Constructed wetland biomass for compost production: Evaluation of effects on crops and soil

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ABSTRACT

This study investigates the suitability of Phragmites australis (reed) biomass deriving from a surface flow constructed wetland (CW) to produce three compost types: reed (RC), reed mixed + potato cuttings (PC) and reed + liquid anaerobic digestate (DC), to promote both resource circularity and soil carbon sequestration. The composts were tested over 60 days on lettuce at two levels in combination or not with NH4NO3 loading, along with NH4NO3 reference (Chem) and an unamended control (Ctrl). The plant tissue dry weight and N load was determined, and the N relative efficiency (N-RAE%) was calculated. On pot soil, total and labile carbon (TOC, Ctot), along with the carbon management index (CMI) and δ13C were evaluated. Pot test showed that PC50 yielded the best (g pot-1) lettuce biomass (3.0) > DC100 and RC100 (2.5 and 1.6) ≈ chemical reference (3.8). A similar pattern was detected at 50% (g pot-1): PC50 (2.9) > DC50 (2.7) > RC50 (2.4). N-RAE (%) reflected this pattern: PC100 (60) > DC100 (21) > RC100 (10) and PC50 (76) > DC50 (53) > RC50 (52). Pot soil analyses showed composts well performed in TOC and CMI, in comparison to Ctrl (+42% and +13%), suggesting a positive impact on soil C amelioration. No significant differences were observed for δ13C distribution, suggesting the composts did not influence the microbial metabolism differently. These results indicated that the biomass harvested from the CWs can represent an interesting material for composting, combining carbon sequestration and nutrients recycling potential of these system, in addition to their wastewater treatment capacity.

1. Introduction

Human activities, including urban, agricultural and industrial processes, produce a number of wastes, including wastewater (Peters, 2011). In particular, agricultural drainage water (ADW) is a type of wastewater which contains different contaminants (e.g. fertilisers, pesticides) used in crop production and it represents a major non-point pollution source. Since it potentially contains various harmful compounds, it requires a proper treatment prior to the discharge in the water cycle (Braschi et al., 2022; Larsen et al., 2016). In facts, ADW pollutants have been found to cause various disturbances in the receiving ecosystems, including eutrophication, groundwater contamination, soil degradation, risks to human and animal health, and loss of biodiversity (Ansari and Gill, 2014; Johannesson et al., 2017).

One of the primary research areas in the effort to treat ADW are Nature-Based Solutions (NBSs), systems where natural processes are applied to remediate contaminated water resources (Nan et al., 2023). Amongst the NBSs, constructed wetlands (CWs) are an example of NBSs that leverage the interactions between selected plants, associated microorganisms and specific substrates for wastewater treatment from various sources (Mancuso et al., 2024; Milani et al., 2019; Parde et al., 2021). Particularly effective for treating ADW, CWs commonly host low-maintenance plants species with low fertilisation, tilling and irrigation needs (Parde et al., 2021). Nitrogen (N) and phosphorus (P) are two nutrients that are essential to soil health and fertility, and that can be retained by CWs from ADW through the aboveground biomass (Kasak et al., 2020; Li et al., 2021). To avoid their return in the effluent, regular harvest is suggested, and the collected CW biomass can be employed as alternative feedstock for biogas and biochar production, and for material preparation (Cui et al., 2022; Pinho and Mateus, 2023). Another possible option is to compost the harvested plants enabling nutrient reuse and recovery (Grigatti et al., 2012; Reyes-Torres et al., 2018). The result is a product that can benefit soil quality and fertility, and whose application can reduce the reliance on mineral fertilisers (Amelung et al., 2020; Pereira et al., 2022).

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parameter for assessing soil quality: its labile fraction (C\textsubscript{f}), in facts, feeds the microbiota responsible for the nutrients cycling within the pedosphere. The distribution of C fractions, represented by the C management index (CMI), can thus be indicative of the soil functionality and health, and can provide useful information about the impact of different agricultural practices (Blair et al., 1995). Furthermore, through isotopic 13C analysis and its abundance in comparison to 12C, it is possible to evaluate the soil microbial activity, and to collect information supporting the nutrients dynamics study (Wang et al., 2015).

