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Use of Biochar for the production of Biofertilizers

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Abbreviation list

°C: Degrees Celsius

cm: Centimeters

CM: Mean square

CO: Carbon Monoxide

Ddl: Degrees of Freedom

F: F-value in ANOVA (*Fisher* test)

FAO: Food and Agriculture Organization

ITGC: Technical Institute of Great Crops

mm: Millimeters

NH: Nitrogen Hydride

P: Probability value in statistics

PGPR: Plant Growth-Promoting Rhizobacteria

PSB: Phosphate-Solubilizing Bacteria

SCE: Sum on Squares

Sig: Significance (Statistical)

SO: Sulfur Oxide

SPSS: Statistical Package for the Social Sciences (Software)

Figure list

Figure 1: Different biofertilizer classifications	4
Figure 2: The the ecosystem colonized by microorganisms	6
Figure 3: Complete process of formulation of biofertilizers	9
Figure 4: Types of biofertilizers formulations.....	10
Figure 5: Main modes of biofertilizers application nowadays.	13
Figure 6: Example site profiles for Oxisol (b) and Terra Preta (a).	14
Figure 7: Images of sawdust biomass (A) and biochar biochar (B)	15
Figure 8: The low-temperature pyrolysis bioenergy concept utilizing sequestration of biochar.	16
Figure 9: Deseases (A), Leaf morphology (B), Flowering (C) Spacing and varieties (D) and deseases (E) in Barley.	20
Figure 10: Plant morphology (A), flowering (B), cosse and seeds morphology (C) and plant life cycle (D)..	23
Figure 11 : Steps of Biochar production.	29
Figure 12 : Steps of Agar-based biochar macerate production.	30
Figure 13 : Steps of determination of germination and growth parameters.....	31
Figure 14 : Based-Biochar produced products.....	32
Figure 15 : Steps of preparation of liquid biofertilizer.....	33
Figure 16: Steps of crop installation.....	35
Figure 17: Variation of Biochar Yields According to Pyrolysis Temperatures and Residence Times.	36
Figure 18: Variation of barley germination percentages on gelose biochar macerate after 20h (A), 44h (B), and 68h (C).....	38
Figure 19 : Variation in root length of barley growing on gel-seeded biochar macerate, on the third (A) and eighth (B) days of germination.....	40
Figure 20 : Variation in the number of roots of barley growing on gel-seeded biochar macerate, on the third (A) and eighth (B) days of germination.	42
Figure 21: Variation in coleoptile and aboveground parts length of barley growing on gel- seeded biochar macerate on the third and eighth days of germination.	43
Figure 22: Variation in average fresh (A) and dry (B) weights of barley seedlings after 8 days of germination.	45

Figure 23: Variation in stem hight of pea plants under the influence of two types of biofertilizers (solid and liquid) and three treatment concentrations (0.5%, 1%, 2%), along with a control.....	46
Figure 24: Variation in number of leaves of pea plants under the influence of two types of biofertilizers (solid and liquid) and three treatment concentrations (0.5%, 1%, 2%).	48
Figure 25: Variation in number of branches on pea plants under the influence of two types of biofertilizers (solid and liquid) and three treatment concentrations (0.5%, 1%, 2%).	49

Table liste

Table 1 : Laboratory materials used during the experiments.	26
Table 2 : General information on the studied varieties.	26
Table 3 : Selected carbonization temperatures and different residence times of biochar production.....	28
Table 4 : Préparation of the set concentrations of soil-biochar	34
Table 5 : Analysis of variances results for barley germination in gelose biochar macerate. ..	37
Table 6 : Analysis of variances of root length in barley grown on biochar macerate.....	39
Table 7 : Table: Analysis of variance of root number in barley growing on biochar macerate.	41
Table 8 : Analysis of variance of coleoptile and aboveground parts length in barley seedlings growing on biochar macerate.	42
Table 9 : Analysis of variance for average fresh and dry weights of barley seedlings growing on gelatinized biochar slurry.	44
Table 10 : Variance nalysis of stem height of pea plants	46
Table 11 : Variance analysis of number of leaves per plant.	47
Table 12 : Variance analysis of number Branches per plant.	48

Contents

Acknowledgement	i
Dedications	ii
Abbreviation list	iii
Figure list	iv
Table liste	vi
Contnents	vii
Introduction	1
Biliographical section	3
1. Bio-Fertilizers	3
1.1. General Overview	3
1.2. History of Biofertilizers	3
1.3. Classification of Biofertilizers	4
1.3.1. Classification based on the type of microorganism	4
1.3.2. Classificationbased on the ecosystem colonized.....	5
1.3.3. Classification based on function	6
1.4. Production and formulation of Bio-fertilizers	9
1.5. Types of formulations.....	9
1.5.1. Powder formulations	9
1.5.2. Granular formulations	9
1.5.3. Liquid formulations.....	10
1.5.1. Cell Immobilization.....	10
1.5.2. Fluid bed-dried formulation	11
1.5.3. Mycorrhizal Formulations.....	11
1.6. Stickersand dditives.....	11
1.7. Packaging.....	11
1.8. Application of biofertilizers.....	11
1.8.1. Field application of biofertilizers	12
1.8.2. Seed application of biofertilizers.....	12
2. Biochar as Bio-fertilizers	13
2.1. History of biochar	13

2.2.	The Biochar	14
2.3.	Characteristics of biochar	14
2.4.	Biochar production	16
2.5.	Benefits of biochar.....	17
2.5.1.	Biochar and soil.....	17
2.5.2.	Water-retention.....	17
2.5.3.	Biochar and climate change	18
3.	Plantes'technical cards	19
3.1.	Barley technical card	19
3.2.	Pea technical card	21
	Experimanetal section	26
	Chapter 1: Materials and Methods	26
1.	Work objectives	26
2.	Workplace	26
3.	Materials	26
3.1.	Laboratory material	26
3.2.	Biological material	26
4.	Methods	27
4.1	Biochar production	27
4.1.1	Biomass collection (sawdust).....	27
4.1.2.	Cleaning Phase	27
4.1.3.	Grinding Phase	28
4.1.4.	Pyrolysis.....	28
4.2.	Assessment of the phytotoxicity of biochar	29
4.3.	Formulation of Biofertilizers	32
4.3.1.	Solid Biofertilizer	33
4.3.2.	Liquid Biofertilizer.....	33
4.4.	Evaluation of the produced Biofertilizers.....	33
4.4.1.	Seeds preparation	33
4.4.2.	Installation of crops	34
4.4.3.	Parameters measured.....	34
5.	Statistical analysis	35
	Chapter 2 Results and discussions	36

1. Results	36
1.1. Biochar yield :	36
1.2. Results of Biochar Phytotoxicity Evaluation.....	37
1.2.1. Barley germination on gelose biochar macerate	37
1.2.2. Growth parameters of barley growing on gelose biochar macerate	39
1.3. Evaluation of the effectiveness of biochar-based biofertilizers.....	46
1.4.1 Evaluation of the effectiveness of biochar-based biofertilizers on Pea Plant.....	46
2. Discussion	50
Conclusion	53
References	56
Abstract	

Introduction

Introduction

In recent years, agriculture has gradually struggled to meet the challenge of producing more food to ensure global food security amidst a growing population(Katiyar *et al.*,2017). This leads to a growing demand for food and agricultural products. There is a reliance on chemical fertilizers, known for their ability to boost productivity to meet this demand. Pesticides also play a significant role in increasing food production and yield(Santos *et al.*, 2012).

Chemical fertilizers are produced and manufactured in industrial setting. These primarily comprise of nitrogen (N), phosphorus (P), and potassium (K) in the defined concentrations. Although, on one hand, the constant use leads to high productivity and the increased yield; however, on the other hand, their use also leads severe threat to environmental problems including degradation in quality of soil, ground water and water at the surface, air pollution, diminished biodiversity, and a crushed ecosystem. It also causes mistreatment and misuse of restricted resources of phosphorus, groundwater pollution with nitrates, and damage to the aquatic ecosystems (Sharma and Sharma, 2021). In 2021, world agriculture used 109 million tons of nitrogen, 46 million tons of phosphorus, and 40 million tons of potassium fertilizers (FAO., 2023).

However, in recent years, there has been a growing recognition of concepts such as "soil health," which emphasizes the importance of viewing soil as an ecosystem that requires balance to sustain plant yield. Additionally, the One Health Concept, which integrates the health of people, animals, and ecosystems, aligns with current trends in soil preservation (FAO., 2023). Based on this philosophy, a decrease in the use of chemical pesticides and fertilizers is mandatory at a universal scale due to their negative effects on environmental pollution, as we mentioned earlier, and also human health-associated risks(Ibáñez *et al.*, 2023a).

Indeed, climate change presents ecological challenges to crop stability, including sudden temperature fluctuations, prolonged periods of rainfall and drought, and the emergence or geographical spread of new pests (Ibáñez *et al.*, 2023b). Soil degradation, primarily through erosion and nutrient loss, poses a significant threat to farmers, affecting both agricultural business and land productivity, compounded by the low regeneration of professional farmers. Thus, there is an urgent need for innovative agricultural technologies utilizing local resources and organic materials (Katiyar *et al.*,2017). In response, biofertilizers are emerging as a promising alternative for sustainable crop production in the 21st century. They have been proposed as enhancers of plant resilience and the rhizosphere against both biotic and abiotic stresses (Ibáñez *et al.*, 2023b).

Agricultural activities generate various types of waste, such as rice husk, straw, and chaff, this waste is notable for its ability to restore soil quality and retain nutrients. It can be processed into biochar, which is a valuable resource for enhancing soil health and fertility. It enhances soil quality by increasing organic matter content, stabilizing and balance pH, and boosting crop yield. It improves soil water-holding capacity, and microbial activity, significantly affecting soil characteristics by enhancing water and nutrient retention, reducing evaporation, and suppressing soil-borne pests and diseases (Katiyar *et al.*,2017). It contains high levels of carbon (C) with small amounts of N, P, K, Ca, Mg, Na, Al, and Fe (Ammal *et al.*, 2020).

In light of this invention, and with the aim of participating in the improvement of soil fertility and the improvement of plant production, our study focused on addressing and researching the following questions:

- Firstly, how is biochar produced?
- Secondly, which protocol should be selected for the production of our biofertilizers and how should it be implemented?
- Finally, how do these fertilizers influence plant growth and soil fertility?

These questions guided our research and formed the basis of our investigation into the potential benefits and applications of biochar-based biofertilizers.

In our memorandum project, our objective was to contribute to the improvement of plant production through:

- ✓ The production of Biochar from sawdust, using various pyrolysis temperatures and durations;
- ✓ Assessing the phytotoxicity of the biochar produced in order to select the best;
- ✓ Formulation of two types of biofertiliser based on biochar (liquid and dry).
- ✓ and finally, evaluation of these biofertilisers on two strategic crops (barley and peas).

Bibliographical section

1. Bio-Fertilizers

1.1. General Overview

The term “Biofertilizer”, also named as bioinoculants or bioformulations has been defined in different ways over the past 20 years, which refers to substances containing living microorganisms that promote plant growth by enhancing nutrient supply. Over time, the definition of biofertilizers has evolved to include microorganisms' role in controlling plant pathogens. They mobilize important nutrients, restore soil health, and enhance plant growth sustainability. While initially focused on nitrogen and phosphorus, efforts are ongoing to identify organisms mobilizing other nutrients (Ibáñez *et al.*, 2023b).

Biofertilizers include bacteria, algae, and fungi that establish symbiotic relationships with plants, enhancing nutrient quality and uptake. They are seen as low-cost, renewable sources of plant nutrients and are gaining importance in integrated nutrient management practices (Reddy *et al.*, 2020).

1.2. History of Biofertilizers

Noble and Hiltner, German scientists, are credited with the development of Nitragin in 1896, considered the starting point for commercial biofertilizers. Nitragin was a laboratory culture of *Rhizobium* bacteria, which aid legumes in fixing atmospheric nitrogen in their root nodules (Oke *et al.*, 2021). It was patented in 1898 (British Patent No. 11460 and US Patent No. 570813) and there were 17 different formulations. On the market, these formulations were available in bottles of 8–10 ounces, containing a substrate composed of sugar, asparagine, gelatin, and aqueous extract of legumes. Starting from 1910, formulations began to utilize substrates such as dry sand, soil, peat, coal, silica, calcium carbonate, and calcium phosphate (Anriquez *et al.*, 2019).

Following the success of Nitragin®, scientists identified other beneficial microbes with agricultural potential. This included *Azotobacter*, another nitrogen-fixing bacteria, and cyanobacteria (blue-green algae) known for their nitrogen fixation abilities in paddy fields. Research continued on various types of biofertilizers, such as mycorrhizal fungi that form symbiotic relationships with plant roots, enhancing nutrient uptake, and phosphate-solubilizing bacteria that unlock unavailable phosphorus in the soil. As concerns about the environmental impacts of chemical fertilizers grew, biofertilizer research gained momentum. Efforts focused on improving the effectiveness, shelf life, and large-scale production of various biofertilizer formulations (Ibáñez *et al.*, 2023a).

1.3. Classification of Biofertilizers

The classification of biofertilizer can be based on diverse parameters resulting in different groups (Fig. 1). We can distinguish:

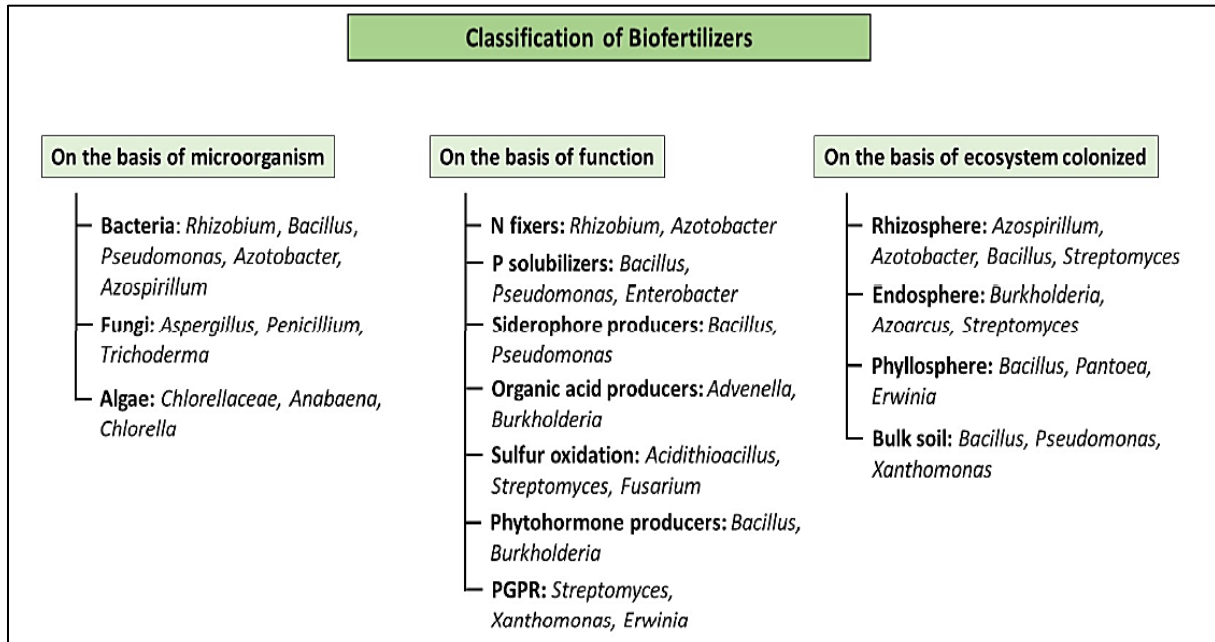


Figure 1: Different biofertilizer classifications (Ibáñez et al., 2023a).

1.3.1. Classification based on the type of microorganism

The simplest classification of biofertilizers is based on the type of microorganism employed, primarily bacteria and fungi (Berg, 2009). Although, the use of microalgae has been on the rise in recent times (Kapoore., 2021). This type of biofertilizers can include:

a) **Bacterial biofertilizers**

A biofertilizer consists of selected efficient living microbial cultures. When applied to plant surfaces, seeds, or soil, these microbes can colonize the rhizosphere or the interior of the host plant, promoting growth by increasing the availability, supply, or uptake of primary nutrients (Thomas and Singh, 2019).

Bacterial biofertilizers play a crucial role in nitrogen fixation. Well-known examples include *Rhizobium* and *Azospirillum*, which convert atmospheric nitrogen (N_2) into a form readily absorbed by plants (NH_4^+) (Nosheen et al., 2021).

b) **Fungal biofertilizers**

Fungi-based biofertilizers are particularly popular. They reduce plant diseases by inhibiting pathogen growth and biological processes, enhancing soil nutrient uptake, producing

bioactive compounds, and stimulating plant growth through hormones and enzymes. Despite challenges like environmental sensitivity, limited shelf life, and slower action compared to chemical fertilizers, biofertilizers offer significant benefits. Mycorrhizal biofertilizers are especially valued for their versatility and environmental friendliness. Ongoing research and advanced technology are expected to further boost their global use and profitability for small and marginal farmers (Odoh *et al.*, 2020).

c) Algal biofertilizers

Algal biofertilizers act as natural recyclers and nutrient reservoirs, enhancing plant growth and offering numerous advantages. Recent research has explored various algae for their beneficial impacts on cultivation, soil, and the environment. Novel industrial processes have been developed for the large-scale cultivation of algae and the production of algal biofertilizers (Iqbal *et al.*, 2021).

