Validation of Biochar as a Soil-Amending Substrate

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Introduction

Residual biomass valorization is a central principle of circular bioeconomy systems, and biochar, produced through thermochemical conversion routes including hydrothermal processing, has emerged as a promising value-added product for the productive reuse of biomass side-streams in agronomic and environmental applications. This concept is usually applied to olive pruning residues and wheat straw fractions generated during the production of advanced biofuels, as part of a zero-waste strategy and efficient use of resources (Mathanker, 2021; Alburquerque, 2013; Supraja, 2023).

In this framework, this study presents the physicochemical characterization and greenhouse validation of biochar as a soil-amending substrate, with emphasis on agronomic performance, environmental functionality, and carbon sequestration potential.

Materials and Methods

The experimental work was carried out in greenhouse pot trials using lettuce seedlings as the model crop. Lettuce was selected because it is suitable for winter cultivation and has relatively low water requirements, allowing irrigation to be adjusted according to substrate dryness. The methodological framework was developed through two consecutive experimental cycles: a preliminary greenhouse optimization stage and a subsequent biochar validation stage.

A broad analytical scheme was applied for both cycles. Initial, intermediate, and final soil measurements included pH, electrical conductivity, total solids (TS), volatile solids (VS), humidity, total organic carbon (TOC), total nitrogen (TN), water dynamics, heavy metals, and nutrient content.

After harvest, leaves and roots were analyzed for TS, humidity, TOC, TN, total chlorophyll, heavy metals, and nutrient content, while optical and developmental indicators were also recorded to assess plant growth. Root development was further examined through dimensional observations, allowing evaluation of the capacity of the root system to expand within each substrate.

Prior to the agronomic trials, both types of biochar used underwent physicochemical analysis to determine their pH, EC, TS and VS, TOC, total nitrogen, heavy metals and nutrient content.

Experimental Cycle 1: Preliminary Greenhouse and Substrate Assessment

The first experimental cycle was designed as a preliminary trial without biochar, with the primary aim of ensuring the proper operation of the greenhouse system. This cycle lasted one month and included two soil substrates, Ericaceous Potting Soil and Enriched Garden Soil, under two treatment conditions: chemical fertilization and no fertilization (control). Each condition was tested in duplicate. The role of this cycle was therefore methodological, providing an initial basis for substrate selection, irrigation management, sampling design, and analytical workflow development before the introduction of biochar.

Experimental Cycle 2: Biochar Validation Trial

The second experimental cycle constituted the first dedicated biochar validation trial, aiming to assess the effectiveness of biochar as a soil amendment and to contribute to the estimation of its carbon sequestration potential. The experimental comparison employed Enriched Garden Soil, amended with biochar, chemical fertilizer, and no fertilization (control). Two biochar types were tested: Olive Branch biochar (B1) and Wheat Straw biochar (B2), each applied at 0, 2, and 5 t/ha equivalent dosages. The planned cultivation period was three months and each condition was duplicated.

Results and Discussion

The available measurements indicated that both biochars had very low moisture content (approximately 2.7–3.2%) and were slightly acidic (pH 5.66-5.10). Wheat-straw biochar showed a lower pH and higher EC than olive-branch biochar, suggesting a higher content of soluble salts. In terms of carbon-related properties, olive-branch biochar showed higher TOC (81.12%) and a higher C/N ratio (73.08), whereas wheat-straw biochar presented lower TOC (71.70%) and relatively greater nitrogen participation (1.36% over 1.11%). Nutrient and mineral analysis showed that olive-branch biochar was particularly rich in Ca (29.51 g/kg), while wheat-straw biochar contained markedly higher Mg (11.83 g/kg) and somewhat higher K. In terms of trace metals, both biochars showed elevated Ni (423.36 and 477.35 mg/kg), while wheat-straw biochar also contained substantially higher Cr (106.15 mg/kg), indicating that the two materials may differ not only in agronomic performance but also in environmental suitability and handling requirements.