Within this context, the goal of the study was to evaluate the suitability of Phragmites australis (reed) aerial biomass derived from an ADW-treating CW, along with two other agricultural by-products, potato cuttings and anaerobic digestate, to produce soil amendments that can be suitable for the application in agriculture and that can be an alternative resource to chemical fertilisers, in line with former research (Arab et al., 2022; Kwarciak-Kozłowska, 2019; Fergola et al., 2020; Thu and Loan, 2024). Therefore, a Lactuca sativa (lettuce) pot test was performed in a controlled environment with different amendment application levels (ALs) to assess the compost capacity to influence and improve the crop growth, as well as its effect on soil characteristics.

2. Materials and methods

2.1. Study area

The test site was an experimental farm in the vicinity of Budrio village (Emilia Romagna Region, Italy) that also hosts a surface flow CW which treats ADW from the farm (Lavrnić et al., 2018). As specified by Regional Agency for Environmental Protection (ARPAN) from 30-year normal values, the area is characterised by a subtropical humid climate (Cfa) according to the Köppen classification of climate, with a mean annual temperature of around 13.7°C and a mean annual rainfall of 771 mm, with most of precipitations during spring and fall. The soil of the CW establishment is Udifluventic Haplustept (Soil Survey Staff, 2014), while the system has an overall surface of 0.4 ha and a storage capacity of 1477 m\textsuperscript{3}, and hosts mainly Phragmites australis, but also Iris spp. and Carex spp. (Lavrnić et al., 2020a). This CW was extensively studied in past, focusing on the aspects such as water quality (Braschi et al., 2022; Buscaroli et al., 2024; Canet-Martí et al., 2022), hydraulic behaviour (Lavrnić et al., 2020b) and sustainability assessment (García-Herrero et al., 2022). Due to its specific nature and big biomass availability, this study aimed to further explore the circularity of the system and its potential to provide soil production.

2.2. Compost preparation and analysis

As a raw compost material, fresh reed was cut from the CW, chopped to 2 cm and placed in ad-hoc constructed compost bins of 1 m\textsuperscript{3}. Three different composts were consequently produced: reed alone (compost “RC”); 25% (wt./wt.) reed added with 75% of potato cuttings (compost “PC”) obtained from the same farm; and 63% (wt./wt.) reed added with 37% of digestate (compost “DC”). The proportions chosen for DC and PC were based on the suggested C:N ratio between 20 and 30 (Hu et al., 2020). The three systems were maintained at a proper moisture level and periodically mixed for 2.5 months. Once ready, the compost aliquots were air-dried at room temperature, ground to 2 mm and stored for further analyses and for the pot test. A commercial green waste compost (“GWC”) was also used to provide a treatment reference.

Fresh compost samples were prepared according to the standard BS EN 13038:2011 for pH and electrical conductivity (EC) measurements, Total Solids (TS), volatile solids (VS) and oxygen uptake rate (OUR). Elemental composition was assessed following the procedure indicated by EPA (Campisano et al., 2017), with an Arcos-AMetek 160 nm – 780 nm ICP-OES. Soil Olsen P was measured on air-dried, milled soil samples according to the Olsen method readapted (Watanabe and Olsen, 1965).

2.3. Pot test

The soil used for the experiment was air-dried and ground to 2 mm. The characteristics are reported in Table 1. It was then mixed with the produced experimental composts, to meet 50% (AL\textsubscript{50}) or 100% (AL\textsubscript{100}) of the theoretical N required in its available form for crop cultivation, namely 280 kg N ha\textsuperscript{-1} (Ghosh et al., 2019). Thirty pots of 2 L were prepared by adding sand as drainage layer, which was covered with soil until reaching a height of 20 cm.

Six of the pots were prepared with unamended soil (without compost addition), of which three were fertilised with a first half of NH\textsubscript{4}NO\textsubscript{3} fertilising solution (“Chem” samples), and three were left unfertilised (“Ctrl” samples). Other twenty-four pots were filled with the soil-compost mixes, with the same substrate layering.

After planting the lettuce, the pots were randomly placed in a growth chamber, with controlled humidity (45%), temperature (22–26°C) and light exposure (approximately 15,000 lx), and equally irrigated with tap water at regular intervals. The pots were randomly repositioned every 15–20 days in order to ensure homogeneous conditions for all the samples. After 30 days of growth, the AL\textsubscript{50} pots were added with a chemical fertiliser to reach 280 kg N ha\textsuperscript{-1}, and the Chem pots received the second half of the N supplementation.

On the 60th day, the plants were harvested, and their aerial parts, roots and soil were separated for differentiated analyses. The plant parts were rinsed when necessary and dried in a ventilated oven at 60°C for 2 days. The soil was placed in an aerated room and dried at room temperature (c.a. 22°C). Once dried, the samples and the composts were ball-milled and collected.

