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# Use of biomass-derived biochar as a sustainable material for carbon sequestration in soil: recent advancements and future perspectives



Basanta Kumar Biswal & Rajasekhar Balasubramanian✉

The application of biomass-derived carbon materials (e.g., biochar) into soil is considered as an attractive and sustainable strategy to enhance carbon sequestration in soil and to mitigate climate change. Our comprehensive literature analysis shows that the carbon sequestration potential of biochar in soil systems varies between 0.7 and 1.8 Gt CO<sub>2</sub>-C<sub>(eq)</sub>/year. Biochar with high stability and C/N ratios is effective to achieve significant carbon sequestration in soil. Furthermore, carbon sequestration is usually favourable at high biochar application rate in soil with high porosity and alkaline pH (>7.5). The dominant bacterial communities enriched in the biochar-amended soil include *Proteobacteria* and *Acidobacteria*, while *Ascomycota* dominates the fungal communities. The impact of biochar amendment on soil microbial biomass and communities depends on the biochar particle size, porosity and application rate. Life cycle assessment (LCA) of biochar-amended soil reveals that biochar produced from waste biomass is found to be environmentally friendly with the acceptable level of economic feasibility in terms of large-scale applications. The recommended future research directions to seek practical applications of biochar amendment in soil include (1) development of biochar-microbe co-engineering strategies to stabilize labile carbon fractions in soil, (2) exploration of machine learning tools to optimize biochar properties for adoption of biochar treatment under region-specific soil conditions, and (3) standardization of carbon accounting methodologies to address and resolve discrepancies in LCA studies. We believe that this comprehensive review would help for development of novel biochar to achieve optimum carbon sequestration efficiency in soil and to develop practical climate change mitigation strategies.

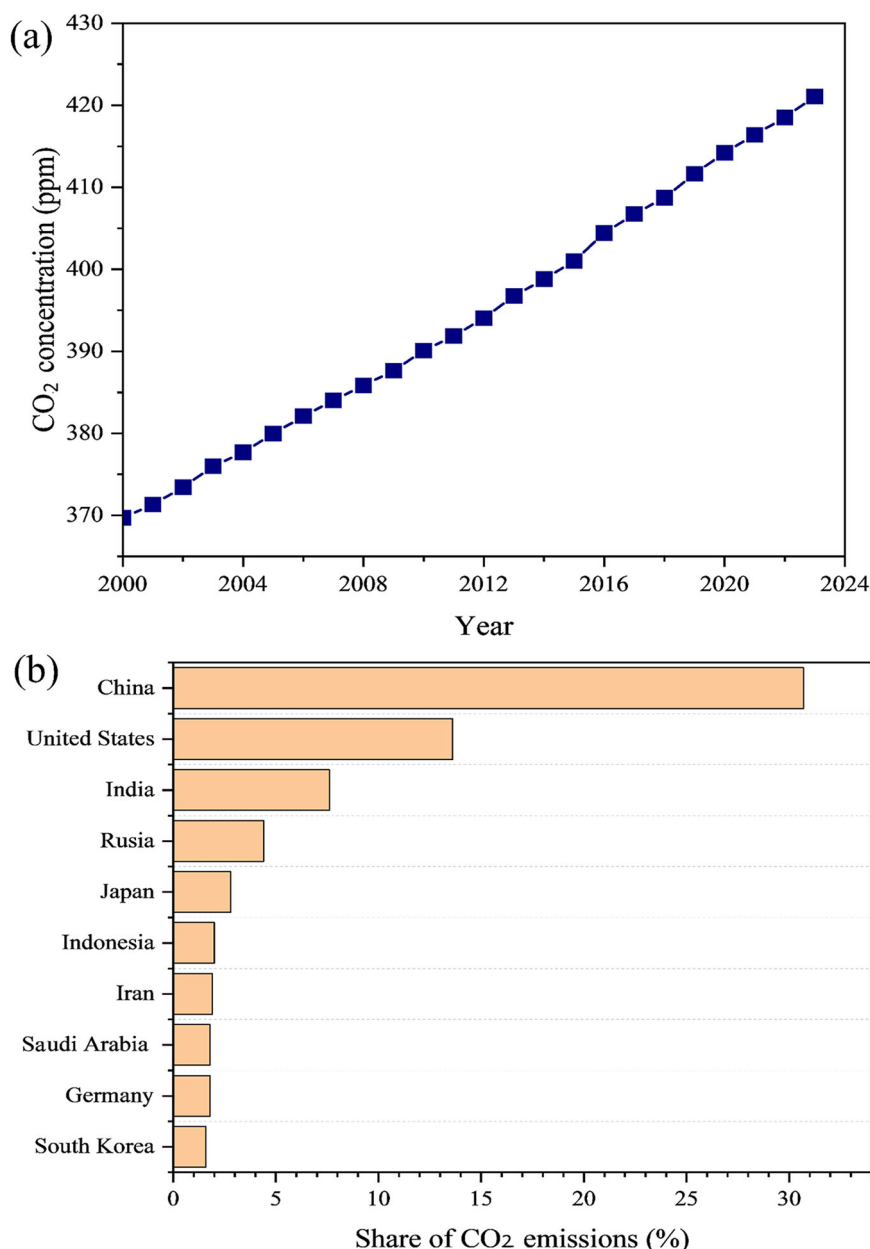
According to the latest Intergovernmental Panel on Climate Change (IPCC) assessment report<sup>1</sup>, human activities have caused a considerable increase in greenhouse gases (GHG) emissions to the atmosphere with the carbon dioxide (CO<sub>2</sub>) concentration reaching up to 422 ppm in 2024. It is predicted that the concentration of CO<sub>2</sub> in the atmosphere will reach to 600–700 ppm by the end of this century, if no concrete actions are taken to reduce their emissions at the global level<sup>2</sup>. The increasing trend in global average annual CO<sub>2</sub> concentrations in the atmosphere from 2000 to 2023 is presented in Fig. 1a, and the distribution of CO<sub>2</sub> emissions among top 10 countries is given in Fig. 1b. The data shows that CO<sub>2</sub> levels in the atmosphere have been constantly increasing over the last 24 years from its value of 369.71 ppm in 2000 by nearly 12% increase in 2023 (421.08 ppm). Among various countries,

China is the top emitter of CO<sub>2</sub> (30.7% share of global CO<sub>2</sub> emissions), which can be attributed to its higher population and industrial growth. The continual rise of GHG emissions will present a critical challenge to meet the international goal of limiting global warming to less than 2 °C compared to the pre-industrial time<sup>3</sup>. Thus, in recent years, significant efforts have been made worldwide to develop novel strategies to reduce GHG emissions/carbon footprints to combat global warming, control climate change and protect the planet.

Carbon capture and storage (CCS) is an efficient strategy to reduce the accumulation of anthropogenically-derived CO<sub>2</sub> in the atmosphere<sup>4</sup>. One promising way to reduce CO<sub>2</sub> levels in the atmosphere is by sequestering CO<sub>2</sub> into soil since soil has the CO<sub>2</sub> storage capacity of 2–3

Department of Civil and Environmental Engineering, National University of Singapore, Singapore, Singapore. ✉e-mail: [ceerbala@nus.edu.sg](mailto:ceerbala@nus.edu.sg)

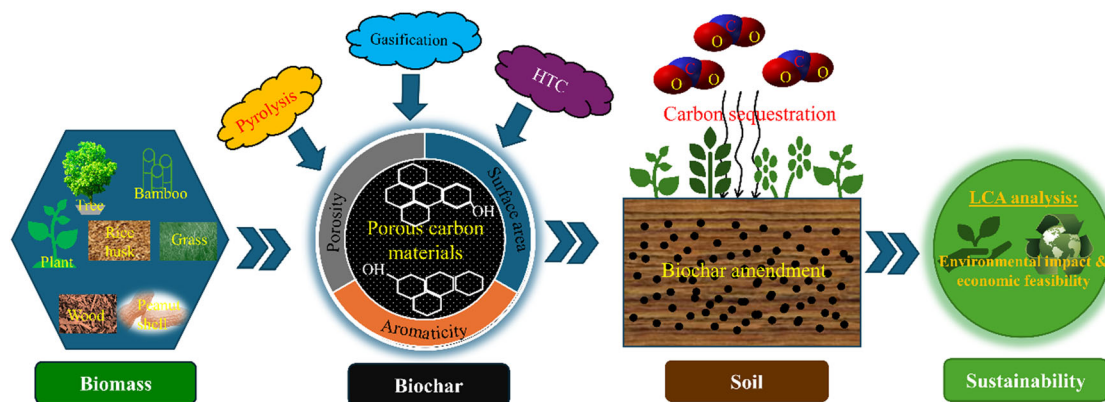
**Fig. 1 | Global atmospheric CO<sub>2</sub> concentration.** Average CO<sub>2</sub> concentrations in the atmosphere worldwide from 2000 to 2023<sup>192</sup> (a). Distribution of CO<sub>2</sub> emissions among the top 10 countries in 2022 (b)<sup>193</sup>.



times higher than the atmosphere and 3–4 times higher than the vegetation<sup>5</sup>. Moreover, CO<sub>2</sub> is nearly 50 times more concentrated in soil compared to in air<sup>6</sup>. Carbon sequestration is mainly associated with the accumulation of carbon in a stable solid form<sup>7</sup>. In recent years, biomass-derived carbon materials (e.g., biochar) are receiving much attention as they can provide a nature-based green solution to enhance the carbon sequestration capacity of soil and thus the mitigation of climate change<sup>8</sup>. Biochar amendment to soil for carbon sequestration has garnered global attention as an eco-friendly strategy<sup>3</sup>. Adsorption of soil organic carbon (SOC) onto biochar results in the reduction of soil CO<sub>2</sub> fluxes i.e., CO<sub>2</sub> release into the atmosphere<sup>9</sup>. Biochar, produced from plant biomass, usually demonstrates higher carbon sequestration potential compared to biochar synthesized from other biomass sources due to higher C/N ratio in plant biomass<sup>10</sup>. Moreover, biochar with greater stability and higher carbon retention capacity can contribute to achieving carbon neutrality, thus addressing the pressing global issue of climate change. The stability/residence time of biochar in soil depends on both the physicochemical properties of biochar and soil conditions<sup>11</sup>.

Biochar (also called black carbon) is a porous carbonaceous material which is obtained from plant biomass, woody materials, plant-based residues, etc. by thermal processes<sup>7,12</sup>. The key thermal processes used for the synthesis of biochar include slow and fast pyrolysis, and these processes are conducted in a sealed container at an elevated temperature (100–1000 °C) under limited oxygen environment<sup>13–16</sup>. Pyrolysis of biomass results in the preservation of 50% of carbon in its original substrate which is highly stable and present in soil for longer time<sup>7</sup>. The physiochemical characteristics of biochar depend on many factors including the properties of biomass/feedstock materials and adopted thermal synthesis conditions (temperature and residence time)<sup>13</sup>. Biomass-based biochar is recognized as a carbon negative material due to low GHG emissions during its production and long-term carbon sequestration<sup>13</sup>. The carbon sequestration potential of biochar in soil varies in the range of 0.7–1.8 Gt/Pg CO<sub>2</sub>-C<sub>(eq)</sub>/year<sup>11,17</sup>.

In addition to carbon sequestration, biochar amendment also improves soil health and fertility<sup>18</sup>. Furthermore, biochar is used for reclamation of degraded soil<sup>19</sup>. Biochar positively impacts the physicochemical properties (e.g., pH and cation exchange capacity) as well as the microbial



**Fig. 2 | Schematic presentation on the overall scope and objectives of this comprehensive review.** HTC hydrothermal carbonization.

communities dynamics and enzymatic activities in the soil ecosystem<sup>9</sup>. Biochar amendment can enhance gross SOC by nearly 27%<sup>20</sup>. From the agronomy perspectives, biochar amendment into soil improves soil fertility and decreases nutrient leaching, thus enhancing plant growth and crop productivity/agronomy yield<sup>19,21</sup>. Moreover, biochar amendment reduces soil bulk density and increases soil productive capacity and plant growth<sup>17,22</sup>. Amendment of biochar into soil could also help to achieve United Nations sustainable development goals (SDGs) such as climate action (SDG 13), clean water and sanitation (SDG 6), etc<sup>20</sup>.

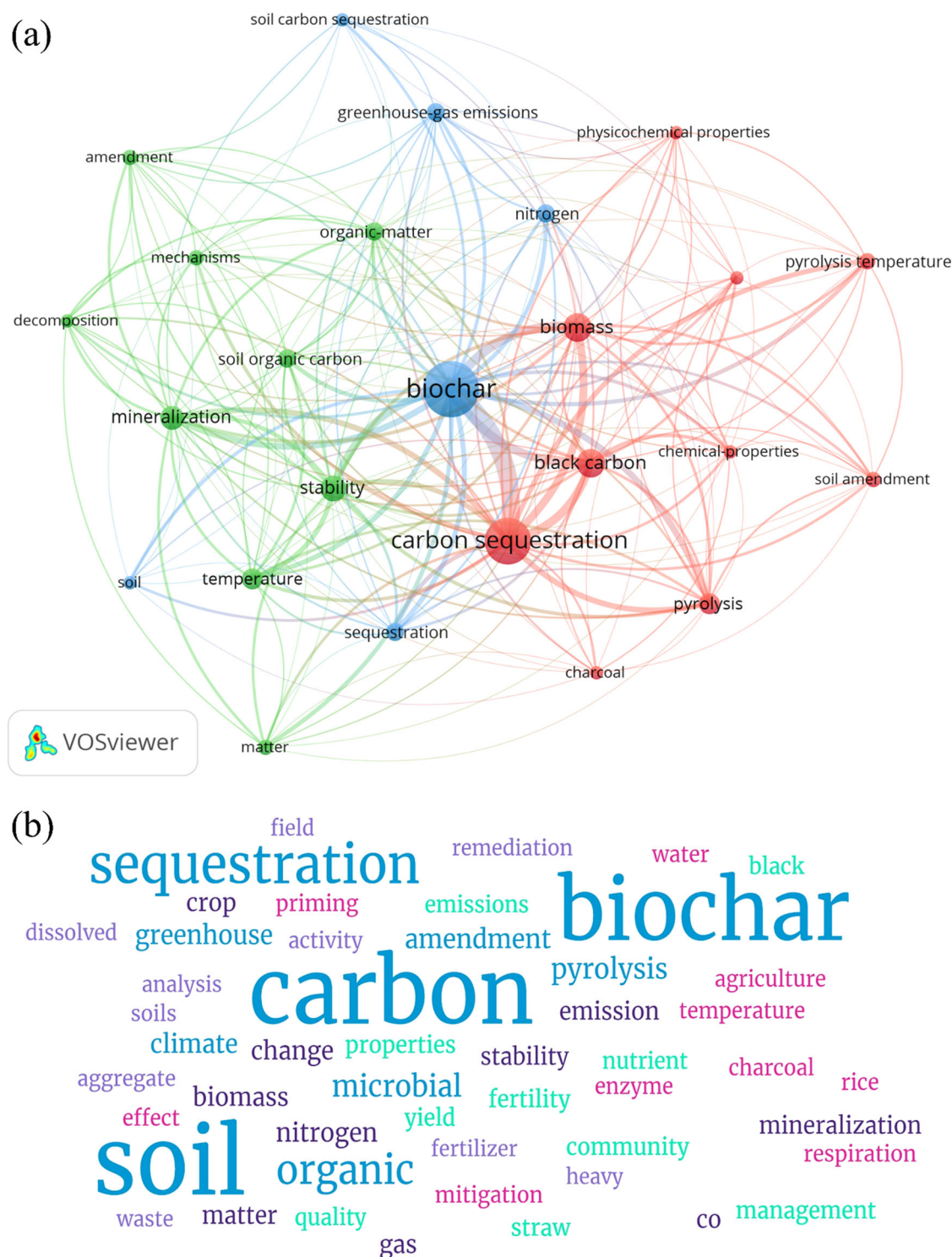
The scope of this review differs from that of previously published review articles as explained below. Our literature review over the last 11 years (2015–2025) reveals that numerous laboratory-scale and field-scale studies have been conducted to assess the capability of biochar for carbon sequestration in soil<sup>15,6,10,11,23–25</sup>. A few review articles were published on topics related to carbon sequestration in soil using biochar<sup>10,21,26,27</sup>. However, there is a lack of a comprehensive analysis on changes of biochar-based carbon sequestration in soil based on laboratory-scale tests and long-term field-scale experiments. In recent years, machine learning-based models are used for better understanding of biochar production processes and optimization of process parameters to improve biochar yield and properties<sup>28–30</sup>. However, a critical review on the application of different machine learning approaches in biochar synthesis is found to be lacking in previous reviews. In addition, limited information is available on the variations of carbon sequestration rates in response to the changes of biochar characteristics (carbon content, surface area, porosity, aromaticity/stability, etc) and soil properties (soil size/texture, pH, initial SOC content, etc). The impact of various biotic and abiotic factors on biochar aging and its impact of carbon sequestration potential of biochar specifically in long term application in various soil systems is not well documented. It is well-known that the physicochemical properties of the biochar are improved after chemical modifications (called modified biochar). However, critical analysis on the carbon sequestration potential of the modified biochar in soil system has not been done yet to the best of our knowledge. The interactions between biochar-soil particles, and their effects on microbial dynamics/enzymatic activities and carbon sequestration rate should be considered as part of this critical analysis. The potential mechanisms that drive the carbon sequestration by biochar in various soil environments are not clearly explained in the past reviews. For large-scale production of biochar and its field-scale application in soil for carbon sequestration, the sustainability aspects including potential negative environmental impacts during production as well as economic feasibility using life cycle assessment (LCA) need to be critically evaluated.

