

Abstract

# Effect of Humic Acids from Biomass Biostimulant on Microalgae Growth <sup>†</sup>

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**Abstract:** In this work, HAs were obtained by humification of lignin that was extracted from beer spent grain (BSG) obtained from beer fabrication with deep eutectic solvents (DESs). The humification process used in this case was reacting the BSG-extracted lignin with hydrogen peroxide in the presence of ferric sulphate heptahydrate. The biostimulant effect of HAs was tested on the *Chlorella Sorokiniana* microalgae species. Additionally, biostimulant tests were conducted using a commercial product containing HAs (BlackJak, BJK) and a coal-extracted lignosulphonate (LsNa).

**Keywords:** humic acids; lignin; microalgae; biostimulant effect



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## 1. Introduction

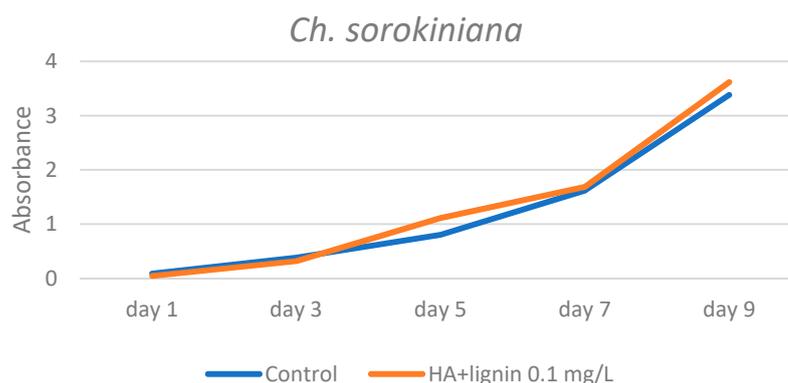
Humic substances (HSs) are formed by chemical and biological transformations of vegetal and animal biomass through microbial metabolism, representing a major organic carbon source at the soil’s surface. They contribute to the adjustment of many major ecological processes. For example, HSs enhance plant growth and terrestrial life in general, adjust carbon and nitrogen cycles in the soil, enhance plant and microorganism growth, improve the fate and transportation of anthropogenic compounds and heavy metals and stabilize the soil [1,2]. Scientists define humic acids (HAs) as humic materials that are soluble in aqueous alkaline solutions and that precipitate when the pH is brought to 1–2 [3].

## 2. Materials and Methods

For lignin extraction from BSG, various DESs were used. Humification of the extracted lignin was carried out through a reaction with hydrogen peroxide in the presence of ferric sulphate heptahydrate. Biostimulant tests were conducted at  $25 \pm 2$  °C, illuminating with a fluorescent light lamp at  $100 \mu\text{mol}/\text{m}^2 \cdot \text{s}$  ( $\mu\text{Einstein}$ ), with a light/darkness period of 14/10 h for 9 days up to 2 weeks. Parameters such as turbidity, optic density and chlorophyll content were studied. Concentrations of 10 mg/L and 1 mg/L in the case of reference products BJK and LSNa and of 1 mg/L in the case of the obtained HAs were used.

## 3. Results

Lignin was extracted with various yields. HAs were identified using FT-IR spectra. Has derived from lignin had the best biostimulant activity in the period of 9 days for the 0.1 mg/L concentration (Figure 1).



**Figure 1.** Optical density of *Ch. sorokiniana* in the presence of humic acids derived from lignin.

#### 4. Conclusions

The biostimulant effect of various HAs showed promising results in every case, and the HAs obtained from lignin had a better effect than the commercial product containing HAs after 9 days.

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