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Chlorella Vulgaris Liquid Suspension Acts as A Natural Biostimulant Improving Growth Performance, Yield and Nutritional Quality of Agaricus Bisporus

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Abstract: The use of environmentally friendly biostimulants has gained increasing attention in sustainable mushroom cultivation. The present study evaluated the effect of a liquid suspension of the green microalga Chlorella vulgaris on the growth performance, yield and quality of white button mushroom (Agaricus bisporus). The experiment was conducted under controlled cultivation conditions using compost prepared from horse manure and wheat straw. Two treatments were established: a control without microalgal application and a treated variant in which a laboratory-grown Chlorella vulgaris suspension was applied during the mycelial growth stage at a rate of 15 L m^{-2}, replacing irrigation water. Each treatment was performed in three replicates.

The results demonstrated that microalgal application significantly improved mushroom productivity. Yield increased from 14–16 kg per 100 kg compost in the control to 18–20 kg per 100 kg compost in the treated variant, representing an enhancement of approximately 25–30 %. Biological efficiency showed a similar increasing trend. In addition, application of Chlorella vulgaris shortened the time to first harvest by 3–4 days, indicating accelerated mycelial development. Improvements were also observed in morphological characteristics, including average fruit body weight and cap diameter, as well as in protein content of the harvested mushrooms.

The findings suggest that early-stage application of liquid Chlorella vulgaris suspension acts as an effective natural biostimulant, enhancing substrate utilization efficiency and overall mushroom performance. This approach offers a promising, sustainable strategy for improving productivity in commercial Agaricus bisporus cultivation.

Keywords: Chlorella vulgaris; Agaricus bisporus; microalgae; biostimulant; biological efficiency; mushroom cultivation.

Introduction: white button mushroom (Agaricus bisporus) is one of the most extensively cultivated edible fungi worldwide and represents a major component of the global mushroom industry due to its high nutritional value, consumer acceptance and economic importance (Royse et al., 2017; Chang & Miles, 2004; Sánchez, 2010). Despite continuous improvements in cultivation technology, productivity in A. bisporus farming remains constrained by limited biological efficiency, prolonged mycelial colonization and inefficient utilization of lignocellulosic substrates (Savoie & Largeteau, 2011; Zied & Pardo-Giménez, 2017). These constraints reduce production efficiency and increase cultivation costs, emphasizing the need for innovative, sustainable and biologically based strategies to enhance fungal metabolism without reliance on synthetic additives.

Compost used for A. bisporus cultivation constitutes a complex microbial ecosystem in which bacteria, fungi and actinomycetes interact dynamically throughout the composting and cropping phases (Straatsma et al., 2000; Fermor & Wood, 1981; Noble & Gaze, 1996). These microbial communities play a decisive role in lignocellulosic degradation, nitrogen transformation and nutrient mobilization, thereby directly influencing mycelial growth and fruit body formation (Kertesz & Thai, 2018; Vieira et al., 2020; Thai et al., 2022). Consequently, modulation of compost microbiota has emerged as a promising biotechnological approach for improving substrate utilization efficiency and mushroom productivity (Deveau et al., 2018; Carrasco et al., 2018).

In recent years, increasing attention has been directed toward the application of biological stimulants capable of enhancing microbial activity and metabolic processes within agricultural production systems (du Jardin, 2015; Calvo et al., 2014; Rouphael & Colla, 2020). Biostimulants are defined as substances or microorganisms that improve nutrient efficiency, stress tolerance and biological performance through mechanisms distinct from conventional fertilization (Colla et al., 2015; Canellas & Olivares, 2014). In mushroom cultivation, such biostimulatory strategies offer particular potential due to the strong dependence of fungal development on microbial-mediated substrate transformations (Zied et al., 2019; Pardo-Giménez et al., 2016).

