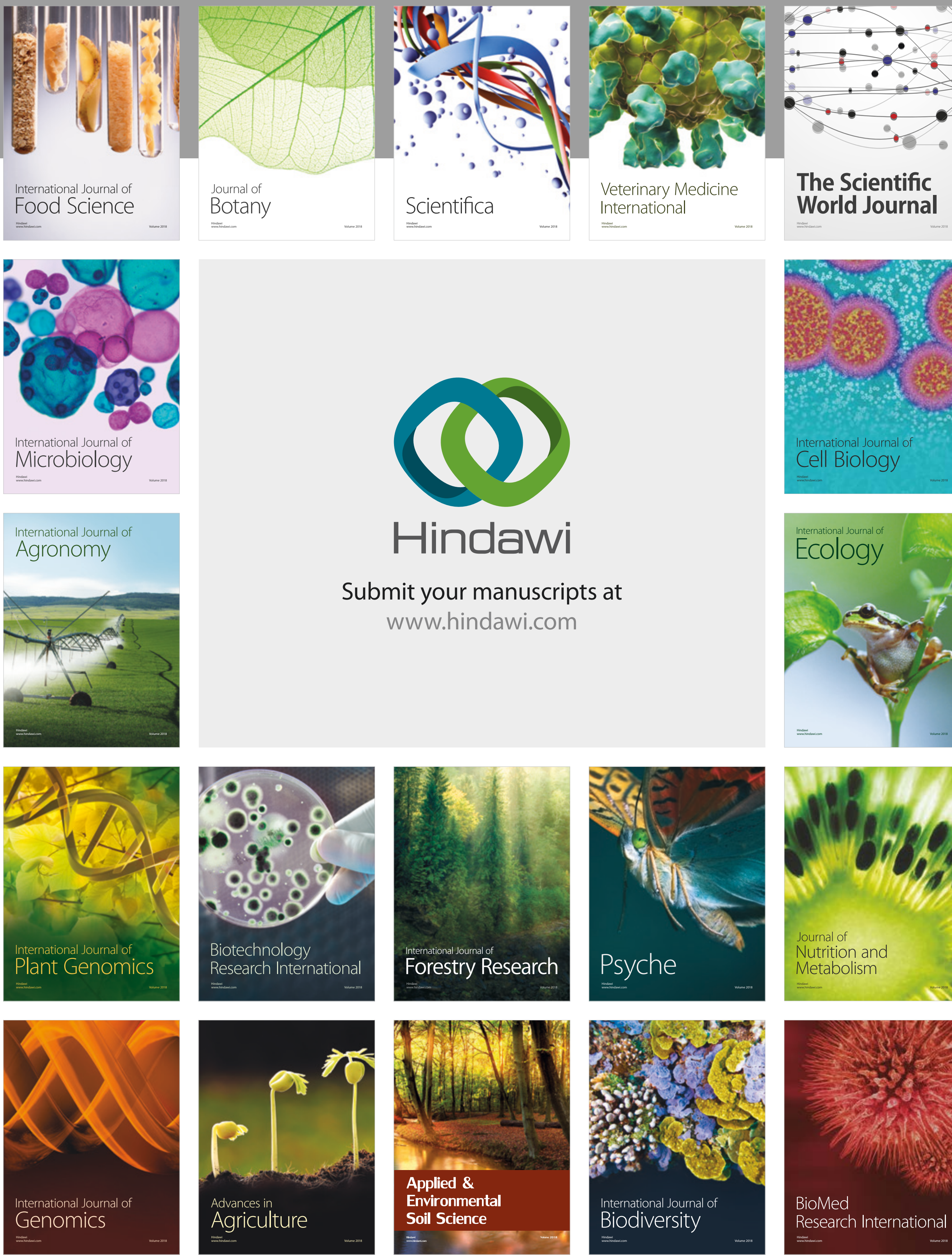
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