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Short Communication

Enhancement of Corn Seed Germination in Calcareous Soil: Combined Effect of Biochar and Calcium Carbonate

Noor Atiqah Zulkifli[1,2], Shahidah Md Nor[3]*

[1]Faculty of Agriculture, Universiti Putra Malaysia, Serdang, 43000, Malaysia, nooratiqahzulkifli@yahoo.com

[2]School of Distance Education, Universiti Sains Malaysia, Gelugor, 11800, Penang, Malaysia.

[3]Department of Technology and Natural Resources, UTHM Kampus Pagoh, Hab Pendidikan Tinggi Pagoh, KM 1, Jalan Panchor, 84600 Panchor, Johor. shahidahmn@uthm.edu.my; shahida.mdnor@gmail.com

Corresponding author: Shahidah Md Nor, Department of Technology and Natural Resources, UTHM Kampus Pagoh, Hab Pendidikan Tinggi Pagoh, KM 1, Jalan Panchor, 84600 Panchor, Johor; shahidahmn@uthm.edu.my

Abstract: Calcareous soil is reported to reduce seed germination performance due to its soil properties, like low moisture content, weak soil structure, and nutrient leaching. Biochar could enhance soil properties by modifying the physiochemical properties of the soil, which aids in accelerating seed germination and plant growth. Due to biochar's alkaline properties, it is rarely recommended for use on calcareous soil, which is naturally alkaline. Nevertheless, the water-holding capacity of biochar still has the potential to improve seed germination and thus could be suitable for working as a soil nourisher in calcareous soil. Till now, the effect of biochar on calcareous soil is lacking information. Thus, the current study aimed to evaluate the combined effects of biochar and calcareous soil on corn seed germination performance. Calcium carbonate of different rates (0, 10, 25% w/w) was mixed in a mixed soil with an initial pH of 6.8 to form the calcareous soil. Then biochar of different rates (0, 5, 10% w/w) was applied to the calcareous soil. After a week, the corn seed was sown, and seed germination performance was evaluated. The experiment was laid out in a completely randomized design (CRD). This study showed that the combination of calcareous soil at 0% and biochar at 10% gave the best seed germination, which supports the idea that biochar nourishes typical soil, as reported in many studies. However, this study also showed the potential of biochar in appropriate amounts to amend alkaline soil. The calcareous soil with the highest calcium carbonate rate (25%) has been shown to reduce seed germination significantly. However, the combined effects of calcium carbonate at 25% and biochar at 5% have significantly increased seed germination. However, the combined effect of calcareous soil at 25% and biochar at 10% portrayed the weakest seed germination. This suggests that 5% of BC is the appropriate amount to amend calcareous soil.

Keywords: biochar; calcium carbonate; calcareous soil; seed germination; corn

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1. Introduction

Biochar is the product of pyrolysis of agricultural waste disposals like rice husk, empty fruit brunch, hardwood, straw, and many others. Its carbon-rich properties allow it to be widely used as a soil nourisher to improve soil properties by increasing water-holding capacity, thereby reducing nutrient leaching and improving nitrogen use efficiency (Hossain *et al*., 2020). In addition, biochar is highly stable and can remain in various soil types for decades to thousands of years (Zhu *et al*., 2023). Despite plenty of research on biochar's effects on soil fertility and plant growth, there is limited information on alkaline soil, like calcareous soil (Bu *et al*., 2020). Calcareous soil can be challenging for agricultural use due to its poor soil properties like low water holding capacity, low nutrient availability, low organic matter and highly alkaline (Bolan *et al*, 2023).

A previous study reported the challenging condition of calcareous soil for optimal growth and germination performance of corn seeds (Mandic *et al.*, 2023; Shahid *et al*., 2023). Alkaline soils cause root poisoning by ions and osmotic imbalance, reducing seed germination performance (Mandic *et al*., 2023). Biochar's alkaline properties are reported to increase soil pH; hence, it is less recommended to be used on calcareous soil (Bu *et al*., 2020). However, biochar still has the potential to enhance calcareous soil properties by improving its organic matter, water content, nutrient availability, soil porosity, bulk density, and microbial activity, aiding in seed germination and enhancing plant growth (Khan *et al.*, 2024) Therefore, in this study, biochar amendment in calcareous soil is expected to improve corn seed germination by nourishing the alkaline soil properties.