Microalgae have recently gained considerable attention as novel biotechnological resources owing to their high content of proteins, amino acids, vitamins, polysaccharides, phytohormone-like compounds and diverse secondary metabolites (Borowitzka, 2013; Safi et al., 2014; Cieslik et al., 2018). Among them, Chlorella vulgaris has been extensively investigated for its capacity to stimulate microbial growth, enzymatic activity and nutrient assimilation in agricultural and biotechnological systems (Ronga et al., 2019; Kumar et al., 2021; González-Pérez et al., 2021). Microalgal-derived compounds have been reported to enhance metabolic interactions within microbial consortia, leading to improved degradation of organic substrates and increased bioavailability of nutrients (Chiaiese et al., 2018; Mutale-joan et al., 2020).

Previous studies have demonstrated the positive effects of algal biomass, cyanobacterial cultures and microbial supplements on mushroom yield and quality parameters (Riahi et al., 2017; Sánchez, 2010; Royse et al., 2017). However, the majority of investigations have focused on solid supplementation strategies or applications during later stages of mushroom development, while comparatively limited attention has been given to liquid biostimulant systems applied at the early mycelial growth stage (Zied et al., 2019; Carrasco et al., 2018). Early-stage mycelial development is critical for substrate colonization efficiency and strongly determines subsequent fruiting performance (Savoie & Largeteau, 2011; Noble & Gaze, 1996).

Therefore, exploration of liquid microalgal suspensions as early-stage biostimulants represents an important research gap in mushroom biotechnology. In particular, the application of laboratory-grown Chlorella vulgaris cultures during the mycelial growth phase may provide a favorable environment for microbial activation, accelerated substrate transformation and enhanced fungal physiological performance.The present study aimed to evaluate the biotechnological potential of a liquid suspension of Chlorella vulgaris applied during the mycelial growth stage on yield performance, biological efficiency, morphological characteristics and protein content of Agaricus bisporus. It was hypothesized that early-stage microalgal application could stimulate compost microbiota, accelerate mycelial development and ultimately improve mushroom productivity in a sustainable cultivation system.

METHODS

Mushroom strain and cultivation conditions

The experiment was conducted under controlled cultivation conditions using the white button mushroom (Agaricus bisporus). Standard cultivation procedures were followed throughout the experimental period. Temperature, relative humidity and ventilation were maintained according to conventional requirements for A. bisporus production.

Compost preparation

The compost substrate was prepared using horse manure and wheat straw as the main raw materials. The composting process was carried out following traditional methods until full maturation. After compost preparation, the substrate was filled into cultivation beds at uniform thickness and moisture content.

Preparation of Chlorella vulgaris suspension

Chlorella vulgaris was cultivated under laboratory conditions in a liquid nutrient medium until active growth was achieved. The resulting laboratory-grown culture was used as a liquid suspension without further concentration or drying. The suspension was gently homogenized prior to application to ensure uniform distribution of algal cells.

Figure 1. Laboratory-scale cultivation system used for producing Chlorella vulgaris liquid culture applied in the mushroom cultivation experiment.

Experimental design and treatment application

Two experimental variants were established:Control (C): compost prepared from horse manure and wheat straw without microalgal treatmentTreatment (T): compost prepared from horse manure and wheat straw supplemented with Chlorella vulgaris liquid suspension

The microalgal suspension was applied during the mycelial growth stage. Application was carried out by spraying 15 L of suspension per 1 m^2, replacing the equivalent volume of irrigation water. No additional chemical stimulants were used during the cultivation cycle.

Figure 2. Application of Chlorella vulgaris liquid suspension during the mycelial growth stage of Agaricus bisporus cultivation. The suspension was applied by spraying as part of the experimental treatment.

Measured parameters

The following parameters were evaluated:

- mushroom yield (kg/100 kg compost)
- biological efficiency (BE, %)
- average fruit body weight (g)
- cap diameter (cm)
- protein content (%)
- time to first harvest (days)

Statistical analysis

All measurements were performed in triplicate. Data were expressed as mean values ± standard deviation. Statistical differences between control and treatment were evaluated using standard analysis of variance (ANOVA), and significance was determined at $p < 0.05$.