2.4. Agronomic parameters calculation

N uptake, percentile Relative Agronomic Efficiency (% N-RAE) and percentile N Apparent Recovery Factor vs. the chemical control (% N-ARF) were calculated according to the following formulas.

N Uptake was calculated as shown in the Eq. 1.

\[
N \text{ Uptake}_i = \frac{Tissue \text{ TN}}{Tissue \text{ dry weight}_i}
\]

Where “i” is the considered sample; “Tissue TN” is the total N measured in either the aerial parts or the roots of the sample “i” (mg g\textsuperscript{-1}); tissue dry weight is the harvested and dried aerial or root part of the sample “i” (g).

N-RAE (%) was calculated as follows:

Table 1 Properties of the soil employed for the pot trial.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>8.25</td>
</tr>
<tr>
<td>EC</td>
<td>0.21 dS m\textsuperscript{-1}</td>
</tr>
<tr>
<td>Sand</td>
<td>25%</td>
</tr>
<tr>
<td>Silt</td>
<td>54%</td>
</tr>
<tr>
<td>Clay</td>
<td>21%</td>
</tr>
<tr>
<td>TOC</td>
<td>9.8 mg kg\textsuperscript{-1}</td>
</tr>
<tr>
<td>Organic Matter</td>
<td>1.6%</td>
</tr>
<tr>
<td>Total Carbonates</td>
<td>5.3%</td>
</tr>
<tr>
<td>Active Carbonates</td>
<td>2.2%</td>
</tr>
<tr>
<td>TN</td>
<td>0.9 mg kg\textsuperscript{-1}</td>
</tr>
<tr>
<td>Ammonia N</td>
<td>87.2 mg kg\textsuperscript{-1}</td>
</tr>
<tr>
<td>Nitric N</td>
<td>5.4 mg kg\textsuperscript{-1}</td>
</tr>
<tr>
<td>Olsen P</td>
<td>5.2 mg kg\textsuperscript{-1}</td>
</tr>
<tr>
<td>Cation Exchange Capacity</td>
<td>22.2 cmolc kg\textsuperscript{-1}</td>
</tr>
</tbody>
</table>
followed an opposite trend: GWC presented the highest content, analogous to PC; DC and RC were characterised by a lower, similar content. Consequently, the C:N ratio resulted significantly higher in RC (24) than in the other three composts (12–20). Regarding the δ^{13}C characterisation, no statistical differences were observed, with values ranging between −28.64 and −27.94%.

3.2. Biomass yield and nitrogen balance

After the two-months period of the pot test (Fig. 1 and Fig. 2), the plant aerial parts, roots and soils were separated. The weights of the dry biomasses harvested, together with their N content, are reported in Table 3. The visual assessment identified the Chem samples as the healthiest specimens (less or no aerial part chlorosis and necrosis, highest leaves number and surface), followed by GWC_{100} and PC_{100}. Despite not having received any fertilisation, the Ctrl samples appeared in the same range of PC_{100} and GWC_{100}, and healthier than DC_{100} and RC_{100}. Amongst the AL_{100} samples, PC_{100} yielded the highest average biomass, reaching the same range of the Chem reference and GWC_{100}. It was followed by DC_{100}; RC_{100} showed the worst average biomass yield, lower than DC_{100}. Considering the AL_{50}, the potato-reed compost mix resulted again to be the most performing on average, with PC_{50} and DC_{50} yielding even more than the standardised GWC_{50} and remaining nearby the Chem range. RC_{50} showed the same yield as GWC_{50}.

N-ARF and N-RAE are reported in Table 4. Chem issues showed the highest N uptake value amongst the unamended references. Amongst the AL_{100} amended samples, the detected trend is the following, from the highest to the lowest values: GWC_{100}, PC_{100}, DC_{100}, RC_{100}. At AL_{50}, potato-reed mix compost showed again the highest level, higher than GWC_{50}, that resulted similar to DC_{50} and RC_{50}. Considering N-ARF, all the treatment performed worse than Chem both at AL_{100} and AL_{50}. Regarding N-RAE, PC performed the best in both the application levels, outscoring the other treatments, especially at AL_{50}.