Previous studies reported that using 1 to 2% of biochar in calcareous soil positively impacted the germination rate and seedling growth of *Robinia pseudoacacia* (Bu *et al*., 2020). In earlier years, a study by Abrishamkesh et al. (2015) showed that the amendment of rice husk biochar in alkaline soil led to significant improvement in lentil growth, soil organic carbon, cation exchange capacity (CEC), available potassium and soil bulk density. These two studies noted that the liming effect of biochar led to dynamic soil pH properties and thus affected the seed germination performance. Limited data possess the effect of biochar directly on soil pH. Thus, it is essential to investigate the effect of biochar on soil pH properties, which should be translated into better seed germination. Corn seed germination is optimal at a favourable pH range between 5.5 and 7.5 and is limited by alkaline conditions (Hanifa *et* *al*., 2019). Hence, in this study, corn seed germination is expected to be improved via biochar amendment in calcareous soil.

In this study, corn seed germination performance was evaluated using specific parameters, such as germination percentage (GP), which measures the percentage of total seed germinated, the germination index (GI), which measures the rate of germination and mean germination time (MGT) which counts the time taken for all the seeds to germinate. Additionally, the coefficients of the velocity of germination (CVG) that analyze the rapidity of germination can be taken as seed germination performance. In summary, the current research aimed to evaluate the potential of rice husk biochar amendment in calcareous soil for improving corn seed germination.

2. Materials and Methods

2.1 Soil Preparation

A mixed soil (Clay: silt: sand) with an initial pH of 6.8 was purchased from a local store (Serdang, Malaysia). The soil was sieved (2 mm sieve), and the pH was measured in suspension 1:3 (soil: water) by pH meter. The pH meter was calibrated using buffer pH 4, 7 and 9 before measurement. The mixed soil was then rigorously mixed with $CaCO_3$ powder (Kaolin, Malaysia) at three different rates (0, 10, and 25% w/w were labelled as CC0, CC1, and CC2, respectively) and incubated for a week to form calcareous soil. After a week, the soil pH of all the samples was measured. Then, each calcareous soil sample was treated with rice husk biochar at three different rates (0, 5, and 10 % w/w labelled as BC0, BC1, and BC2, respectively) and incubated further for another week. Then, the soil pH of all the samples was re-determined. Nine samples of soil (Table 1) were transferred to nine seedling trays (9 experimental units) in three replications (3BC X 3CC X 3 rep=27), making a total of 27 trays. Each tray has 32 holes, making a total of 243 holes of sampling units (32 X 27=243 holes). Corn seeds (variety sweetcorn SS 63) were soaked for 12 hours before being sown in the 27 seedling trays of 9 soil samples and observed for 10 days for seed germination performance.

Table 1. Calcium carbonate (CC) and biochar (BC) rates were applied in the mixed soil.

CaCo3 (CC)/ Biochar(BC)	**CC0 (0%w/w)**	**CC1 (10%w/w)**	**CC2 (25%w/w)**
BC0 (0%w/w)	CC0BC0	CC1BC0	CC2BC0
BC1 (5%w/w)	CC0BC1	CC1BC1	CC2BC1
BC2 (10%w/w)	CC0BC2	CC1BC2	CC2BC2

2.2 Statistical Analysis

The experiment was laid out in a completely randomized design (CRD) with two factors, calcium carbonate (CC) and biochar (BC), with three levels (3 rates of both CC and

BC) in three replications. The data were analyzed using two-way analysis of variance (ANOVA) in the Statical Software Package (SAS version 9.2) and GraphPad Prism 8. Means were separated with the least significant difference (LSD) test at a 0.01 and 0.05 probability level.

2.3 Germination Parameters

The number of seeds germinated in each seedling tray (32 holes) was recorded every day for 10 days after sowing. The germination parameters were measured based on the equation of germination percentage (Equation 1), mean germination time (Equation 2), coefficient velocity of germination (Equation 3), and germination index (Equation 4) following the method conducted by Shahriari (2013). The formula is elaborated below:

2.3.1 Equation 1: Germination percentage

$$GP\ (\%) = \frac{\text{Final number of seeds germinated}}{\text{Total number of seeds}} \times 100 \quad (1)$$

Where n is the number of seeds germinated daily, D is the day after sowing, and N is the total number of seeds germinated.