1.3.2. Classification based on the ecosystem colonized

While the type of microorganism is a common classification system, another, less prevalent approach, categorizes biofertilizers based on the environments they colonize (Bhattacharyya *et al.*, 2012). This system identifies four main groups:

a) Rhizospheric microorganisms

These microorganisms colonize the soil directly surrounding the roots of plants, forming a crucial zone of interaction for nutrient exchange and plant growth promotion. They include nitrogen-fixing bacteria like *Rhizobium* and phosphate-solubilizing bacteria (PSB) like *Pseudomonas* (Dennis *et al.*, 2010.)

b) Endospheric microorganisms

Endophytes, microorganisms that live inside plant tissues without causing harm, play a significant role in promoting plant growth and enhancing stress resistance. They form symbiotic relationships with their host plants, which can lead to improved nutrient uptake, disease resistance, and tolerance to abiotic stresses such as drought or salinity (Reinhold-Hure *et al.*, 2011).

Mycorrhizae fungi are a prime example. They colonize plant roots and facilitate nutrient uptake, particularly phosphorus (Lesueur *et al.*, 2016).

c) **Phyllosphere microorganisms**

These colonize the aerial parts of plants, such as the stem or leaves, and may play a role in nutrient acquisition, disease suppression, or plant growth regulation. Bacterial and fungal species can fall into this category, influencing plant health through mechanisms like nitrogen fixation or production of antimicrobial compounds (Lindow and Brandl., 2003).

d) **Free-living microorganisms**

These are found throughout the bulk soil and contribute to overall soil health and nutrient cycling, indirectly benefiting plant growth (Fig. 2). They decompose organic matter, making nutrients available for plants, and may also influence soil structure and water retention (Gupta *et al.*, 2015). These include free-living amoebae like *Naegleria spp.*

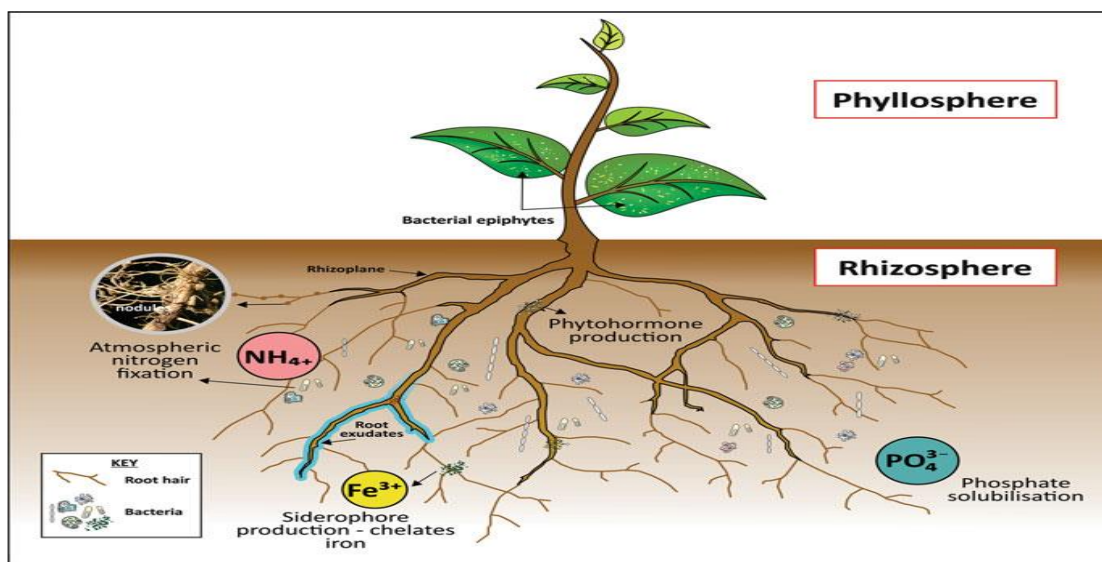


Figure 2: The the ecosystem colonized by microorganisms (Gupta *et al.*, 2015).

1.3.3. Classification based on function

A traditional classification based on the function of microorganism can be considered. Biofertilizers encompass a diverse array of microorganisms, including nitrogen-fixing bacteria, microorganisms capable of solubilizing essential nutrients like phosphorus, potassium, or zinc, siderophore producers, organic acid originators, sulfur oxidizers, phytohormone producers, and plant growth-promoting rhizobacteria (PGPR). However, categorizing them based solely on function isn't always straightforward, as many microorganisms can perform multiple functions. Moreover, their synergistic combinations often result in beneficial effects for plants (Ibáñez *et al.*, 2023a). The main functional groups of biofertilizers are:

a) *Nitrogen-fixing bacteria*

These bacteria, like *Rhizobium* and *Azospirillum*, have the remarkable ability to convert atmospheric nitrogen (N_2) into a form usable by plants (NH_4^+). This plays a vital role in plant growth, especially for crops like legumes (beans, peas, etc.) that can establish a symbiotic relationship with these bacteria (Vessey *et al.*, 2005).

b) *Nutrient solubilizing microorganisms*

This group includes bacteria and fungi that can unlock nutrients like phosphorus, potassium, or zinc from unavailable forms in the soil. For example, phosphate-solubilizing bacteria (PSB) can make phosphorus more accessible for plants by converting insoluble phosphates into soluble forms (Thomas and Singh, 2019).

c) *Siderophore producers*

These microbes produce special molecules called siderophores that chelate iron from the soil. Iron is an essential nutrient for plants, but often gets bound to soil particles in a form unavailable for uptake. Siderophores help plants acquire this crucial nutrient (Thomas and Singh, 2019).

d) *Organic acid originators*

Certain bacteria and fungi secrete organic acids that can dissolve minerals in the soil, making nutrients like phosphorus and potassium more plant-available. Organic acids such as malate, citrate and oxalate play key roles in rhizosphere processes - nutrient acquisition, metal detox, stress relief, mineral weathering, and pathogen attraction (Jones, 1998).

e) *Sulphur oxidizers*

Plants absorb sulphate, the oxidized form of elemental sulphur (S^0), from soil. These bacteria transform elemental sulfur (S^0) into a form usable by plants (sulfates, SO_4^{2-}). This process improves plant access to sulfur, a vital nutrient for various plant functions (Joshi *et al.*, 2021). Species as **Thiobacillus**, **Thiomicrospira**, and **Thiosphaera** are classified as sulphur oxidizing bacteria. However, heterotrophs, such as some species of **Paracoccus**, **Xanthobacter**, **Alcaligenes**, and **Pseudomonas**, can also exhibit chemolithotrophic growth on inorganic sulphur compounds (Vidyalakshmi *et al.*, 2009).

f) *Phytohormone producers*

Some biofertilizers harbor microbes that can produce plant growth hormones like auxins and cytokinins. These hormones stimulate root development, cell division, and overall plant growth (Spaepen and Vanderleyden., 2011).

g) Plant growth-promoting rhizobacteria (PGPR)

This broad category encompasses a diverse group of bacteria that colonize plant roots and promote growth in various ways. They can improve nutrient uptake efficiency, suppress plant diseases, and enhance stress tolerance (Lugtenberg and Kamilova., 2009).

h) Other mineral-solubilizing Biofertilizers

Soil-dwelling microorganisms serve as biofertilizers, providing essential nutrients like potassium, zinc, iron, and copper in addition to nitrogen and phosphorus. Certain rhizobacteria, such as *Bacillus edaphicus*, *Paenibacillusglucanolyticus*, and *Bacillus mucilaginosus*, enhance potassium uptake, resulting in higher biomass yields for crops like wheat, black pepper, eggplant, pepper, and cucumber. Other microbes like *Bacillus subtilis*, *Thiobacillus thiooxidans*, and *Saccharomyces spp.* can solubilize cheaper zinc compounds found in the soil, reducing the need for expensive zinc sulfate. Additionally, microorganisms can hydrolyze silicates and aluminum silicates, releasing nutrients for plant uptake (Thomas and Singh, 2019).

i) Biofertilizers improving compost quality

These biofertilizers contain microorganisms that accelerate the composting process. They can break down organic matter more efficiently, leading to faster production of high-quality compost rich in nutrients for plants (Thomas and Singh, 2019).

j) Biopesticides

This category includes biofertilizers containing microorganisms that can suppress plant diseases or pests. They might act as antagonists to harmful pathogens or produce compounds with insecticidal properties (Singh *et al.*, 2019).

k) Stress Tolerance Enhancers

Plant-microbe interactions influence plant diversity and survival. Stress in plants affects microbial communities. Drought impacts crop productivity. Plants and microbes develop strategies for drought resistance. Bacterial communities can enhance water stress tolerance in plants. Utilizing microbial consortia is crucial for drought-resistant bacterial inoculants. For example, *Achromobacterpiechaudii* enhances water stress resistance in pepper and tomato plants (Adeleke *et al.*, 2019).

1.4. Production and formulation of Bio-fertilizers

Different producers manufacture various biofertilizers based on their physical nature and carrier materials. These include carrier-based inoculants, agar-based, broth, and dried cultures. New developments in biofertilizer production, such as freeze-dried inoculants, Rhizobium-paste, granular inoculants, pelleting, polyacrylamide-entrapped rhizobia, and pre-coated seeds, show promise for successful inoculation (Reddy *et al.*, 2020). The production of Bio-fertilizers involves several steps, which are illustrated in Figure 3.

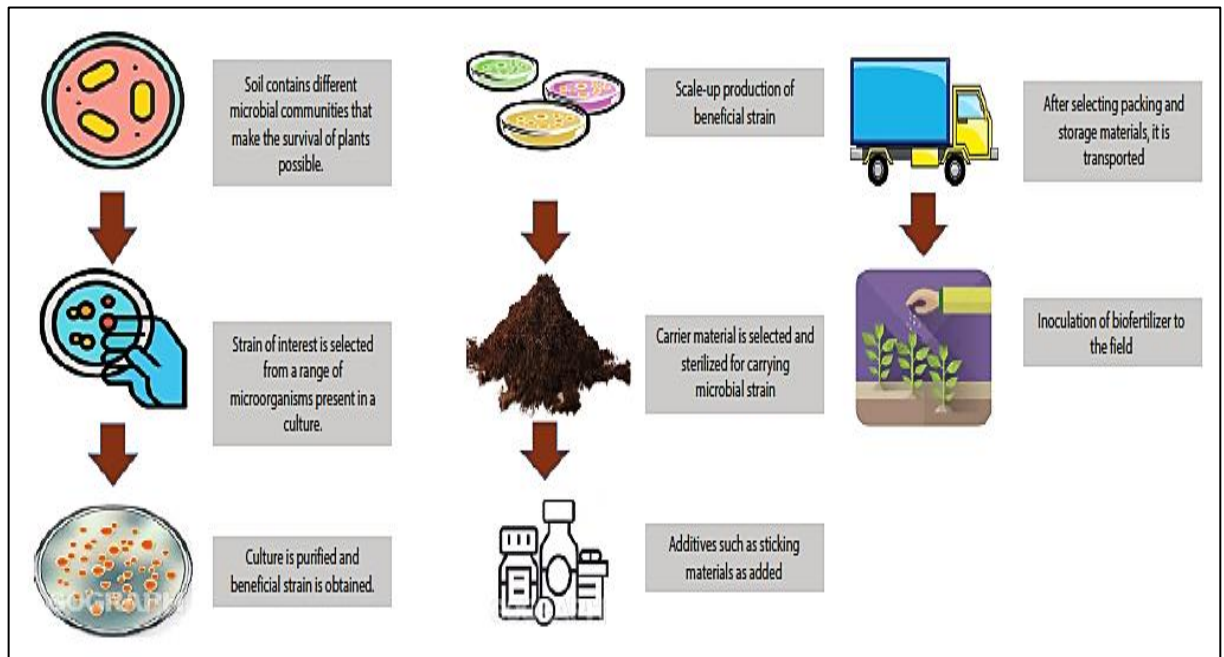


Figure 3: Complete process of formulation of biofertilizers (Saif *et al.*, 2021).

1.5. Types of formulations

Biofertilizers, are crucial for promoting plant growth and improving soil health. To ensure their effectiveness and ease of application in various agricultural settings, biofertilizers come in a variety of formulations. Here's a breakdown of the common types (Fig. 4):

1.5.1. Powder formulations

Powder formulations are dry and stable biofertilizers that often use peat or lignite as a carrier material (Fig. 4A). The carrier protects the delicate microbes during storage, transport, and application until they reach the soil environment (Reddy *et al.*, 2020).

1.5.2. Granular formulations

Resembling to conventional chemical fertilizers, granular biofertilizers are produced by granulating a carrier material like vermicompost or press cake, which is then inoculated with

beneficial microbes (Fig. 4B). Granules offer ease of handling and spreading in fields, making them a user-friendly option for large-scale applications (Reddy *et al.*, 2020).

1.5.3. Liquid formulations

This readily available format suspends live microbes in a carrier solution (Fig. 4C). Liquid bio-fertilizers are often applied directly to soil or seeds, making them convenient for targeted application. However, they may have a shorter shelf life compared to some other formulations (Reddy *et al.*, 2020).

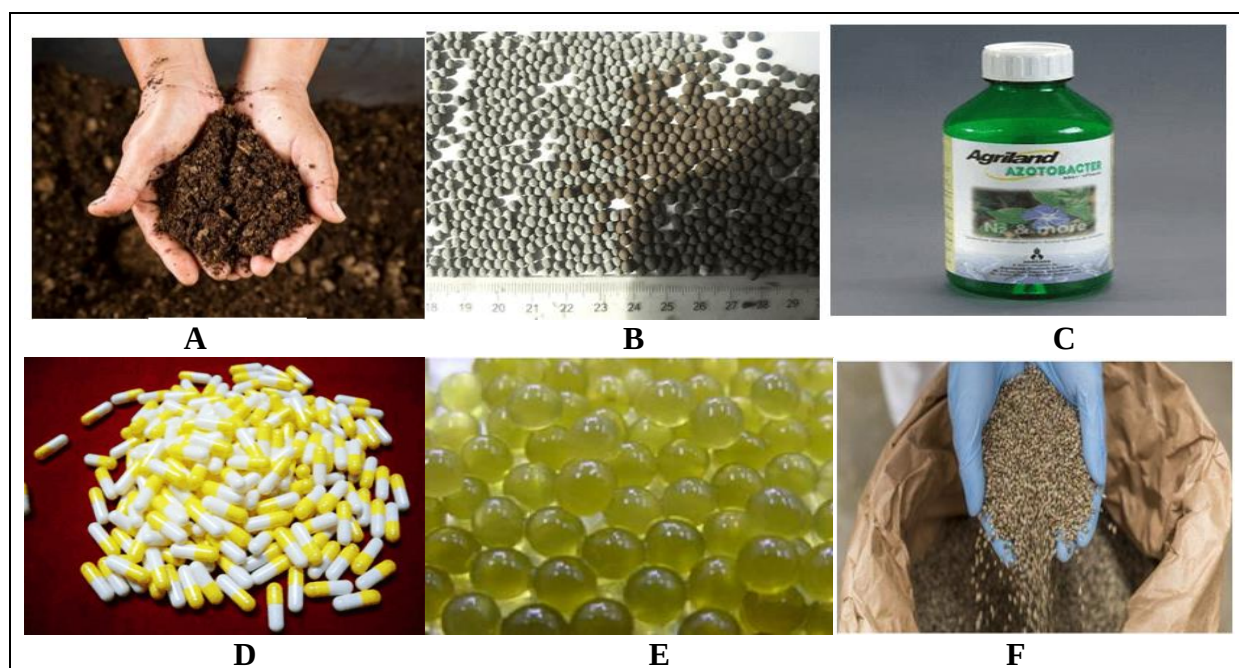


Figure 4: Types of biofertilizers formulations: A: Powder, B: Granules, C: Liquid, D: Encapsulated freeze-dried powders, E: Cell Immobilization, F: Fluid bed-dried formulation.

Sources: A: (<https://www.trees.com/gardening-and-landscaping/peat-moss>); B, C, D and F: (<https://www.bio-fit.eu/hu/q5/lo5-production-of-biofertilizers?showall=1>), E: (https://www.researchgate.net/figure/mobilization-of-microbial-cells-in-calcium-alginate-beads-by-cross-linking-technique_fig1_236119384).

1.5.1. Cell Immobilization

This technique goes beyond simply using a carrier material. It involves immobilizing microbes within a gel matrix or carrier to enhance their survival and effectiveness in the soil (Fig. 4E). Cell immobilization can protect the microbes from harsh environmental conditions and extend their lifespan in the soil, potentially leading to longer-lasting benefits (Saif *et al.*, 2021).

1.5.2. Fluid bed-dried formulation

This method utilizes a stream of air to rapidly dry bio-fertilizers in a fluidized bed. This rapid drying process helps preserve the viability of the microbes, ensuring they remain effective when applied to the soil (Saif *et al.*, 2021).

1.5.3. Mycorrhizal Formulations

These biofertilizers specifically contain mycorrhizal fungi, which form symbiotic relationships with plant roots. These fungi help plants access essential nutrients from the soil, promoting plant growth and overall soil health (Saif *et al.*, 2021).