3.3. Soil carbon fractioning

The results of the analyses for TOC, C_{ML} and C_{L} are reported in Table 5. At the end of the two-months pot trial, the unfertilised and chemical references presented a TOC content similar to the starting soil. Regarding the AL_{100} series, RC_{100} showed the highest TOC content, DC_{100} and PC_{100} were statistically equal, while GWC_{100} had a TOC content slightly lower than the others. The samples of AL_{50} series presented an overall lower TOC content in respect to AL_{100}, with comparable values amongst samples that ranged from 16.85 to 17.39 g kg⁻¹. Regarding C_{ML}, the Chem and Ctrl references were in the range between 13.30 and 14.22 g kg⁻¹ (93.4–94.7 TOC). At AL_{100}, RC_{100} showed a significantly higher content (20.22 g kg⁻¹, 94.7% TOC), while DC_{100}, PC_{100} and GWC_{100} presented similar levels, between 18.66 and 19.09 g kg⁻¹ (94.7–95.0% TOC). At AL_{50}, the amended samples showed no significant difference and ranged between 15.39 and 16.56 g kg⁻¹ (94.7–95.3% TOC). Considering the C_{L} concentrations, the unfertilised references showed no statistical difference, with Ctrl accounting for 0.80 g kg⁻¹ (5.3% TOC). Chem had an average value of 0.90 g kg⁻¹ C_{L} (6.6% TOC). Amongst the amended samples, RC_{100} presented the highest C_{L} fraction (1.13 g kg⁻¹, 5.3% TOC), significantly different than

Table 2

<table>
<thead>
<tr>
<th>Compost</th>
<th>pH</th>
<th>EC</th>
<th>OUR</th>
<th>TS</th>
<th>VS</th>
<th>TOC</th>
<th>TN</th>
<th>TP</th>
<th>C:N</th>
<th>δ^{13}C</th>
</tr>
</thead>
<tbody>
<tr>
<td>DC</td>
<td>7.88</td>
<td>0.35</td>
<td>15</td>
<td>41.1</td>
<td>86.2</td>
<td>335</td>
<td>17.6</td>
<td>1.5</td>
<td>20</td>
<td>−27.94</td>
</tr>
<tr>
<td>PC</td>
<td>8.62</td>
<td>0.55</td>
<td>22</td>
<td>36.4</td>
<td>63.1</td>
<td>243</td>
<td>20.1</td>
<td>2.2</td>
<td>12</td>
<td>−28.19</td>
</tr>
<tr>
<td>RC</td>
<td>7.65</td>
<td>0.51</td>
<td>18</td>
<td>48.4</td>
<td>88.9</td>
<td>412</td>
<td>17.0</td>
<td>1.2</td>
<td>24</td>
<td>−28.56</td>
</tr>
<tr>
<td>GWC</td>
<td>8.91</td>
<td>0.13</td>
<td>7</td>
<td>59.0</td>
<td>52.5</td>
<td>308</td>
<td>20.8</td>
<td>3.1</td>
<td>15</td>
<td>−28.64</td>
</tr>
</tbody>
</table>

DC: digestate-reed compost; PC: potato-reed compost; RC: reed only compost; GWC: green waste compost.
the other treatments, that ranged between 0.93 (5.0% TOC) and 11.1 (5.6% TOC). However, considering the relative concentration of C in relation to TOC, the samples presented similar values, ranging between 5.0 and 5.6 g kg⁻¹. Regarding the AL group, the samples resulted more clustered, with no significative difference in between and with values ranging between 0.82 and 0.88 g kg⁻¹ (4.7–5.3% TOC), with PC and GWC on the lower level of the range. Regarding the CMI, in reference to Chem, the Ctrl samples presented an index of 79%. Considering the AL series, DC₅₀, RC₅₀ and GWC₅₀ had similar values, higher than Chem and ranging between 108% and 113% while PC₁₀₀ presented the