2.3.2 Equation 2: Mean germination time

$$MGT = \frac{{}^{O}\Sigma Dn}{N} \quad (2)$$

2.3.3 Equation 3: Coefficient velocity of germination

$$CVG = \frac{(N1+N2+\cdots.N9)}{(N1T1+\cdots.NxTx)} \times 100 \quad (3)$$

Where N is the number of seeds germinated each day, and T is the number of days corresponding to N.

2.3.4 Equation 4: Germination index

$$GI=(10\times n1)+(9\times n2)\pm\cdots\ldots(1\times n10) \quad (4)$$

Where n is the number of germinated seeds from day one until the ten days, the day is the weight given to the number of germinated seeds on the subsequent days.

3. Results and Discussions

The ANOVA shows that both treatments, CC and BC, have single and interaction effects on soil pH and seed germination performance as rated by germination percentage (GP), mean germination time (MGT), coefficient velocity (CVG) and germination index (GI) at p<0.01 (Table 2).

Table 2. Mean squares from ANOVA of seed germination parameters of sweet corn under treatments of different rates of $CaCO_3$ and biochar.

Sources	df	Soil pH [CC+BC]	GP	MGT	CVG	GI
CC	2	4.20****	2108.00****	2.84****	25.41****	98948.00****
BC	2	0.50****	765.60***	0.77****	0.86**	17128.00****
CC*CC	4	0.083****	551.70***	0.77****	12.94****	9498.00****
error	18	0.0004	60.78	0.03	0.10	300.40
CV		5.30	126.00	0.36	0.66	146.00

**CC: Calcium Carbonate; BC: Biochar; GP: Germination Percentage; MGT: Mean Germination Time; CVG: Coefficient Velocity of Germination; GI: Germination Index; df: Degree of freedom (*, ** significant difference at p<0.05 and 0.01)

3.1. The Combined effect of $CaCO_3$ and Biochar on Soil pH

In this study, the soil pH significantly increased with the sole application of CC and BC. The soil pH altered by the incubation of CC and BC is presented in Table 3. Application of CC1 increased the soil pH to about 6% (7.2±0.02) while CC2 to about 22% (8.3±0.02). CC1 raised the soil pH but is still within for agricultural use (pH 7.2), while CC2 makes the soil too alkaline (8.3) and might affect plant performance. The corn seed, for instance, is only favourable to be germinated at a pH between 5.6 and 7.5 (Hanifa *et al*., 2019). Applying BC1 and BC2 raised the soil pH to about 5.8% (7.2±0.02) and 7.3% (7.3±0.02), respectively, adequately rising within a suitable pH range for agricultural use.

Table 3. The soil pH after application of $CaCO_3$ and rice husk biochar.

Treatment	Initial pH	[CC]added (%w/w)	pH after [CC]	[BC] added (%w/w)	pH after [CC+BC]
CC0BC0	6.8	0	6.8±0.02	0	6.8±0.02
CC0BC1	6.8	0	6.8±0.02	5%	7.2±0.02
CC0BC2	6.8	0	6.8±0.02	10%	7.3±0.02
CC1BC0	6.8	10%	7.2±0.02	0	7.2±0.02
CC1BC1	6.8	10%	7.2±0.02	5%	7.3±0.02
CC1BC2	6.8	10%	7.2±0.02	10%	7.4±0.02
CC2BC0	6.8	25%	8.3±0.02	0	8.3±0.02
CC2BC1	6.8	25%	8.3±0.02	5%	8.3±0.02
CC2BC2	6.8	25%	8.3±0.02	10%	8.3±0.02

The combination of CC and BC at a specific rate increased the soil pH even more than their sole application. For instance, CC1BC1 was raised to (7.3±0.02) and CC1BC2 at (7.4±0.02). Nevertheless, this pH range was still within the range for agricultural use. However, the combination of biochar and CaCO3 observed in CC2BC1 (pH 8.3) and CC2BC2 (pH 8.3) did not raise the soil pH. This data is in sync with the previous study by Liu and Zhang (2012), which reported that biochar had not increased the soil pH of five alkaline soils with an initial pH of 7.9. This effect could be assumed to be a biochar liming effect that was prevented in highly alkaline soil due to buffering capacity, thus eliminating the concern that biochar application is detrimental to alkaline soil pH.