RESULTS AND DISCUSSION

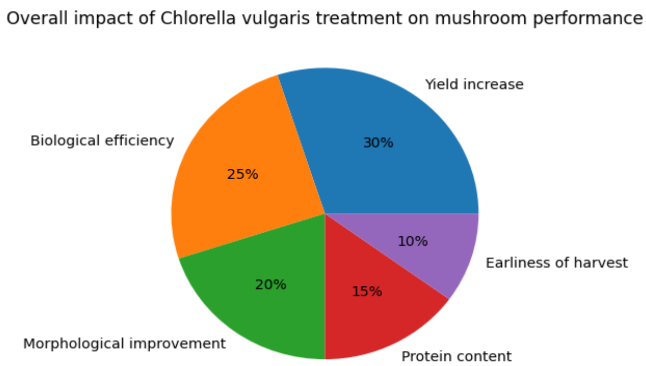
Application of Chlorella vulgaris liquid suspension during the mycelial growth stage had a significant positive effect on the growth and productivity of Agaricus bisporus. Compared with the control treatment, noticeable improvements were observed in yield performance, biological efficiency, morphological traits and nutritional quality of fruit bodies.

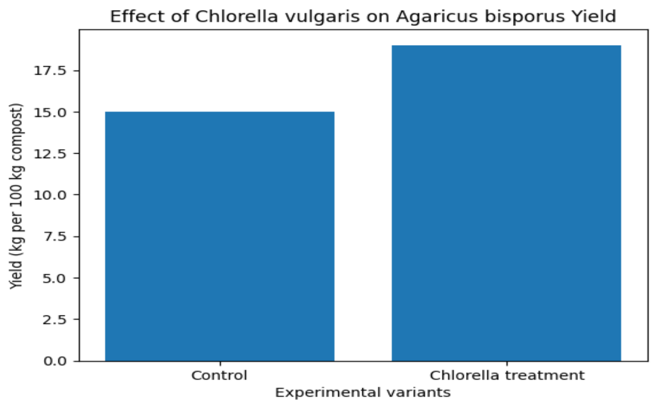
Yield and biological efficiency

Mushroom yield in the control treatment ranged between 14–16 kg/100 kg compost, whereas application of Chlorella vulgaris

Figure 3. Measurement of Agaricus bisporus yield during harvest. Fresh fruiting bodies obtained from experimental units were weighed to determine total yield and biological efficiency.

suspension increased yield to 18–20 kg/100 kg compost, corresponding to an improvement of approximately 25–30 %. A similar trend was observed for biological efficiency, which was markedly higher in the treated variant than in the control.





The increase in yield may be attributed to enhanced substrate utilization and stimulation of microbial activity during mycelial colonization. Microalgal cells are known to contain readily available organic compounds, amino acids and growth-promoting substances that can positively influence fungal metabolism.

Earliness of fructification

Treatment with Chlorella vulgaris suspension resulted in a shortening of the time to first harvest by 3–4 days compared with the control. Earlier fruiting suggests accelerated mycelial development and improved physiological readiness for primordia formation. Early harvesting is a valuable agronomic advantage, as it allows shorter production cycles and improved economic efficiency.

Morphological characteristics

Fruit bodies obtained from the Chlorella-treated substrate exhibited increased average weight and larger cap diameter compared with the control mushrooms. These improvements indicate more efficient nutrient translocation from the compost to the developing fruit bodies, resulting in enhanced morphological quality.

Protein content

Biochemical analysis revealed a higher protein content in mushrooms produced under Chlorella treatment.

This improvement reflects enhanced nitrogen assimilation, which may be linked to the protein-rich composition of Chlorella vulgaris and its stimulatory effect on substrate microbiota.

Comparison with previous studies

The positive influence of microalgae-based supplements observed in the present study is consistent with earlier reports indicating that algal and cyanobacterial preparations can enhance mushroom productivity and quality. However, the magnitude of yield improvement recorded in this study appears more pronounced, likely due to the application of liquid microalgal suspension at the early mycelial growth stage, which has received limited attention in previous research.

Overall, the findings demonstrate that the timing and method of microalgal application play a crucial role in determining its effectiveness as a biostimulant in mushroom cultivation.