Fig. 1. Status of the plants at the end of the pot test, 100% application level. Ctrl: unamended control soil; Chem: chemical reference; DC: digestate-reed compost; PC: potato-reed compost; RC: reed only compost; GWC: green waste compost. Acronym subscript refers to amendment application level. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Fig. 2. Status of the plants at the end of the pot test, 50% application level. Ctrl: unamended control soil; Chem: chemical reference; DC: digestate-reed compost; PC: potato-reed compost; RC: reed only compost; GWC: green waste compost. Acronym subscript refers to amendment application level. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)
Table 3
Biomass yield and N content of leaves and roots of the harvested plants collected at the end of the pot trial from the pots cultivated at 0%, 50% and 100% organic amendment application levels.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Aerial Biomass</th>
<th>Roots Biomass</th>
<th>Total Biomass</th>
<th>Aerial N</th>
<th>Roots N</th>
<th>Total N</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(g pot⁻¹)</td>
<td>(g pot⁻¹)</td>
<td>(mg pot⁻¹)</td>
<td>(mg pot⁻¹)</td>
<td>(mg pot⁻¹)</td>
<td>(mg pot⁻¹)</td>
</tr>
<tr>
<td>Ctrl</td>
<td>2.1±</td>
<td>0.3±</td>
<td>2.6±</td>
<td>39.0±</td>
<td>3.9±</td>
<td>42.9±</td>
</tr>
<tr>
<td>Chem</td>
<td>3.1±</td>
<td>0.7±</td>
<td>3.8±</td>
<td>76.5±</td>
<td>7.2±</td>
<td>83.7±</td>
</tr>
<tr>
<td>DC100</td>
<td>1.6±</td>
<td>0.9±</td>
<td>2.5±</td>
<td>14.3±</td>
<td>5.3±</td>
<td>19.5±</td>
</tr>
<tr>
<td>PC100</td>
<td>2.8±</td>
<td>0.3±</td>
<td>3.0±</td>
<td>40.7±</td>
<td>1.8±</td>
<td>42.5±</td>
</tr>
<tr>
<td>RC100</td>
<td>0.9±</td>
<td>0.1±</td>
<td>1.0±</td>
<td>6.6±</td>
<td>4.6±</td>
<td>11.2±</td>
</tr>
<tr>
<td>GWC100</td>
<td>2.3±</td>
<td>0.5±</td>
<td>2.8±</td>
<td>39.0±</td>
<td>4.9±</td>
<td>44.0±</td>
</tr>
<tr>
<td>DC50</td>
<td>2.0±</td>
<td>0.7±</td>
<td>2.7±</td>
<td>40.3±</td>
<td>2.3±</td>
<td>42.6±</td>
</tr>
<tr>
<td>PC50</td>
<td>2.4±</td>
<td>0.2±</td>
<td>2.6±</td>
<td>57.9±</td>
<td>4.2±</td>
<td>62.1±</td>
</tr>
<tr>
<td>RC50</td>
<td>1.8±</td>
<td>0.6±</td>
<td>2.4±</td>
<td>39.5±</td>
<td>4.3±</td>
<td>43.8±</td>
</tr>
<tr>
<td>GWC50</td>
<td>1.6±</td>
<td>0.8±</td>
<td>2.4±</td>
<td>40.2±</td>
<td>7.7±</td>
<td>47.9±</td>
</tr>
</tbody>
</table>

Table 4
Tissues N apparent recovery fraction (N-ARF) and relative agronomic efficiency (RAE) of the harvested plants leaves, collected from the pots cultivated at 0%, 50% and 100% organic amendment application levels.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Leaves</th>
<th>Roots</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N-ARF (%)</td>
<td>N-RAE (%)</td>
</tr>
<tr>
<td>Ctrl</td>
<td>Chem</td>
<td>DC100</td>
</tr>
<tr>
<td>N-ARF</td>
<td>20.6±</td>
<td>11.4±</td>
</tr>
<tr>
<td>N-RAE</td>
<td>19.8±</td>
<td>0.9±</td>
</tr>
</tbody>
</table>