3.2 The Single Effect of $CaCO_3$ on Corn Seed Germination Performance.

The current study found that only CC2 (CC2BC0: 25% $CaCO_3$, 0% biochar) affected seed germinated percent (GP) in the absence of BC (BC0 treatments) (Figure 1). The mean germination time (MGT) was neither affected by CC content, as all the CC treatments, including control (CC0), took around 7±1 days to germinate fully. The mean germination index was reported to be lowest by CC2. The coefficient velocity of germination (CVG), which measured the speed or rapidity of germination, also showed no significant difference between all the treatments, ranged 13.8 to 14.3. This data proved that the $CaCO_3$ amendment has a limitation of up to 5% to improve seed germination. Supplying $CaCO_3$ beyond that limit can cause a reduction in GP, GI and CVG. The reduction could be attributable to the presence of high Ca^{2+} cation. The high Ca^{2+} salt concentration can reduce seed water imbibition needed for germination (Kolodziejek & Patykowski, 2015).

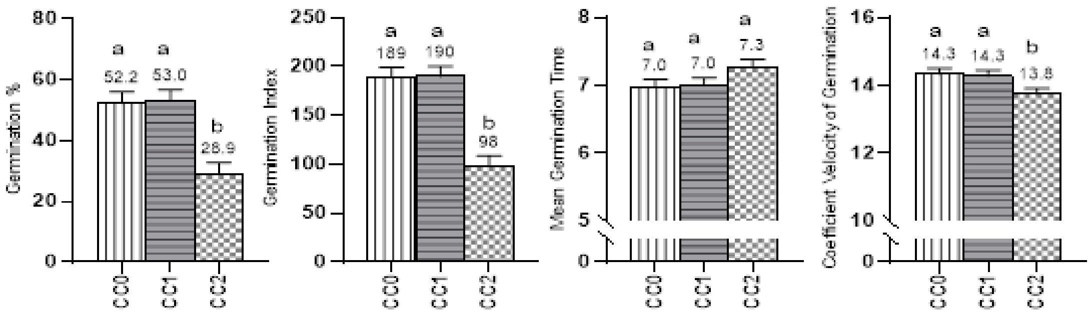

Figure 1. Seed germination as affected by Calcareous soil in BC0 treatments (absence of Biochar) within treatments, means with the same letter are not significantly different at $p>0.05$ using LSD. CC0: [0 Biochar]; CC1: [5% w/w Calcium carbonate]; CC2: [5% w/w Calcium carbonate].

3.3 The Single Effect of Rice Husk Biochar on Corn Seed Germination Performance

Rice husk biochar application without CC significantly improved seed germination (Figure 2). The germination percentage was reported at 52% in control (CC0BC0), raised to 61% in CC0BC1 (5% biochar) and high up to 80% in CC0BC2 (10% Biochar). A similar pattern was observed in GI, increasing from 189±10 in control (CC0BC0) to 268±10 in

CC0BC1 and 342±10 in CC0BC2. The MGT observed no significant difference between control (CC0BC0) and CC0BC1 at 7±0.05 days but only sped up to 5.8±0.05 days in CC0BC2. The CVG also observed a similar pattern as the control (CC0BC0), and CC0BC1 showed no significant difference at 14±1 but significantly increased to 17±1 in CC0BC2. In short, the current study found that 10% of BC (BC2) enhanced seed germination performance better than 5% of BC (BC1).

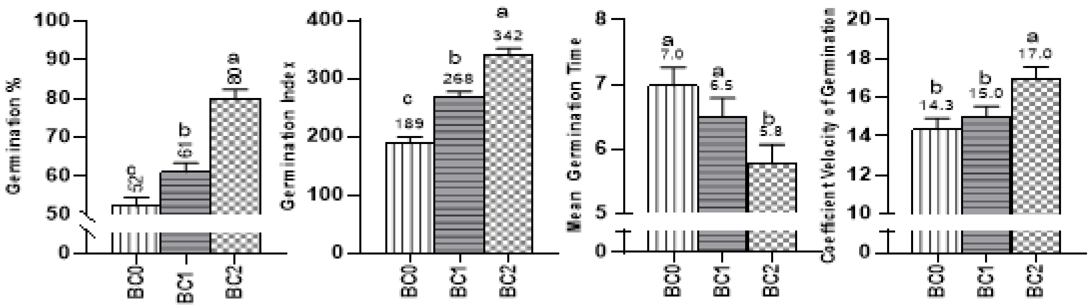

Figure 2. Biochar amendment in calcareous soil. Within treatments, means with the same letter are not significantly different at p>0.05 using LSD. BC0: [0 Biochar]; BC1: [10% w/w Biochar]; BC2: [10% w/w Biochar].