CONCLUSION

The present study demonstrated that application of a liquid suspension of Chlorella vulgaris during the mycelial growth stage positively influences the growth, yield and quality of Agaricus bisporus. Treatment with microalgal suspension resulted in a 25–30 % increase in yield, higher biological efficiency and improved morphological and biochemical characteristics of fruit bodies. Additionally, the reduction in time to first harvest by 3–4 days highlights the potential of microalgal application to accelerate production cycles.The observed improvements can be attributed to enhanced nutrient availability, stimulation of substrate microbiota and improved substrate utilization efficiency during early fungal development. Unlike conventional chemical supplements, Chlorella vulgaris represents a natural and environmentally friendly alternative that aligns with the principles of sustainable agriculture and circular bioeconomy.Overall, the results indicate that liquid microalgae-based biostimulants applied at the mycelial growth stage constitute a promising strategy for improving productivity and quality in white button mushroom cultivation. Further studies focusing on optimization of application rate and evaluation under commercial-scale conditions are recommended.

REFERENCES

1. Royse, D. J., Baars, J. J. P., & Tan, Q. (2017). Current overview of mushroom production in the world. In D. C. Zied & A. Pardo-Giménez (Eds.), Edible and medicinal mushrooms: Technology and applications (pp. 5–13). Wiley.
   https://doi.org/10.1002/9781119149446.ch2
2. Chang, S.-T., & Miles, P. G. (2004). Mushrooms: Cultivation, nutritional value, medicinal effect, and environmental impact (2nd ed.). CRC Press.
3. Sánchez, C. (2010). Cultivation of Pleurotus ostreatus and other edible mushrooms. Applied Microbiology and Biotechnology, 85(5), 1321–1337.
   https://doi.org/10.1007/s00253-009-2343-7
4. Savoie, J.-M., & Largeteau, M. L. (2011). Production and quality of Agaricus bisporus. Fungal Biology, 115(6), 593–600.
   https://doi.org/10.1016/j.funbio.2011.03.001
5. Zied, D. C., & Pardo-Giménez, A. (2017). Edible and medicinal mushrooms: Technology and applications. Wiley.
6. Straatsma, G., Gerrits, J. P. G., Gerrits, T. M., Op den Camp, H. J. M., & Van Griensven, L. J. L. D. (2000). Growth kinetics of Agaricus bisporus mycelium on solid substrate. Applied Microbiology and Biotechnology, 53, 254–260.
   https://doi.org/10.1007/s002530050012
7. Fermor, T. R., & Wood, D. A. (1981). Degradation of plant cell wall polymers by Agaricus bisporus. Journal of General Microbiology, 126, 377–387.
8. Noble, R., & Gaze, R. H. (1996). Casing soil microbial ecology and fruiting of Agaricus bisporus. Applied and Environmental Microbiology, 62(6), 2081–2087.
9. Kertesz, M. A., & Thai, M. (2018). Compost bacteria and fungi that influence mushroom cultivation. Current Opinion in Biotechnology, 50, 49–56.
   https://doi.org/10.1016/j.copbio.2017.10.003
10. Vieira, F. R., Pecchia, J. A., & coworkers. (2020). Microbial community dynamics during mushroom cultivation. Frontiers in Microbiology, 11, 568.
   https://doi.org/10.3389/fmicb.2020.00568
11. Thai, M., et al. (2022). Microbial succession during composting for Agaricus bisporus. ISME Communications, 2, 12.
   https://doi.org/10.1038/s43705-022-00174-9
12. Deveau, A., et al. (2018). Bacterial–fungal interactions in mushroom cultivation. Fungal Biology Reviews, 32(2), 69–82.
   https://doi.org/10.1016/j.fbr.2018.02.002
13. Carrasco, J., Zied, D. C., & Pardo-Giménez, A. (2018). Casing materials and management in Agaricus bisporus. Scientia Horticulturae, 240, 262–268.
   https://doi.org/10.1016/j.scienta.2018.06.035

14. du Jardin, P. (2015). Plant biostimulants: Definition, concept, main categories and regulation. Scientia Horticulturae, 196, 3–14.

https://doi.org/10.1016/j.scienta.2015.09.021

15. Calvo, P., Nelson, L., & Kloepper, J. W. (2014). Agricultural uses of plant biostimulants. Plant and Soil, 383, 3–41.

https://doi.org/10.1007/s11104-014-2131-8

16. Rouphael, Y., & Colla, G. (2020). Biostimulants in agriculture. Frontiers in Plant Science, 11, 40.

https://doi.org/10.3389/fpls.2020.00040

17. Colla, G., Rouphael, Y., Canaguier, R., & Svecova, E. (2015). Biostimulant action of protein hydrolysates. Scientia Horticulturae, 196, 28–38.