Table 5
Results for the analyses of total organic carbon (TOC), non-labile (C₄₀), labile carbon (C₃₀) and carbon management index (CMI) of the soil samples collected at the end of the pot trial. Soil total nitrogen (TN) and plants to soil N:N ratio are also reported.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>TOC (g kg⁻¹)</th>
<th>C₄₀ (g kg⁻¹)</th>
<th>C₃₀ (g kg⁻¹)</th>
<th>CMI (%)</th>
<th>TN (g kg⁻¹)</th>
<th>N:N (Tissues to Soil)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ctrl</td>
<td>14.8±</td>
<td>14.2±</td>
<td>0.8±</td>
<td>79±</td>
<td>1.8±</td>
<td>13.8±</td>
</tr>
<tr>
<td>Chem</td>
<td>15.0±</td>
<td>13.9±</td>
<td>0.9±</td>
<td>100±</td>
<td>2.2±</td>
<td>19.7±</td>
</tr>
<tr>
<td>DC100</td>
<td>19.7±</td>
<td>18.6±</td>
<td>1.1±</td>
<td>110±</td>
<td>2.1±</td>
<td>11.6±</td>
</tr>
<tr>
<td>PC100</td>
<td>20.0±</td>
<td>19.0±</td>
<td>0.9±</td>
<td>90±</td>
<td>2.2±</td>
<td>13.2±</td>
</tr>
<tr>
<td>RC100</td>
<td>21.3±</td>
<td>20.2±</td>
<td>1.1±</td>
<td>112±</td>
<td>2.2±</td>
<td>7.3±</td>
</tr>
<tr>
<td>GWC100</td>
<td>19.3±</td>
<td>18.9±</td>
<td>1.0±</td>
<td>108±</td>
<td>2.1±</td>
<td>12.8±</td>
</tr>
<tr>
<td>DC50</td>
<td>16.5±</td>
<td>15.9±</td>
<td>0.8±</td>
<td>87±</td>
<td>2.0±</td>
<td>13.4±</td>
</tr>
<tr>
<td>PC50</td>
<td>17.3±</td>
<td>16.5±</td>
<td>1.0±</td>
<td>81±</td>
<td>2.1±</td>
<td>13.2±</td>
</tr>
<tr>
<td>RC50</td>
<td>16.5±</td>
<td>15.6±</td>
<td>0.8±</td>
<td>86±</td>
<td>1.9±</td>
<td>12.4±</td>
</tr>
<tr>
<td>GWC50</td>
<td>15.5±</td>
<td>14.7±</td>
<td>0.8±</td>
<td>73±</td>
<td>1.9±</td>
<td>12.2±</td>
</tr>
</tbody>
</table>

Table 6
The Olsen P assessed in the soils collected after the two months pot trial. The unfertilised Ctrl and the Chem reference resulted to have, respectively, the lowest and the highest labile Olsen P content. Regarding AL100, GWC100 presented the highest value, similar to PC100, DC100 a slightly lower and RC100 presented the lowest value. Concerning the AL50 samples, RG50 was characterised by the highest Olsen P value, significantly higher than GWC50, DC50 and PC50 presented similar values.

4. Discussion

4.1. Main composts characteristics

The different materials employed for composting, namely the reed, the potato cuttings and the digestate, played a critical role on the final characteristics of the organic products which was in line with previous research (Oviedo-Ocana et al., 2019). In particular, the substances obtained by composting reed alone (RC) and reed mixed with the digestate (DC) showed higher OM, TOC and C:N ratio than the PC and GC. All the products microbial stability (OUR) complied with the maximum threshold imposed by the European Fertiliser Regulation of 25 mmol O₂ kg⁻¹ organic matter h⁻¹ and could, therefore, be considered as safe for agricultural applications (Council of the European Union, 2019).

4.2. Biomass yield and nitrogen balance

The higher C:N ratios could have caused an imbalance in the nutrient dynamics within the soil of RG100 and DC100, which showed worse health, biomass yield and N uptake levels in comparison to PC100 and GWC100. This is consistent with former studies, where compost C:N ratios higher than 20 caused N immobilisation by microorganisms and lead to a decreased biomass production, while lower values were shown to favour microbial N mineralisation and, consequently, higher bioavailability and biomass outputs (Brust, 2019; van der Sloot et al., 2022). In fact, a reduced N availability was shown to cause biomass reduction due to the plants diversion from development towards survival (Mu and Chen, 2021). This aspect is also reflected by the patterns observed for N-ARF and N-RAE, and by the ratio between the tissue N and the soil N content in relation to the C:N ratio of the applied AL100 group and ranging between −0.65 and −0.39.

3.5. Soil phosphorus fractioning

Table 6 reports the Olsen P assessed in the soils collected after the two months pot trial. The unfertilised Ctrl and the Chem reference resulted to have, respectively, the lowest and the highest labile Olsen P content. Regarding AL100, GWC100 presented the highest value, similar to PC100, DC100 a slightly lower and RC100 presented the lowest value. Concerning the AL50 samples, RG50 was characterised by the highest Olsen P value, significantly higher than GWC50, DC50 and PC50 presented similar values.

As reported in Fig. 3, Ctrl and Chem presented a statistically similar isotopic δ¹³C signature (−24.12 and −24.13%, respectively). The AL100 samples were found to have a significant depletion of δ¹³C. GWC100 presented the lowest value (−25.50%), followed by RC100 (−25.36%), PC100 (−25.23%) and DC100 (−25.04%). The chemical fertilisation applied to AL50, caused the samples to have a lower depletion in comparison to the unamended Ctrl, with no significant reciprocal difference and values ranging between −24.78 and −24.52%.