Based on our current research, we can conclude that biochar amendment alone can enhance corn seed germination. The effectiveness of seed germination performance increased with the high rate of single biochar amendment. Anyanwu et al. (2018) also documented that higher biochar application rates (1–10%) led to faster seed germination in *Oryza sativa* and *Solanum lycopersicum*. Bu *et al*. (2020) concluded that applying biochar at a higher rate improves seed germination due to its high surface area porosity and carbon sequestration that enhances the soil's water-holding capacity, aiding the seed to attain more vigour.

3.4 The Combined Effects of $CaCO_3$ and Biochar on Corn Seed Germination Performance

The result showed that the combined effect of CC at low concentrations between 0–10% and BC at a concentration of 5–10% increased the GP, GI and CVG (Figure 3). Nevertheless, using CC at 25% in combination with all biochar concentrations of 5, 10 and 15% reduced the germination performance rated by GP, GI and CVG. A combination of 25% $CaCO_3$ with 10% biochar produced the lowest GP, GI and CVG. Regarding MGT, a high concentration of CC, especially at 25% (CC2) in combination with 10% biochar (BC2) concentrations, has lengthened the germination time. In summary, these results support the idea that applying high $CaCO_3$ in combination with high biochar concentration will not improve the corn seed germination performance. This data reveals a contrasting trend compared to the findings of Liu and Zhang (2012), where highly alkaline soil demonstrated improved seed germination with the application of high biochar concentrations.

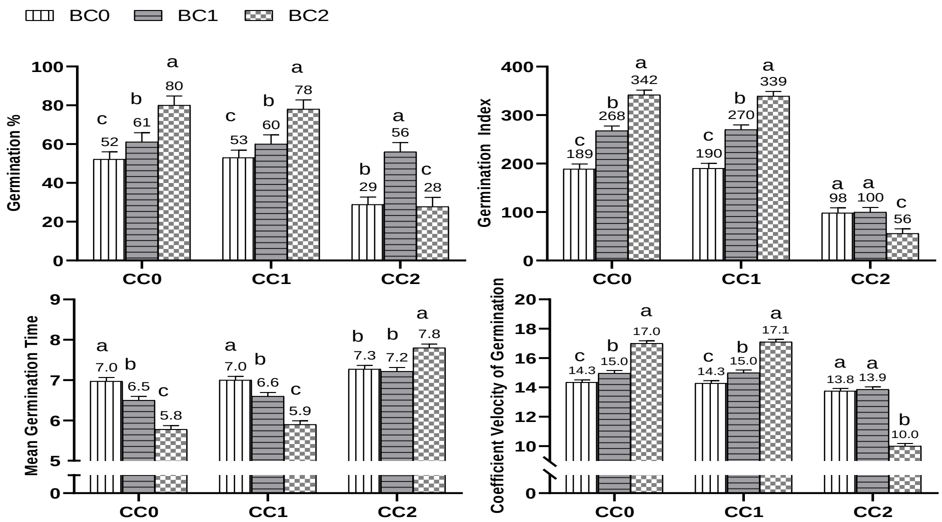

Figure 3. Biochar in combination with $CaCO_3$ amendment. Within treatments, means with the same letter are not significantly different at $p>0.05$ using LSD. CC0: [0 $CaCO_3$]; CC1: [10% w/w $CaCO_3$]; CC2:[25% w/w $CaCO_3$]; BC0: [0 Biochar]; BC1: [5% w/w Biochar]; CC2:[10% w/w Biochar].