The overarching objective of this review is to conduct a critical analysis of recent advancements in the application of biochar for carbon sequestration in soil and to explore climate change mitigation on a large scale. The overall scope of this review is presented in Fig. 2. Specifically, the commonly used thermal-based synthesis methods (e.g., pyrolysis), the activation

process (physical and chemical activation) and other modifications applied for improvement of biochar properties are discussed in detail. Recent research findings of laboratory-scale and field-scale experiments on carbon sequestration capacity of biochar in soil ecosystems are critically analyzed. Moreover, how biochar characteristics and soil properties impact the rate of carbon sequestration are explained. Insights into the potential mechanisms for biochar-based carbon sequestration in soil environment are elucidated. The impact of biochar application on soil health (e.g., fertility and productivity) as well as microbial communities in soil is outlined. The sustainability and circular economy aspects on the use of biomass-derived biochar for carbon sequestration in soil are evaluated using life-cycle assessment (LCA). Future perspectives to increase carbon sequestration capacity of biochar and key challenges involved with the large-scale production of biochar, long-term stability, and carbon sequestration capability of biochar are also highlighted. This comprehensive review enhances our fundamental knowledge on the different aspects of the biochar-derived carbon sequestration in soil systems. Moreover, this insightful review would help to develop novel biochar with high stability and high carbon sequestration capacities in soil environment. Moreover, it may offer guidance to environmental regulatory agencies for development of novel strategies (e.g., selection of specific biomass substrates for biochar production, exploration of biochar applications based on soil properties, co-application of biochar with N/P fertilizers or agricultural residues/compost, etc.) to enhance carbon sequestration rate, reduce CO<sub>2</sub> levels in the ambient air and mitigate global climate change.

## Literature Trends

To better understand the current state-of-knowledge in literature regarding the significance of biochar-based carbon sequestration in soil, we conducted a critical analysis of recent publications using a mainstream and reputed scientific database, Web of Science (WOS) (Fig. S1). Three important keywords/terms were used in the search engine of WOS database namely “Biochar”, “Soil” and “Carbon Sequestration”. The literature review was carried out to comprehend the publication trend in the last 11 years (2015–2025) with specific reference to the application of biomass-derived biochar for carbon sequestration in soil. Our search using the above three keywords in the WOS website resulted in a total of 2529 publications including 2076 (82%) research articles, 377 (15%) reviews and 76 (3%) other publications (e.g., conference proceedings, book chapters, letters, etc.). As can be seen in Fig. S1a, the quantity of publications appears to be constantly increasing in the past 10 years, for example, 105 articles were published in 2015, but the number of publications increased by about 3.4 times in 2024 (354 publications). This rise of the publication trend suggests that an increasing interest is given worldwide for the development of novel biomass-derived biochar for enhancement of the carbon sequestration capabilities in soil having complex physicochemical and microbial characteristics. Further



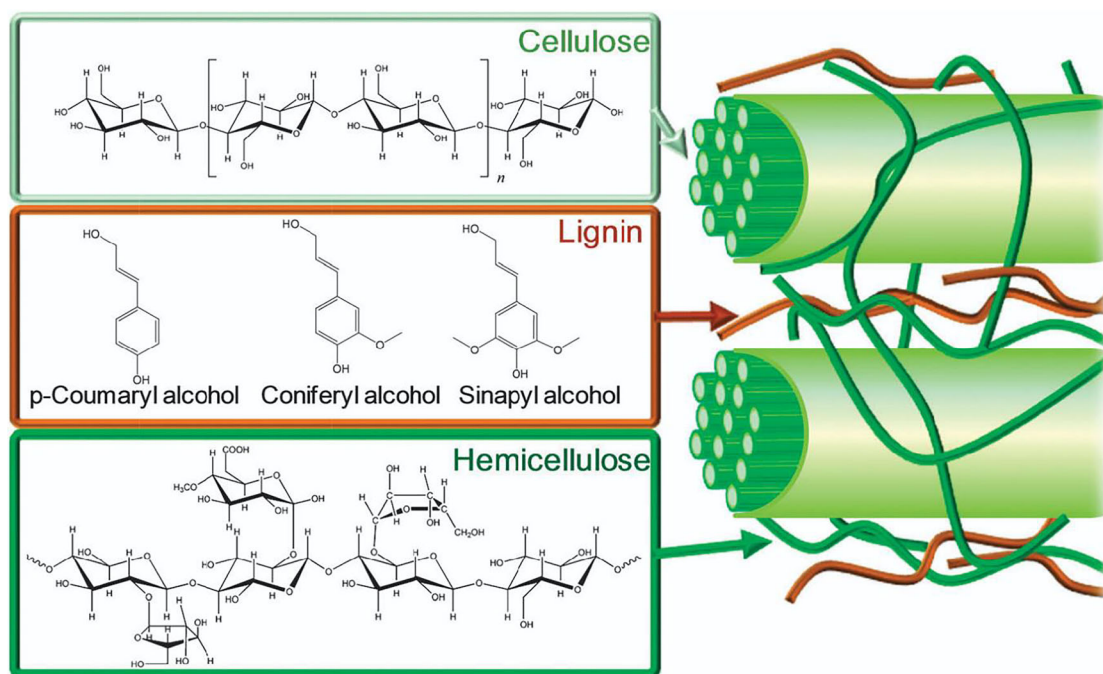
**Fig. 3 | Literature trends.** Bibliometric analysis on the cooccurrence of keywords reported in literature **(a)**. The node size indicates the frequency of occurrence of a specific keyword. The curves connecting between the nodes denotes keywords cooccurrence in the same publication. If the distance between the two keywords are

shorter, it signifies greater number of their cooccurrence. **(b)** The font size of a keyword in the word cloud represents its frequency of occurrence. Keywords with larger font size indicates their greater occurrence.

analysis about the distribution of publications among different countries/regions reveals that the published articles were from 114 countries. It is thus clear that climate change has been recognized as a major global issue, and hence worldwide contributions are explored to combat the global warming. As displayed in Fig. S1b, among various countries, China has published 978 papers, accounting for 38.7% of the total publication quantity. This impressive research output could be due to

large agriculture-related activities in China, and agricultural activities could also emit significant levels of GHG which need to be controlled<sup>31</sup>. Most of the publications were contributed by experts from three disciplines including environmental science/ecology, agriculture and engineering, suggesting that multidisciplinary knowledge is required to mitigate climate change by employing negative emission approaches and strategies such as biochar amendment in soil for carbon sequestration.





**Fig. 4 | Structure of key components of lignocellulosic biomass.** Reproduced with permission from ref. 194. Copyright (2012) Royal Society of Chemistry.

**Table 1 | Different thermochemical processes used for production of biochar**

Parameter	Slow pyrolysis	Fast pyrolysis	Gasification	Hydrothermal carbonization
Temperature	300–800 °C	400–1000 °C	750–1000 °C	180–260 °C
Heating rate	<10 °C/min	300–800 °C/min	50–100 °C/s	5–10 °C/min
Residence time	5 min–12 h	< 10 s	10–20 s	5 min–12 hr
Main product	Biochar	Bio-oil	Syngas	Hydrochar
Solid product yield	35–50%	15–35%	< 10%	45–70%
Typical reactor	Fixed bed	Fluidized bed	Fluidized bed	Hydrothermal reactor

For deeper understanding about the recent advancements in the biochar-mediated carbon sequestration in soil, a further bibliometric analysis on the WOS database was done using the VOSviewer software<sup>32</sup>. The keywords co-occurrence analysis (Fig. 3a) shows that “biochar” was the most frequently used keyword (total link strength: 171), followed by “carbon sequestration” (total link strength: 139), “black carbon” (total link strength: 80) and “biomass” (total link strength: 78). Moreover, there is a strong link between biochar and carbon sequestration, i.e., these two keywords are frequently used in the scientific publications. A word cloud was also produced using the keywords retrieved from the Scopus database to observe the frequency of usage of important keywords in literature (Fig. 3b). Based on the font size of the keywords, the dominant keywords used in literature include biochar, carbon, soil and sequestration.

To screen and select appropriate references to include in this review, we followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines for systematic reviews and meta-analysis<sup>33,34</sup>. The PRISMA flow diagram is presented in Supplementary Materials (Fig. S2). Three main scientific databases namely WOS, Scopus and Google Scholar were chosen to collect the references for this review. By employing various inclusion and exclusion criteria, for example, the articles published in English (2015–2025) that are most relevant to the scope and objectives of this review are selected for this review. Furthermore, the quality of articles was further evaluated by checking the reputation of the journal, the number of citations, indexing databases, etc. The relevant quantitative and qualitative information of a particular study were extracted by carefully reading the full text of the article. Finally, 195 articles related to lab- scale investigations

and full-scale studies meta-analysis reports and modeling-based works pertaining to biochar-based carbon sequestration in soil are critically evaluated and discussed in this review.

## Biochar production and characterization

International Biochar Initiative (IBI) has provided detailed guidelines on consideration of different aspects of biochar including selection of feedstocks, technologies biochar production, characterization and utilization of biochar for various purposes (<http://www.biochar-international.org/>). The global biochar market is anticipated to reach US\$368.85 million by 2028<sup>35</sup>. The main components of a plant-based biomass include cellulose (30–50%), hemicellulose (25–30%) and lignin (10–30%)<sup>36</sup>. The chemical structure of lignocellulosic biomass is presented in Fig. 4. Biomass with higher lignin content is beneficial for biochar production since it results a higher biochar yield with improved physicochemical properties such as greater porosity and higher aromaticity<sup>37,38</sup>.

Various thermochemical methods used for biochar production include pyrolysis (fast and slow pyrolysis), gasification, and hydrothermal carbonization (HTC) (Table 1)<sup>37,39</sup>. Moreover, emerging technologies such as microwave-assisted pyrolysis are used due its advantages such as achievement of uniform heating with reduced energy consumption<sup>40,41</sup>. To improve biochar yield and functionality, co-pyrolysis of biomass feedstocks with waste materials (e.g., plastics wastes) has been explored in recent years<sup>42–44</sup>. Waste plastics are usually rich in C and O which may change the surface functionality of biochar<sup>45</sup>. Moreover, co-pyrolysis strategies offer new pathways for sustainable waste management and energy production<sup>46</sup>

**Table 2 | Biochar synthesis conditions and its properties reported in previous studies**

Biomass	Process	Temperature (°C)	pH	C (%)	N (%)	O (%)	H (%)	References
Corn cob	Pyrolysis	360	8.22	65.7	0.91	-	-	73
Rice straw	Pyrolysis	350	8.9	46.9	0.5	-	-	177
Corn silage	Pyrolysis	500	9.73	77.88	1.99	-	-	66
Oak pellets	Pyrolysis	550	10.5	56.07	0.22	0.7	0.85	178
Maize straw	Pyrolysis	300	7.6	58.12	1.7	28.29	4.84	88
Maize straw	Pyrolysis	450	9.7	68.51	1.88	18.19	3.74	88
Maize straw	Pyrolysis	600	10	71.78	1.16	12.25	2.42	88
Fruit tree branches	Pyrolysis	300	7.2	40.74	1.03	-	4.32	117
Fruit tree branches	Pyrolysis	450	9.99	53.42	1.24	-	2.29	117
Fruit tree branches	Pyrolysis	600	9.96	58.95	0.98	-	1.24	117
Peanut shells	Pyrolysis	300	8.81	52.02	1.13	-	3.37	117
Peanut shells	Pyrolysis	450	10.51	56.06	1.04	-	2.19	117
Peanut shells	Pyrolysis	600	10.35	57.35	0.95	-	1.17	117
Rice straw	Pyrolysis	300	9.19	72.1	1.55	21.3	5.03	179
Rice straw	Pyrolysis	400	9.96	77.2	1.74	17	4.01	179
Rice straw	Pyrolysis	500	10.48	82.8	1.77	12.1	3.25	179
Rice straw	Pyrolysis	600	10.84	87.1	1.52	8.8	2.51	179
Rice straw	Pyrolysis	700	10.77	90.6	1.41	6.2	1.8	179
Rice straw	Pyrolysis	-	10.2	65.7	0.6	-	-	74
Rice straw	Pyrolysis	300	8.9	46.97	1.1	30.64	3.22	71
Rice straw	Pyrolysis	500	10.43	51.61	0.88	25.13	1.68	71
Rice straw	Pyrolysis	700	11.06	52.02	0.94	20.05	1.71	71
Wheat straw	Pyrolysis	450	10.4	47.1	0.77	-	-	91
Tree ( <i>Pinus massoniana</i> )	Pyrolysis	450	6.87	66.66	0.41	30.09	2.83	98
Rice husk	Pyrolysis	300	8.15	24.86	1.13	-	-	152
Empty fruit bunches	Pyrolysis	300–350	8.53	52.11	0.38	-	-	152
Rice husk	Pyrolysis	650	-	74.37	1.02	-	1.78	173
Corn stover	Pyrolysis	550	-	77.51	1.5	-	2.21	173
Wheat straw	Pyrolysis	350	7.78	65.7	0.5	-	-	180
Poplar wood	Gasification	1200	9.6	70.5	1.7	-	-	181
Maize silage	Gasification	1200	10.1	69.6	1.7	-	-	181
Maize silage	HTC	180–230	3.6	52.2	1.5	-	-	181
Corn silage	HTC	180–230	4.15	51	1.9	19	5.7	182

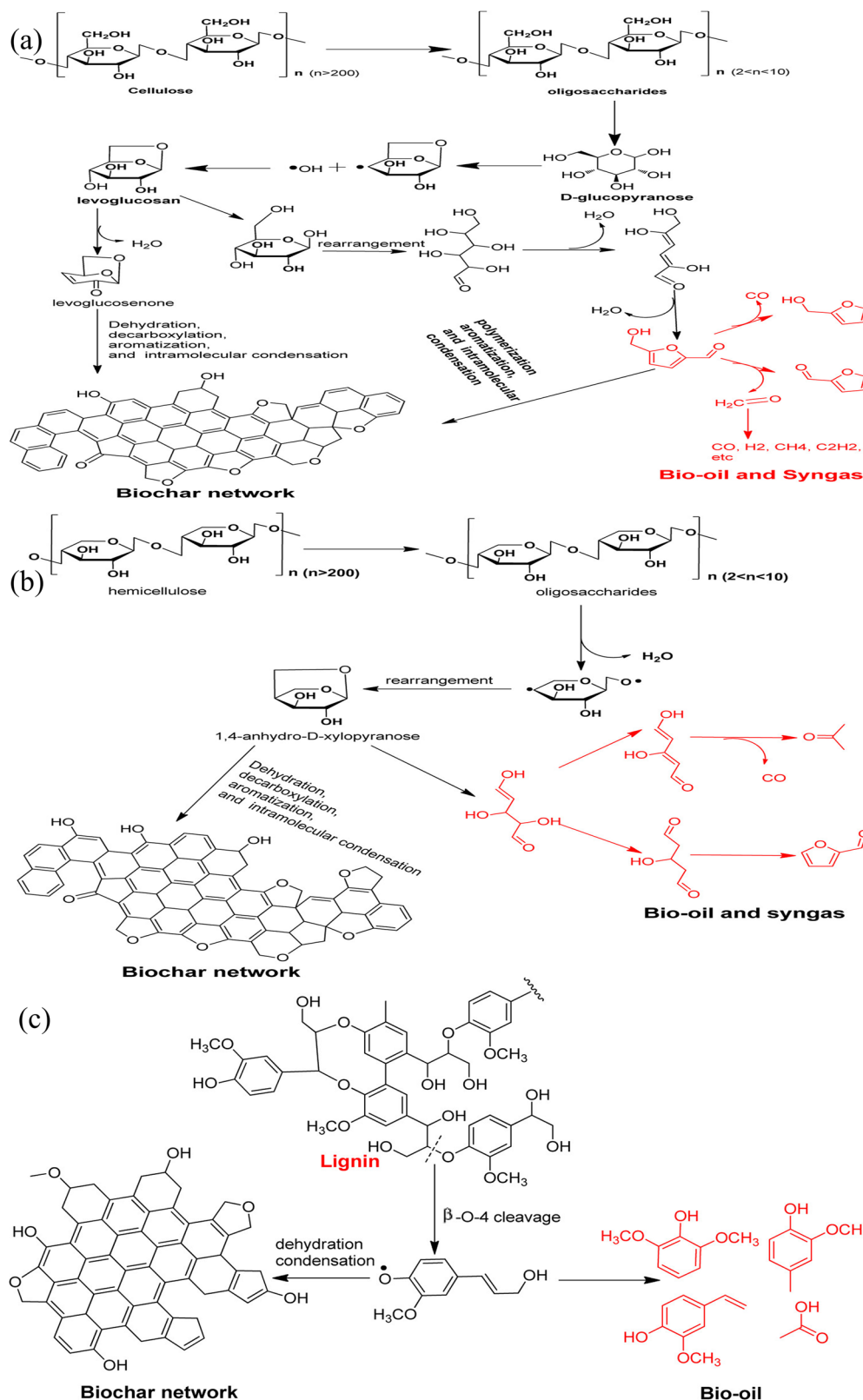
HTC hydrothermal carbonization.

Although HTC has emerged as a promising technology for conversion of biomass with high moisture content (e.g., >20 wt%) into value-added products (e.g., biochar)<sup>47</sup>, there are certain challenges that still exist, for example, reactor design, process optimization and treatment of process water from reactors for the upscaling of HTC technology from lab-scale to large-scale industrial applications<sup>48</sup>. To achieve sustainability with reduction of energy use, solar-driven pyrolysis processes are used for conversion of biomass to biochar<sup>49,50</sup>.