Fig. 3 shows the Δ¹³C in respect to Chem as well. Ctrl resulted to have no significant difference in respect to Chem, with a δ¹³C of 0.01. All the amended samples presented a negative Δ¹³C, with the AL100 series showing lower average values, and no significant differences between each other, ranging between −1.37 and −0.91. Similarly, at AL50, no significant difference was recorded, with average values higher than the lowest value. The samples resulted more clustered at AL50, with no significant difference between DC50, PC50 and RG50, and with values ranging between 81 and 87%. GWC50 presented the lowest value, 76%.

3.4. Soil ¹³C signature

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composts. It emerged that tissues-to-soil N ratio tended to increase with lower compost starting C:N ratio, suggesting a higher bioavailability with lower ratios, as previously stated. (Table 2 and Table 5).

The chemical supplementation of the AL samples seemingly boosted the health of plants, that produced aerial parts in similar quality and quantity and that presented similar levels of N uptake. An exception was PC, that presented higher levels in all the considered parameters, arguably due to the lower C:N ratio (~26.7%) (van der Sloot et al., 2022). Chemical N fertilisation in addition to organic amendment, especially with high C:N ratios, was shown to shift the plants dependence from the microbial community composition towards the soil composition. In facts, AL samples were more clustered, since the chemical N supplementation arguably reduced the variability caused by the different amendment composition, rendering the plants more dependent upon the soil composition (Bossolani et al., 2022). Noteworthy, such supplementation produced specimens with outputs similar to the Chem samples: a partial substitution of the chemical fertiliser can induce yields comparable to a chemical-only fertilisation, thus presenting the opportunity to reduce the consumption of mineral resources and to increase the overall sustainability (Jin et al., 2022).

### 4.3. Soil carbon fractioning

The rise observed in C content within the amended samples (between 2% and 48% in relation to the Ctrl soil) indicated the positive effect of employing reed as compost feedstock in terms of C sequestration and soil enrichment in TOC content, especially in contrast to a chemical-only fertilisation, as shown by previous research (Wu et al., 2023). Most of the amended samples (except for RC and GWC) presented higher TOC and CMI than the unamended soils. This was in accordance with previous research, underlining the sustainability of GWs not only as a remediation technology, but also as carbon-mitigating systems, which allow the sequestration of atmospheric C and integration into the pedosphere (Cooper and DeMarco, 2023; Wijesekara et al., 2021).

Despite the differences observed in the TOC concentrations, the samples had similar C to TOC percentages, that ranged between 5.0% and 5.6%. The organic C added through amendment apparently presented a low labile fraction, and did not seem to contribute to almost any extent to the labilisation of the overall C. Conversely, it was observed that the chemical fertilisation of the Chem samples somewhat increased the C fraction in comparison to the other treatments, presenting a 27% higher relative C content, suggesting that a high chemical supplementation may render the already present C more bioavailable (Mayer et al., 2022). These aspects should be further explored, e.g. by studying the effect of different organic-chemical fertiliser proportions on the labilisation of the carbon, especially in relation to the added TOC. In contrast to the previously presented results, regarding the plants health and N management, Chem and GWC soil samples featured lower CMI values than DC and RC samples. The soils amended with DC and RC could be characterised by a higher C stability but a lower N bioavailability, due to their C:N ratio, as discussed before. In fact, since CMI is an index that focuses on the soil health, it does not consider other critical factors (e.g. nutrients bioavailability). Consequently, even at high CMI, the plants growth conditions may be sub-optimal, e.g. due to N availability reduction caused by microbial immobilisation (Brust, 2019; van der Sloot et al., 2022). On the other hand, this problem could be addressed by preparing the amendments of different composition. In particular, it would be important to reach lower C:N ratios, that can facilitate N labilisation and, consequently benefit plants nutrition (Brust, 2019; van der Sloot et al., 2022).

### 4.4. Soil $^{13}$C signature

Considering the negligible variation of $\delta^{13}$C amongst the samples
within each application level, it can be hypothesised that the applied composts did not particularly influence the biotic metabolism within the soil. This is consistent with the starting statistically similar compost δ13C values, as well as with the lower isotopic abundance found for the AL60 samples when compared to the AL100 ones. The difference observed in comparison to the baseline and between the two application levels are probably related to the addition of C through the composts and not to differences in the biotic metabolism (Menichetti et al., 2013).