1.6. Stickers and additives

Various additives can enhance biofertilizer performance and characteristics. Sticking agents, commonly assimilated with peat-based materials, improve the formulation's ability to achieve maximum coverage over seeds. These adhesive materials, often polysaccharides like carboxymethylcellulose or gum, caseinate salts, and polyalcohol derivatives, must be nontoxic, easily dispersible, and exhibit better adhesion to ensure the survival of microbes on seeds. While sticking agents for rhizobia aim to maintain bacterial viability, the mechanism by which viability is increased remains unclear. Cryoprotectants, such as glycerol or sugars, can be added to biofertilizers requiring cold storage to protect microbes during freezing and thawing processes (Saif *et al.*, 2021).

1.7. Packaging

The packaging of biofertilizers serves multiple vital functions. Firstly, it protects microbes from physical damage, controls moisture levels, and shields against light and oxygen exposure. All of which can affect microbial viability, it regulates moisture by offering different permeability levels, catering to various biofertilizer formulations. Lastly, packaging materials are selected based on their ability to act as barriers against light and oxygen, ensuring the viability of the microbes (Reddy *et al.*, 2020).

1.8. Application of biofertilizers

Various methods are employed to introduce biofertilizers into the soil. These methods include the sprinkling method, which entails moistening seeds with a small amount of water before blending them with peat powder, as well as inoculating seeds with powder formulations. Additionally, biofertilizers can be mixed with dry seeds in the seed hopper or treated directly onto seeds. The slurry method involves suspending biofertilizers in water before mixing with

seeds, while a peat-in-water mixture can be sprayed into furrows during planting. Other methods include seed pelleting, seedling root dipping, soil application, and coating a slurry mixture of biofertilizers and adhesive onto seeds (Bashan, 1998). Some of the applications of the biofertilizers are (Fig. 5):

1.8.1. Field application of biofertilizers

a) Soil application of biofertilizers

This method involves the direct application of biofertilizers to the soil, either separately or in combination with other biofertilizers. For example, a phosphate-solubilizing biofertilizer can be combined with and rock phosphate, and stored overnight with moisture content at 50%, before applying to the soil (Krishnaprabu, 2020).

Soil application of biofertilizers offers advantages such as eliminating seed mixing, reducing direct contact with treated seeds, increasing delivery rates, providing more rhizobia per unit area, and better tolerance to low moisture conditions compared to powder form. Biofertilizers applied via soil include *Rhizobium* for trees or leguminous plants, and *Azotobacter* for various crops (Amenaghawon *et al.*, 2021).

b) Foliar application of biofertilizers

While less common than seed-based or soil applications, foliar application presents an intriguing approach for introducing biofertilizers into the plant system. This method involves directly spraying biofertilizers onto the leaves of plants, bypassing the root zone entirely. However, the effectiveness of this method for biofertilizers remains an area of active research (Sharma *et al.*, 2013).

1.8.2. Seed application of biofertilizers

a) Seed Treatment

Seed treatment is the most commonly used technique for applying various types of inoculants due to its effectiveness and economy. In this method, seeds are uniformly coated with a mixture of inoculants in a slurry, then dried in the shade and planted within 24 hours. Seed treatment allows for the use of different combinations of bacteria without negative effects and ensures adequate delivery of the required number of bacteria to achieve desirable results (Thomas and Singh, 2019).

b) Seedling root dipping

The seedling root dipping involves dipping of the seedling roots in a watersuspension made up of biofertilizers for a particular period of time before transplanting. The treatment time required for the dipping from crop to crop. For example, paddy crops require a much longer dipping period (about 8–12 h) than vegetable crops (about 20–30 min) (Amenaghawon *et al.*, 2021).

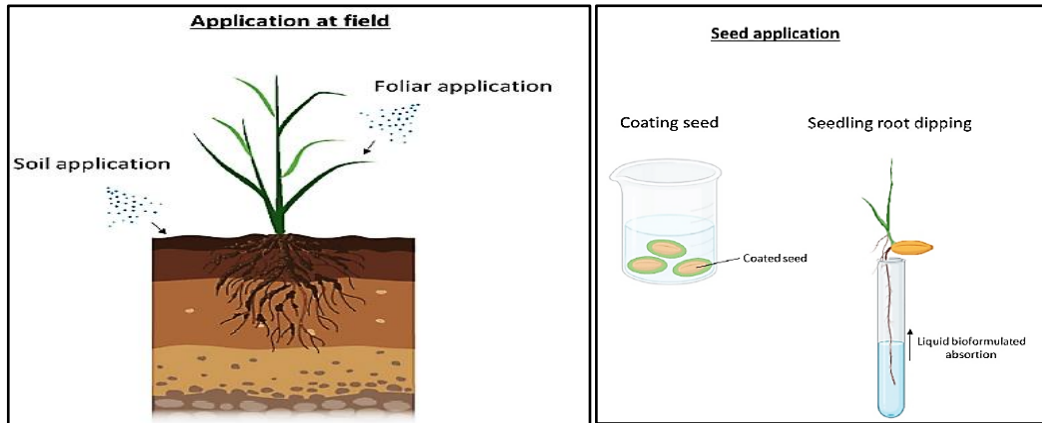


Figure 5: Main modes of biofertilizers application nowadays. A) Field application (Soil application and foliar application); B) Seed application (coating seeds; seedling root dipping).

(Source : https://www.researchgate.net/figure/Main-modes-of-biofertilizers-application-nowadays-Created-by-BioRendercom_fig2_376250829).

2. Biochar as Bio-fertilizers

2.1. History of biochar

The origin of biochar can be traced back to the agricultural practices of the inhabitants of the Amazon, who incorporated large quantities of charcoal into the soil along with manure and other organic fertilizers to improve crop yields (Sohi, 2012). The Amazonian landscape holds extensive evidence of human activities before Columbus' arrival, significantly impacting natural resources. Among this evidence are dark anthropogenic soil matrices spread across various regions of the Amazon. These soils, characterized by a darkened A horizon (**Fig. 6**) and containing archaeological remnants, are referred to as archaeological black earth (terra pretaarqueológica), Indian black earth (terra preta de índio), or simply black earth (terra preta). They serve as indicators of dense or prolonged human presence, marking cultural histories of the past. Additionally, there are soils known as terra mulata, likely resulting from pre-Columbian agricultural practices. Terra mulata soils are brownish in color and contain more charcoal than surrounding soils but exhibit lower chemical fertility compared to terraspretas (Kern *et al.*, 2009).

Biochar has a longstanding history of use in Asian agriculture, particularly in Japan and Korea, across various regions. In the mid-1990s, scientists began recognizing its potential for carbon sequestration and reducing emissions after studying Terra Preta soils. This coincided with global efforts to address climate change by reducing atmospheric carbon dioxide levels. The inaugural meeting of the International Biochar Advocacy Organization in Australia in 2007 prompted the establishment of National Biochar Societies in many countries. These societies aimed to drive biochar research and organize demonstration conferences, leading to a steady increase in research efforts focused on biochar (Han *et al.*, 2020).



Figure 6: Example site profiles for Oxisol (b) and Terra Preta (a). (Glaser *et al.*, 2001)

2.2. The Biochar

Biochar is a carbon-rich substance produced from the controlled heating of biomass, such as wood, manure, or leaves, in a closed container with limited accessible air. This process, known as thermal decomposition, occurs under limited oxygen supply ($<700^{\circ}\text{C}$), leading to the change of organic material into a stable form of carbon. Biochar production reflects old industrial methods, analogous to the manufacture of charcoal, and involves the controlled pyrolysis of biomass. This technology uses the principles of thermochemistry to convert organic matter into a valuable resource with broad applications in agriculture, environmental remediation, and renewable energy (Lehmann and Joseph, 2024). The type of biomass used, the temperature at which the pyrolysis occurs, and residence time all significantly impact the physical and chemical characteristics of biochar (Bruun *et al.*, 2010).

2.3. Characteristics of biochar

The chemical composition of biochars is highly heterogeneous, comprising both stable and labile compounds. The proportions of these compounds vary significantly depending on the pyrolysis process and the type of biomass used (Verheijen *et al.*, 2010). The presence of micropores in biochar, defined as pores with diameters less than 2 nm (Fig.7), contributes to its high adsorption properties (Lehmann and Joseph, 2024). A longer residence time during pyrolysis fosters the polymerization process and facilitates the formation of a porous structure within biochar. Slow pyrolysis, characterized by a residence time surpassing 1 hour, is recognized as a prevailing technology for biochar production. This approach is favored for its enhanced economic viability and advanced technological readiness (Chen *et al.*, 2019).

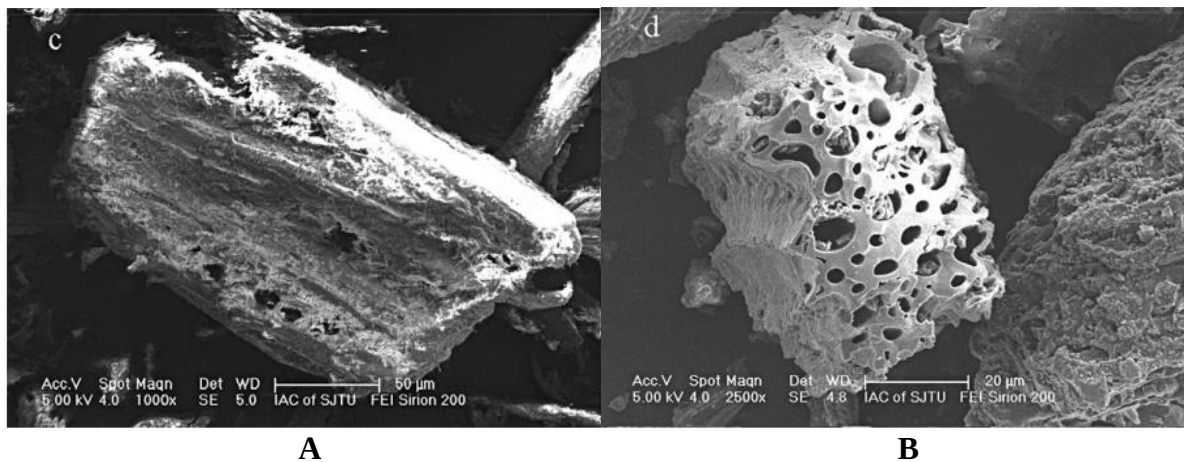


Figure 7: Images of sawdust biomass (A) and biochar (B) (Wang *et al.*, 2014).

The stability and aromaticity of produced biochar are influenced by the ratios of (H/C) and (O/C). Research indicates that the carbon content in biochar increases with higher pyrolysis temperatures. As the temperature rises further, there is a reduction in hydrogen- and oxygen-containing functional groups due to dehydration and deoxygenation processes (Zhou *et al.*, 2021). This increase in carbon content and decrease in hydrogen content led to a decline in the H/C ratio, indicating a more stable biochar structure. Moreover, the proportion of molten aromatic ring structures in biochar tends to increase with higher pyrolysis temperatures, while the content of unstable non-aromatic ring structures tends to decrease (Zhang *et al.*, 2020).

The substantial specific surface area of biochar acts as a sanctuary for soil microorganisms and a repository for soil nutrients, fostering an ideal environment for beneficial microorganisms to establish and flourish within biochar. This attribute facilitates essential soil biological processes such as the decomposition of organic matter and the release of nutrients. Moreover, biochar's capacity to retain soil nutrients extends its availability over time, enhancing

plant absorption and mitigating losses through leaching. Consequently, these advantages culminate in enhanced soil fertility and amplified crop yields (Chan and Xu., 2009).

2.4.Biochar production

Pyrolysis is the process of thermally decomposing biomass without the presence of oxygen and high temperatures (Jahirul *et al.*, 2012). Greater pyrolysis process temperature leads to an enhancement in various biochar properties, such as surface area, toxic metal stabilization, accessible essential nutrients, pH, and carbon content, whereas its cation exchange capacity, overall nitrogen content, total yield, and water adsorption capacity are diminished. Biochar generated at elevated temperatures ameliorates its porousness, subsequently augmenting its efficaciousness as sorbents for capturing pollutants in the soil. In contrast, at lower temperatures (Fig. 8), it is desirable for agrarian purposes (Agrafioti *et al.*, 2013). In the process of pyrolysis, the deliberate exclusion of oxygen plays a pivotal role in orchestrating the controlled decomposition of biomass, resulting in the production of biochar, bio-oil, and syngas. This meticulously regulated environment facilitates the conversion of biomass into these valuable products without engaging in combustion, thereby preserving their chemical composition (Bridgwater, 2012). Slow pyrolysis is favored for its ability to yield biochar with enhanced physical and chemical properties, making it suitable for various agricultural and environmental applications (Fryda and Visser, 2015).

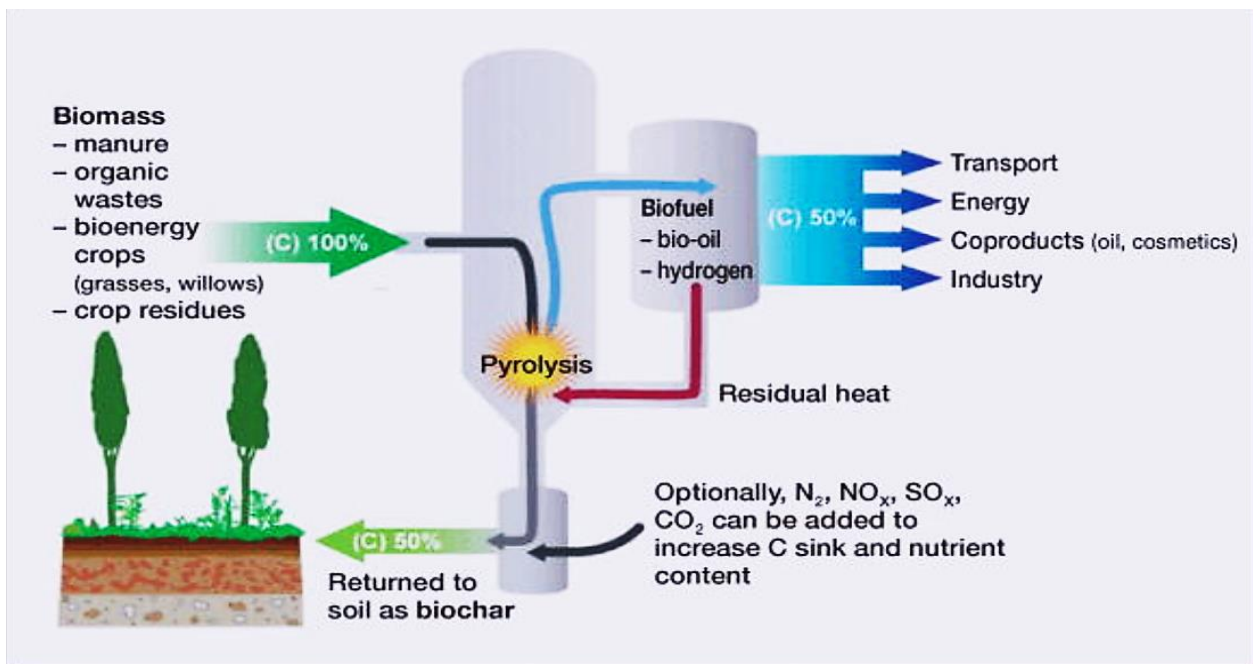


Figure 8: The low-temperature pyrolysis bioenergy concept utilizing sequestration of biochar (Lehmann and Joseph, 2024).

2.5. Benefits of biochar

2.5.1. Biochar and soil

Biochar significantly influences soil fertility by improving water retention, providing habitat for soil microorganisms, enhancing plant nutrient availability, minimizing nutrient leaching, and mitigating nitrogen losses. Moreover, it can elevate soil pH levels and enhance crucial soil properties like aggregation capacity, cation exchange capacity, and nutrient cycling. The effectiveness of biochar in enhancing soil fertility is influenced by various factors including its unique properties, application methods, and soil characteristics. However, to fully comprehend the benefits of biochar application on soil fertility, extensive long-term field trials are essential (Ding *et al.*, 2016). The addition of biochar to soil offers numerous benefits, primarily attributed to its high carbon content, which contributes both organic and inorganic matter to the soil. One of the main advantages is the enhancement of soil organic matter, which improves soil quality in several ways. Biochar increases the soil's capacity to retain water, reduces soil density, facilitates easier plowing and better root development, and enhances nutrient availability for plants. Additionally, the porous structure of biochar enables it to store water and nutrients, promoting their efficient uptake by plants and reducing nutrient loss through runoff and leaching. Emerging evidence also suggests that biochar aids in soil aggregation, promoting soil health and plant productivity by facilitating better nutrient retention and root penetration. Overall, the incorporation of biochar into soil holds great potential for improving soil fertility, water retention, nutrient availability, and overall plant growth (Scott *et al.*, 2014).

According to (Wang *et al.*, 2014), the addition of 4% biochar derived from rice husks to the soil of tea gardens significantly improved the acidic soil properties. Soil pH levels, as well as the levels of potassium (K), calcium (Ca), magnesium (Mg), sodium (Na), total carbon (C), and total nitrogen (N), increased. At the same time, the contents of aluminum (Al) and lead (Pb) decreased.