Biochar characteristics change with the changes of synthesis methods and biomass properties (Table 2). Since the variations of physicochemical properties of biochar (e.g., C/N ratios and pH) could considerably impact its carbon sequestration potential, a statistical analysis was done by creating a box plot (Fig. S3) to show the changes of C/N ratios and pH of biochar reported in the literature. For the box plot analysis, the data presented in Table 2 were used. The biochar C/N ratios have large variations between 22–255 (mean: 66), while the pH ranged from 3.6 to 11.1 (mean: 9.2). The variations of biochar properties could be related to the various feedstock materials used for the synthesis of biochar and pyrolysis conditions employed in different studies. The biochar formation mechanisms using

cellulose, hemicellulose and lignin biomass as the feedstock by pyrolysis are shown in Fig. 5. Among different thermochemical methods, HTC is considered as the cost-effective method since it is normally conducted at relatively lower temperature than other thermochemical methods<sup>51</sup>.

Key physicochemical properties (e.g., surface area and porosity) of the biochar can be further improved by employing activation agents (physical or chemical), and the resulting carbon materials are referred to as modified biochar/engineered biochar<sup>38,52,53</sup>. Acid-modified biochar was reported to be highly suitable for soil amendment<sup>52</sup>. Liu et al.<sup>54</sup> synthesized biochar using rice husk as the feedstock at the pyrolysis temperature of 700 °C. The obtained pristine biochar was modified by using phosphoric acid (H<sub>3</sub>PO<sub>4</sub>) followed by treatment with nanoscale zero-valent iron (nZVI). In lab-scale incubation tests, the addition of nZVI-based biochar composite to soil at a dose of 2% (w/w) resulted in reduction of CO<sub>2</sub> release from soil by 80.29–91.60% at the end of 60 days incubation period. Among the three types of biochar systems, the highest CO<sub>2</sub> release reduction was observed on phosphoric acid-treated biochar (91.60%) followed by nZVI-based biochar (88.28%) and pristine biochar (80.29%). The incorporation of P increases biochar stability (i.e., amount of stable C), thus enhances biochar's carbon



**Fig. 5 | Biochar formation mechanisms by pyrolysis process using lignocellulose biomass as the feedstock material. a** Cellulose, **b** hemicellulose and **c** lignin. Reproduced with permission from ref. 195. Copyright (2015) American Chemical Society.

sequestration efficiency<sup>54</sup>. A recent study reported that the application of nZVI-based biochar into soil significantly enhanced carbon sequestration by reducing SOC mineralization<sup>55</sup>. The concentration of Fe-oxides in the soil system increases with the application of nZVI-based biochar. Several potential abiotic and biotic mechanisms are reported in literature to explain

the enhancement of carbon sequestration in soil with nZVI biochar treatment. Researchers have proposed an “iron gate” mechanism, in which organic carbon (e.g., SOC) are adsorbed/co-precipitated with the newly generated Fe-oxides from nZVI<sup>55,56</sup>. The Fe-bound organic carbon (Fe-OC) complexes exhibit increased chemical stability, and play a critical role in

SOC conservation. With the addition of nZVI biochar, the soil microbial community structures and enzymatic activities are considerably changed, for example, reduced activities of carbon mineralization related enzymes namely  $\beta$ -glucosidase and phenol oxidase and a shift in bacterial community composition from unstable carbon-dominant communities (*Actinobacteria* and *Proteobacteria*) to stable carbon-dominant communities (*Firmicutes*)<sup>55</sup>. Another study also reported that potassium incorporation (2%, w/w) into biochar synthesized from *Miscanthus* biomass pyrolyzed at 450 °C enhanced carbon sequestration potential of the modified biochar by 45% which is equivalent to the rise of carbon sequestration potential of modified biochar to over 2.6 G tons CO<sub>2</sub>-C(eq)/year<sup>11</sup>. The findings of these studies suggest that chemically modified biochar has greater carbon sequestration potential than pristine biochar.

The carbon sequestration potential of biochar changes with the changes of its physicochemical properties and surface chemistry. In addition to the classical techniques (e.g., SEM, TEM, XRD, BET, XPS, FT-IR, etc.), several advanced synchrotron-based techniques such as X-ray absorption near edge structure spectroscopy (XANES) and scanning transmission X-ray microscopy (STXM)-based near edge X-ray absorption fine structure (NEXAFS) are used to provide insights into physicochemical properties and surface chemistry of biochar<sup>57,58</sup>.

In recent years, several machine learning (ML)-based models/approaches (e.g., artificial neural network (ANN), random forest (RF), support vector machine (SVM), etc.) are used for optimization of biochar production processes to increase biochar yield and improvement of its properties and surface chemistry<sup>59,60</sup>. These ML-based approaches are useful to achieve multi-objectives optimization to synthesize biochar with desired qualities. Moreover, they can make considerable contribution to a sustainable future with development of a circular bio-based economy<sup>60</sup>. Sensitivity analysis is found to be an effective technique for determination of key feedstock properties and optimization of biochar synthesis process and its properties. Tee et al. used artificial neural network model (Levenberg-Marquardt algorithm) for prediction of the carbon sequestration potential of biochar prepared from biomass by pyrolysis process<sup>61</sup>. The ANN models demonstrated good performances with high correlation coefficient ( $R$ : >0.96) and coefficient of determination ( $R^2$ : >0.92), indicating good alignment between the predicted and actual values of yield and surface area of biochar. The proximate analysis of feedstocks was found to have considerable effects on the biochar yield, while ultimate analysis had greater impact on the surface area<sup>61</sup>. A recent study reported that among the five machine learning models investigated for prediction of biochar yields and its properties (e.g., C and N contents), multi-layer perceptron neural network and Gaussian process regression models shows good performance ( $R^2$ : 0.92–0.97)<sup>62</sup>. In the context of reducing environmental footprints, LCA is employed to assess the carbon sequestration potential of biochar in soil under two different scenarios such as replacement of the N content in biochar by two types of fertilizers, (1) urea ammonium nitrate and (2) calcium ammonium nitrate. The carbon sequestration potential (GWP) of the biochar in two scenarios was found to be −1323 and −1355 kg CO<sub>2</sub>-eq/t biomass, respectively. The negative value of GWP suggests that biochar is an environmentally friendly material. Overall, ML is an important tool for the production of biochar with desired characteristics for different environmental applications including soil carbon sequestration.

### Impact of biochar application in soil on carbon sequestration

In recent years, biochar amendment into soil has been increasingly advocated as an effective strategy for carbon sequestration and climate change mitigation<sup>63,64</sup>. Biochar is one of the negative emission technologies with a very high technology readiness level (TRL)<sup>11,65</sup>. Biochar amendment in soil impacts SOC mineralization rates which is referred to as priming effects<sup>66</sup>. The application of biochar to soil can either increase SOC mineralization (positive priming, increase of CO<sub>2</sub> emissions) or decrease SOC mineralization (negative priming effects, reduction of CO<sub>2</sub> emissions)<sup>67</sup>. The biochar-treatment enhanced the mineralization of SOC in a sandy soil but

suppressed the mineralization of SOC in clay soils<sup>67</sup>. Biochar application usually reduces soil CO<sub>2</sub> fluxes in fertilized soils (agricultural soils)<sup>68</sup>. Moreover, biochar application to soil reduces 1/8 of CO<sub>2</sub> emissions and sequesters 2.5 gigatons of CO<sub>2</sub> annually<sup>35</sup>. Biochar produced at higher temperatures ( $\geq 600$  °C) with high C/N, low O/C and low H/C ratios usually shows greater carbon sequestration potential than that produced with other characteristics<sup>69,70</sup>. Low H/C and O/C ratios indicate an increase of aromaticity in the biochar which indicates high stability (i.e., recalcitrant nature to chemical and microbial degradation) of the biochar and could show positive effects in terms of carbon sequestration performance<sup>71,72</sup>.

Several laboratory-scale and field-scale studies were conducted to evaluate the carbon (CO<sub>2</sub>) sequestration potential of biomass-derived biochar in various soil conditions (Table 3). A global meta-analysis of 64 publications reported that field-scale experiments were carried out over a time period of 1 and 10 years with biochar application rate varying between 1 and 100 Mg/hectare which corresponds to the increase of the SOC amount by 29% (15 Mg/hectare)<sup>10</sup>. However, lab-scale pot experiments were conducted in incubation time ranging from 1 and 1278 days with the biochar incorporation rate varying between 5 and 200 g/kg soil, corresponding to an increase of SOC by 75% (6.3 g/kg)<sup>10</sup>. Moreover, biochar synthesized from plant biomass demonstrated greater carbon sequestration potential compared to that prepared from fecal matter, which is primarily due to the high C/N ratio. Biochar was produced in a study employing two different thermochemical methods (slow pyrolysis and HTC) from corn silage used as feedstock, and their effects were then compared in terms of the soil CO<sub>2</sub> sequestration potential in lab-scale tests<sup>66</sup>. Pyrolyzed biochar (1% w/w) reduced CO<sub>2</sub> emissions from the agricultural soil while HTC-derived biochar (1% w/w) considerably increased CO<sub>2</sub> emissions from soil. The larger CO<sub>2</sub> emissions could be due to the unstable nature of HTC-based biochar since nearly 50% decomposition of the HTC-derived biochar was noticed at the end of incubation period (50 days)<sup>66</sup>. Moreover, the changes of priming effects/SOC mineralization could be due to a shift of soil microbial communities from limited available SOC to more labile SOC from HTC-based biochar. In a recent study<sup>73</sup>, soil samples were collected from an agricultural site which received the biochar treatment of 9.0 tons/hectare/year for nearly a decade. Then, the biochar amended soil was processed in microcosm mode with and without (control) plant biomass (wheat/soybean straw) added to the soil. Compared to the control system, biochar-amended soil reduced CO<sub>2</sub> emissions by nearly 11% in 56 days of incubation time with nearly a 2 fold increase of the SOC amount. Accumulation of SOC onto microbial biomass and direct adsorption and blocking of SOC by biochar particles could cause a decrease of SOC release<sup>73</sup>. A mesocosm study was conducted to compare the application of rice straw blended with urea versus rice straw-derived biochar blended with urea onto agricultural soil on the global warming potential (GWP)<sup>74</sup>. Both rice straw-derived materials were applied in the field at a rate of 6 tons/hectare. Notably, the rice straw-amendment reduced the CO<sub>2</sub> uptake rate by 27.1%, whereas the use of biochar enhanced the CO<sub>2</sub> uptake by 43.5% and reduced the GWP by 375.6 g CO<sub>2</sub>-eq/m<sup>2</sup>. The results of this study suggest that the decrease of GWP by the use of biochar amendment is due to the increase of CO<sub>2</sub> uptake in the soil system. Moreover, rice fields act as CO<sub>2</sub> sink due to the increased CO<sub>2</sub> uptake by stimulation of plant photosynthesis<sup>74</sup>. In a microcosm-based study, a comparative evaluation of the GHG emission rates was made in the coastal saline soil with the application of cork stalk-derived biochar (dose: 0–32 tons/hectare) and untreated corn stalk (7.8 tons/hectare) used as the control system<sup>75</sup>. The findings revealed that the soil treated by biochar enhanced the global warming mitigation potential (GWMP) (−3.84 to −3.17 ton CO<sub>2</sub>-eq/hectare/tonne C) compared to the corn stalk-treated soil system (−0.11 ton CO<sub>2</sub>-eq/hectare/tonne C). Specifically, the GWMP of biochar was nearly 28–34 times higher than the corn stalk biomass which could be due to higher carbon sequestration potential of biochar than corn stalk. This study indicated that the conversion of agricultural residues into biochar followed by their application to soil could result in a nearly four-fold increase of carbon sequestration. In some studies, the increase of CO<sub>2</sub> emissions was observed at the early stage of tests which could be due to



**Table 3 | Impacts of biochar amendment in soil on the carbon sequestration potential**

Biochar synthesis conditions	Type of studies	Soil type	Biochar application rate	Carbon sequestration potential	References
Corn silage, 500 °C	Lab-scale	Forests and agricultural soils	1% (w/w)	No impact on forest soil, but reduced CO <sub>2</sub> emission from the agricultural soil	66
Swine manure, 600–800 °C	Lab-scale	Rice paddy field	2% (w/w)	Significant reduction of CO <sub>2</sub> emission after biochar treatment	85
Reed straw, 400 °C (nZVI-biochar)	Lab-scale	Saline-alkali soil	0.15–0.45% (w/w)	Significant reduction of CO <sub>2</sub> emission after nZVI-biochar treatment	55
Corn cob, 250 °C	Lab-scale	Acidic sandy soil	~0.84% (w/w)	Reduction of CO <sub>2</sub> emission by 11.8%	86
Rice husk, 700 °C	Lab-scale	Soil from university campus	2% (w/w)	Reduction of CO <sub>2</sub> emission by 80.29%	54
Rice husk, 700 °C (H <sub>3</sub> PO <sub>4</sub> -biochar)	Lab-scale	Soil from university campus	2% (w/w)	Reduction of CO <sub>2</sub> emission by 91.60%	54
Rice husk, 700 °C (H <sub>3</sub> PO <sub>4</sub> -nZVI-biochar)	Lab-scale	Soil from university campus	2% (w/w)	Reduction of CO <sub>2</sub> emission by 88.28%	54
Hard wood, 200–600 °C (Steam & CO <sub>2</sub> activation)	Lab-scale	Topsoil (silt loam)	0.75% (w/w)	Reduction of CO <sub>2</sub> emission by 18%	183
Wood sawdust, 450 °C	Lab-scale	Surface soil	3.2% (w/w)	Negative priming effects was observed with biochar treatment (–0.22 to –23.56 mg-CO <sub>2</sub> -C/g –soil-C)	184
Peanut shells, 400–500 °C	Lab-scale	Soil from an experimental field	~1.4% (w/w)	Reduced CO <sub>2</sub> emission by 23.61%	185
Rice straw, 500 °C	Lab-scale	Saline-Alkaline Soil (sandy loam)	~0.77% (w/w)	Reduction of CO <sub>2</sub> emission by 35.19% with addition of biochar as well as straw and urea	186
Wheat straw, 450 °C	Lab-scale	Irragic Anthrosols	~1.1% (w/w)	Biochar application decreased CO <sub>2</sub> emission by an average of 23%	91
Rice husk, 300 °C	Lab-scale	Soil of Bungor Series	~0.54% (w/w)	Cumulative CO <sub>2</sub> emission reduced by 139.41% compared to control	152
Wheat straw, 500–600 °C	Pot experiments	Clay loam soil	50–95% (w/w)	CO <sub>2</sub> emissions reduced by 8.05–31.46%. Higher CO <sub>2</sub> emissions observed at higher biochar dose.	187
Corn stover, 550 °C	Pot experiments	Garden top soil	3% (w/w)	CO <sub>2</sub> emissions reduced by 15% compared to control soil	173
Pine wood, 500–700 °C	Pot experiments	Olton clay loam soil	1% (w/w)	Reduced CO <sub>2</sub> emission by 66.9–72.4%	188
Corn stalks, 400 °C	Microcosms	Coastal saline soil	16 tons/hectare	Corn stalks-derived biochar showed higher GWMP (–3.84 to –3.17 tonne CO <sub>2</sub> -eq/hectare/tonne C) than control treatment (–0.11 tonne CO <sub>2</sub> -eq/hectare/tonne C).	75
Rice straw	Mesocosm	Rice paddy field	6 tons/hectare	CO <sub>2</sub> uptake increased by 43.5% and decreased the GWP by 375.6 g CO <sub>2</sub> -eq/m <sup>2</sup> /season	74
Multiple feedstocks, 280 °C	Pot experiments	Alkaline clay and acidic sandy soil	4 tons/hectare	Biochar with N fertilizer addition reduced CO <sub>2</sub> emission by 7–12%	189
Corn straw, 360 °C	Microcosm	Agricultural soil	9 tons/hectare	Reduced CO <sub>2</sub> emission by 11%	73
Farm wastes and wood residues, 500–550 °C	Field-scale	Andisol	11 tons/hectare	Biochar amendment reduced CO <sub>2</sub> fluxes. But, no significant differences in CO <sub>2</sub> emission rates among different types of biochar treatments	24
Corn cobs, 500–550 °C	Field-scale	Haplic Acrisols	0–30 tons/hectare	Specific maintenance respiration (qCO <sub>2</sub> ) reduced by 66–73%	114
Maize straw, 350–550 °C	Field-scale	sandy-loam soil	30 tons/hectare	Reduced CO <sub>2</sub> emission by 33%	190
Corn straw, 450 °C	Field-scale	Sandy loam soil	20 tons/hectare	Biochar addition enhanced SOC levels and reduced CO <sub>2</sub> emissions	76

mineralization of labile carbons from biochar and stimulation of carbon mineralization microbial communities<sup>75</sup>. Overall, lab-scale incubation, mesocosm and microcosm-based experiments show that biochar is effective in enhancing the carbon sequestration in soil.