Experimental Cycle 1 provided important methodological and interpretative insights that helped refine the design of the subsequent biochar trial. A slight increase in pH and EC was observed across substrate types, likely reflecting mild salt accumulation and fertilizer-related nitrate dynamics, while the stronger increase in C/N ratio in Ericaceous soils suggested relative nitrogen depletion during cultivation.

Elemental observations showed that several metals accumulated more strongly in roots than in leaves, with Cu and Ni particularly enriched in roots, whereas Cr was rarely detected, and Mn and Zn were present in both organs. Leaves showed higher TOC and TN and lower C/N values than roots, indicating more metabolically active tissues, while roots had a more structural composition. Visual measurements also revealed substrate-dependent growth responses, with one substrate favoring leaf production and another promoting shoot height and root elongation, thereby informing substrate selection for the second cycle.

At the start of Experimental Cycle 2, all soil substrates exhibited a basic pH and relatively moderate electrical conductivity, indicating balanced soils rich in nutrients and suitable for cultivation. At the same time, due to the content of volatile solids (7–10%), it is concluded that there is sufficient organic matter, and water and nutrient retention in the soil is enhanced. In conjunction with this, an increase in total organic carbon is observed in substrates containing any type of biochar, along with increased total nitrogen, which enhances plant growth.

The results suggest that the application of both types of biochar at a rate of 5 t/ha contributed more to the accumulation of Cr, Ni, Mn, and Cu than to the improvement of nutrient availability, compared to the control group soils and those treated with chemical fertilizer. In contrast, biochar at a rate of 2 t/ha appeared to enhance nutrient availability in one replicate, particularly for P-PO_4, K, Ca, Mg, and Fe, although significant variability was observed among the samples, possibly due to the type of biochar. The application of chemical fertilizer did not lead to a clear increase in measured P-PO_4 or K, while Cd remained undetectable and Pb and Zn remained at low to moderate levels in all treatments.

Visual measurements of the lettuce plants were taken to determine their height and the number of leaves for all substrate treatments. The results are summarized as growth rates from the first recording as follows (Table 1).

Table 1. Percentage growth of optical indicators for all substrates

	No. of leaves	Height
Control	48.6%	40.78%
Soil+ Chemical Fertilizer	25%	32.8%
Soil + Biochar 2 t/ha	39%	38.4%
Soil + Biochar 5 t/ha	49.9%	60%

Conclusions

Regarding the visual indicators of plant growth, some key conclusions have emerged from the data recorded. In general, a significantly greater increase in plant height and leaf mass was observed in pots containing a substrate with biochar; specifically, a large increase in the number of leaves, by approximately 39%, for a concentration of 2 t/ha, and more significant leaf development, at approximately 60%, for a concentration of 5 t/ha. The percentages for pots containing chemical fertilizer are, on average, 25% and 32.8%, respectively.

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References

Alburquerque, J.A., Salazar, P., Barrón, V., Torrent, J., del Campillo, M.C., Gallardo, A. and Villar, R., 2013. Enhanced wheat yield by biochar addition under different mineral fertilization levels. *Agronomy for Sustainable Development*, 33, pp.475–484. https://doi.org/10.1007/s13593-012-0128-3

Mathanker, A., Das, S., Pudasainee, D., Khan, M., Kumar, A. and Gupta, R., 2021. A review of hydrothermal liquefaction of biomass for biofuels production with a special focus on the effect of process parameters, co-solvents, and extraction solvents. *Energies*, 14(16), p.4916. https://doi.org/10.3390/en14164916

Supraja, K.V., Kachroo, H., Viswanathan, G., Verma, V.K., Behera, B., Doddapaneni, T.R.K.C., Kaushal, P., Ahammad, S.Z., Singh, V., Awasthi, M.K. and Jain, R., 2023. Biochar production and its environmental applications: Recent developments and machine learning insights. *Bioresource Technology*, 387, p.129634. https://doi.org/10.1016/j.biortech.2023.129634