2.5.2. Water-retention

Water retention is a fundamental consideration in agricultural soil management, influencing irrigation practices and plant growth dynamics (Sim *et al.*, 2021). It is critical for crop productivity as it directly impacts water uptake and transport by plants, thus influencing plant physiology and yield. Experimental research indicates that the addition of biochar can modify soil water retention. This effect is likely due to biochar's high porosity, presence of

hydrophilic domains, and large specific surface area, which enable it to retain water in the soil effectively (Lateef *et al.*, 2019; Razzaghi *et al.*, 2020).

2.5.3. Biochar and climate change

The effectiveness of using biochar to mitigate climate change depends mostly on the amount of carbon from biomass that is stored in the biochar and the speed at which it is released back into the atmosphere (Wang *et al.*, 2015). If un-pyrolyzed biomass that decomposes easily is exposed to fire or left to disintegrate, it will quickly release most of its carbon back into the atmosphere. Consequently, the amount of carbon not converted into minerals is greater for biochar than raw biomass that would have naturally decomposed or burnt. This occurs when the total amount of carbon released through biomass decay exceeds that released through pyrolysis and biochar decomposition (Whitman *et al.*, 2010). The process of converting biomass into biochar can prevent the release of N₂O and CH₄ gases that would have been produced through the decomposition or burning of the biomass. Pyrolysis results in the emission of volatile and gaseous organic chemicals, accounting for over 50% of biomass carbon. A well-engineered contemporary pyrolysis plant guarantees that organic molecules are thoroughly combusted to CO₂ (Woolf *et al.*, 2021).

3. Plantes' technical cards

3.1. Barley technical card

Category	Details
Common Name	Barley (El-Hashash and El-Absy, 2019)
Scientific Name	<i>Hordeum vulgare</i> (El-Hashash and El-Absy, 2019)
Family	Poaceae (El-Hashash and El-Absy, 2019)
Origin	Fertile Crescent region of the Middle East (El-Hashash and El-Absy, 2019)
Climate	Cool-season crop, temperate climates (JACOBS, 2016).
Soil Requirements	Well-drained loamy soil, pH 6.0-8.5 (JACOBS, 2016).
Watering	Moderate water, avoid waterlogging (JACOBS, 2016).
Light Requirements	Full sun (JACOBS, 2016).
Propagation	Seeds (JACOBS, 2016).
Planting Time	Spring or autumn (climate dependent) from March to June (JACOBS, 2016).
Spacing	12.5 cm between rows and 2.5 cm between seeds (JACOBS, 2016).
Harvest Time	90-120 days from planting (variety dependent) (JACOBS, 2016).
Common Varieties (cultivated in Algeria)	Rihane 03 - Saida 183 - El fouara 97 (Boufenar and zaghouan, 2006)
Nutritional Value	Rich in fiber, vitamins B1 and B3, minerals (magnesium, phosphorus) (JACOBS, 2016).
Common Pests	Aphids, wireworms, armyworms (JACOBS, 2016).
Common Diseases	Powdery mildew, rust, smuts, leaf blight montana barley (JACOBS, 2016).
Fertilization	Nitrogen-rich fertilizer montana barley (JACOBS., 2016).
Companion Plants	Clover, beans, corn (JACOBS, 2016).
Non-Companion Plants	Garlic, onions (JACOBS, 2016).
Uses	Animal feed, brewing, food products (flour, soups, stews) (Kevin Young., 1995)(JACOBS, 2016).
Special Considerations	Legume rotation, sensitive to soil acidity (JACOBS, 2016).
Plant Parts (Edible)	Grains (Kevin Young, 1995)
Plant Parts (Non-Edible)	Leaves (narrow, lanceolate), Stems (hollow, jointed) (Kevin Young, 1995)
Leaf Shape	Long and narrow, lanceolate (SALMANIA, 2023)
Leaf Arrangement	Alternate, emerging from nodes along the culm (SALMANIA, 2023)
Stem Structure	Jointed and hollow, up to 1 meter tall (SALMANIA, 2023)
Root System	Fibrous, spreading widely near the surface (SALMANIA, 2023)
Flower Characteristics	Small, inconspicuous flowers in dense spikes (single flower per spikelet) (JACOBS, 2016).
Seed Description	Small, elongated grains (hulled or hulls) (SALMANIA, 2023)
Germination	3-7 days in optimal conditions (moist soil) (JACOBS, 2016).
Seedling Stage	First true leaves emerge, needs consistent moisture (JACOBS., 2016).
Vegetative Growth	Rapid development of leaves and stems (JACOBS, 2016).
Maturity	Grains reach full size and harden (90-120 days) (JACOBS, 2016).

Bolting	Not applicable (heads out to flower) (JACOBS, 2016).
Harvest	Grains harvested when hard and plants turn golden brown (combine harvester) (JACOBS, 2016).

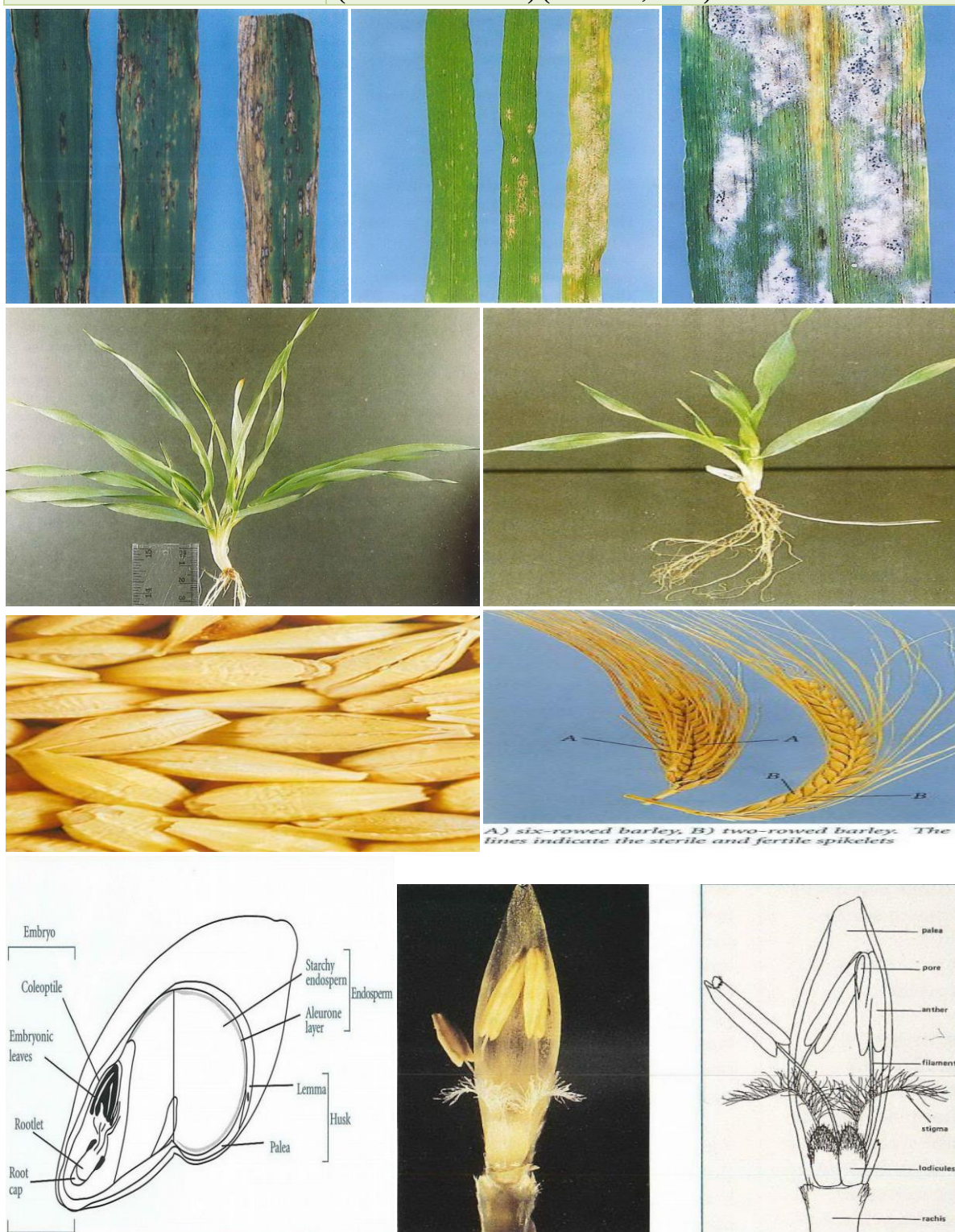


Figure 9: Diseases (A), Leaf morphology (B), Flowering (C) Spacing and varieties (D) and diseases (E) in Barley. (kevin young., 1995)

3.2. Pea technical card

PeaTechnicalcard	
Parameter	Details
Common Name	Pea (Field pea production technology, 2017).
Scientific Name	<i>Pisum sativum</i> (Field pea production technology, 2017).
Family	Fabaceae (Pavek,2012)
Origin	Eastern Mediterranean and Western Asia (Field pea production technology, 2017).
History	Both <i>P. sativum</i> and <i>P. fulvum</i> were domesticated in the Near East around 11,000 years ago, likely from <i>P. humile</i> , while <i>P. abyssinianum</i> emerged from <i>P. sativum</i> in ancient Egypt 4,000–5,000 years ago. Subsequent breeding has led to thousands of pea varieties today. for people eating peas is that of starch grains founded embedded in the calculus (plaque) on Neanderthal teeth at Shanidar Cave and dated about 46,000 years ago, with undomesticated pea remains found in Palestine 23,000 years ago. The deliberate cultivation of peas began in the Near East around 11,300 years ago at Jerf el Ahmar, Syria. Ahihud, a Neolithic site in Palestine, had domesticated peas stored alongside other legumes (Hagenblad <i>et al.</i> , 2014).
Climate	Cool-season crop grows best in temperatures between 13-18°C (53-65°F) (Field pea production technology, 2017).
SoilRequirements	Well-drained loamy soil with pH between 6.0 and 7.5 (Field pea production technology, 2017).
Watering	Moderate water, requires consistent moisture but avoid waterlogging (Field pea production technology, 2017).
Light Requirements	Full sun to partial shade (Pavek,2012).
Propagation	Seeds (Pavek,2012).
Planting Time	Early spring or late summer/early fall for cooler climates (Elzebroek and Wind, 2008).
Spacing	5-10 cm (2-4 inches) between plants and 45-60 cm (18-24 inches) between rows (Elzebroek and Wind, 2008).
Harvest Time	60-70 days from planting for shelling peas; 50-60 days for snap peas (Elzebroek and Wind, 2008).
Common Varieties (Cultivated in Algeria)	Garden peas (shelling peas), snow peas, and snap peas (Field pea production technology, 2017).
Nutritional Value	Rich in vitamins A, C, and K; good source of protein, fiber, and iron (Field pea production technology, 2017).
Common Pests	Aphids, pea weevils, and thrips (Field pea production technology, 2017).
Common Diseases	Powdery mildew, root rot, and fusarium wilt (Field pea production technology, 2017).
Fertilization	Benefits from a balanced fertilizer; inoculating seeds with rhizobium bacteria for nitrogen fixation (Field pea production technology, 2017).
Companion Plants	Carrots, radishes, cucumbers, and beans (Field pea production technology, 2017).

Non-Companion Plants	Onions, garlic, and gladiolus (Field pea production technology, 2017).
Uses	Fresh eating, soups, stews, casseroles, and canning (Pavek,2012).
SpecialConsiderations	Provide Cultivation in saltesoil-borne diseases (Egybte agricluter gov, 2003).
Leaves	Pinnately compound leaves with tendrils at the tips used for climbing (Fig.10A) (Pavek,2012).
Stems	Hollow and green; can be climbing or bushy depending on the variety (Fig.10A) (Pavek,2012).
Roots	Taproot with secondary fibrous roots capable of nitrogen fixation (Fig.10A) (Pavek. ,2012).
Leaf Shape	Pinnately compound with ovate leaflets, tendrils at the tips (Fig. 10A) (Pavek. ,2012).
Leaf Arrangement	Alternate (Fig. 10A) (Pavek. ,2012).
Stem Structure	Herbaceous, hollow, and can be either climbing or bushy (Fig.10 A) (Pavek,2012).
Root System	Taproot with secondary fibrous roots (Pavek. ,2012).
Flower Characteristics	White, pink, or purple flowers bilateral symmetry, typically self-pollinating (Fig.10 B) (Pavek,2012).
Cosse	The ovary houses as many as 15 ovules. The resulting fruit is a closed pod, ranging from 1 to 4 inches in length, frequently characterized by a rough inner membrane. (Fig.10 C) (Pavek. ,2012).
Seed Description	Round, smooth or wrinkled seeds depending on the variety (Fig. 10 A) (Pavek,2012).
Life Cycle (Fig. 10D)	
Germination	Seeds sprout within 10-14 days in optimal conditions, requires moist soil (Elzebroek and Wind, 2008).
Seedling Stage	Emerges with the first set of true leaves; requires consistent moisture (Pavek,2012)
VegetativeGrowth	Rapid leaf and stem development critical for supporting the plant structure (Elzebroek and Wind, 2008).
Maturity	Spring peas bloom 30 to 50 days after planting, while fall peas bloom about 250 days later. Flowering lasts 2 to 4 weeks. Spring peas grow for 60 to 150 days and fall peas for 300 to 320 days (Elzebroek and Wind, 2008).
Bolting	Not applicable (peas do not bolt; they flower and produce pods directly) (Elzebroek and Wind, 2008).
Harvest	Pods are harvested when they are plump and before they become over-mature; best done in the morning (Elzebroek and Wind, 2008).

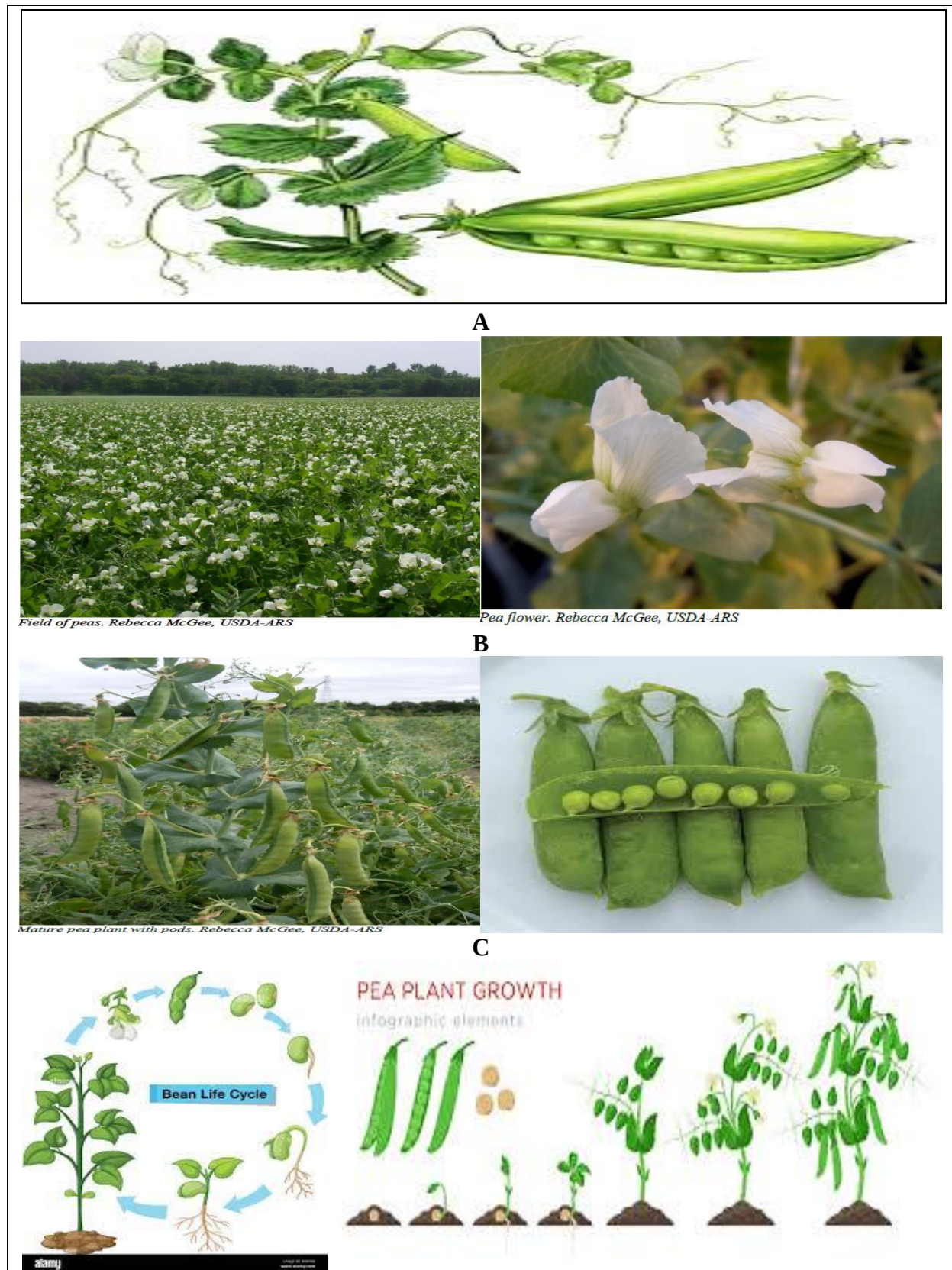


Figure 10: Plant morphology (A), flowering (B), cosse and seeds morphology (C) and plant life cycle (D). (Source: Pavek, 2012 ; Field pea production technology, 2017).