Several field-scale experiments were conducted to assess the effectiveness of biochar amendment to soil on the carbon sequestration potential/GHG emission rates<sup>24,64</sup>. A year-long field-scale experiment was conducted on Humic Haploxerands (Andisols) using different biochar materials (manures-/wood residue-derived biochar) applied at a rate of 1% which is equivalent to 11 tons/hectare<sup>24</sup>. Andisols are the soils formed in volcanic ash with short-range-order minerals containing little orderly crystalline structure. It was observed that after an initial increase of soil CO<sub>2</sub> fluxes in 45 days, a decrease of CO<sub>2</sub> emission was recorded in biochar amended soil. However, no significant difference in the CO<sub>2</sub> emission rates was found among different types of biochar-treated soil systems<sup>24</sup>. The initial rise of CO<sub>2</sub> emissions could be due to the mineralization of labile/oxidizable organic carbon fraction leached from the biochar and/or decomposition of soil organic matter by possible microbial activities<sup>24</sup>. Moreover, the study was conducted at the soil temperature of 9.6–25.7 °C with the moisture level of 42.7–59.7% which are quite favorable for microbial aerobic processes that results CO<sub>2</sub> as the end product<sup>24</sup>. In another work conducted by Liu et al.<sup>64</sup>, SOC mineralization rates and carbon sequestration potential of two types of soil environments (upland soil and paddy soil) were investigated. The soil samples were blended with crop straw-derived biochar (rate: 20 tons/hectare) and crop straw (rate: 7.5–9 tons/hectare). Biochar amendment reduced the SOC mineralization rates in both upland soil (19.7–20.1%) and paddy soil (9.2–12.2%). Additionally, the biochar application decreased CO<sub>2</sub> emission rates in both upland (15.2–18.6%) and paddy (8.9–12.5%) soils. The difference in the SOC mineralization and CO<sub>2</sub> emissions rates among two types of soil systems could be related to their physicochemical properties including SOC content, nutrient level and pH<sup>64</sup>. In a recent field-scale experiment with the application of corn straw-derived biochar (20 tons/hectare) together with a low dose of nitrogen fertilizer (e.g., 255 kg/hectare) added to soil was effective in increasing the SOC content (30.91%) and reducing the CO<sub>2</sub> release. The reduction of CO<sub>2</sub> emissions could be due to the SOC immobilization and the decrease of soil microbial activities due to the adsorption of nutrients onto the surface of biochar<sup>76</sup>. Nevertheless, a higher dose (300 kg/hectare) of nitrogen fertilizer application showed negative effects, i.e., increase (up to 48%) of CO<sub>2</sub> emissions. The interactions between biochar and nitrogen fertilizers played a critical role in the abundance of soil microbial communities since the bacterial abundance was lower in biochar treated soil, but considerably higher in the co-treatment system (biochar plus N fertilizer). The results of this study reveal that the addition of N fertilizers with an appropriate dose is required to reduce the CO<sub>2</sub> release from the agricultural soil. Altogether, findings of the above studies indicate that the biochar treatment of soil is effective in terms of carbon sequestration and reduction of GHG emissions under field-scale conditions. Furthermore, these long-term field experimental datasets could be useful for the validation of biotechnical-based models that are used for the prediction of the carbon sequestration potential of biochar in soil.

In addition to experimental works, a few modeling studies were carried out to predict carbon sequestration potential of biochar-amended soil<sup>77,78</sup>. Using a biogeochemical model, Yin et al.<sup>77</sup> investigated the long-term (5, 50, and 500 years) carbon sequestration potential of biochar application to soil by considering the complex biochar-soil-plant interactions. Through simulations by considering the biochar application rate of 7.5–75 tons/hectare in rainfed cropland soil (50 cm depth) planted with corn, it was observed that biochar could sequester 483–557 kg C/tons biochar-C after 500 years. Moreover, biochar tends to reduce the SOC degradation by 44–265 kg C/tons biochar-C as well as promote photosynthesis by providing nutrients to plants by capturing 66–1039 kg C/tons biochar-C. This study indicates that biogeochemical reactions/processes need to be considered while evaluating the long-term carbon sequestration potential of biochar addition to soil. In another study, the modified Rothamsted Carbon

(RothC) model was used to evaluate the carbon sequestration potential of sugarcane residues-derived biochar in soil in São Paulo State in Brazil<sup>78</sup>. With the biochar application rate of 4.2 tons/hectare/year, the model predicted an increase of the SOC content by  $2.35 \pm 0.4$  tons C/hectare/year. By considering the total sugarcane area of the São Paulo State of 5.77 M hectare, the total carbon sequestration potential was found to be 49.5 M tons of CO<sub>2</sub>/year which is 31% of the total yearly emissions (159 M tons CO<sub>2</sub>/year). The results of these modeling-based works supported the experimental (lab-scale and field-scale work) findings of higher carbon sequestration potential of biochar in soil. Moreover, this modeling-based studies may help decision makers with the development of novel strategies to achieve sustainable agricultural production with high carbon sequestration in soil.

Several meta-analysis-based studies were carried out to quantify the impact of biochar incorporation into soil on the carbon sequestration<sup>79–81</sup>. Bu et al.<sup>79</sup> conducted the meta-analysis of 15 studies published before November 2021, and reported that the carbon sequestration potential in paddy soils varied between 0.0066 and 2.0 Pg C with the change of biochar application rate from 2 to 40 tons/hectare. Another global meta-analysis work involving 169 publications reported that biochar amendment considerably improved different fractions of soil carbon including total carbon (64.3%), organic carbon (84.3%), labile carbon (22.9%) and microbial biomass carbon (20.1%)<sup>81</sup>. Similar findings were reported from a recent work with a critical analysis of 44 publications specifically related to the application of straw-derived biochar<sup>82</sup>. Straw-based biochar amendment to soil considerably enhanced the quantity of dissolved organic carbon (24.9%) as well as microbial biomass carbon (16.7%). A meta-analysis of 64 global studies by Gross and co-workers<sup>10</sup> indicated an average increase of the SOC amount in biochar-amended soil by 13.0 Mg/hectare which is equivalent to a 29% increase with biochar application in field-scale tests. In contrast, in the pot and incubation-based experiments (biochar dose: 5–200 g/kg soil), the increase of SOC was nearly 6.3 g/kg soil, which corresponds to a 75% increase of SOC after biochar addition. The authors stated that more SOC accumulation occurs in long term studies. For example, pot/incubation tests are typically carried out over a period of more than 500 days, while field experiments are conducted spanning from 6–10 years<sup>10</sup>. These meta-analysis-based studies are important to better understand the carbon sequestration potential of biochar under various experimental conditions, and its potential long-term field-scale applications for climate change mitigation.

Several factors including soil chemistry (e.g., soil pH) and characteristics of biochar feedstocks could significantly influence the SOC dynamics and carbon sequestration rate in biochar-amended soil systems. It has been observed that acidic soil has the tendency to release more CO<sub>2</sub> (1.5–3.5 times higher) compared to alkaline/neutral soils which could be due to degradation/disintegration of SOC and biochar in acidic environments<sup>71</sup>. Biochar shows liming effects. In other words, in alkaline soil systems, the bioavailability of SOC is decreased by adsorption of SOC into biochar which leads to the reduction of CO<sub>2</sub> emissions<sup>9</sup>. Li et al. reported that intense forest management and climate change considerably impact the forest soil properties by different processes namely soil acidification, decrease of SOC content, decrease of soil biodiversity and degradation of soil biological activities<sup>83</sup>. However, biochar amendment had complex effects (positive, negative, or negligible) on soil CO<sub>2</sub> release rates.

The physicochemical properties of biochar can be affected by various technological and operational parameters including feedstock types<sup>84</sup>. For example, biochars synthesized from animal litters/solid wastes as feedstock materials lead to less carbon contents, specific surface area, volatile matters, but higher cation exchange capacity (CEC) than biochars prepared from wood biomass/crop residues. The difference in the variation of these properties is potentially due to changes in the quantity of lignin, cellulose and moisture contents in the feedstocks<sup>84</sup>. In laboratory tests using different types of biochars (swine manure and barley stover-derived biochars) and soil systems (rice paddy and pasture), the variations of CO<sub>2</sub> emissions was observed with changes in types of biochars/soils<sup>85</sup>. In rice paddy soil, a

significant reduction of CO<sub>2</sub> emissions was recorded with treatment (2%, w/w) of swine manure-based biochar. However, in the pasture soil, there was an increase of CO<sub>2</sub> emissions in the early stage of incubation (7 days), but no significant differences of CO<sub>2</sub> evolution were observed at the later stage of incubation (i.e., after 20 days). The differences in the CO<sub>2</sub> evolution rate between two different biochar materials could be due to the difference in their physicochemical properties such as surface area (75.63 m<sup>2</sup>/g in swine manure biochar vs 40.60 m<sup>2</sup>/g). Moreover, the variations of greenhouse gas emissions (e.g., CO<sub>2</sub>) from two different soil systems could be related to differences in their properties and microbial communities. Since paddy soil usually receives N fertilizer and is dominated by N-transforming microbial communities, biochar addition could suppress metabolic activities of microbial communities, thus reduced CO<sub>2</sub> emissions<sup>85</sup>. Similar findings were reported by Wu et al. who investigated two types of biochar materials (corn cob and olive pulp-based biochar) in two soil types (acidic sandy soil and alkaline clay soil) on GHG emissions (e.g., CO<sub>2</sub> and N<sub>2</sub>O)<sup>86</sup>. In acidic sand soil, 11.8% reduction of CO<sub>2</sub> emissions was found with corn cob-derived biochar treatment, while olive biochar enhanced the CO<sub>2</sub> emission by nearly 2-folds. Nevertheless, in alkaline clay soil conditions, the addition of both types of biochars had no significant effects on the CO<sub>2</sub> release. Windeatt et al. reported that the carbon sequestration potential of 8 different types of biochars produced from agricultural crop residues (palm shell, sugarcane bagasse, rice husk, coconut shell, coconut fiber wheat straw, cotton stalk, and olive pomace) varied between 21.3–32.5%<sup>72</sup>.

Overall, the variations among findings that emerged from short term lab-scale and long term field-scale studies on the impacts of biochar amendment into soil on the SOC mineralization and carbon sequestration rate could be due to the differences in the adopted experimental conditions such as (i) soil types (i.e., variations of soil types with different physicochemical characteristics), (ii) biochar types (biomass feedstocks used to prepare biochar) and biochar application rate/dosage, (iii) incubation conditions (e.g., incubation time and water saturation)<sup>10,68,87</sup>.

## Impact of biochar and soil properties on carbon sequestration

The experimental conditions maintained during the synthesis of biochar and its characteristics such as pyrolysis temperature and the quantity of biochar-derived dissolved organic carbon (BDOC) could impact its carbon sequestration potential<sup>88,89</sup>. Specifically, the biochar synthesized at lower temperature (e.g., ≤300 °C) is likely to contain a high proportion of BDOC, and hence may not have a favorable impact on the carbon sequestration potential and the priming effect in soil environment<sup>89</sup>. The key components of BDOC include protein-, lipid- and carbohydrate-like compounds<sup>89</sup>. The biochar synthesized at higher temperature (e.g., ≥ 600 °C) normally possesses high porosity compared to the low temperature-based biochar, therefore, and thus have the capabilities to retain more SOC<sup>90</sup>. Yang et al.<sup>88</sup> synthesized biochar from maize straw at three different temperatures (300, 450 and 600 °C), and evaluated the amendment of biochar on soil properties as well as the carbon sequestration potential. In general, the biochar use (2% wt%) improves soil nutrients contents including N (8–36%) and P (19–69%) and promotes soil aggregation. However, the carbon sequestration potential is different among biochars synthesized at different temperatures. For example, biochar produced at 300 °C had lower carbon sequestration potential as evident from the increased CO<sub>2</sub> emission rate by nearly 45% at the end of 180 days incubation period. However, biochar prepared at higher temperatures (450 and 600 °C) reduced CO<sub>2</sub> emissions rate by nearly 10% and 15%, respectively. The higher CO<sub>2</sub> emission rate from soil blended with low temperature (300 °C) pyrolyzed biochar was mainly due to the release of leachable/labile organic carbons from the biochar since a significant (34–69%) increase of DOC amount was noticed after incubation of 180 days<sup>88</sup>. In a recent laboratory incubation-based tests in paddy soil system, the removal of BDOC from biochar (specifically biochar synthesized at 300 °C) prior to use in soil showed positive effects towards enhancement of carbon sequestration potential<sup>89</sup>. The priming effects of BDOC free biochar significantly changed from the positive (i.e., 3.7 mg

CO<sub>2</sub>–C/kg paddy soil) to negative state (i.e., –14.4 mg CO<sub>2</sub>–C/kg paddy soil). Moreover, the biochar produced at higher temperature (450 °C) caused further increase of negative priming effect by 31% as well as reduced biochar's mineralization rate by 41–65%. The increase of carbon sequestration potential by biochar amendment could be due to the formation of soil aggregates by biochar and reduction of degradation of SOC<sup>89</sup>.

Types of biochar, for example, fresh vs aged biochar show contrasting effects on carbon sequestration rate in soil ecosystem<sup>91</sup>. In a field-scale system under long-term operation, several biotic and abiotic factors such as variations of temperature, precipitation events and microbial activities could cause aging/weathering of biochar materials as well as changes of their physicochemical properties<sup>92</sup>. Moreover, potential mechanisms involved in biochar aging include dissolution, fragmentation/disintegration, interactions with soil minerals, biological degradation and abiotic oxidation<sup>92</sup>. The aging process can cause various changes of biochar properties including a considerable degradation of its molecular structure, an increase of specific surface area, and that of oxygen-containing functional groups as well<sup>93</sup>. In general, biochars synthesized at higher temperature are not much affected by the aging process (i.e., biochar carbon mineralization) due to the presence of highly recalcitrant aromatic carbons, thus less susceptible to oxidation<sup>94</sup>. Aging may also impact agronomic effectiveness and the carbon sequestration potential of biochar<sup>95</sup>. A long-term (13 years, biochar application rate: 31.5 Mg/ha) experiments shows that soil properties were significantly changed due to biochar aging<sup>95</sup>. For example, the soil pH was reduced from 7.4 to 6.8, while the electrical conductivity was decreased from 217 to 81.1 μS/cm during the long-term aging. Fresh biochar demonstrates no priming effect on SOC specifically in the early stage of incubation. However, the use of both fresh and aged biochar decrease the carbon loss from soil<sup>96</sup>. Reduction of CO<sub>2</sub> emissions was observed in a recent field-scale study following the aging (1 year) of two types of biochars (rice biochar (RB) and maize biochar (MB)). 2%, w/w in both acidic (paddy soil) and alkaline (fluvisol) soil systems<sup>93</sup>. In paddy soil, the biochar amendment reduced CO<sub>2</sub> emissions by 3.09 (RB)–17.05% (MB), while a higher reduction (16.38 (RB)–37.88% (MB)) of CO<sub>2</sub> emissions was observed in biochar treated fluvisol system. The findings of this study suggest that aged biochars have higher carbon sequestration potential, and both biochar and soil types impact the CO<sub>2</sub> release rate. Another recent study reported that biochar aging considerably impacts the soil respiration, biochar wettability and CO<sub>2</sub> adsorption rate<sup>97</sup>. After 1 month of the application of biochars (poplar and pine biochars) to soil, soil respiration decreased by 11.1–13.4%, while a complete disappearance of soil respiration was observed after 1 year of soil treatment. Moreover, biochars change from being hydrophobic to hydrophilic after the aging process. The disappearance of soil respiration could be due to abiotic or biotic oxidation processes which transform biochar from being a water-repellent material to a hydrophilic material<sup>97</sup>. Wang et al.<sup>91</sup> found that the glucose mineralization rate in soil amended with aged biochar was enhanced by 1.4–2.0%, nevertheless a reduction of glucose mineralization rate by 0.1–1.9% was recorded in 120 days incubation with fresh biochar (dose: 10.6 g biochar/kg soil). Several studies have also reported an increase of soil microbial biomass carbon (MBC) with the use of fresh biochar<sup>98–100</sup>. Zhao et al.<sup>98</sup> studied the changes of MBC quantity with the addition of fresh (*Pinus massoniana* tree-based) biochar (dose: 2%) and aged biochar (ages for 10 years, dose: 2%), to infertile soil under an incubation period of 42 days. Compared to the control system without biochar (MBC: 24.64 mg/kg), the MBC amount was higher in soil added with fresh biochar (42.09 mg/kg) than the soil containing aged biochar (36.38 mg/kg). Notably, with the increase of fresh biochar dose from 2 to 5%, the MBC quantity was significantly increased by nearly 5-folds (117.54 mg/kg). This study indicates that biochar application rate/dose considerably influences the mineralization rate of SOC, and the blending of soil with higher amount biochar is often effective to reduce the degree of SOC mineralization rate<sup>101</sup>. Several mechanisms are proposed to explain the reduction of CO<sub>2</sub> emissions from soil treated with aged biochar. In the aging process, dissolved organic matter/soluble organic matter in biochar are decomposed, and organic-mineral complexes are formed. Thus, it inhibits the biodegradation of



soluble organics and the release of CO<sub>2</sub><sup>93</sup>. Furthermore, the abundance and diversity of microbial communities in the aged biochar is different than that of the pristine biochar, with an enrichment of CO<sub>2</sub>-fixing bacteria, which could decrease CO<sub>2</sub> emissions<sup>93</sup>.

The ash content in biochar could impact its potential for soil carbon storage. Biochar with a low amount of ash shows higher potential for long-term soil carbon storage compared to biochar having more ash content<sup>102</sup>. The low ash biochar contains a high amount of stable organic carbon which contributes to the carbon storage in soil system. Variations of CO<sub>2</sub> release rate from soil were found with the change of biochar particle size<sup>103</sup>. Biochar with larger particle size is effective in reducing CO<sub>2</sub> emissions from soil since Windeatt et al.<sup>103</sup> reported that soil added with dust-sized biochar (size: <0.42 mm) had higher CO<sub>2</sub> release (281 mg/kg) than soil amended with pellet-sized biochar (size: >2 mm) (226 mg/kg). Biochar with the smaller particle size could be easily disintegrated in the complex soil system during different processes (e.g., microbial attack), and this disintegration may be the potential reason for the greater CO<sub>2</sub> emissions from the soil blended with smaller particle size biochar<sup>103</sup>.

Among various soil properties, pH is one of the important factors which considerably influences the carbon sequestration potential of biochar. In general, the carbon sequestration potential of biochar decreases with a decrease of soil pH (acidic environment), i.e., more CO<sub>2</sub> release (e.g., 1.5–3.5 times higher) is observed in acidic soil than alkaline or neutral soil systems<sup>71</sup>. The acidic environment also accelerated the biochar aging processes. Biochar amendment decreases soil acidity which is due to the presence of oxides, hydroxides and carbonates of various metals including Ca, Mg, Na and K in biochar<sup>104</sup>. Moreover, soil pH buffering capabilities of biochar depends on its ash content and alkalinity<sup>104</sup>.