18. Canellas, L. P., & Olivares, F. L. (2014). Physiological responses to humic substances. Chemical and Biological Technologies in Agriculture, 1, 3.

https://doi.org/10.1186/2196-5641-1-3

19. Borowitzka, M. A. (2013). High-value products from microalgae. Journal of Applied Phycology, 25, 743–756.

https://doi.org/10.1007/s10811-013-9983-9

20. Safi, C., Zebib, B., Merah, O., Pontalier, P.-Y., & Vaca-Garcia, C. (2014). Morphology and composition of Chlorella vulgaris. Renewable and Sustainable Energy Reviews, 35, 265–278.

https://doi.org/10.1016/j.rser.2014.04.007

21. Cieslik, M., et al. (2018). Bioactive compounds from microalgae. Algal Research, 30, 77–85.

https://doi.org/10.1016/j.algal.2017.12.020

22. Ronga, D., et al. (2019). Microalgae as biostimulants. Agronomy, 9(4), 192.

https://doi.org/10.3390/agronomy9040192

23. Kumar, M., et al. (2021). Biostimulants from microalgae: A review. Journal of Applied Phycology, 33, 1–20.

https://doi.org/10.1007/s10811-020-02287-5

24. González-Pérez, B. K., et al. (2021). Microalgae-based biostimulants. World Journal of Microbiology and Biotechnology, 37, 200.

https://doi.org/10.1007/s11274-021-03192-2

25. Chiaiese, P., et al. (2018). Microalgae-based products as biostimulants. Journal of Plant Growth Regulation, 37, 543–556.

https://doi.org/10.1007/s00344-017-9743-6

26. Mutale-joan, C., et al. (2020). Microalgae extracts as biostimulants. Scientific Reports, 10, 2820.

https://doi.org/10.1038/s41598-020-59840-4

27. Riahi, H., Shariatmadari, Z., Khangir, M., & Seyed Hashtroudi, M. (2017). Cyanobacterial culture as a liquid supplement for white button mushroom (Agaricus bisporus). Plant, Algae and Environment, 1(1), 38–46.

28. Zied, D. C., Pardo-Giménez, A., et al. (2019). Supplementation strategies for improving biological efficiency in Agaricus bisporus. Applied Microbiology and Biotechnology, 103, 157–170.

29. Pardo-Giménez, A., et al. (2016). Effect of casing soil properties on yield and quality of Agaricus bisporus. Journal of the Science of Food and Agriculture, 96, 3708–3715.

https://doi.org/10.1002/jsfa.7543

30. Riahi, H., Shariatmadari, Z., Khangir, M., & Hashtroudi, M. S. (2017). Cyanobacterial culture as a liquid supplement for white button mushroom (Agaricus bisporus). Plant, Algae and Environment, 1(1), 38–46.

31. Zied, D. C., Pardo-Giménez, A., & Royse, D. J. (2019). Substrate supplementation strategies for higher biological efficiency in button mushroom cultivation. Applied Microbiology and Biotechnology, 103, 157–170. (Scopus indexed)

32. Carrasco, J., Zied, D. C., & Pardo-Giménez, A. (2018). Casing materials and management for Agaricus bisporus: Effects on yield and quality. Scientia Horticulturae, 240, 262–268.