Experimental section

Chapter 1:

Materials and methods

Chapter 1: Materials and Methods

1. Work objectives

The main objective of this study is to test the potential of biochar produced from pyrolysis on plant growth, plant elongation, and the change that occurs in soil fertility through:

- ✓ The production of Biochar from sawdust, using various pyrolysis temperatures and durations;
- ✓ Assessing the phytotoxicity of the biochar produced in order to select the best;
- ✓ Formulation of two types of biofertiliser based on biochar (liquid and dry).
- ✓ and finally, evaluation of these biofertilisers on two strategic crops (barley and peas).

2. Workplace

Our work was carried out in the laboratories of plant biotechnology, plant physiology, animal ecology, plant ecology, plant protection and microbiology of the Faculty of Natural and Life Sciences, Ibn Khaldoun university of Tiaret.

3. Materials

3.1. Laboratory material

The material (equipment, glassworks, chemical products, and others) used during the experiments are listed in the table below:

Table 1 : Laboratory materials used during the experiments.

Equipment	Oven; accurate balance; airtight paper; magnetic stirrerbar; Autoclave; hotplate; stirrer.
Glassworks	Beakers; erlenmeyer flask; vial (50ml, 100ml); Petri dishes; filter funnel.
Chemical products	Agar agar
Others	Sand; cups; distilled water; ruler

3.2. Biological material

During our experimentation, barley was used to assess the phytotoxicity of produced biochar and barley and pea were used for the evaluation of the Biochar based Biofertilizer. The information of the varieties studied are represented in the following table.

Table 2 : General information on the studied varieties.

Plantes	Varieties	Origine	Sources
Barley	Saida 183	Local (Algeria)	ITGC
Pea	Onward	France	Agricultural supplies store Tiaret

4. Methods

4.1. Biochar production

In order to produce our biochars, we went through several steps:

4.1.1. Biomass collection (sawdust)

The biochar utilized in this study was derived from sawdust, specifically post-consumer feedstock obtained from a local wood carpenter in Tiaret, Algeria.

Sawdust, a byproduct of woodworking processes, consists of fine particles of wood that are generated during cutting, grinding, drilling, or sanding of wood. The selection of sawdust as a biomass source for biochar production offers several advantages like: abundance and availability; environmental benefits, energy efficiency (sawdust has a high surface area and low moisture content, making it an efficient pyrolysis material, and its richness in cellulose (source of carbone).

The local origin of the sawdust underscores the importance of utilizing regional resources, thereby minimizing transportation costs and associated carbon emissions. This practice exemplifies a closed-loop system in waste management and resource utilization, contributing to the overall sustainability of the biochar production process.

4.1.2. Cleaning Phase

In the cleaning phase, the collected sawdust underwent a thorough purification process to ensure the material's integrity and suitability for biochar production. This phase involved the meticulous removal of any contaminants, waste materials, and appendages that could affect the quality of the biochar.

The cleaning process included initial screening to separate larger debris and foreign objects from the sawdust, manual sorting to eliminate non-wood materials, such as plastic, metal fragments, and other impurities, and fine sieving to ensure uniform particle size and to remove any remaining small debris (Fig.11).

This comprehensive cleaning ensured that the sawdust used in the subsequent phases was of high purity, enhancing the consistency and quality of the biochar produced.

4.1.3. Grinding Phase

During this phase, the cleaned sawdust was processed to achieve a uniform particle size and optimizing it for pyrolysis. The sawdust was grounded using a mechanical grinder (Fig.11) designed to achieve a precise particle size of 1 mm by 1 mm. Grinding was followed by sieving to ensure uniform particle size.

4.1.4. Pyrolysis

The process of producing high-quality biochar began with the careful storage of the ground sawdust in metal containers (Fig.11). These containers were specifically chosen for their ability to be tightly sealed, creating an anaerobic environment essential for the pyrolysis process. This step was fundamental in maintaining the integrity of the material and ensuring the optimal conditions for biochar production.

Pyrolysis was carried out under controlled conditions, with three selected carbonization temperatures (300°C, 400°C, and 500°C) and three different residence times (3 hours, 4 hours, and 5 hours). This experimental design resulted in nine (09) distinct types of biochar, each produced under a unique combination of temperature and time parameters as flowing (**Table 3**):

Table 3 : Selected carbonization temperatures and different residence times of biochar production.

	3h	4h	5h
300°C	300°C/ 3h	300°C /4h	300°C /5h
400°C	400°C /3h	400°C /4h	400°C /5h
500°C	500°C /3h	500°C /4h	500°C /5h

When the furnace hit the set carbonization temperature, residence time started. After the required time(pyrolysis), samples were taken out, cooled, and placed in plastic-sealed containers (Fig.11). These conditions were chosen to study temperature and time effects on biochar quality. By varying temperature and time, the study aimed to produce diverse biochar samples, offering insights into optimal production conditions. These steps ensured the final product met the specifics of our study. Biochar yield was calculated as:

$$\text{Biochar yield (\%)} = [(\text{initial weight} - \text{final weight})/\text{initial weight}] * 100$$



Figure 11 : Steps of Biochar production.

4.2. Assessment of the phytotoxicity of biochar

In order to assess the phytotoxicity of the produced biochars, germination tests were done in Agar-based biochar macerate. Biochar are characterized by their richness in mineral, so their salinity can be high. Selected plants were chosen regarding their resistance against salt. Barley is known for its resistance to salt but the selected variety is sensitive to carbon.

Biochar macerate was prepared by stirring 20 g of biochar in 250 ml of distilled water for 24h. Total concentration of this macerate was 8%. 1% of (w:v) Agar-based biochar was prepared by adding 0.8 g of Agar Agar to 80 ml of each concentration (Fig.12). Solutions were boiled, autoclaved and then cooled in Petri dishes and kept until it became solid. Each dish was repeated three times.



Figure 12 : Steps of Agar-based biochar macerate production.

Seeds' surface was sterilized using 5.25% to 6.0% sodium hypochlorite (Fig.12). for this, 10% of bleach solution was prepared by mixing one part household bleach with nine parts distilled water.

The surface of the seeds was sterilised for 5 minutes, then the seeds were rinsed with distilled water 5 times. The seeds were then placed in Petri dishes containing the previously prepared biochar macerate, at a rate of 21 seeds per dish. Petri dishes were placed in the incubator at a temperature of 21°C (Fig.12).

Seed germination was monitored daily and growth parameters were measured on days three and eight of germination.

The percentage of germination was determined after 20h, 44h and 68h using the following formula:

$$\text{Germination percentage} = (\text{Number of germinated seeds} / \text{Total number of seeds}) * 100$$

At 68 h of germination, the majority of Petri dishes had a germination percentage of 100%, which led us to stop calculating the number of germinated seeds and to start determining the growth parameters.

The growth parameters measured on the third day of germination were rootlet number, rootlet length and coleoptile length. The lengths were determined in mm using a double decimetre. The number of rootlets was counted manually (Fig. 13).



Figure 13 : Steps of determination of germination and growth parameters.

The growth parameters measured on the third day of germination were rootlet number, rootlet length and coleoptile length. At the eighth day of germination, the number of roots, the

length of the roots, sheaths and leaves and the fresh and dry weights of the seedlings were measured for barley.

The lengths were determined in mm using a double decimetre. The number of rootlets was counted manually. The weights were determined using a precision balance. Dry weights were obtained after drying the seedlings in a ventilated oven at 80°C for 72 hours (Fig.13).

4.3. Formulation of Biofertilizers

As our theme is listed as an innovative project (Startup) under Decree 1275, we have named our company "Bio-Phoenix". The concept of our company is to restore agricultural waste and use it to manufacture biochar.

We manufacture two main products: a solid biochar-based fertiliser called "Phoenix fertilizer" and a liquid fertiliser called "Phoenix liqui-fertiliser".

The biochar and the biochar waste resulting from the manufacture of the liquid biochar were used to manufacture seven other products (company with 0 waste). The by-products produced are (Fig.14): "Phoenix ruminant's care" biochar sticks for animal feed, "Phoenix land" mineral cultivation substrate, "Phoenix for Mashroom" mushroom cultivation substrate, "Phoenix compressed discs" charcoal discs, "Phoenix plants' care" discs for protecting ornamental and potted crops, "Phoenix dehumidifier" moisture absorption discs and "Phoenix Deodorizer" bad odour absorption discs.



Figure 14 : Based-Biochar produced products.

4.3.1. Solid Biofertilizer

Solid biochar-based fertiliser was produced by pyrolysis of biomass (sawdust) at 300°C for 3 hours. The choice of this temperature and duration was justified by the fact that these factors (temperature and duration) had no significant effect on germination and growth parameters. For this reason, we thought it wise to choose the lowest temperature and duration to reduce energy consumption.

4.3.2. Liquid Biofertilizer

Liquid biofertiliser was produced by maceration of 800 g of solid biochar in 10 litres of distilled water for 24 hours at room temperature. This stage was followed by filtration (Fig.15). The filtrate was recovered and then diluted to obtain different concentrations of 0.5%, 1% and 2%. The 4 and 8% concentrations were avoided because they had some undesirable effects in the phytotoxicity tests.



Figure 15 : Steps of preparation of liquid biofertilizer.

4.4. Evaluation of the produced Biofertilizers

To assess the effectiveness of the biofertilisers produced, two crops of peas and barley were planted in the presence of the biofertilisers and their growth parameters were monitored for three months.

4.4.1. Seeds preparation

Seeds' surface (barley and pea) was sterilized using 5.25% to 6.0% sodium hypochlorite for 5 minutes, then the seeds were rinsed with distilled water 5 times. The seeds were then placed in Petri dishes containing three slides of filter paper, watered and placed in an incubator at 20°C. At the fifth day of germination, seeds were transferred into trays containing a mixture of soil and compost (1:1; w: w) until the development of seedlings.

4.4.2. Installation of crops

Soil collected from the Tiaret region was used for this experiment. It was cleaned of large particles and sieved to a diameter of 2 mm (Fig.16). It was placed in three-litre plastic pots. Each pot contained 3 kg of soil.

To evaluate the solid biofertiliser, a soil-biochar mixture was prepared to obtain concentrations of 0%, 0.5%, 1% and 2% as shown in Table 4. The mixture, with a total weight of 3kg, was placed in the plastic pots one week before planting to allow good interaction between the biochar and the soil. These pots were irrigated with tap water.

Table 4 :Préparation of the set concentrations of soil-biochar

Concentration	Mixture
0%	3000 g of soil only.
0.5%	Mix 15 g of biochar with 2.985 g of soil.
1%	Mix 30 g of biochar with 2.970 g of soil.
2%	Mix 60g of biochar with 2.940g of soil.

To evaluate the liquid biofertiliser, 3kg of soil was placed in the plastic pots and biochar macerate in concentrations of 0%, 0.5%, 1% and 2% were prepared by diluting the previously prepared liquid biofertiliser. These macerates were used to irrigate the crops.

The seedlings (barley and peas) were then transferred to pots pre-filled with soil or the soil-biochar mixture at a rate of 3 seedlings per pot. Each pot was repeated 3 times.

Control pots were prepared under the same conditions. They contained 3 kg of soil, 3 pea or barley seedlings and were irrigated with tap water.

The trial was set up under cover, in semi-controlled conditions, using a total randomisation system.

4.4.3. Parameters measured

After the crops were established, growth parameters were measured every week. For barley, the parameters measured were stem height, number of leaves and leaf length, and for peas, the parameters measured were stem height, number of branches and number of leaves.



Figure 16: Steps of crop installation

5. Statistical analysis

Statistical processing was carried out using SPSS software, and the data obtained was subjected to an analysis of variance. The student's *t* test was applied to reveal the difference between the means of the treatments. Homogeneous groups were compared using the *Tukey* test at the 95% safety level.

Chapter 2 :

Results and discussion

Chapter 2 : Results and discussions

1. Results

1.1. Biochar yield

During this experiment, biochar was produced by pyrolysis of sawdust using different temperatures (300°C, 400°C, and 500°C) and durations (3 hours, 4 hours, and 5 hours).

The results of biochar yields obtained are illustrated in (Fig.17) Biochar yields varied from 28.95% to 58.05%. Student's t-test revealed significant differences ($p < 0.05$) among the temperatures and durations of biochar production.

There is a clear negative correlation between, on one hand, the temperatures and durations of biochar production, and on the other hand, the biochar yield. As the pyrolysis temperature increases, the biochar yield decreases. Similarly, as the residence time increases, the biochar yield decreases. The highest biochar yields were obtained at a temperature of 300°C and a residence time of 3 hours. The lowest biochar yields were obtained at a temperature of 500°C and a residence time of 4 hours.

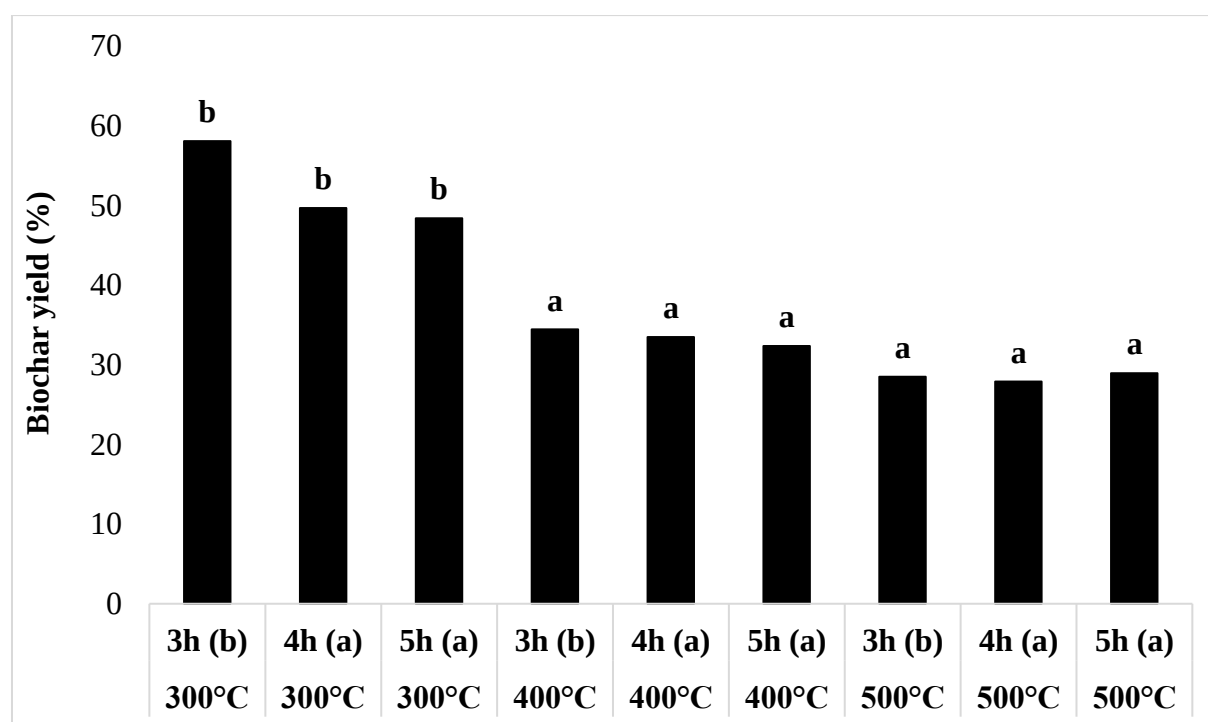


Figure 17: Variation of Biochar Yields According to Pyrolysis Temperatures and Residence Times.

1.2.Results of Biochar Phytotoxicity Evaluation

1.2.1. Barley germination on gelose biochar macerate

The results of the analysis of variances, illustrated in the table below, reveal significant differences ($P \leq 0.05$) among the biochar production temperatures after 20 hours and 68 hours. This indicates that they have a significant effect on barley germination. However, there is no significant effect ($P > 0.05$) observed between the biochar production duration on barley germination.

Table 5 : Analysis of variances results for barley germination in gelose biochar macerate.

Germination	Sources	SCE	ddl	CM	F	Sig.
After 20h	Temperatures	622.159	2	311.079	4.116	0,019*
	Durations	228.118	2	114.059	1.509	0,227 ns
After 44h	Temperatures	58.791	2	29.396	1.807	0,17 ns
	Durations	44.681	2	22.341	1.373	0,258 ns
After 68h	Temperatures	60.807	2	30.404	3.776	0,027 *
	Durations	2.352	2	1.176	0.146	0,864 ns

The graphs in (Fig. 18 A, B, and C) illustrate the percentages of barley germination in gelose biochar macerate after 20h, 44h, and 68h, respectively.

After 20h, it is notable that seeds germinating in gelose water alone (control) have the highest germination percentages (79.365%) compared to seeds germinating in gelose biochar macerate (Fig. 18A). The barley germination rates are similar across the three temperatures, averaging 60.847% at 400°C, 60.635% at 300°C, and 56.191% at 500°C. Regarding biochar production durations, gelose biochar macerate produced over 3 hours yielded the highest germination percentages with an average of 61.058%. The durations of 4 hours and 5 hours showed similar results (58.413% and 58.201%, respectively).