Soil types/textures (e.g., sandy loam soil, sandy clay loam soil, etc) impact the biochar stability as well as its carbon sequestration potential<sup>96</sup>. The variations of key soil properties including pH, SOC/TC and TN contents reported in past studies are presented elsewhere (Table S1 and Fig. S4). The stability of biochar is usually higher in sandy clay loam soil compared to sandy loam soil which could be due to the presence of different minerals (e.g., kaolinite, quartz, illite and goethite) and metals (Fe and Al) in clay-based soil<sup>96</sup>. Higher amount of clay and amorphous Fe in sandy clay loam soil reduces the SOC degradation<sup>105</sup>. Yang et al.<sup>96</sup> reported that with the amendment of fresh biochar (2%), more carbon mineralization (13 to 47%) was found in the sandy loam soil compared to sandy clay loam soil system. Nevertheless, changes of soil types have a negligible impact on the carbon mineralization rate when used aged biochar<sup>96</sup>. Bi et al.<sup>105</sup> also reported similar results of soil having high clay content in terms of being effective for the reduction of carbon mineralization since only 1.16% loss of carbon from the applied fresh biochar was observed in quaternary red clay paddy soil (clay content: 32.6%). However, 11.7% biochar carbon loss was found in Yellow River alluvium paddy soil system (clay content: 20.5%). Moreover, the soil SOC was higher in biochar amended soil with higher clay content (64.5 Mg C/hectare) than the soil with lower amount of clay (57.6 Mg C/hectare). Together, biochar properties (type: fresh vs aged, BDOC content, C/N ratio, pH and application rate) and soil characteristics (pH, SOC and clay content) considerably influence on the carbon sequestration potential of biochar in soil (Table 4). Among these properties, soil pH and biochar C/N ratio are considered as the most critical factors which impact the biochar efficiency of the SOC mineralization rate, carbon sequestration potential and crop productivity<sup>106</sup>. A recent meta-analysis study reported that the biochar C/N ratio of ≤50 considerably impacts the change of soil pH<sup>107</sup>.

### Impact of biochar on soil health and fertility

Soil pH, SOC, and aggregate stability are the basic indicators for soil health/soil quality in biochar-treated soil<sup>108</sup>. Biochar amendment positively impacts the soil health, fertility, restoration of degraded soil and crop growth and development<sup>108–111</sup>. In agriculture, “soil health” refers to the capacity of soil to sustain and promote plant growth and development<sup>112</sup>. Overall, biochar provides multifaceted solutions to address issues related to soil health and fertility as well as crop yield<sup>113</sup>. The key physical properties of soil which are

influenced by soil amendment include porosity, water holding capacity and bulk density, whereas pH, salinity and cation exchange capacity are the major chemical properties which are changed by biochar amendment<sup>112</sup>. In biological properties, in addition to changes of soil microbial diversity by biochar use, the changes of MBC and microbial biomass nitrogen (MBN) are also evaluated<sup>112</sup>. Amoakwah et al.<sup>114</sup> reported that with the treatment of soil with corn cob-based biochar (30 tons/hectare) resulted in a significant increase of MBC (i.e., nearly 8 folds, rose from 39.7 to 324.6 mg/kg) and MBN (i.e., nearly 3 folds, rose from 20.5 to 55.1 mg/kg) compared to the control without biochar. Additionally, further increase of MBC (328.5 mg/kg) and MBN (55.7 mg/kg) was recorded with co-treatment of soil with biochar and phosphate fertilizer. Biochar application considerably enhanced the abundance and diversity of both bacterial and fungal communities with higher fungal-to-bacteria ratios being found at higher biochar application rate (30 tons/hectare)<sup>114</sup>. A meta-analysis work involving 97 research publications found an overall increase of MBC and MBN quantity by 25% in biochar treated soil<sup>115</sup>. Another meta-analysis study reported that labile fractions in biochar account for only 3% with mean residence time (MRT) of 108 days while the recalcitrant carbon pool accounts for 97% with MRT of 556 years<sup>105</sup>. Additionally, the biochar addition reduced the SOC mineralization by 3.8%. The negative priming effects typically varied between 8.6–20.3% depending on the nature of feedstocks used for the synthesis of biochar, the pyrolysis temperature maintained during the synthesis and the amount of biochar added to soil. Liu et al.<sup>80</sup> also conducted meta-analysis of 50 publications, and reported that the biochar addition to soil significantly increases the SOC and MBC amounts by 40% and 18%, respectively. The impact of biochar amendment on the soil physicochemical properties is summarized in Table 5. Soil pH, soil aggregate stability and SOC are the three important parameters that predominantly impacts the soil functions<sup>108</sup>. Biochar application to soil enhances crop productivity due to augmentation of soil structure, greater nutrient use efficiency, increase of aeration, porosity and water-holding capacity<sup>110</sup>. The rise of crop productivity by biochar application is mainly found in the coarse-textured and sandy soils than the fine-textured and fertile soils<sup>110</sup>. Moreover, other factors such as soil characteristics, biochar properties and its application rate/dose and type of planted crop species considerably impact on the crop growth and productivity<sup>109</sup>. The findings of a previous study indicate that biochar amendment enhances the crop yield by nearly 20% with the application rate of 10 tons/hectare<sup>111</sup>. Another meta-analysis study reported that biochar application in soil enhances phosphorous availability by 4.6 folds as well as improves the crop yield by 10–42%<sup>116</sup>.

Biochar addition considerably changes different soil properties. For example, it usually increases soil pH, but decreases soil exchangeable acidity, hydrogen (H) and metallic elements (e.g., Al)<sup>117</sup>. In a pot-based experiment in laboratory conditions using agricultural soil, it was observed that the use of biochar (2%, w/w) prepared under different pyrolysis temperatures (300–600 °C) considerably enhanced soil pH by 8.48–79.25% (i.e., pH changed from ~4.0 to ~7.0). Also, biochar amendment reduced soil exchange acidity by 56.94–94.95% and exchangeable H by 58.72–93.27%<sup>117</sup>. Biochar synthesized under different temperatures had different pH values with a higher increase of soil pH noticed following the addition of biochar produced at higher temperature (e.g., 600 °C). In field-scale experiments on agricultural soil using rice husk-based biochar (10 tons/hectare), Sing et al.<sup>118</sup> found that biochar application improved soil nutrients and rice productivity. Compared to control field tests without biochar, rice grain yield was increased by nearly 44% (i.e., rose from 2.57–4.55 tons/hectare) in the agricultural field applied with rice husk-derived biochar. Total nitrogen and total phosphorus contents in the soil were enhanced by 58.3% (increased from 0.05–0.12 g/kg soil) and 14.3% (rose from 0.12–0.14 g/kg soil), respectively.

Different mechanisms have been proposed about the impact of biochar on soil health and fertility. Biochar is considered as a redox active material which could facilitate microbial and abiotic transformations<sup>119</sup>. The electrochemical properties of biochar impact soil redox properties as well as various soil biotic and abiotic processes<sup>120</sup>. Biochar contains various



**Table 4 | Impact of biochar and soil properties on soil organic carbon mineralization and carbon sequestration potential**

Biochar/soil properties	Differential effects	References
Aged/fresh biochar	Addition of aged and fresh biochars decreased soil CO <sub>2</sub> emissions	91
Biochar with low amount of ash	Low-ash containing biochar shows higher potential for long-term soil carbon storage compared ash having high amount of ash	102
Biochar synthesized at lower temperature	Release of biochar-derived dissolved organic carbon, and SOC mineralization	89
Biochar synthesized at higher temperature	Higher porosity and retain more SOC	90
Biochar particle size	Lower CO <sub>2</sub> release in larger particle size biochar (>2 mm)	103
Acidic soil	Accelerated biochar aging and SOC mineralization rate	71
Sandy loam soil	Higher SOC mineralization	96,105
Sandy clay loam soil	Lower SOC mineralization	96,105

**Table 5 | Impact of biochar application on soil biophysical and chemical properties**

Biochar source	Soil type	Effect on soil properties/soil quality changes
Different feedstock types	Different soil types	Increase in soil pH, cation exchange capacity (CEC), available K, Ca and Mg, total N and available P; decrease in Al saturation of acid soils.
Wood charcoal	Anthrosol and Ferralsol	Increase in soil C content, pH value and available P; reduction in leaching of applied fertilizer N, Ca and Mg and lower Al contents.
Eucalyptus logs, maize stover	Clay-loam Oxisol; silt loam	Increase in total N derived from the atmosphere by up to 78%; higher total soil N recovery with biochar addition.
Charcoal site Soil	Haplic Acrisols	Increase in total porosity from 46% to 51% and saturated soil hydraulic conductivity by 88% and reduction in bulk density by 9%.
Peanut hulls, pecan shells, poultry litter	Loamy sand	Biochars produced at higher pyrolysis temperature increased soil pH, while biochar made from poultry litter increased available P and Na.
Wood and peanut shell – Chicken manure – wheat chaff	Sandy soils	Increase in P availability from 163 to 208%, but decreased AMF (arbuscular mycorrhizae fungi) abundances in soils from 43 to 77%.
Wood and manure-derived biochars	Different soil types	Increase the soil's saturated hydraulic conductivity and plant's water accessibility, as well as boost the soil's total N concentration and CEC, improving soil field capacity, and reduce NH <sub>4</sub> -N leaching.
Manure, corn stover, woods, food waste	Alfisol	Tissue N concentration and uptake decreased with increasing pyrolysis temperature and application rate, but increased K and Na content.
Different biochar sources	Different soil types	Increased crop yield, improved microbial habitat and soil microbial biomass, rhizobia nodulation, plant K tissue concentration, soil pH, soil P, soil K, total soil N, and total soil C compared with control conditions.
Peanut hull	Ultisols	Increased K, Ca, and Mg in the surface soil (0–15 cm). Increased K was reflected in the plant tissue analysis.
Simoca, activated wundowie	Loamy sand –clay	Increased soil microbial activity more in clay than loamy soil
Acacia whole tree green waste	Planosol	Increase in porosity either direct pore contribution, creation of accommodation pores or improved aggregate stability
Wheat straw	Fimi-Orthic Anthrosols	Increase in soil pH, organic carbon, total nitrogen and reduction in yield scaled N <sub>2</sub> O emissions

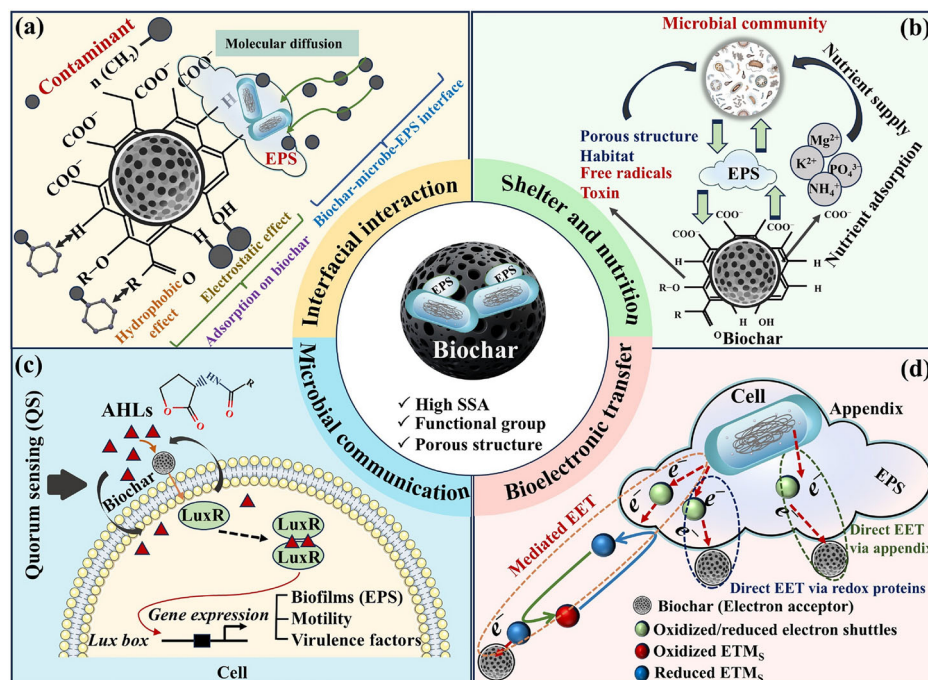
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components such as mineral components as well as carbon in different forms (e.g., labile organic carbon, amorphous carbon, graphitic carbon, etc.) which can act as either electron donors or acceptors for different metabolic pathways in soil ecosystem<sup>120</sup>. Specifically, the presence of functional groups such as phenols can act as an electron donor (i.e., reducers), while quinones and polycondensed aromatic functional groups can accept electrons (oxidants)<sup>121</sup>. Both pyrolysis conditions and feedstock properties impact the redox capacity of biochar. Biochar synthesized at higher temperature (e.g., 400–500 °C) possesses greater electron exchange capacity (EEC)<sup>122</sup>. Additionally, biomass having a high amount of lignin produces biochar with higher EEC. When biochar is applied to soil, it interacts with many materials including SOC and forms organo-mineral-biochar complexes. These complexes are mainly formed by redox reactions. Biochar has been shown to accept/donate electrons from/to their environment (e.g., soil) through abiotic or microbial processes<sup>123</sup>. Yu et al. reported that with the incubation of *Geobacter sulfurreducens* with biochar as the only terminal electron

acceptor under anaerobic conditions, the bacterial biomass was considerably increased by 31-fold<sup>124</sup>. Using *Geobacter metallireducens* (GS-15) as the microbial agent, Saquing et al. calculated the bioavailable electron storage capacities (ESCs) of biochar on the basis of acetate oxidation and nitrate reduction. The ESCs were found to be 0.85 and 0.87 mmol e<sup>-</sup>/g, respectively which are comparable to those of humic substances (e.g., 0.822 mmol e<sup>-</sup>/g for Leonardite Humic Acid)<sup>125</sup>.

Biochar having pore structures (e.g., micro- and macropores) can provide habitat/physical shelter to soil microorganisms for their conducive growth and metabolic activities and protect them from the predators (e.g., exogenous organisms)<sup>126,127</sup>. Moreover, microbes and their secreted extracellular polymeric substances (EPS) play a critical role in biofilm formation on the biochar surface which increases the resilience of microorganisms to different stresses caused by harsh environmental conditions and improves their environmental preformance. EPS can act as a mediator to facilitate electron transfer between microbes and biochar to further enhance the

**Fig. 6 | Potential mechanisms of microorganisms–EPS–biochar interactions.** **a** interfacial interaction, **b** shelter and nutrition, **c** microbial communication, and **d** bioelectronic transfer. Reproduced with permission from ref. 126. Copyright (2025) Springer.



electrochemical properties of biochar<sup>126</sup>. The synergistic interactions between microorganisms–EPS–biochar enhance the soil fertility and contaminants removal capability. The potential mechanisms of microorganisms–EPS and biochar interactions include (1) adhesion and interfacial interactions, (2) shelter and nutrition transfer from biochar to microbes, (3) microbial communication and (4) bioelectronic transfer<sup>126</sup>. These mechanisms are shown on Fig. 6. The surface functional groups (e.g., hydroxyl, carboxyl, ketone and quinone) in biochar show high affinity for adsorption of nutrients which are required for microbial survival and growth.

Biochar aging changes its physicochemical properties; thus, it may also impact the soil health and fertility specifically in the long-term. Aged biochar usually shows different effects on soil qualities. For example, compared to fresh biochar treatment, a decrease of pH (i.e., liming effects, 0.09–0.55 units) and an increase of the SOC content (2.59–10.75 g/kg) was noticed with the application of aged biochars (acidification, dry and wet, and freezing–thawing aging biochars)<sup>128</sup>. Moreover, the activities of two key enzymes namely dehydrogenase and urease were reduced by 10.74–20.99% and 3.37–24.12%, respectively with the treatment of aged biochars. These biochar properties may change over its long-term applications in soil, which may in turn impact soil health and quality. In another study involving the aging of biochar under field conditions over a period of 24 months, an increase of the physical fragmentation of biochar particles was noticed with an increase of aging time<sup>129</sup>. Moreover, FESEM (field emission scanning electron microscope) micrographs shows that biochar pores were filled with microbial biomass and soil materials. The biochar surface functionalities changed with an increase of O-alkyl C and alkyl C in the aged biochars, which could be due to the disintegration of aromatic hydrocarbons. The formation of organo-mineral complexes with enrichment in oxygenated functional groups in pores and biochar surfaces during aging were also found in a previous work<sup>130</sup>. Soil total carbon and nitrogen contents tend to increase with the biochar age, which suggests negative priming effects (an increase of carbon sequestration)<sup>131</sup>.