4.5. Soil phosphorus fractioning

P, distribution reflected the pattern observed for the plant health and N management, with PC100 and GWG100 presenting the highest Olsen P content amongst the amended samples. This was arguably another aspect that could have contributed to such plants performances and have favoured their metabolism (Lizzano-Toledo et al., 2021). In contrast to the labile vs. total C (C2 vs. TOC) ratios, the labile vs. total P (Olsen P vs. TP) ratios followed the distribution of Olsen P itself, indicating that the P labile fraction remained apparently unaltered and proportional to the TP variation observed ex post. This might be related to aspects analogous to the ones influencing the C lability, as previously observed. Chemical N fertilisation may have promoted the internal soil dynamics, promoting the labilisation of the mineralised nutrients and a better absorption capacity of plants (Ciui and Delgado, 2021).

5. Conclusions

The agricultural sector is one of the most important polluters of the water resources and constructed wetlands (CWs) are a valid method to treat surface runoff or drainage water from agricultural areas. One of the most common plants applied in CWs, reed was found to be unfit for direct application, if composted alone, requiring a partial chemical fertilisation. While a reed-digestate mixture showed results similar to the reed-only, potato cuttings seemed to be a promising material for increasing the compost quality, yielding healthier lettuce specimens, both in appearance and in terms of N management.

The reed-only and reed-digestate composts presented higher values (labile C and C Management Index) than the reed-potato mix. This aspect, however, did not account for other fundamental factors for plants growth, such as N. A partial chemical N fertilisation granted a considerable boost for all the amended samples, indicating a possibility to use CW-derived organic compost as a partial substitution of the chemical fertilisation and as a way for C relocating from the atmosphere into the soil. Additional analyses are suggested to explore further conditions, such as different composting and cultivation times, re-wastes mixes and organic vs. chemical proportions, as well as to better comprehend the dynamics taking place within the soil amongst different components (e.g. microorganisms, plants, soils, nutrients).

6. Glossary

13C isotope: naturally occurring carbon isotope useful for the determination of the carbon conservation and stability within soil (Inacio et al., 2018).

Agricultural drainage water: water deriving from precipitations upon and irrigation of cultivated crops, usually collecting in perimetal ditches surrounding the cultivated areas (Braschi et al., 2022).

Apparent Recovery Fraction, nitrogen (N-ARF): plants uptake of nitrogen, calculated in reference to a control sample, e.g. unfertilised, and employed for evaluating the impact of different treatments (Santos et al., 2018).

Bioavailability: availability of a certain element or compound to be readily absorbed and utilised by organisms, generally plants and microbes.

Carbon Management Index (CMI): index calculated from the soil Carbon Pool Index (CPI) and Lability Index (LI), which is used to evaluate soil health in terms of carbon storage and availability (Blair et al., 1995).

Constructed wetland: an engineered system where selected plants, their symbiotic microorganisms and particular substrates are applied for the treatment of polluted water or soil (Parde et al., 2021).

Carbon Pool Index (CPI): index calculated as ratio of total organic carbon (TOC) quantified in a sample and a reference, e.g. a chemically fertilised soil (Blair et al., 1995).

Green waste compost: compost produced from plants parts, in particular from plants and grasses cleaning and pruning byproducts, originating from sources such as municipal parks, domestic dwellings and gardens (Reyes-Torres et al., 2018).

Labile C: readily oxidisable carbon fraction, determined e.g. through the permanganate oxidation method, present in a soil and available to organisms for their metabolism, also in function of the total organic carbon (see “Total Organic Carbon”) (Blair et al., 1995).

Labile P: phosphorus fraction readily available to organisms for their metabolism, determined e.g. through bicarbonate extraction (Watanabe and Olsen, 1965).

Lability (carbon): ratio between the labile carbon present in a soil and the total organic carbon content of the same soil (Blair et al., 1995).

Lability Index (LI): ratio between the carbon lability of a sample soil and the carbon lability of a reference, e.g. a chemically fertilised soil (Blair et al., 1995).

Relative Agronomical Efficiency, nitrogen (N-RAE): index that compares the agronomic effects of an organic fertiliser with the effects of a chemical fertiliser, with focus on nitrogen (Santos et al., 2018).

Soil Organic Carbon (SOC): refer to “Total Organic Carbon (TOC)”. Total Organic Carbon (TOC): total carbon present in a sample and bound to organic molecules, directly influencing soil chemical, physical and biological characteristics, also in function of its labile fraction (see “Labile C”) (Vieira et al., 2007).

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CRediT authorship contribution statement

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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.

Data availability

Data will be made available on request.

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