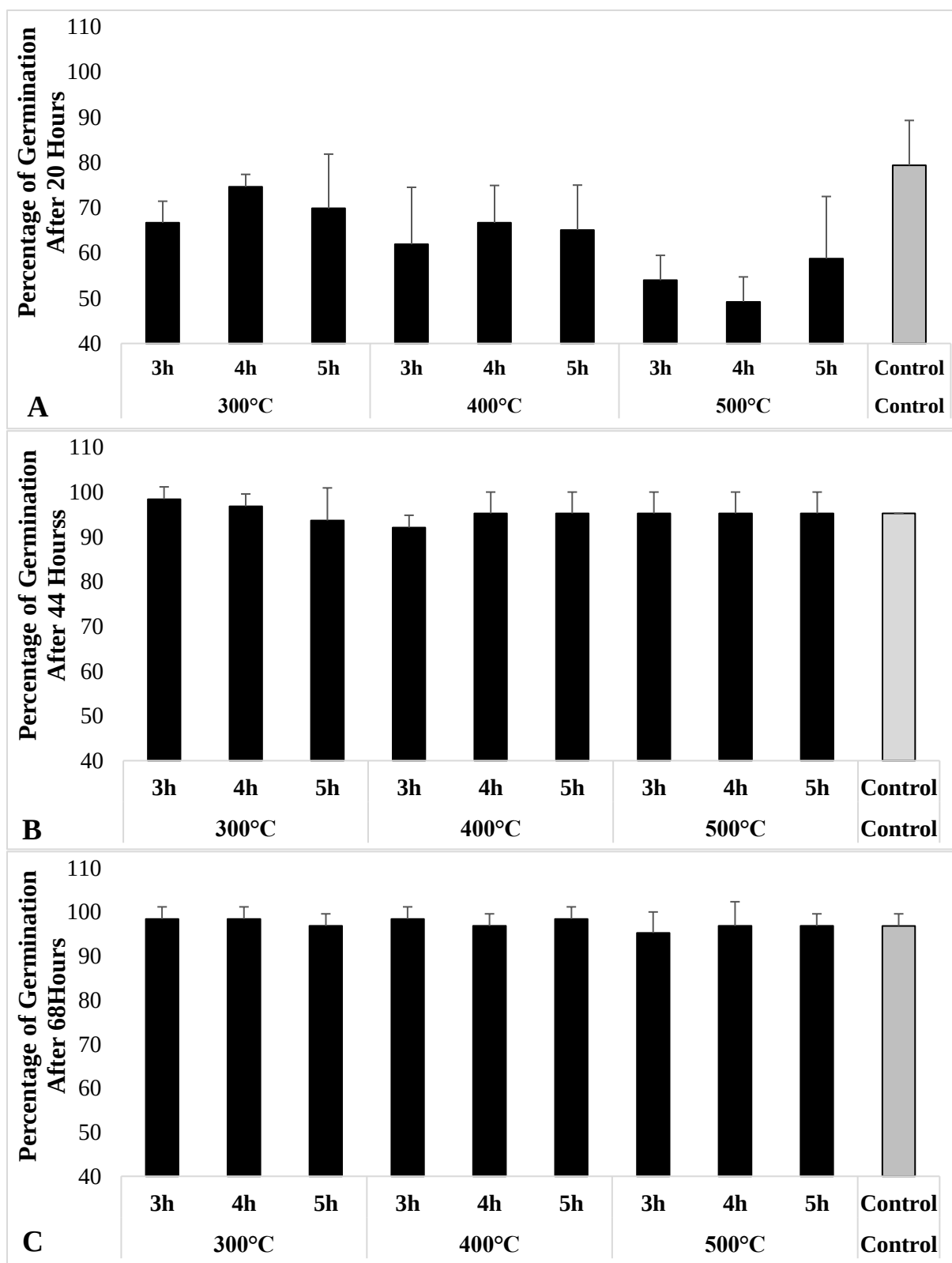


Figure 18: Variation of barley germination percentages on gelose biochar macerate after 20h (A), 44h (B), and 68h (C).

After 44h and 68h, the germination percentages in the biochar macerates are very similar to each other and to those of the control, mostly ranging between 90% and 100% (Fig. 18 A and B).

At 44h, barley germination rates in gelose biochar macerate are nearly identical across the three biochar production temperatures: 96.402% at 300°C, 95.873% at 400°C, 95.238% for the control, and 94.815% at 500°C. Significant results were recorded for barley germination in biochar macerate produced over 3 hours of pyrolysis (96.296%). Production durations of 4 hours and 5 hours, along with the control, yielded very similar results (95.873%, 95.238%, and 94.921%, respectively).

At 68h, biochar production temperatures of 300°C and 400°C reveal the highest germination percentages, with 98.413% and 97.989%, respectively, followed closely by 500°C and the control with 96.825% (Fig. 18 C). Production durations of 3 hours, 4 hours, and 5 hours achieved high germination percentages of 97.884%, 97.778%, and 97.566%, respectively, followed by the control (96.825%). Various concentrations of gelose biochar macerate yielded interesting results.

1.2.2. Growth parameters of barley growing on gelose biochar macerate

a) Root Length

The results of the analysis of variances (Table 6) indicate highly to very highly significant differences between the pyrolysis temperatures ($P \leq 0.01$ and $P \leq 0.001$), and non-significant effects between the durations ($P > 0.05$). This suggests that temperature has a significant effect on the root length of barley.

Table 6 : Analysis of variances of root length in barley grown on biochar macerate.

Days	Sources	SCE	ddl	CM	F	Sig.
3rd day	Temperatures	1750.782	2	875.391	11.438	0***
	Durations	227.742	2	113.871	1.488	0,227 ns
8th day	Temperatures	139197.6	2	69598.801	295.967	0***
	Durations	481.647	2	240.824	1.024	0,36 ns

The histograms in Figure (19) illustrate the root length of barley growing on gel-seeded biochar macerate on the 3rd and 8th days of germination.

On the 3rd day of germination, the control plants had the longest roots (32.133 mm) compared to most plants growing on gel-seeded biochar macerate. Seedlings growing on biochar macerate produced at 500°C had the shortest roots (26.85 mm). Seeds growing on biochar macerates produced at pyrolysis temperatures of 400°C and 300°C showed intermediate values. The pyrolysis durations had no effect on root length on the third day of germination. Germination percentages were very similar (Fig. 19A).

By the eighth day of germination, biochar promoted rapid root development, especially at pyrolysis temperatures of 300°C and 400°C. The temperature of 500°C appeared to have a negative effect on root length growth (Fig. 19B). Biochar residence times had no effect on root length, as their values were very close (53.68 mm at 3h, 53.61 mm at 4h, and 55.44 mm at 5h), but they were better than those obtained by control plants (47 mm).

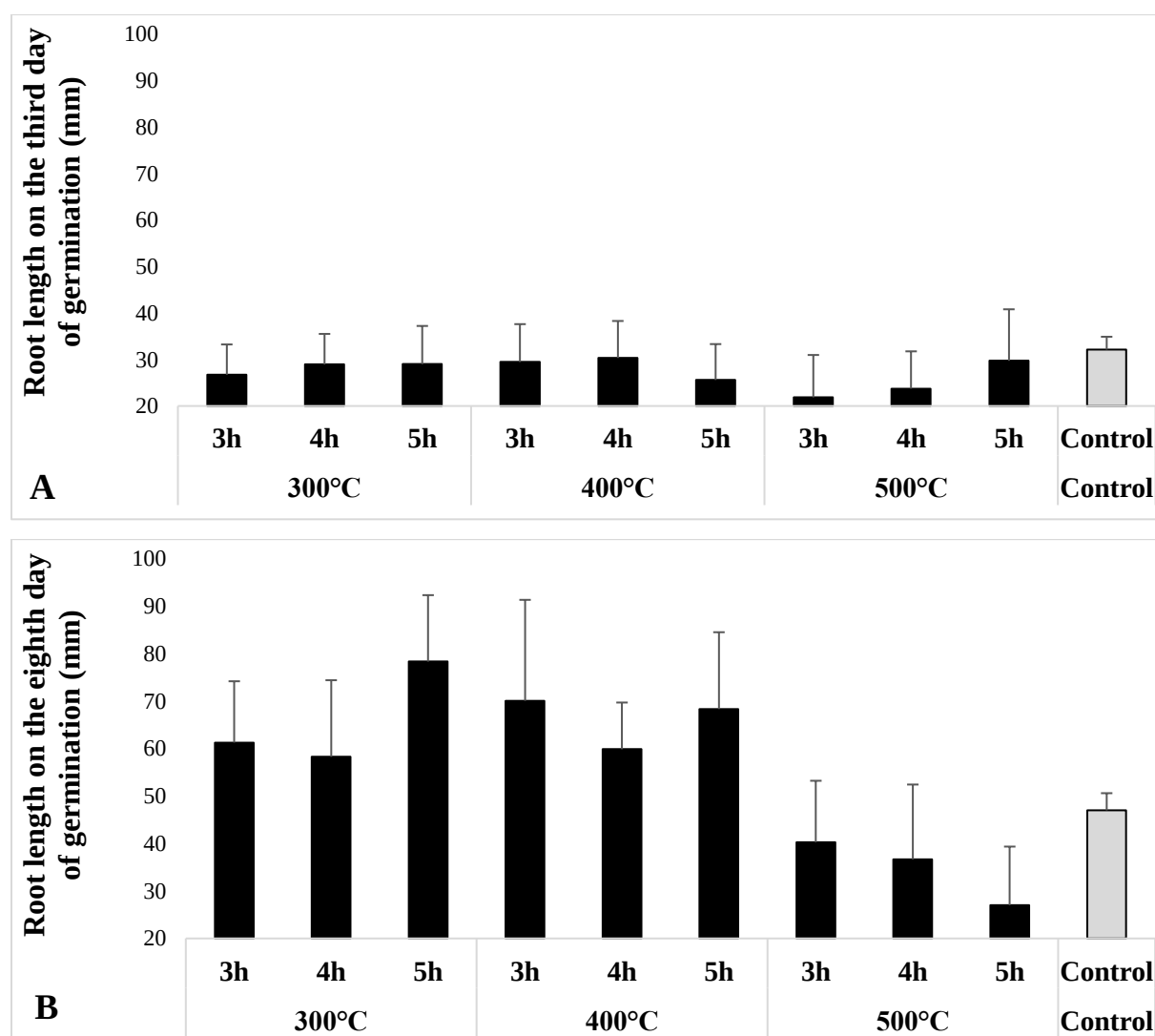


Figure 19 : Variation in root length of barley growing on gel-seeded biochar macerate, on the third (A) and eighth (B) days of germination.

a) Number of roots

The results of the analysis of variance (table 7) indicate that there are no significant differences for temperatures and durations of biochar ($P > 0.05$). This suggests that these factors do not have a significant effect on root number.

Table 7 :Table: Analysis of variance of root number in barley growing on biochar macerate.

Days	Sources	SCE	ddl	CM	F	Sig.
3rd day	Temperatures	0.439	2	0.219	0.482	0,618 ns
	Durations	0.679	2	0.339	0.745	0,475 ns
8th day	Temperatures	1.327	2	0.664	1.208	0,3 ns
	Durations	2.492	2	1.246	2.267	0,104 ns

The histograms in figure (20) illustrate the number of roots of barley seedlings growing on gel-seeded biochar macerate on the third and eighth days of germination. It ranged between 3.8 and 4.667. Control seedlings had an average root number of 4.333 on the 3rd day of germination and 4.666 on the 8th day.

On the third day of germination, the highest root numbers in barley seedlings were observed in those growing on biochar produced at 300°C for 5h (Fig. 20A). The average root numbers for these treatments were 4.6. The lowest root numbers in barley seedlings were observed in those growing on biochar produced at 400°C for 5h, and at 500°C for 3h. The average root numbers for these treatments were 3.93 and 3.93, respectively.

At the eighth day of germination, the number of roots in barley seedlings was very similar between those growing on biochar macerate and control seedlings. A pyrolysis duration of 5h resulted in seedlings having the lowest number of roots at 4.33 (300°C), 4.26 (400°C) and 4.13 (500°C) compared to control plants (Fig. 20B). The 3h and 4h durations showed intermediate results.

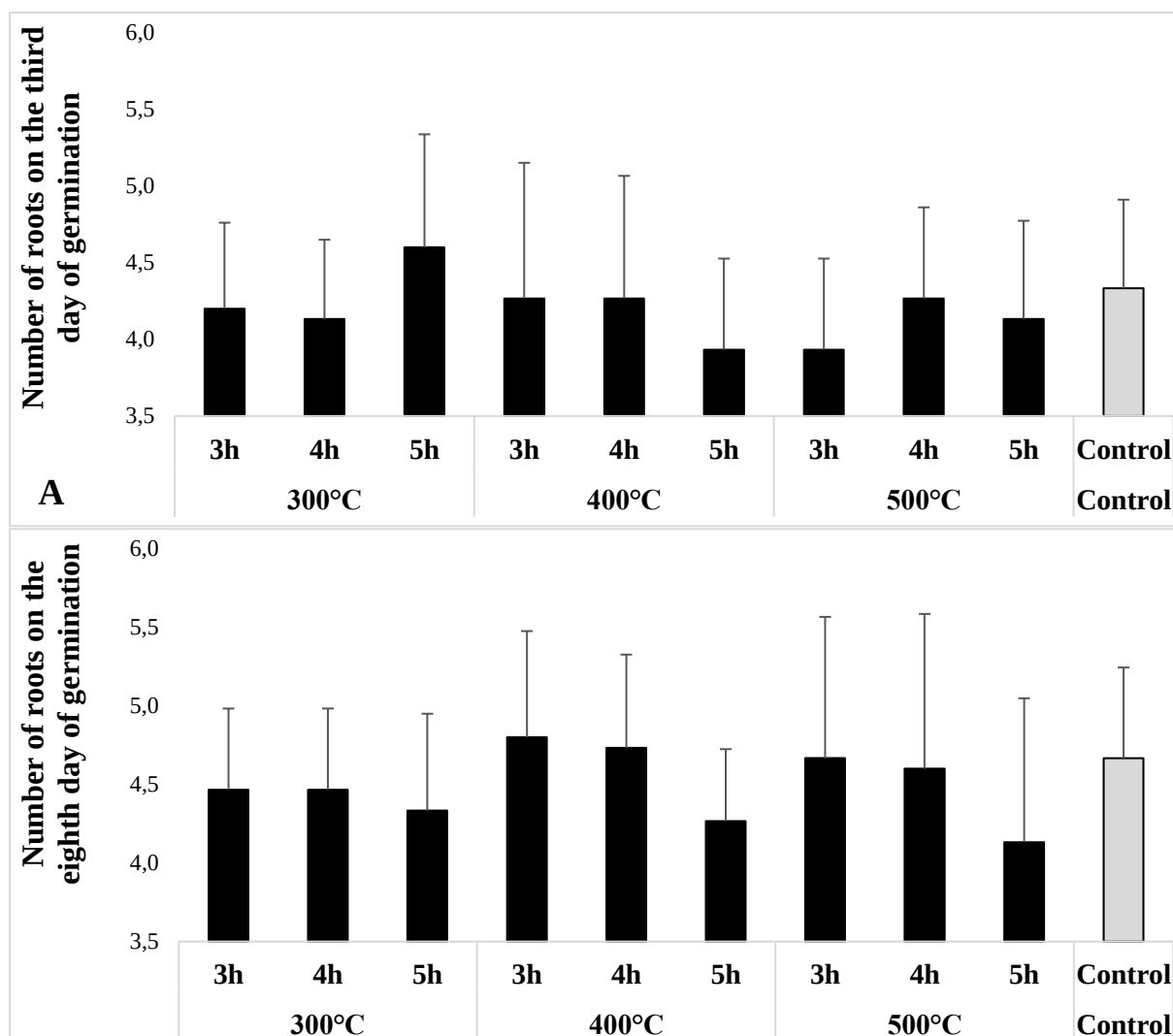


Figure 20 : Variation in the number of roots of barley growing on gel-seeded biochar macerate, on the third (A) and eighth (B) days of germination.

b) Length of coleoptiles and aboveground parts

The results of the analysis of variance (Table 8) reveal highly significant differences between the production temperatures of biochar ($P \leq 0.001$). No significant effect was observed for biochar production durations ($P > 0.05$). This indicates that biochar temperature significantly affects coleoptile length after 3 days of germination.

Table 8 : Analysis of variance of coleoptile and aboveground parts length in barley seedlings growing on biochar macerate.

Days	Sources	SCE	ddl	CM	F	Sig.
3rd day	Temperatures	3139.701	2	1569.85	45.101	0***
	Durations	62.865	2	31.433	0.903	0,406 ns
8th day	Temperatures	47522.85	2	23761.424	77.648	0***
	Durations	898.039	2	449.019	1.467	0,231 ns

The graph in figure (21) illustrates the variation in coleoptile and aboveground parts length of barley on the third and eighth days of germination.

On the third day of germination, most barley seeds growing on gel-seeded biochar macerate had longer coleoptiles compared to control plants. The highest coleoptile lengths were recorded in seeds growing on biochar macerate produced at 300°C for 4h, averaging 15.46 mm, and at 400°C for 4h, averaging 14.53 mm (Fig. 21A). Conversely, the lowest values (4.26 mm) were recorded in barley seeds growing on biochar macerates produced at 500°C, especially those pyrolyzed for 3h and 4h.