Biochar aging considerably impacts on the soil microbial activities and enzymatic activities. The dynamics of soil microbial communities and/or enzyme activities under short-term and long-term biochar applications were explored in past studies<sup>132,133</sup>. Using 16S rRNA sequencing, Nguyen

and co-workers investigated short term (1 year) and long term (9 years, aged biochar) impacts of biochar (application rate: 10 tons/hectare) amendment on the soil bacterial communities<sup>133</sup>. Microbial diversity was remarkably changed in short and long term biochar amendments. Bacterial diversity was considerably increased in the short term, but no significant changes of bacterial diversity and community structure were found in the long term after biochar treatment. Moreover, after 1 year of biochar treatment, the abundance of bacteria involved in carbon cycling/degradation of aromatic hydrocarbons (e.g., *Gemmatimonadetes* and *Actinobacteria*) and N cycling (e.g., nitrifiers: *Nitrospirae*) was increased<sup>133</sup>. Correlation analysis revealed that the bacterial community was impacted by soil qualities, specifically soil pH and the SOC content in soil. In the long term, the bacterial communities may be acclimatized to the biochar treated soil environment, and hence, no remarkable changes of bacterial diversity were noticed. The findings of this study suggest that the impact of biochar on soil biological properties is time dependant. Futa et al. reported that soil physicochemical properties as well as enzymatic activities were altered with natural biochar aging (application rate: 10–30 tons/hectare) over long-term field tests (4–6 years)<sup>132</sup>. The SOC, TN and nitrate contents were significantly enhanced in biochar treated soils in 4–6 years. The enzymatic activities also increased with the rise of biochar application rate with a constant increase of dehydrogenase activity up to 30 tons/hectare, but the highest phosphatase and urease activities were recorded with slightly lower biochar application rate (20 tons/hectare). These results suggest that the biochar application rate controls soil enzymatic activities. In a recent work, the field-scale natural aging (9 years) caused a reduction of surface area (decreased from 46.04 to 38.2 m<sup>2</sup>/g) of biochar which indicates disintegration/deformation of biochar due to aging<sup>134</sup>. Considerable changes of microbial (specifically fungal) communities abundance were also observed, the abundance of arbuscular mycorrhizal fungi (AMF) considerably increased (1.03–1.16 folds) in both aged and fresh biochar treated soil samples. However, there was a slight increase (1.01–1.02-fold) of abundance gram-positive and gram-negative bacteria in the aged biochar-treated soil samples. In field-scale trials, Zhang et al. found that the application of biochar (5.25–42 g/kg), which was aged for 5 years, to sandy soil considerably altered carbon fractions and enzyme activities<sup>135</sup>. The SOC and MBC contents increased by 122 and 900%, respectively, while soil invertase and urease activities were enhanced by 46.76 and 55.81%, respectively.

Taken together, based on the literature analysis, it is found that biochar amendment to soil decreases bulk density, but enhances soil pH, moisture retention/water holding capacity, aggregates stability, soil porosity, infiltration rate, cation exchangeable capacity, nutrient absorption and retention, as well as accelerates microbial and enzymatic activities. These changes create a fertile environment for sustainable agricultural activities including increased crop growth and productivity<sup>112,136,137</sup>. Moreover, the aging of biochar occurring during its long-term applications alters its physico-chemical properties with considerable impacts on the soil health and fertility.

## Impact of biochar on soil microbial communities and enzymes

Biochar amendment into soil significantly impacts both the abundance and diversity of soil microorganisms (bacteria and fungi) (Table 6). An improvement in microbial diversity is reported in many studies<sup>73,117</sup>. Biochar application usually impacts the dynamics of the carbon metabolism-inducing microorganisms including microbial communities responsible for degradation of labile and complex organic carbons<sup>73</sup>. Microorganisms usually release EPS. EPS contain well-developed hyphae (e.g., fungi) which help with the enhancement of biochar-based soil aggregation<sup>73</sup>. The shift of soil microbial communities with the biochar use suggests possible changes of qualities and quantities of soil nutrients by biochar<sup>73</sup>. Biochar has more influences on the changes of the composition of bacterial community rather than those of fungal community in agricultural (vegetable) soil systems<sup>138</sup>. The ratio of gram-negative ( $G^-$ ) (e.g., *Alphaproteobacteria* and *Bacteroidetes*)/gram-positive ( $G^+$ ) bacteria (e.g., *Acidobacteria*) was found to be negatively co-related with the priming effects<sup>71</sup>. Acidic soil environments promote the enrichment of  $G^+$  bacteria which are mainly responsible for the disintegration of biochar as well as mineralization of SOC. An considerable increase (1.5–3.5 folds) of  $CO_2$  emissions was noticed in biochar treated acidic soil<sup>71</sup>. Biochars prepared at different pyrolysis temperatures had different effects on both the soil microbial communities and enzyme activities with the reduction of microbial community abundance and enzyme activities reported, following the application of biochar produced at higher temperatures. This means that the presence of recalcitrant aromatic hydrocarbons in biochar shows negative effects on microbial growth and enzymatic activities<sup>139</sup>. Zhang et al.<sup>139</sup> stated that the treatment of soil with biochar (2.5% w/w) produced at higher temperatures (400 and 600 °C) caused a decline of bacterial quantity by 6.7–27%, while much higher reduction of fungal quantity (19–35%) was noticed. The enzymatic activities (ligninolytic and catechol 2,3-dioxygenase) were also reduced (13–42%) with the increase of pyrolysis temperature. The high porosity of biochar synthesized at high pyrolysis temperatures causes adsorption of water and nutrients from soil, making the soil with low moisture and nutrient contents which hinders microbial growth<sup>139</sup>. The dominant bacterial communities (abundance: 6.1–52%) enriched in the biochar treated soil include *Actinobacteria*, *Proteobacteria*, *Chloroflexi*, and *Acidobacteria*. No consistent trend was found when compared the abundance of these bacteria with soil without biochar addition. In fungal community, *Ascomycota* was the major candidate with its abundance varying between 73–96%, and its abundance increased by 6.4–21% due to biochar amendment<sup>139</sup>.

In addition to the impact of biochar on soil microbiological properties (e.g., microbial communities and enzyme activities), changes in soil biogeochemical functions/cycling (e.g., carbon, nitrogen and phosphorous cycling) were observed in the biochar treated soil<sup>140–142</sup>. A comprehensive understanding of microbially driven C, N and P cycling in biochar amended soil is needed to develop novel strategies for the reduction of GHG emissions from soil<sup>142</sup>. Several studies have reported that an increase in enzymatic activities was responsible for C ( $\beta$ -glucosidase) and N cycling (e.g., urease)<sup>143,144</sup>. In a recent short term (45 days) field-scale trials, the biochar application (10 tons/hectare) showed an enhancement of carbon immobilization into bacterial biomass with the reduction of specific microbial respiration ( $qCO_2$ )<sup>140</sup>. Moreover, the abundance of various communities

including the gram negative bacteria and AMF was considerably enhanced in the biochar treated soil with respect to control which received no treatment. In pot-based experiments, based on 454-pyrosequencing analysis of soil samples, Anderson et al. reported that biochar treatment changed the soil microbial communities' structures which influence biogeochemical cycling<sup>142</sup>. Biochar treatment caused the temporal changes in the following bacterial family: *Bradyrhizobiaceae*, *Hyphomicrobiaceae*, *Streptosporangineae* and *Thermomonosporaceae* which are involved in various biogeochemical cycling (e.g., C and N cycling). In a pot-based experiment (372 days), it has been reported that the biochar application (oak wood/bamboo biochar, 0.5–2.0%, w/w) changed carbon fractions and enzymatic activity in red soil<sup>144</sup>. The increase of SOC content indicates an increase of carbon sequestration in soil. The lowest biochar dose (0.5%) was effective in increasing microbial enzymatic activities, SOC and soil stability (macro-aggregates formation). The dehydrogenase activity, which is usually employed as an indicator to measure the degree of recovery of degraded soils, was increased, which shows an increase in the stability of soil. Differential effects on  $\beta$ -glucosidase activities were observed with the use of two different types of biochar (i.e., a decrease with oak wood, but an increase with bamboo biochar), which could be due to differences in their physicochemical properties<sup>144</sup>. In a short-term field-scale test (18 months), changes in the abundance and microbial community structure as well as carbon cycling were found with the application of biochar (40 tons/hectare) to an acidic rice paddy soil<sup>145</sup>. Soil pH, SOC, TN, MBC and MBN were significantly increased, but soil bulk density was decreased with biochar treatment. Biochar applications enhanced the enrichment of total and metabolically active bacteria, but showed negative effects on fungi. Biochar also changed the microbial community structure with an enrichment of microbes that can utilize polymers, phenols and amines as carbon substrates<sup>145</sup>. The reduction of soil respiration and  $\beta$ -glucosidase enzyme activity (involved in carbon mineralization) was recorded, which indicates an increase in soil carbon stability and carbon sequestration potential by biochar treatment<sup>145</sup>. A current meta-analysis work (analysis of 131 articles) reported that short-term (less than 1 year) biochar applications considerably decreased cellulase activity by 4.6% and increased soil carbon sequestration by 87.5%. Nevertheless, long-term ( $\geq 1$  year) biochar treatment considerably increased ligninase activity by 5.2% with relatively smaller enhancement of carbon sequestration by 25.1%<sup>146</sup>. The results from this work suggest that a shift in enzyme activities occurred in soil with biochar applications in different time periods.

Microbially-driven iron cycling could impact soil carbon cycling in biochar treated soil<sup>73</sup>. He et al.<sup>73</sup> reported that the biochar amendment (9.0 tons/hectare/year) considerably changes the dynamics of soil bacterial and fungal communities. The dominant bacterial communities in biochar added soil include *Actinobacteria* and *Firmicutes* phyla, while the major fungal communities enriched were *Ascomycota* and *Basidiomycota* phyla<sup>73</sup>. Biochar addition resulted in nearly 11% reduction of  $CO_2$  emissions in short term period (56 days). The integration of iron cycling into carbon cycling increased microbial extracellular electron transfer reactions and the carbon use efficiency of soil microbes. In a pot-based study<sup>117</sup>, it was found that fruit tree branches-derived biochar amendment to topsoil collected from an agricultural site at the dose of 2% (w/w) caused an enrichment of different microbial communities with the top three bacterial phyla being *Actinobacteriota* (38.0%), *Proteobacteria* (27.0%), and *Chloroflexi* (11.0%), while the top most abundant fungal phyla enriched were *Ascomycota* (50.0%), *Opisthokonta* (26.0%) and *Basidiomycota* (12.0%). The activities of key soil enzymes such as urease and phosphatase increased with the biochar addition. Biochar amendment also appears to promote carbon sequestration since a significant increase (109.3%) of SOC was found. In a lab-scale microcosm-based system using sandy loam Alfisol, the richness and diversity of bacterial community in soil mixed with biochar (4%, w/w, prepared from biogas residues from an anaerobic digester) were not significantly impacted since the reduction of richness and diversity varied between only 4–7%<sup>90</sup>. However, much higher reduction (19–28%) of



**Table 6 | Dynamics of soil microbial communities with the application of biochar**

Biomass for biochar synthesis	Biochar application rate/dose	Soil type	Dynamics of soil microorganisms		References
			Bacteria	Fungi	
Corn straw	9.0 tons/hectare/year	Agricultural soil	Major bacterial phyla include <i>Actinobacteria</i> and <i>Firmicutes</i>	Major fungi phyla include <i>Ascomycota</i> and <i>Basidiomycota</i>	73
Fruit tree branches	2% (w/w)	Agricultural soil	Dominant phyla: <i>Actinobacteriota</i> (38.0%), <i>Proteobacteria</i> (27.0%), <i>Chloroflexi</i> (11.0%), <i>Acidobacteriota</i> (6.1%), and <i>Firmicutes</i> (4.2%)	Dominant phyla: <i>Ascomycota</i> (50.0%), <i>Opisthokonta</i> (26.0%), <i>Basidiomycota</i> (12.0%), and <i>Mortierellomycota</i> (8.9%)	117
Biogas residues	4% (w/w)	Sandy loam Alfisol	Higher relative abundance (29.57–30.41%) of phyla <i>Actinobacteria</i> in biochar treated soil than control systems without biochar (16.64–24.80%)	NA	90
Walnut shells, corn cobs, corn stems, and rice straw	2.5% (w/w, equivalent to 39 tons/hectare)	Surface soil from an indigenous coking area, unfavorable for growth of some plants	Top three phyla: <i>Actinobacteria</i> (29–52%), <i>Proteobacteria</i> (20–34%), <i>Chloroflexi</i> (12–24%)	Top three phyla: <i>Ascomycota</i> (73–96%), <i>Ciliophora</i> (1.0–13%) and <i>Chytridiomycota</i> (0.27–11%)	139
Corn cobs	30 tons/hectare	Haplic Acrisols	Abundance of gram-positive and gram-negative bacteria increased by 1.7 and 1.5 folds, respectively	Abundance of arbuscular mycorrhizal fungi increased by 4.5 folds. Soil fungal abundance rose by 5.4 folds	114
Holm oak chips	20 tons/hectare/year	Soil with a loam texture	<i>Actinobacteria</i> phylum was the most dominant (88.5%). <i>Acidobacteria</i> abundance was significantly reduced by biochar treatment	<i>Ascomycota</i> was the dominant fungal phylum (89.7%). Abundance of <i>Chytridiomycota</i> was significantly reduced by biochar treatment	191
Wheat straw	20 or 40 tons/hectare	Agricultural soil	Dominant phyla: <i>Proteobacteria</i> , <i>Acidobacteria</i> , <i>Chloroflexi</i> and <i>Actinobacteria</i> (total abundance: > 80%). Abundances of <i>Actinobacteria</i> and <i>Chlorobi</i> with 40 tons/hectare were significantly reduced by 21% and 35%	<i>Ascomycota</i> was dominant phylum in the non-treated soils with abundance of 74.5% and reduced to 66.3% in the biochar added soils at 40 tons/hectare. The phylum <i>Zygomycota</i> abundance rose by 43% and 147% in biochar applied soils at 20 and 40 tons/hectare	148
Wheat straw	40 tons/hectare	Rice paddy soil (sandy loam soil)	Changes in abundance of <i>Chloroflexi</i> and <i>Actinobacteria</i> was observed	Changes of <i>Ascomycota</i> and <i>Glomeromycota</i> was observed	145

bacterial richness and diversity was noticed in soil processed with only biogas residues (no biochar). Moreover, the soil amendment with raw biogas residues or biochar derived using this as a feedstock considerably shifted the abundance of bacterial community<sup>90</sup>. The relative abundance of phyla *Actinobacteria* was 16.64% in biogas residues treated soil, but its abundance was nearly doubled (30.41%) in soil treated with biochar pyrolyzed at 600 °C. Moreover, slightly lower abundance (29.57%) of *Actinobacteria* was found in soil treated with biochar produced at 300 °C<sup>90</sup>. The CO<sub>2</sub> emission rate was declined in the initial incubation period (8 days) and remained stable for up to 2 months. These findings suggest that the biochar synthesized at different pyrolysis temperatures shows differential effects on the dynamics of soil microbial communities. A recent global meta-analysis of 24 research articles showed that biochar amendment into soil remarkably impacted mainly two bacterial phyla namely *Acidobacteria* and *Gemmatimonadetes*<sup>147</sup>. Furthermore, the relative abundance of *Acidobacteria* and *Gemmatimonadetes* was decreased by 14.6% and 19.8%, respectively<sup>147</sup>. Enhancement of soil carbon sequestration by 87.5% and 25.1% was reported for short term (less than 1 year) and long term (more than 1 year) biochar application, respectively. Taken together, biochar considerably enhances soil bacteria richness and diversity. However, the key factors including biochar dose, synthesis temperature, biochar properties (pH and C/N ratio) as well as soil properties (pH, SOC and C/N ratio) significantly impact microbial dynamics in biochar amended soil<sup>147,148</sup>.

Effects of biochar amendment on different soil enzyme activities (specifically enzymes which are responsible for the carbon, nitrogen and phosphorus cycling) including urease, phosphomonoesterase, catalase, β-glucosidase, cellobiohydrolase and xylanase were explored in earlier works (Table 7)<sup>117,149</sup>. Overall, the biochar addition changes soil enzyme activities (C, P, and N cycling) with an increase of specific enzyme activities observed with the rise of biochar dose, but biochar quality and soil conditions considerably influence the enzyme activities<sup>138,149</sup>. In a recent publication, carbon degrading soil enzyme activities were analyzed which revealed that the biochar incorporation into soil accelerated the soil ligninase activity (degradation of complex phenolic compounds) by 7.1%, but reduced the cellulase activity (degradation of polysaccharides) by 8.3%<sup>146</sup>. Moreover, the long-term (more than one year) biochar treatment considerably increased the ligninase activity by 5.2% which led to the enhancement of soil carbon sequestration by 25.1%<sup>146</sup>. In another study, Geng et al.<sup>117</sup> reported that the biochar (synthesized using different feedstocks namely fruit tree branches, peanut shells, and cow dung) amendment to soil collected from an agricultural site enhanced β-glucosidase and cellobiohydrolase activity by 74.9–120.4% and 32.8–141.9%, respectively. However, biochar derived from fruit tree branches and peanut shells significantly enhanced the xylanase activity. In a long-term experiment over a period of 2 years on calcareous soil using maize straw-derived biochar, the activities of several enzymes involved in the C, N and P cycling were enhanced with biochar treatment<sup>149</sup>. The urease and phosphomonoesterase activities considerably increased from 0.16 to 0.32 NH<sub>4</sub><sup>+</sup>-mg/g/d, and 166.80 to 176.54 nmol/hr/g, respectively with the rise of biochar dose from 2.5 to 22.5 tons/hectare. Moreover, the β-glucosidase activity rate increased from 61.32 to 70.53 nmol/hr/g<sup>149</sup>. A significant increase in the SOC amount was found after biochar application which suggests an enhancement of carbon question. These findings indicate that biochar application rates remarkably influence the soil enzyme activities. In pot-based experiments using rice husk-derived biochar at a dose of 20 g/kg soil, the phosphatase enzyme activity was enhanced by 28% compared to the control test which consisted soil without biochar<sup>150</sup>. The increase of phosphatase activity could be due to the rise of pH in soil by biochar addition and/or the increase of P content in soil caused by biochar amendment<sup>150</sup>. Moreover, lower urease activity was noticed, i.e., only 4.3% higher than the control system. The lower urease activity could be due to the presence of limited amounts of N-containing compounds in the soil. In summary, the influence of biochar on soil enzyme activities depends on the types of enzymes and biochar qualities<sup>139</sup>.