4. Conclusions

This study concluded that high biochar amendment is unsuitable for improving alkaline calcareous soil with 25% $CaCO_3$. Only a lower biochar (BC1) rate in combination with 0–10% $CaCO_3$ (CC0 and CC1) can effectively improve the seed germination performance. Despite the concern that biochar alkalinity might be unsuitable for alkaline soil, this study found that rice husk biochar application did not raise the soil pH in high calcareous soil at 25% $CaCO_3$ content. Supplying the rice husk biochar on low calcareous soil with 10% $CaCO_3$ content will raise the pH suitable for agricultural use. Since the incubation period is too short, biochar amendment might not achieve its full potential and needs further validation in future experiments.

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Conflicts of Interest: The authors declare no conflict of interest.

References

Abrishamkesh, S., Gorji, M., Asadi, H., *et al*. (2015). Effects of rice husk biochar application on the properties of alkaline soil and lentil growth. *Plant, Soil and Environment*, *62*(11), 475–482. https://doi.org/10.17221/117/2015-PSE

Anyanwu, I. N., Alo, M. N., Onyekwere, A. M., *et al*. (2018). Influence of biochar aged in acidic soil on ecosystem engineers and two tropical agricultural plants. *Ecotoxicology and Environmental Safety*, *153*, 116–126. https://doi.org/10.1016/j.ecoenv.2018.02.005

Bolan, P., Srivastava, P., Rao, C. S., *et al*. (2023). Distribution, characteristics and management of calcareous soils. *Advances in Agronomy*, *182*, 81–130. https://doi.org/10.1016/bs.agron.2023.06.002

Bu, X. L., Xue, J. H., Wu, Y. B. *et al*. (2020). Effect of biochar on seed germination and seedling growth of Robinia pseudoacacia L. in karst calcareous soils. *Communications in Soil Science and Plant Analysis*, *51*(3), 352–363. https://doi.org/10.1080/00103624.2019.1709484

Hanifa, H., Utami, S. N. H., Wulandari, C. *et al*. (2019). Soil chemical properties and corn productivity as affected by application of different types of fertilizer and planting method in acid sulfate soil. *IOP Conference Series: Earth and Environmental Science*, *393*, 012001. https://doi.org/10.1088/1755-1315/393/1/01200

Hossain, M. Z., Bahar, M. M., Sarkar, B., *et al*. (2020). Biochar and its importance on nutrient dynamics in soil and plant. *Biochar*, *2*, 379–420. https://doi.org/10.1007/s42773-020-00065-z

Khan, S., Irshad, S., Mehmoo, K., *et al*. (2023). Biochar production and characteristics, its impacts on soil health, crop production, and yield enhancement: A review. Plants, 13(2), 166. https://doi.org/10.3390/plants13020166

Kolodziejek, J. & Patykowski, J. (2015). Effect of temperture, Light and Calcium Carbonate on Seed Germination and Radicle Growth of the Polycarpic Perennial Galium Cracoviense (Rubiaceae). A narrow Endemic Species from Sourthern Polland. *Acta Biologica Cracoviensia Series Botanica*, *57*(1), 70–81. https://doi.org/10.1515/ABCSB-2015-0006

Liu, X. H. & Zhang, X. C. (2012). Effect of biochar on pH of alkaline soils in the loess plateau: Results from incubation experiment. *International Journal of Agriculture and Biology*, 14(5), 745–750.

Mandic, V., Krnjaja, V., Simic, A., *et al*. (2023). Effect of pH on germination and seedling growth of maize. *Biotechnology in Animal Husbandry*, *39*(2), 195–203, http://doi.org/10.2298/BAH2302195M

Shahid, Z., Ali, M., Shahzad, K., *et al*. (2023). Enhancing maize productivity by mitigating alkaline soil challenges through acidified biochar and wastewater irrigation. *Scientific Reports*, *13*, 20800. https://doi.org/10.1038/s41598-023-48163-9

Shahriari, A. (2013). *Sweet corn (Zea Mays L.) seed germination and production under water and nitrogen deficit under glasshouse conditions*. [Doctoral Dissertation, Universiti Putra Malaysia]. http://psasir.upm.edu.my/id/eprint/49659/1/FP%202013%2069RR.pdf

Zhu, H., An, Q., Mohd Nasir, A. S., *et al*. (2023). Emerging applications of biochar: A review on techno-environmental-economic aspects. *Bioresource Technology*, *388*, 129745.