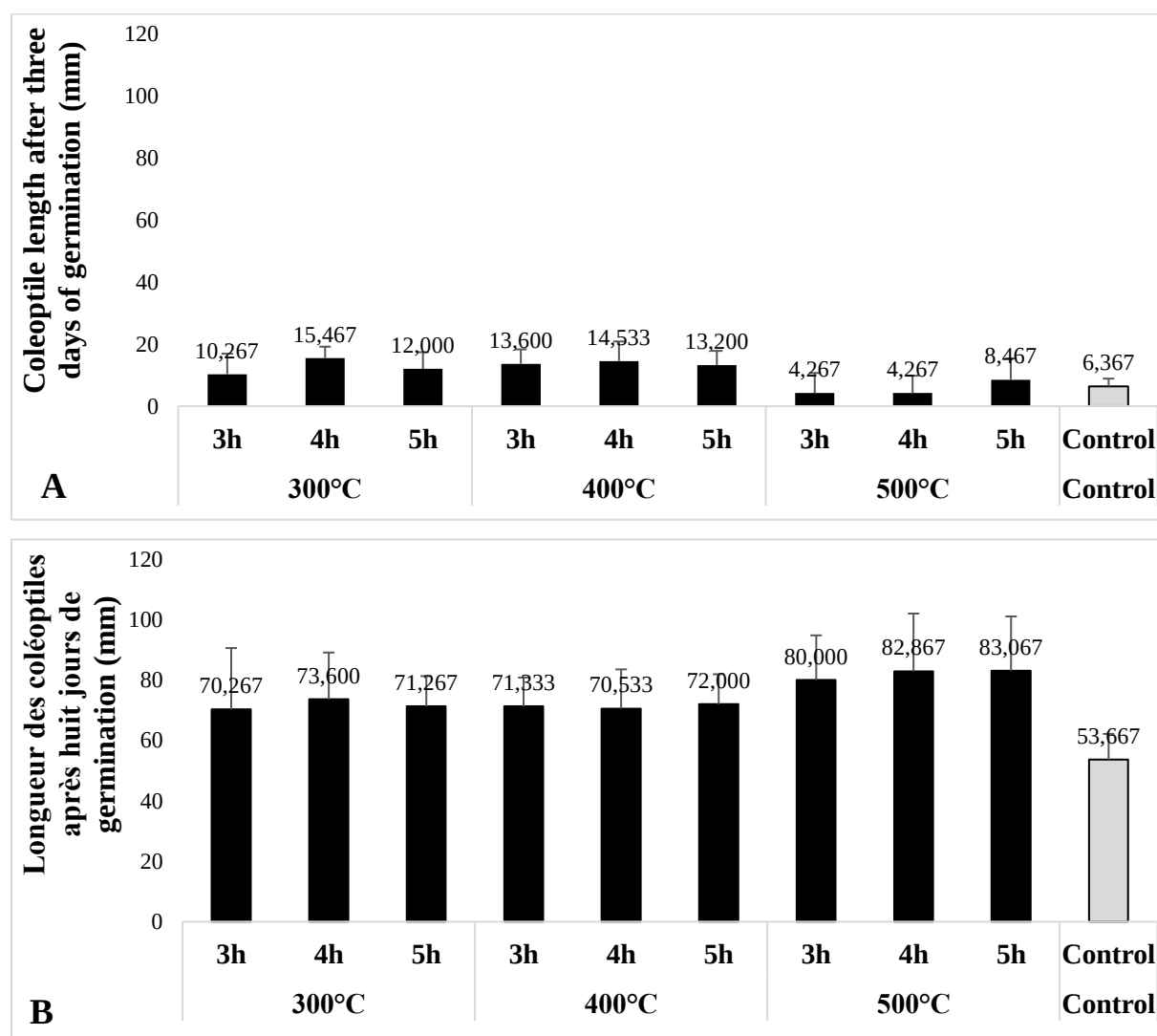


Figure 21: Variation in coleoptile and aboveground parts length of barley growing on gel-seeded biochar macerate on the third and eighth days of germination.

By the eighth day of germination, all biochars stimulated aboveground growth, as the lengths of aboveground parts in control seedlings were the lowest at 53.66 mm. It is also notable

that the temperature of 500°C, which delayed coleoptile growth on the third day, resulted in the best aboveground lengths here.

The highest lengths of aboveground parts were recorded in seeds growing on biochar macerate produced at 500°C for 5h and 4h, with respective averages of 83.067 mm and 82.86 mm (Fig. 21B). In contrast, the lowest values (70.26 mm and 60.53 mm) were recorded in barley seeds growing on biochar macerates produced respectively at 300°C and 400°C and pyrolyzed for 3h and 4h, respectively.

c) Fresh and dry weights of seedlings

The results of the analysis of variance (Table 9) reveal highly significant differences ($P \leq 0.001$) between biochar production temperatures, indicating that temperature has a significant effect on the average fresh and dry weights of plants. In contrast, there was no significant effect ($P > 0.05$) for biochar production duration, indicating that these factors did not significantly influence the average fresh and dry weights of plants.

Table 9 : Analysis of variance for average fresh and dry weights of barley seedlings growing on gelatinized biochar slurry.

	Sources	SCE	ddl	CM	F	Sig.
Fresh Weight	Temperatures	0.063	2	0.031	7.058	0,001***
	Durations	0.002	2	0.001	0.169	0,845 ns
Dry Weight	Temperatures	0.012	2	0.006	8.533	0***
	Durations	0	2	7.44E-05	0.103	0,903 ns

The histograms in figure (22) illustrate the average fresh and dry weights of barley seedlings growing on gelatinized slurry of biochars produced at different pyrolysis temperatures and durations.

It is evident that the control seedlings had the lowest fresh and dry weights, with respective averages of 0.165 and 0.061 g per seedling (Fig. 22A and B). All biochars resulted in seedlings with significantly higher fresh and dry weights, highlighting the beneficial role of biochar in these traits. For fresh weights, biochar produced at 400°C appears most effective, yielding seedlings with an average fresh weight of 0.335 g per seedling. Meanwhile, for dry weights, biochar produced at 500°C yielded the highest averages (0.123 g per seedling).

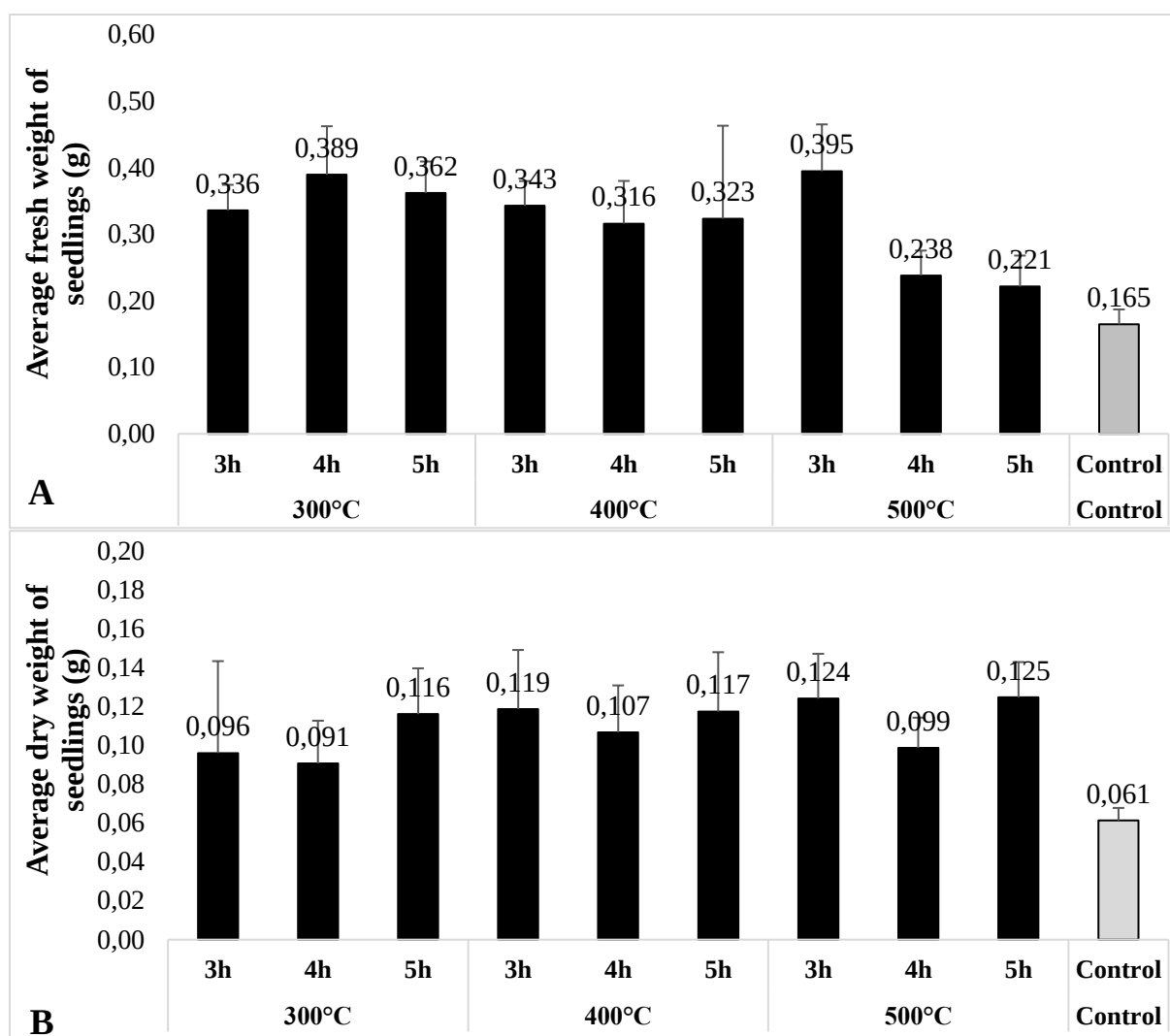


Figure 22: Variation in average fresh (A) and dry (B) weights of barley seedlings after 8 days of germination.

The highest fresh weights of seedlings are recorded for biochars with respective biochar produced at pyrolysis temperatures of 500°C for 3 hours and 300°C for 4 hours, with averages of 0.395 and 0.389 g per seedling, respectively (Fig.22A). The highest dry weights of seedlings are recorded for biochars produced at a pyrolysis temperature of 500°C for 3 hours and 5 hours, with averages of 0.124 and 0.125 g per seedling, respectively (Fig.22 B).

The lowest fresh weights of seedlings are recorded for biochars with respective biochar produced at a pyrolysis temperature of 500°C for 4 hours and 5 hours, with averages of 0.238 and 0.221 g per seedling, respectively (Fig.22 A). The lowest dry weights of seedlings are recorded for biochars with respective biochar produced at pyrolysis temperatures of 300°C for 3 hours and 4 hours, with averages of 0.096 and 0.091 g per seedling, respectively (Fig.22 B).

1.3. Evaluation of the effectiveness of biochar-based biofertilizers

1.4.1 Evaluation of the effectiveness of biochar-based biofertilizers on Pea Plant

a) Stem hight

The analysis of variance results, illustrated in the table below, reveal significant differences ($P \leq 0.05$) for biofertilizer concentrations, indicating that the concentrations have a significant effect on the stem height of pea plants. There is no significant effect ($P > 0.05$) for the types of biofertilizers, meaning these biofertilizers did not influence the stem height of pea plants.

Table 10 : Variance nalysis of stem height of pea plants

Sources of variation	SS	Df	MS	F	Sig.
Biofertilizers	58.07	1	58.07	1.664	0,211 ns
Concentrations	321.774	2	160.887	4.611	0,022*

The graph in the figure (23) illustrates the stem height of pea plants under the influence of two types of biofertilizers (solid and liquid) and three treatment concentrations (0.5%, 1%, 2%), along with a control at the 81st day of planting.

Notably, the control exhibits the greatest stem height compared to the two types of biofertilizers (45 cm). Both the solid and liquid biofertilizers, with the exception of the 2% liquid biofertilizer, show similar results, with stem heights ranging from 30 cm to 34.33 cm. The liquid biofertilizer at 2% results had the shortest stem height compared to the other treatments (20.67 cm).

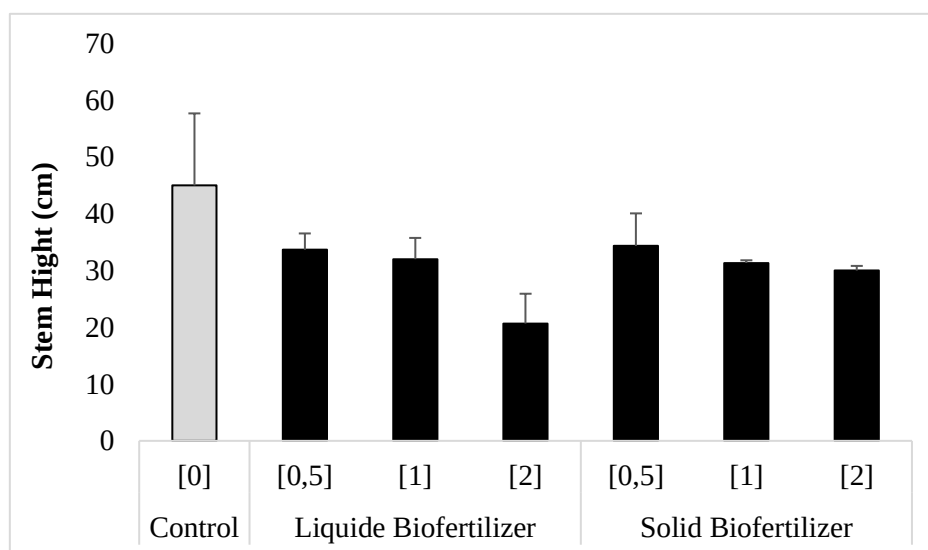


Figure 23: Variation in stem height of pea plants under the influence of two types of biofertilizers (solid and liquid) and three treatment concentrations (0.5%, 1%, 2%), along with a control.

The comparison of means shows significant differences in stem height under various biofertilizer conditions and concentrations. The groups of biofertilizers indicate that the stem height is highest under control conditions (45 cm). Among the tested biofertilizers, the solid biofertilizer (31.89 cm) had a higher stem height than the liquid biofertilizer (28.78 cm).

Regarding concentrations, the 0.5% concentration achieves the highest stem height (34 cm) compared to other concentrations tested. The 1% concentration (31.67 cm) shows a higher stem height than the 2% concentration (25.33 cm) (Fig. 23).

b) Leaves number

The results of the analysis of variance (table 11) reveal a significant difference ($P \leq 0.05$) between biofertilizer concentrations, indicating a notable effect of concentrations on the number of leaves in pea plants. However, there is no significant effect ($P > 0.05$) for the types of biofertilizers, suggesting that these did influence the number of leaves per plant.

Table 11 : Variance analysis of number of leaves per plant.

Sources of variation	SS	df	MS	F	Sig.
Biofertilizers	29.63	1	29.63	0.836	0,371ns
Concentrations	245.484	2	122.742	3.461	0,05*

The graph in the figure (24) illustrates the number of leaves on pea plants under the influence of two types of biofertilizers (solid and liquid) and three treatment concentrations (0.5%, 1%, 2%), along with a control on the 81st day.

Significantly, the control exhibits the highest number of leaves per plant compared to both types of biofertilizers (51 leaves). Both solid and liquid biofertilizers, except for the 2% liquid biofertilizer show similar results, with leaf numbers ranging between 25 and 37.67 leaves per plant. The liquid biofertilizer at 2% displays the lowest number of leaves compared to other types (21 leaves).

The comparison of means reveals significant differences in leaf numbers under different biofertilizer conditions and concentrations. Biofertilizer groups indicate that the highest number of leaves occurs under control conditions (51 leaves per plant). Among the tested biofertilizers, the liquid biofertilizer (31.556 leaves per plant) shows a higher number of leaves compared to the solid biofertilizer (29.333 leaves per plant).

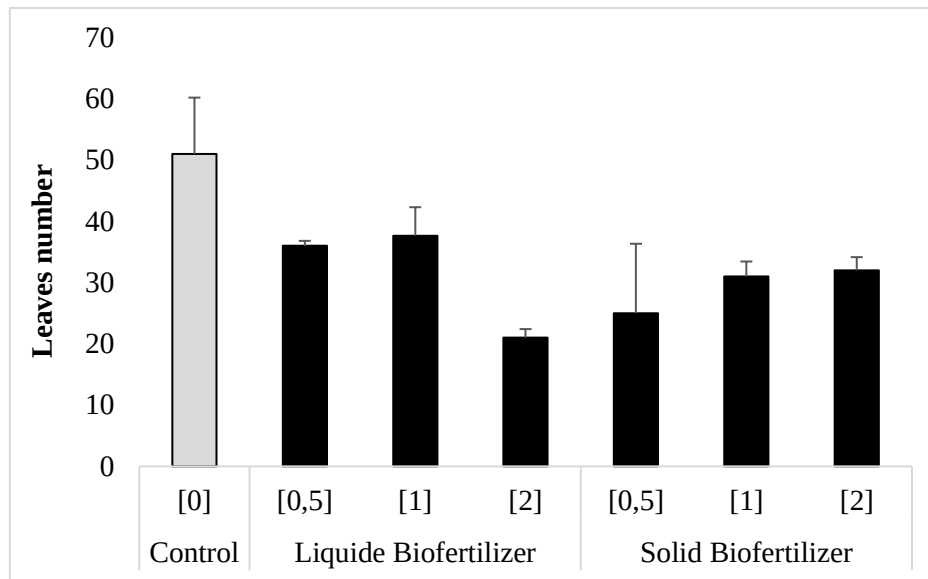


Figure 24: Variation in number of leaves of pea plants under the influence of two types of biofertilizers (solid and liquid) and three treatment concentrations (0.5%, 1%, 2%).