**Table 7 | Impacts of biochar amendment on soil enzyme activities**

Biomass for biochar preparation	Biochar application rate	Soil type	Changes of various soil enzyme activities	References
Fruit tree branches, peanut shells and cow dung	2% (w/w)	Agricultural soil	Activity of $\beta$ -glucosidase and cellobiohydrolase enhanced by 74.91–120.39% and 32.77–141.86%, respectively	117
Maize straw	0–22.5 tons/hectare	Calcareous soil	The activities of C, N, and P cycling enzymes considerably increased with rise of biochar application rate	149
Rice husk	20 g/kg soil	Agricultural soil	Phosphatase and urease activity increased by 28% and 4.3%, respectively	150
Pine wood	10 Mg/hectare	Agricultural soil	$\beta$ -glucosidase and urease activities increased by 6.1% and 1.4%, respectively	143
Walnut shells, corn cobs, corn stems, and rice straw	2.5% (w/w, equivalent to 39 tons/hectare)	Surface soil from an indigenous coking area, unfavorable for growth of some plants	Reduced Lignin-peroxidase activity by 16–58% as well as decreased laccase, Manganese-dependent peroxidase, and C23O activity by 17–36%, 13–34%, and 17–42%, respectively	139
Corn cobs	30 tons/hectare	Haplic Acrisols	Urease activity increased by 1.5-fold, whereas dehydrogenase activity enhanced by 3.2-fold	114
Holm oak chips	20 tons/hectare/year	Soil with a loam texture	$\beta$ -glucosidase activity significantly decreased, while urease activity considerably increased	191
Wheat straw	20 or 40 tons/hectare	Agricultural soil	Dehydrogenase activity reduced by 52% and 40%, while $\beta$ -glucosidase activity decreased by 28% and 49%	148
Wheat straw	40 tons/hectare	Rice paddy soil (sandy loam soil)	$\beta$ -glucosidase activity was decreased, whereas a dehydrogenase and alkaline phosphatase activities were increased	145
Rice husk	2% (w/w)	Field soil (sandy loam texture)	Increase of urease activity by 51%. No significant effects on the $\beta$ -glucosidase and phosphatase activities	88
Wheat straw	13.5 tons/hectare/year	Agricultural soil	Polyphenol oxidase activity increased by 31%	180

## Potential mechanisms of biochar-based carbon sequestration in soil

Biochar-based carbon sequestration in soil is mediated by multiple inter-connected factors. The combined effects of these factors result in the overall increase of carbon sequestration and carbon storage capacity of soil<sup>96,151</sup>. The key factors by which biochar contributes to the increase of carbon sequestration rate in soil environment include (i) increased SOC contents in soil, (ii) protection against microbial degradation, (iii) enhanced soil aggregation, (iv) increased contents of nutrients and water, (v) stabilization of labile/leachable carbon, and (vi) abundance and diversity of soil microbial communities and functional enzymes<sup>151</sup>. These mechanisms are shown in Fig. 7. Biochar amendment in soil induces negative priming effects (i.e., reduced CO<sub>2</sub> release from soil) by a combination of various mechanisms namely substrate switch, dilution, immobilization of substrates, reduction of microbial accessibility to SOC/organic substrates through sorption and soil aggregation, as well as reduction of SOC mineralization and decomposition rates by inhibition of microbial activities and functional enzyme activities due to limited availability of nutrients which are required for microbial growth<sup>96,152,153</sup>. The extracellular enzymes such as  $\beta$ -glucosidase (responsible for degradation of SOC to CO<sub>2</sub>) is one of the key factors that controls SOC mineralization<sup>154</sup>. According to a recent study, a considerable (30%) reduction of  $\beta$ -glucosidase activity in soil was found with the application (4%, w/w) of potassium-modified biochar<sup>154</sup>. Luo and Gu reported that the application (0.5–2%, w/w) of bamboo residues-based biochar to sediment reduced several enzyme activities (e.g., peroxidase, acid phosphatase and N-acetyl-glucosaminidase) and the abundance of microbial communities (bacteria and fungi)<sup>155</sup>. Specifically, the abundance of bacteria and fungi was reduced from  $5.25 \times 10^{10}$  to  $9.18 \times 10^9$ , and from  $1.48 \times 10^8$  to  $2.39 \times 10^7$  copies/gram dry sediment, respectively. In the biochar amended soil, the enrichment of oligotrophic bacteria (e.g., *Actinobacteria* and *Anaerolineae*) could result in the decrease of CO<sub>2</sub> emissions from soil. In a long-term (6 years) field-scale study with 20–40 tons/hectare biochar application, the soil carbon mineralization rate was decreased by 4.2–19.4%<sup>156</sup>. Moreover, the decrease of carbon mineralization was linked to the reduction of various carbon hydrolyzing enzyme activities namely  $\alpha$ -glucosidase (20%), cellobiohydrolase (17%),  $\beta$ -glucosidase (13%) and xylanase (2.5%). In a recent study on a temperate wheat-maize agroecosystem, the amendment of soil with both biochar and straw showed a positive effect on the reduction of SOC mineralization since a DOC decrease of 18.5% was noticed<sup>25</sup>. The dominant bacterial communities detected in the soil systems include phyla of *Actinobacteriota* (22.2–28.2%), *Proteobacteria* (16.9–24.4%), *Acidobacteriota* (11.3–25.1%), and *Chloroflexi* (11.2–13.6%). Importantly, biochar addition caused reduction of microbial (e.g., *Actinobacteriota* by 12.3%) and enzyme activities (e.g.,  $\beta$ -N-acetyl-glucosaminidase by 24.2%).

Key biochar properties including types of feedstocks used for biochar synthesis, pyrolysis conditions, structural properties, incubation time and application rate significantly impact soil properties and soil microbial dynamics, thus changing the carbon sequestration potential rate and overall soil carbon stock<sup>153</sup>. Biochar acts as a stable form of carbon and persists in soil environment for a longer period of time, thus enhancing the SOC input (carbon sequestration)<sup>151</sup>. Biochar prepared at higher temperature (e.g., >500 °C) is usually rich in aromatic hydrocarbons, which are stable and recalcitrant in nature<sup>157,158</sup>. Moreover, biochars synthesized at higher temperature are rich in porosity and possesses high specific surface area which are beneficial to achieve higher sorption of SOC onto biochar surface. In biochar, the proportion of labile carbon pool is only 3%, while the amount of recalcitrant carbon pool is 97%<sup>159</sup>. The decomposition rate of labile carbon (0.0093%/day) is much higher than the recalcitrant carbon (0.0018%/day). However, the mean residence time of recalcitrant carbon (556 years) is significantly greater compared to labile carbon (108 days)<sup>159</sup>. These findings suggest that biochar prepared at higher temperature (e.g., 500–650 °C) should be considered for soil amendment to achieve greater carbon sequestration rate as well as overall soil carbon stock. Several studies have reported that biochar enhances soil CO<sub>2</sub> release specifically at the early stage

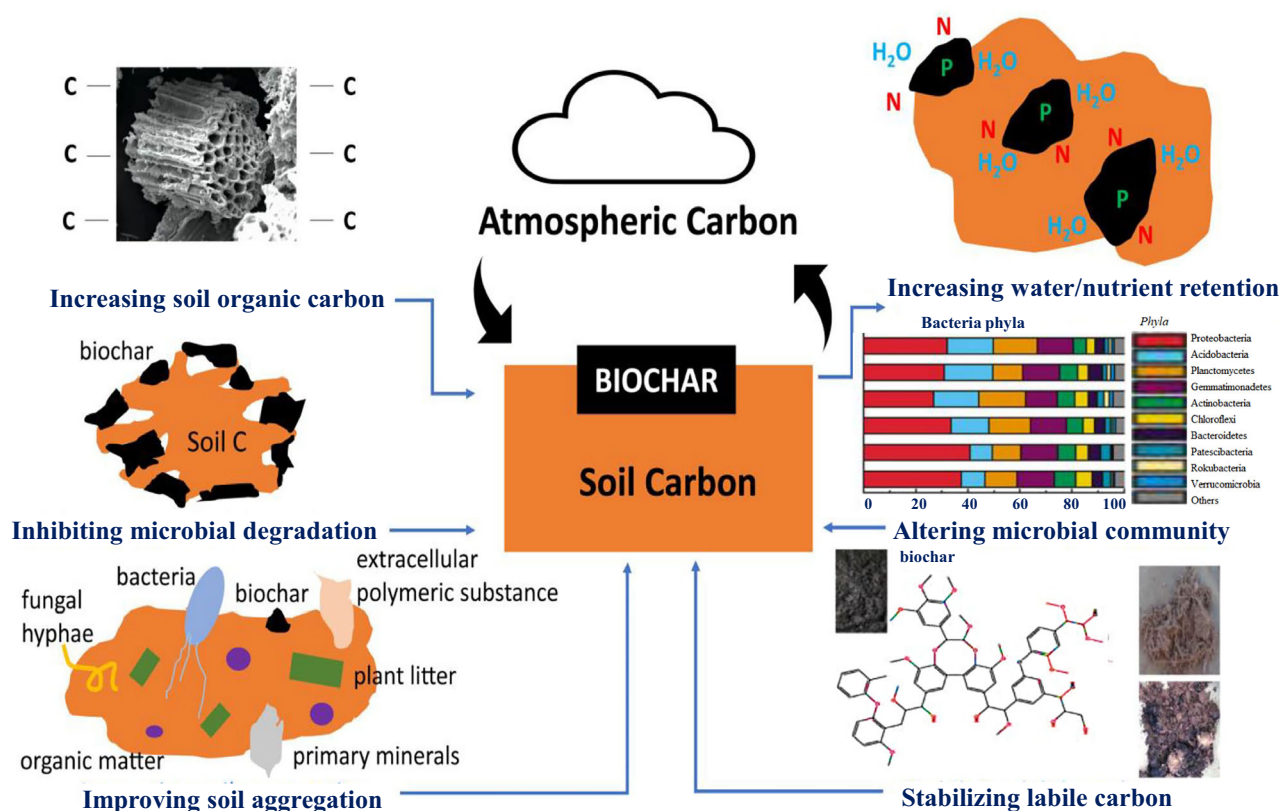


Fig. 7 | Key mechanisms of biochar on enhancing soil carbon sequestration. Reproduced with permission from ref. 151. Copyright (2023) MDPI.

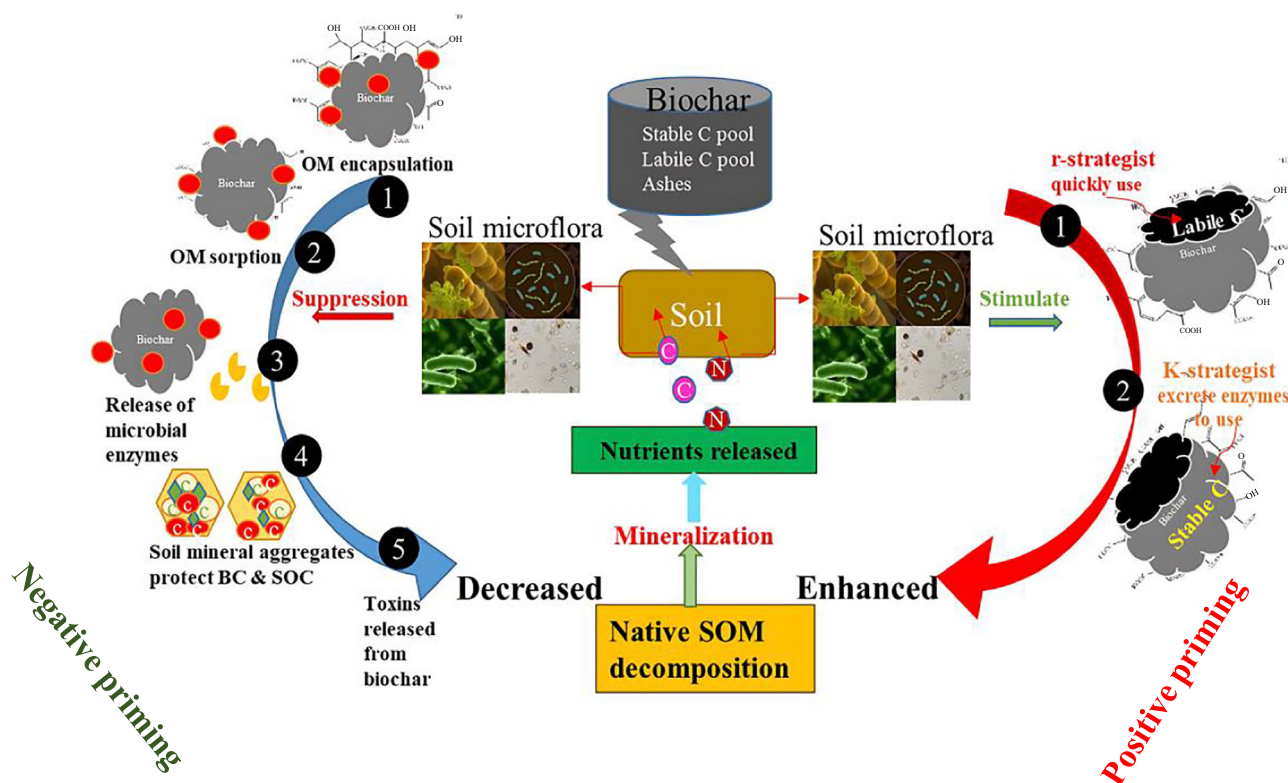
of incubation period (i.e., positive priming effects)<sup>68,160</sup>. Biochar produced at lower temperature (e.g., 250–400 °C) usually increases carbon mineralization rate (positive priming) in soil due to less pores and smaller specific surface area<sup>161</sup>. Thus, there is a weak protection of SOC from degradation due to low degree of sorption of SOC onto biochar with low porosity/specific surface area<sup>162</sup>. A meta-analysis study involving a critical analysis of results from 91 research articles reported an enhancement of CO<sub>2</sub> release by 22.14% with biochar treatment to soil<sup>68</sup>. The increase of soil CO<sub>2</sub> release is mainly due to the increase of microbial activities, the rise of SOC by the input of biochar-derived organic carbon (labile fraction) and abiotic release of inorganic carbon<sup>96,152,160</sup>. The increase in the abundance of copiotrophic bacteria including *Gemmatimonadetes* and *Bacteroidetes* usually promotes the CO<sub>2</sub> release from soil<sup>9</sup>. Moreover, the increase of enzyme activities specifically  $\beta$ -glucosidase could promote SOC degradation and CO<sub>2</sub> emission from soil systems<sup>155</sup>. Wang et al. found that the biochar addition (15.75–47.25 tons/hectare) in maize field enhanced the soil respiration, increasing soil CO<sub>2</sub> emissions by 18.04–73.15%<sup>163</sup>. The increase of CO<sub>2</sub> emissions was linked to the increase of various enzyme activities namely  $\beta$ -glucosidase, sucrase, catalase and urease. A recent field-scale study over a period of 3 years reported that the application of both biochar and nitrogen fertilizer caused a considerable (9–48%) increase of CO<sub>2</sub> emissions from soil compared to a control site which received no treatment by biochar or fertilizer<sup>76</sup>. The biochar treatment considerably influenced the soil microbial functional diversity with higher bacterial-to-fungal ratios were found in soil which received both biochar and N fertilizer treatment than soil with biochar treatment alone. Jing et al. reported that the presence of N in the biochar-amended soil can considerably impact the CO<sub>2</sub> emission rate as well as the microbial functional genes involved in carbon degradation<sup>164</sup>. For example, when N was present at a lower concentration (3–6 g N/m<sup>2</sup>/y) in the biochar-amended soil, it enhanced CO<sub>2</sub> emission rate by accelerating the SOC-degrading enzyme activities and increased the abundance of labile carbon degrading genes (e.g., *amyA*, *glucoamylase* and *pula*). However, higher concentrations of N (9 g N/m<sup>2</sup>/y) showed inhibitory effects, and

decreased CO<sub>2</sub> emissions due to reduction in the abundance of functional genes involved in the degradation of both labile carbon (*amyA*, *glucoamylase* and *pula*) and recalcitrant carbons (*vana*, and *phenol-oxidase*)<sup>164</sup>. These findings indicate that nitrogen addition to biochar-treated soil changes the abundance and diversity of functional genes involved in the carbon cycling as well as the CO<sub>2</sub> emission rate. Microbial degradation of various organic fractions in soil and/or biochar such as starch, cellulose, hemicellulose and lignin could contribute to CO<sub>2</sub> emissions<sup>165</sup>. The key functional genes that drive the biodegradation of these compounds include *sga* for starch, *abfA*, *manB* and *xylA* for hemicellulose, *cex* for cellulose and *lig* and *mnp* for lignin<sup>165</sup>.

The potential mechanisms of negative and positive priming effects by biochar application in soil are illustrated in Fig. 8. Overall, inhibition/alteration of soil microbial communities/functional enzymes (specifically C, N and P cycling) and sorption of SOC/organic substrates onto biochar enhances the carbon sequestration by soil, but the increased SOC mineralization caused by soil microbial activities can lead to the release of CO<sub>2</sub> from soil. Specifically, the carbon sequestration potential and priming effects of biochar in soil largely depend on both the biochar and soil properties.