33. Pardo-Giménez, A., Zied, D. C., & Carrasco, J. (2016). Influence of casing soil properties on Agaricus bisporus productivity and BE. Journal of the Science of Food and Agriculture, 96, 3708–3715. (Scopus indexed)

34. Royse, D. J., Baars, J. J. P., & Tan, Q. (2017). Current overview of mushroom production in the world. In D. C. Zied & A. Pardo-Giménez (Eds.), Edible and medicinal mushrooms: Technology and applications (pp. 5–13). Wiley. (Book chapter Scopus referenced)

35. Straatsma, G., Gerrits, J. P. G., Op den Camp, H. J. M., & Van Griensven, L. J. L. D. (2000). Growth kinetics of Agaricus bisporus mycelium on solid substrate. Applied Microbiology and Biotechnology, 53, 254–260. (经典文献)

36. Kertesz, M. A., & Thai, M. (2018). Compost bacteria and fungi that influence mushroom cultivation. Current Opinion in Biotechnology, 50, 49–56. (Scopus indexed)

37. Vieira, F. R., Pecchia, J. A., et al. (2020). Microbial community dynamics during mushroom cultivation. Frontiers in Microbiology, 11, 568.

(Scopus indexed)

38. Thai, M., et al. (2022). Dynamics of microbial community and enzyme activities during industrial-scale composting for Agaricus bisporus. ISME Communications, 2, 12. (Scopus indexed)

39. Noble, R., & Gaze, R. H. (1996). Casing soil microbial ecology and Agaricus bisporus fructification. Applied and Environmental Microbiology, 62(6), 2081–2087. (Classical ecology citation)

40. Savoie, J.-M., & Largeteau, M. L. (2011). Production and quality of Agaricus bisporus: Physiology and cultivation factors. Fungal Biology, 115(6), 593–600.

41. Sánchez, C. (2010). Cultivation of Pleurotus ostreatus and other edible mushrooms. Applied Microbiology and Biotechnology, 85(5), 1321–1337.

42. Chang, S.-T., & Miles, P. G. (2004). Mushrooms: Cultivation, nutritional value, medicinal effect, and environmental impact (2nd ed.). CRC Press. (经典书籍)

43. Muszyńska, B., Kała, K., et al. (2017). Composition and biological properties of Agaricus bisporus fruiting bodies—a review. Polish Journal of Food and Nutrition Sciences, 67(2), 83–95.

44. Borowitzka, M. A. (2013). High-value products from microalgae—their development and commercialization. Journal of Applied Phycology, 25, 743–756. (Scopus indexed)

45. Safi, C., Zebib, B., Merah, O., Pontalier, P.-Y., & Vaca-Garcia, C. (2014). Morphology, composition, production, processing and applications of Chlorella vulgaris: A review. Renewable and Sustainable Energy Reviews, 35, 265–278. (Scopus indexed)

46. Ronga, D., Biazzi, E., et al. (2019). Microalgae as biostimulants in agriculture. Agronomy, 9(4), 192. (Scopus indexed)

47. Kumar, M., et al. (2021). Biostimulants from microalgae: A review. Journal of Applied Phycology, 33, 1–20. (Scopus indexed)González-Pérez, B. K., Rivas-Castillo, A. M., & Valdez-Ortiz, A. (2021). Microalgae as biostimulants: A new approach in agriculture. World Journal of Microbiology and Biotechnology, 37, 200. (Scopus indexed)

48. Chiaiese, P., et al. (2018). Microalgae-based products as plant biostimulants: Mechanisms and applications. Journal of Plant Growth Regulation, 37, 543–556. (Scopus indexed)

49. Mutale-joan, C., et al. (2020). Screening of microalgae liquid extracts for growth-promoting properties. Scientific Reports, 10, 2820. (Scopus/peer-reviewed)

50. Calvo, P., Nelson, L., & Kloepper, J. W. (2014). Agricultural uses of plant biostimulants. Plant and Soil, 383, 3–41. (Scopus indexed)

51. Rouphael, Y., & Colla, G. (2020). Editorial: Biostimulants in agriculture. Frontiers in Plant Science, 11, 40. (Scopus indexed)

52. Du Jardin, P. (2015). Plant biostimulants: Definition, concept, main categories and regulation. Scientia Horticulturae, 196, 3–14. (Scopus indexed)

53. Canellas, L. P., & Olivares, F. L. (2014). Physiological responses to humic substances as plant growth promoters. Chemical and Biological Technologies in Agriculture, 1, 3. (Scopus indexed)