Regarding concentrations, the 1% concentration achieves a higher number of leaves (34.333 leaves per plant) compared to the other tested concentrations. The 0.5% concentration (30.5 leaves per plant) had a higher number of leaves than the 2% concentration (26.500).

c) *Branch number*

The results of the analysis of variance (table 12) indicate significant differences ($P \leq 0.05$) between both biofertilizers forms and biofertilizer concentrations, suggesting that these factors have a notable effect on the number of branches in pea plants.

Table 12 : Variance analysis of number Branches per plant.

Sources of variation	SS	df	MS	F	Sig.
Biofertilizers	24	1	24	5.6	0,028*
Concentrations	35.705	2	17.852	4.166	0,03*

The graph in the figure (25) illustrates the number of branches of pea plants under the influence of two types of biofertilizers (solid and liquid) and three concentrations (0.5%, 1%, 2%), along with a control on the 81st day after planting.

Significantly, the control exhibits the highest number of pea plant branches compared to both types of biofertilizers (14 branches per plant). Both solid and liquid biofertilizers show close results, with branch numbers ranging between 5.33 and 9.67 branches per plant. The liquid biofertilizer shows slightly higher branch numbers than the solid biofertilizer.

The comparison of means reveals significant differences in branch numbers under different biofertilizer conditions and concentrations. Biofertilizer groups indicate that the highest number of branches occurs under control conditions (14 branches per plant). Among the tested biofertilizers, the liquid biofertilizer (8.222 branches per plant) shows a higher number of branches than the solid biofertilizer (6.222 branches per plant).

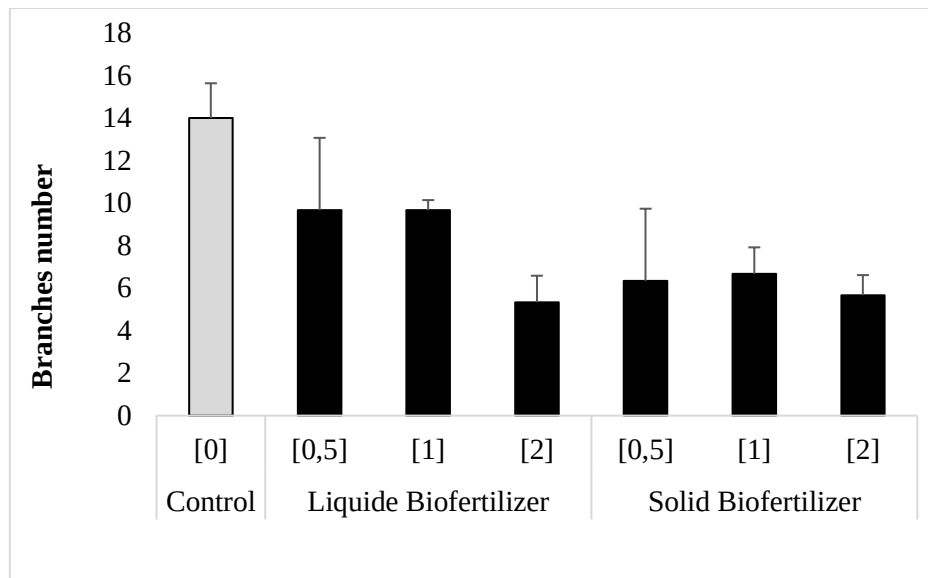


Figure 25: Variation in number of branches on pea plants under the influence of two types of biofertilizers (solid and liquid) and three treatment concentrations (0.5%, 1%, 2%).

Regarding concentrations, the 1% concentration achieves a higher number of branches (8.167 branches per plant) compared to other concentrations tested. The 0.5% concentration (8.000) results in a higher number of branches than the 2% concentration (5.5 branches per plant), while the control exhibits the highest number of branches (14 branches per plant).

2. Discussion

The performances and mechanisms of biochar in improving soil fertility can be used as a source of nutrients to increase soil fertility, improve the soil's physical and chemical properties, store nutrients, and act as a slow-release fertilizer, enhancing soil biological properties (Ding *et al.*, 2016).

In our study, we observed that biochar produced at 300°C for 3 hours showed the highest yield percentage compared to other types. The more the pyrolysis' temprature and duration increase, the more the biochar yield decrease. This result is in agreement with the work of Demirbas (2004) and Zhang *et al.*, (2019) who recorded that the yields of straw biochars showed a steadier decrease as the pyrolysis temperature continued increasing. (Demirbas, 2004). These results confirmed that the increase in temperature enhanced the stability of biochar and the loss of volatile fractions (Zornoza *et al.*, 2015).

To determine the potential impact of toxic substances and salt stress, this study focused on the effect of sawdust biochar, produced in several pyrolysis temperature (300°C, 400°C, and 500°C) and duration (3h, 4h and 5h) on seed germination tests, particularly using barley as resistant plante. The tests were conducted on biochar-based macerate and sand using several concentrations of biochar (0%, 0,5%, 1%, 2%, 4% and 8%).

For both culture substrate, in the first test experiment, the germination of barley was delayed after 20h but recovered within 68 h. These results correspond to those of (Bargmann et al. (2013) who performed germination tests with barely. At an application rate of 10 %, the germination was delayed within the first week in some treatments but recovered within 14 days whereas biochar had no effect, and we observed similar barley plant growth across different treatments. Barley is indeed sensitive to various carbon sources and soil conditions, and this sensitivity can impact its growth and development (Boufenar & Zaghouan, 2006).

Based on our results, it is clear that biochar shows a positive effect on the growth parameters of barley in both culture substrates. There was no particular effect of temperatures and the duration of pyrolysis on the growth parameters of barley.

The obtained results align with previous studies demonstrating the beneficial effects of biochar on plant growth. Carter *et al.* (2013) showed that biochar application significantly increased various plant growth parameters and root morphological characteristics of crops compared to the control group. Additionally, studies such as those of Uzoma *et al.* (2011) and Usman *et al.* (2016) reported an increased in growth, yield, and nutrient uptake of corn and

tomato plants, respectively, with biochar application. no negative effect of peanut hull biochar on the germination of barley was reported by Busch *et al.* (2012).

The fresh and dry weight of barley plants was significantly affected by biochar application, both weights increased compared to the control. The increase in shoot fresh and dry weights caused by biochar application can be attributed to a decrease in the toxic element by the pyrolysis process. Previous studies reported improved plant growth performance with biochar application (Naeem et al., 2017).

The results obtained from the two phytotoxicity tests revealed no negative effects of sawdust biochar on germination, providing a preliminary indication that it could be safely used for agriculture.

While sand plays a crucial role in providing drainage for certain crops, macerate biochar stands out as a superior soil amendment due to its unique ability to nutrients effectively. This property not only enhances plant growth but also improves overall soil health by fostering beneficial microbial activity and enhancing nutrient availability over time. (Lehmann and Joseph, 2009).

Based on our phytotoxicity results, we formulated two types of biofertilizer: a solid biochar and a liquid-based fertilizers.

Solid biochar-based fertilizer was made at 300°C for 3 hours. Liquid biofertilizer was produced from solid biochar in distilled water at room temperature for 24 hours, with three concentrations (0.5%, 1%, and 2%).

Analysis of variance did not show significant differences between the two types of biofertilizers. They had close effects and were better than unfertilized barley plants, but fewer unfertilized pea plants, which is inconsistent with previous studies.

According to Berihun *et al.* (2017), biochar application did not decrease germination. This is due to the fact that biochar has a certain degree of adsorption and contains a certain level of mineral elements of soils, which may provide nutrients for seed germination. Similarly, Agboola and Moses (2015) reported that biochar's sorptive capacity for allelochemicals may increase plant germination.

The number of leaves per plant increased with increasing rates of biochar and cow dung. Kamara *et al.* (2014) stated that the number of maize or rice seeds germinated on biochar-treated soils was higher than the control.

Roots serve as the interfaces between biochar particles and growing plants. The application of biochar can influence root growth and characteristics, potentially impacting overall plant performance (Xiang *et al.*, 2017). Therefore, in appropriate use of biochar may have an effect opposite to that anticipated (Zimmerman, 2011).

The difference in the growth results of barley and peas is due to the difference in the lower part of the plants. Roots are taproot to the pea and fibrous root to the barley (Yong, 1995; Pavek, 2012). The biochar can absorb within the pore structure and between particles significant amounts of water and dissolved nutrients (Conte & Schmidt., 2017). The high water retention in biochar treatments could be due to the highly porous nature of the biochar (Shaheen and Bukhari, 2017), resulting in water deficiency in the pea roots and then they can't absorb the water.

We concluded that biochar gradually improves soil health, leading to enhanced long-term plant growth. The macerate biochar had rapid response in plant growth and health due to the rapid availability of nutrients. Both biofertilizers have a positive effect on improving the growth.

Conclusion

Conclusion

Biofertilizer refers to substances with living microorganisms promoting plant growth by enhancing nutrient supply and controlling plant pathogens. They mobilize nutrients, restore soil health, and boost plant growth sustainability, they are considered a sustainable alternative to chemical fertilizers, contributing to more environmentally friendly agriculture.

Agricultural activities generate various types of waste, such as rice husk, straw, and sawdust. These wastes are notable for their ability to restore soil quality and retain nutrients. It can be processed into biochar which is a product of biomass pyrolysis, also recognized for its agricultural benefits. It emerges as a promising solution by enhancing soil quality, retaining nutrients, improving water retention, and promoting microbial activity. This sustainable alternative aims to mitigate the negative impacts of traditional fertilizers and pesticides on both the environment and human health.

Our project focused on producing biochar from sawdust using various pyrolysis temperatures and durations, assessing the phytotoxicity of the produced biochar to select the best one, formulating two types of biofertilizer (liquid and dry) based on biochar, and evaluating these biofertilizers on barley and peas.

The techniques used in our study included producing biochar, assessing the phytotoxicity of biochar using barley germination tests in agar biochar macerate, followed by tests in sand and sand-biochar mix. Solid biochar-based fertilizer was made at 300°C for 3 hours to avoid negative effects on germination and growth, while liquid biofertilizer was produced by mixing solid biochar in distilled water at room temperature for 24 hours, using three different concentrations. We prepared soil substrates with various biochar concentrations, inoculated seeds with biofertilizers, and monitored germination and plant growth parameters.

Germination results showed a significant improvement in germination rates. Seeds germinated faster and more uniformly, indicating a more favorable soil environment.

Growth parameters such as plant height, biomass, and root development of barley seedlings all benefited from the application of biochar and biofertilizers. Plants showed more vigorous growth.

It is important to note that no signs of phytotoxicity were observed in the treated plants. This confirms that the use of biochar and biofertilizers at appropriate concentrations is safe for crops.

The second part of the study aimed to specifically evaluate the effect of the based biochar biofertilizers on the growth of pea (*Pisum sativum*). The pea variety Onward, originating from France, was obtained from an agricultural supplies store in Tiaret.

The addition of biochar led to a significant increase in both plants growth. Biofertilizers further enhanced this improvement. Growth parameters such as plant height and total dry weight all showed notable improvements. This may be due to the improvement of water retention and nutrient availability in the soil.

In conclusion, the combined use of biochar and biofertilizers offers considerable potential for improving agricultural productivity sustainably, without the risk of phytotoxicity for the studied crops.

Overall, this study provides valuable insights into the positive effects of biochar on soil and plant health, suggesting its potential as a sustainable agricultural practice.

Given the diversity of effects that biochar may induce in soil, guidelines for future biochar use should adopt a structured and holistic approach that considers all positive and negative effects of biochar.

We recommended as perspective for further study:

- ✓ Studying the long-term effects of biochar on soil properties and ecosystems is crucial for understanding its full potential.
- ✓ Examining its role in carbon sequestration and its potential to mitigate greenhouse gas emissions.
- ✓ Determining the optimal application rates and investigating different methods of applying biochar for various soil types and crops is necessary.
- ✓ Studying the differences in biochar produced from various biomass materials is important.
- ✓ Investigating biochar's potential to immobilize heavy metals and other contaminants in soil is vital.
- ✓ Exploring the integration of biochar with other organisms, such as beneficial microbes and fungi, can enhance its positive effects on soil and plant health.
- ✓ When incorporated into the soil, biochar acts like a sponge, capable of holding onto water and nutrients due to its porous structure. Further studies on this aspect can help optimize water usage in agriculture, particularly in arid and semi-arid regions.

This study should also be compared with other studies on the same subject and completed by the evaluation of C/N ratio in soil and the evaluation of the physicochemical proprieties of the set biochars.

Overall, this study provides valuable insights into the positive effects of biochar on soil and plant health, suggesting its potential as a sustainable agricultural practice.

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Abstract

Biofertilizers enhance plant growth and promote sustainable agriculture. Agricultural waste, can be processed into biochar through biomass pyrolysis.

This project produced biochar from sawdust at various pyrolysis temperatures (300, 400, and 500°C) and durations (3, 4, and 5h), assessed biochar phytotoxicity on barley seedling. Biochar produced at 300°C for 3 hours was used to formulate two biofertilizer solid and liquid. The effectiveness of these biofertilizer was tested on barley and peas.

Biochar significantly improved germination rates and plant growth parameters without showing phytotoxicity. Both biofertilizers showed an improvement of growth of barley and pea due to enhanced water retention and nutrient availability from biochar and biofertilizers. Barley showed more improved growth parameters compared to pea. Overall, the use of biochar as biofertilizers offers sustainable improvements in agricultural productivity.

In conclusion, biochar presents a promising sustainable agricultural practice that can be used for enhancing soil and plant health.

Keywords: Biochar, Biofertilizers, Barley, Pea, Plant Growth, phytotoxic, liquid biofertilizer, solid biofertilizer.

المخلص

تعزز الأسمدة الحيوية نمو النباتات وتعزز الزراعة المستدامة. يمكن معالجة النفايات الزراعية وتحويلها إلى فحم حيوي من خلال الانحلال الحراري للكتلة الحيوية.

تم من خلال هذا المشروع إنتاج الفحم الحيوي من نشارة الخشب في درجات حرارة مختلفة للتحلل الحراري (300 و 400 و 500 درجة مئوية) ومدد مختلفة (3 و 4 و 5 ساعات)، وتم تقييم السمية النباتية للفحم الحيوي على شتلات الشعير، كما تم صياغة نوعين من السماد الحيوي (السائل والصلب). تم استخدام الفحم الحيوي المنتج عند درجة حرارة 300 درجة مئوية لمدة 3 ساعات لصياغة نوعين من السماد الحيوي الصلب والسائل. تم اختبار فعالية هذه الأسمدة الحيوية على الشعير والبازلاء.

لقد أدى الفحم الحيوي إلى تحسين معدلات الإنبات ونمو النبات دون أن يظهر سمية نباتية. وأظهر كلا المخصبين الحيويين تحسناً في نمو الشعير والبازلاء بسبب تعزيز احتباس الماء وتوافر المغذيات من الفحم الحيوي والمخصبين الحيويين. وأظهر الشعير نتائج أفضل في معايير النمو مقارنة بالبازلاء. وبشكل عام، يوفر استخدام الفحم الحيوي كمخصبات حيوية تحسينات مستدامة في الإنتاجية الزراعية.

وفي الختام، يقدم الفحم الحيوي ممارسة زراعية مستدامة واعدة يمكن استخدامها لتعزيز صحة التربة والنبات.

الكلمات المفتاحية: الفحم الحيوي، والأسمدة الحيوية، والشعير، والبازلاء، ونمو النبات، والسماد الحيوي السائل، والسماد الحيوي الصلب.

Résumé

Les biofertilisants améliorent la croissance des plantes et favorisent l'agriculture durable. Les déchets agricoles peuvent être transformés en biochar par pyrolyse de la biomasse.

Ce projet a permis de produire du biochar à partir de sciure de bois à différentes températures de pyrolyse (300, 400 et 500°C) et pendant différentes durées (3, 4 et 5 heures), et d'évaluer la phytotoxicité du biochar sur l'orge. Le biochar produit à 300°C pendant 3 heures a été utilisé pour formuler deux biofertilisants solide et liquide. L'efficacité de ces biofertilisants a été testée sur l'orge et le petit pois.

Le biochar a amélioré de manière significative les taux de germination et les paramètres de croissance des plantes sans montrer de phytotoxicité. Les deux biofertilisants ont montré une amélioration de la croissance de l'orge et des pois grâce à une meilleure rétention de l'eau et à la disponibilité des nutriments du biochar et des biofertilisants. Les paramètres de croissance de l'orge se sont améliorés davantage que ceux du pois. Dans l'ensemble, l'utilisation du biochar et des biofertilisants permet d'améliorer durablement la productivité agricole.

En conclusion, le biochar est une pratique agricole durable prometteuse qui peut être utilisée pour améliorer les sols et les potentialités des plantes.

Mots-clés : Biochar, biofertilisants, orge, pois, croissance des plantes, phytotoxique, biofertilisant liquide, biofertilisant solide.