### Sustainability aspects of biochar amendment in soil for carbon sequestration

Biochar amendment into soil is considered as a sustainable solution to address the emerging global climate change issues by enhancing carbon sequestration from the atmosphere and reducing greenhouse gas emissions from soil<sup>69,166</sup>. Several LCA-based studies were conducted to assess the environmental impacts of biochar soil amendment systems, or to evaluate the potential of biochar-treated soil as a negative emission strategy<sup>26,167–170</sup>. Direct comparison of results from different LCA-based studies is not possible due to the different scope of LCA studies and the difference in biochar characteristics, soil conditions, etc.<sup>167</sup>. A cautious analysis of the whole system of biochar synthesis, and its application to soil are required. Prior large-



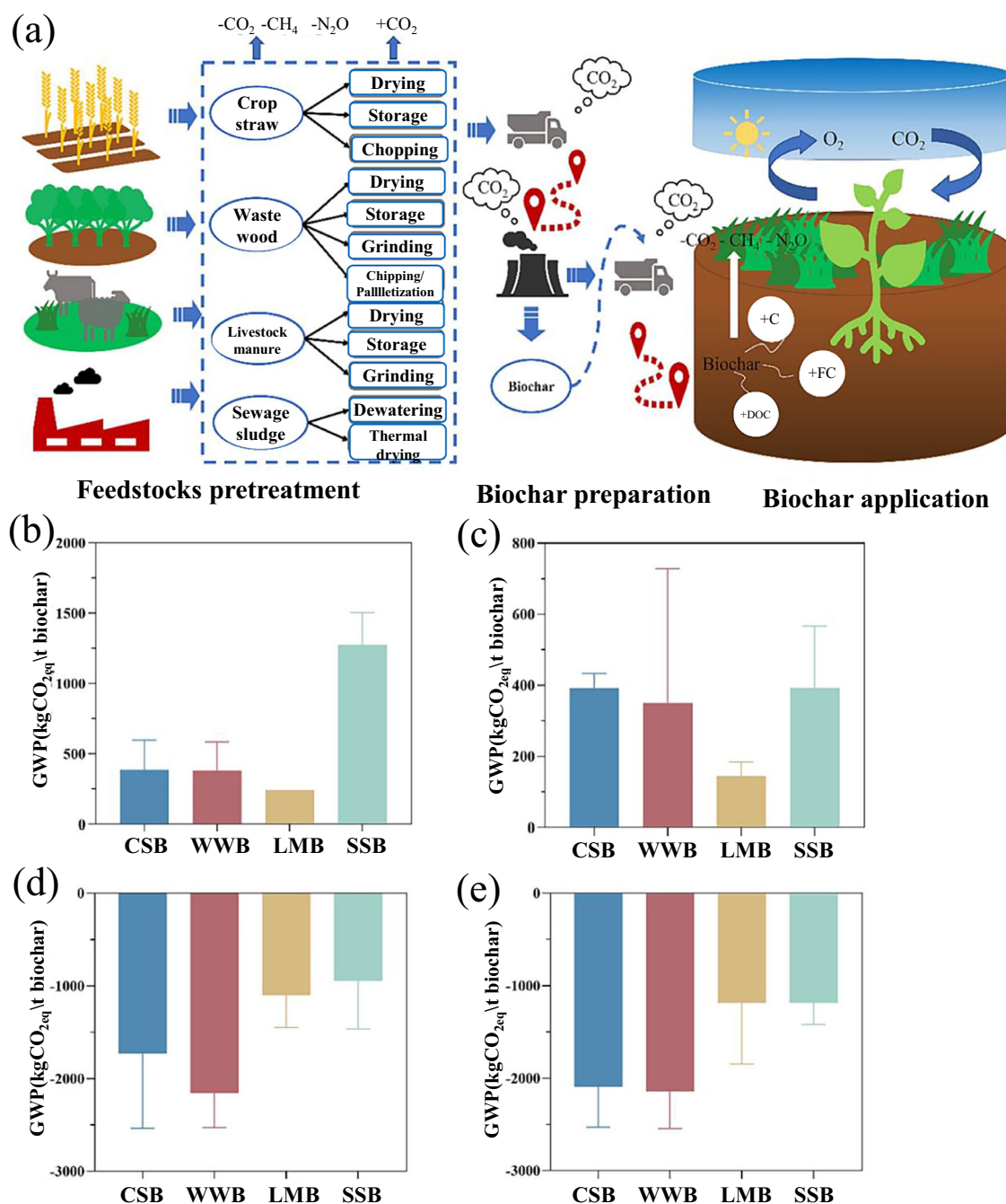
**Fig. 8 | Key mechanisms of negative and positive priming effects induced by the biochar amendment in soil.** Reproduced with permission from ref. 153. Copyright (2022) Elsevier.

scale biochar production with desired characteristics and its use in soil in a field-scale are needed to evade making greater negative environmental impacts<sup>171</sup>. The LCA tool is efficient in evaluating the effects of the different stages of the biochar production and use over its life cycle on the GHG emissions<sup>170</sup>. The key steps involved in the biochar life cycle include feedstock collection and/or pre-treatments, pyrolysis of selected feedstock for biochar production, transportation of produced biochar to the place of application, and soil amendment<sup>167,170</sup>. These are the four potential sources/stages that contribute to GHG emissions to the atmosphere in biochar life cycle. Soil amendment with pyrolyzed biochar results in the following important benefits namely (i) long-term carbon sequestration from the atmosphere by the stable carbon present in the biochar, (ii) generation of renewable energy (e.g., using the by-products such as bio-oil and syngas) during the production of biochar, and reuse of biomass wastes<sup>168</sup>. Burning of agricultural residues is a common practice in several Asian countries which causes severe air pollution episodes and negative effects on human health<sup>172</sup>. However, converting agricultural residues to biochar could be a one of the sustainable solutions for better management of agricultural-based residues and stabilization of global climate through CO<sub>2</sub> sequestration from ambient air as well as to achieve circular economy<sup>173</sup>.

Based on the LCA analysis of biochar production from four different types of biomass/feedstocks, Xia et al.<sup>170</sup> reported that biochar has a negative effect on the life cycle of GHG emissions. Moreover, the order of carbon sequestration capacity of biochars produced from four different feedstock materials was: waste wood biochar > crop straw biochar > livestock manure biochar > sewage sludge biochar. The biochar derived from waste wood shows higher carbon sequestration potential in soil since waste wood is mainly rich in lignocellulose biomass (i.e., C, H and O as the major elements), and hence it contains higher amount of carbon than other feedstocks<sup>170</sup>. The higher carbon content in waste wood biochar accelerates the carbon fixation and conversion processes in soil. The life cycle of biochar produced from four different feedstocks is illustrated in Fig. 9a. The impacts of different stages of biomass processing including feedstock pre-treatments,

biochar synthesis stage and their application stage are shown in Fig. 9b–d, and the carbon sequestration potential among various biochar materials is presented in Fig. 9e. In another study, LCA analysis was carried out on biochar systems to estimate the climate change effects, economic viability and net energy generation for preparation of biochar using three different feedstocks (yard waste, corn stover and switchgrass energy crops)<sup>168</sup>. A considerable reduction (62–66%) of GHG emissions was found for both yard waste (−885 kg CO<sub>2</sub>e/tons dry biomass) and corn stover (−864 kg CO<sub>2</sub>e/tons dry feedstock) biomass. Notably, switchgrass pyrolysis system results in the increase of GHG emissions (+36 kg CO<sub>2</sub>e/tons dry biomass). The net increase of GHG emissions from the switchgrass pyrolysis system could be due to the contribution from different sources including land-use for its production, applied fertilizers and cultivation-based GHG emissions<sup>168</sup>. The overall economic feasibility of the biochar systems largely depends on the feedstocks production cost, pyrolysis cost and the value of carbon offsets<sup>168</sup>. Feedstocks that require for waste management (e.g., yard wastes) have the maximum potential for economic viability (i.e., \$69/tons dry biomass). Among the various steps, feedstock transportation distance and storage of feedstock materials to a centralized facility are usually expensive. Thus, these steps represent the major obstacles that affect the economic viability for a large-scale production of biochar<sup>171,174</sup>. Using LCA, Gievers et al. assessed the potential negative environmental impacts of sewage sludge-derived biochar to use as a soil carbon sequestrator<sup>175</sup>. The LCA was evaluated under four different scenarios (i.e., use of biochar (1) in agriculture, (2) in horticulture, (3) cascade use in biogas plant and agriculture, and (4) co-incineration of biochar in lignite-fired power plants). Among the four different scenarios evaluated, the best scenario was found to be the application of biochar in horticulture activities with net emissions (GWP) of 2 g CO<sub>2</sub> eq./kg sewage sludge. Compared to the conventional method of biochar treatment using incineration process, the use of biochar in horticulture could reduce 78% of CO<sub>2</sub> eq. emissions. Additionally, no negative ecological impacts including ecotoxicity or eutrophication was found<sup>175</sup>. The findings of this study shows that use of sewage sludge derived





**Fig. 9 | Sustainability of biochar-based carbon sequestration in soil.** Life cycle of biochar produced from four different feedstocks (a), effect of GHG emissions at biochar feedstock pre-treatment stage (b). Effect of GHG emissions at biochar production stage (c). Effect of GHG emissions at biochar application stage (d), and

carbon sequestration capacity of different biochars (e). CSB crop straw-based biochar, WWB waste wood-based biochar, LMB livestock manure-based biochar, SSB sewage sludge-based biochar. Reproduced with permission from ref. 170. Copyright (2024) Elsevier.

biochar as a soil carbon sequester could be an environmentally benign option. In summary, LCA-based studies reveal that biochar amendment to soil is a sustainable strategy to enhance carbon sequestration in soil and to achieve climate change mitigation. Biochar synthesized from waste biomass (e.g., agricultural/crop residues) is found to be environmentally friendly and economically profitable<sup>176</sup>.

### Conclusions and future perspectives

Biochar, a sustainable solid material derived from biomass pyrolysis, is receiving increased attention for sequestration of atmospheric CO<sub>2</sub> in soil and to achieve carbon neutrality. This review comprehensively examined the recent developments on the use of biomass-derived biochar as a

sustainable material for carbon sequestration in soil. The key conclusions are highlighted below.

- Biomass/feedstocks (e.g., agricultural-based residues) having more lignocellulosic contents are beneficial for the synthesis of biochar with a high proportion of carbon that in turn can enhance carbon sequestration in soil.
- Biochar produced at high temperatures (e.g., > 500 °C) through pyrolysis of biomass feedstocks usually contains a high quantity of aromatic hydrocarbons which make the biochar as a stable and recalcitrant material. Biochar with enriched carbon shows high sorption capacity to CO<sub>2</sub>, and it is also highly resistant to microbial as well as physico-chemical degradations.



- Several studies have reported either a decrease or an increase in the SOC mineralization and in CO<sub>2</sub> release rates after biochar amendment to soil. The difference in findings among these studies could be due to the difference in biochar qualities (pH, aromaticity, etc.) and biochar application rates (low vs high dose), soil characteristics (pH, SOC contents, etc.), soil textures (i.e., variations in clay contents) and incubation periods (short term vs long term).
- Most studies reported that biochar amendment to soil enhances the carbon sequestration potential by decreasing soil mineralization and thus CO<sub>2</sub> release rates. Hence, soil can act as a sink for the sequestration and storage of atmospheric carbon.
- Biochar incorporation into soil changes soil microbial characteristics including a shift of bacterial and fungal communities as well as enzymatic activities (specifically enzymes responsible for C, N and P cycling).
- The key mechanisms that facilitate the acceleration of carbon sequestration in soil after biochar treatment include the decrease of SOC mineralization by sorption of SOC onto biochar as well as that of the abundance and diversity of carbon-metabolizing soil microbial communities.
- LCA-based studies revealed that biochar (specifically produced from waste biomass) production and its addition to soil are environmentally friendly and economically viable.

### Future perspectives

The following knowledge gaps identified by the critical analysis of literature findings should be considered in future studies to make further advancements in the use of biochar-amended soil for carbon sequestration.

- At present, most of the experiments relating to carbon sequestration in soil are carried out in the lab-scale pot experiments under controlled conditions, and limited field-scale work has been performed in real environmental settings. Therefore, in-depth, well-designed field-scale investigations are required to better understand the potential of biochar for increasing the carbon sequestration rate in the complex soil system. The field-scale experimental data should be used in conjunction with modeling-based investigations for validation of biogeochemical models and accurate prediction of biochar performance in varying soil/environmental systems as well as for realistic estimation of soil carbon stock (e.g., SOC).
- In recent years, modified biochar/engineered biochar materials are widely used for environmental pollution remediation. Thus, it is expected that modified biochar materials would show better carbon sequestration potential compared to pristine biochar. Machine learning-based approaches can be used for preparation of biochar with unique properties to be suitable for a specific soil type/climatic condition to achieve greater carbon sequestration rate. However, limited information is available on the carbon sequestration potential of engineered biochar. Thus, future studies should focus on the development of novel engineered biochar, followed by the evaluation of its carbon sequestration potential by conducting both lab-scale and field-scale experiments.
- Understanding of biochar-soil-microbial interactions is crucial since this will elucidate the biogeochemical cycling (e.g., C cycling) involved in soil systems and its impacts on the GHG (CO<sub>2</sub>) emissions. Moreover, biochar-soil interactions could impact the stability, integrity, and carbon sequestration capacity of biochar during its long-term operation in the field. However, these interactions have not been explored yet in sufficient depth. Furthermore, the potential underlying mechanisms for biochar aging in long-term period and its impact on carbon sequestration rate remain poorly understood. Consequently, efforts are needed to provide deep insights into biochar-soil-microbial interactions in the context of understanding carbon sequestration potential of biochar-amended soil.
- Carbon sequestration in biochar treated soil systems is mainly driven by various microbial processes. Genetic engineering approach can be applied to engineer the soil microbial system and change metabolic pathways to enrich more CO<sub>2</sub> fixing microbial communities, e.g., autotrophs that can use CO<sub>2</sub> as the carbon source or engineering heterotrophic bacteria that capable of using CO<sub>2</sub> as the sole carbon source for their growth.
- The use of biochar for carbon sequestration in soil provides additional benefits from the agricultural perspectives. To use biochar for better management of degraded/infertile soils and to enhance crop growth and productivity, long-term field trials are necessary to monitor the soil health/fertility and to understand the changes of soil microbial processes influenced by biochar characteristics and its application rate to soil. The molecular level investigations are particularly needed to better understand the changes of carbon cycle-related functional genes and their correlation with CO<sub>2</sub> emissions. Development of biochar-microbe co-engineering strategies should be explored to stabilize labile carbon fractions and reduce CO<sub>2</sub> emission from soil.
- Biochar-based soil carbon sequestration is considered as a negative emission process. However, in-depth LCA -based analysis is required prior to exploring large-scale applications of biochar in different soil systems (natural, fertile, infertile, saline soils, etc.). Moreover, LCA would provide insights into whether or not the large-scale biochar preparation and its field-scale applications would be environmentally benign and economically feasible. However, limited work has been done on LCA-based studies in the context of biochar-based carbon sequestration in soil. Standardizing carbon accounting methodologies would help to resolve discrepancies among various LCA-based studies that were conducted to evaluate the sustainability of biochar-based carbon sequestration in soil.
- More conceptual modeling-based studies are required by integrating different biogeochemical interactions as well as simulating different environmental conditions at different time scales (incubations periods). The outcome of such studies would help to better understand the long-term performance of biochar for carbon sequestration in soil. In recent years, machine learning (ML)-based models are increasingly applied to resolve various environmental problems. Thus, a novel ML-based model can be developed for accurate prediction of changes of CO<sub>2</sub> emission from soil after biochar amendment.

### Data availability

No datasets were generated or analyzed during the current study.

### Code availability

Not applicable since there is no computer programming/coding was used for this work.

### List of abbreviations

AMF	Arbuscular mycorrhizae fungi
ANN	Artificial neural network
BDOC	Biochar-derived dissolved organic carbon
BET	Brunauer–Emmett–Teller
C/N ratio	Carbon-to-nitrogen ratio
CCS	Carbon capture and storage
CEC	Cation exchange capacity
CO <sub>2</sub>	Carbon dioxide
CSB	Crop straw-based biochar
EEC	Electron exchange capacity
EPS	Extracellular polymeric substances
ESCs	Electron storage capacities
Fe-OC	Fe-bound organic carbon
FT-IR	Fourier transform infrared spectroscopy
G <sup>−</sup>	Gram-negative
G <sup>+</sup>	Gram-positive
Gg	Giga-gram
GHG	Greenhouse gases
GWMP	Global warming mitigation potential

GWP	Global warming potential
H/C ratio	Hydrogen-to-carbon ratio
H <sub>3</sub> PO <sub>4</sub>	Phosphoric acid
HTC	Hydrothermal carbonization
IBI	International Biochar Initiative
IPCC	Intergovernmental Panel on Climate Change
LCA	Life cycle assessment
LMB	Livestock manure-based biochar
M tons/ hectare	Metric tons/hectare
MB	Maize biochar
MBC	Microbial biomass carbon
MBN	Microbial biomass nitrogen
ML	Machine learning
MRT	Mean residence time
NEXAFS	Near edge X-ray absorption fine structure
NMR	Nuclear magnetic resonance
nZVI	Nanoscale zero-valent iron
O/C ratio	Oxygen-to-carbon ratio
Pg	Peta-gram
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
RB	Rice biochar
RF	Random forest
SDGs	Sustainable development goals
SEM	Scanning electron microscopy
SOC	Soil organic carbon
SSB	Sewage sludge-based biochar
STXM	Scanning transmission X-ray microscopy
SVM	Support vector machine
TEM	Transmission electron microscope
TN	Total nitrogen
TRL	Technology readiness level
w/w	Weight/Weight
WOS	Web of Science
WWB	Waste wood-based biochar
XANES	X-ray absorption near edge structure spectroscopy
XPS	X-ray photoelectron spectroscopy
XRD	X-ray diffraction

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## Author contributions

B.K.B.'s contributions include Conceptualization, Investigation, Methodology, Writing—original draft, and Writing—review & editing. R.B.'s contributions include Conceptualization, Funding acquisition, Supervision, Writing—review & editing.

## Competing interests

The authors declare no competing interests.

## Additional information

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**Correspondence** and requests for materials should be addressed to Rajasekhar Balasubramanian.

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