

Sewage sludge derived biochar accelerates toluene removal by *Pseudomonas plecoglossicida*

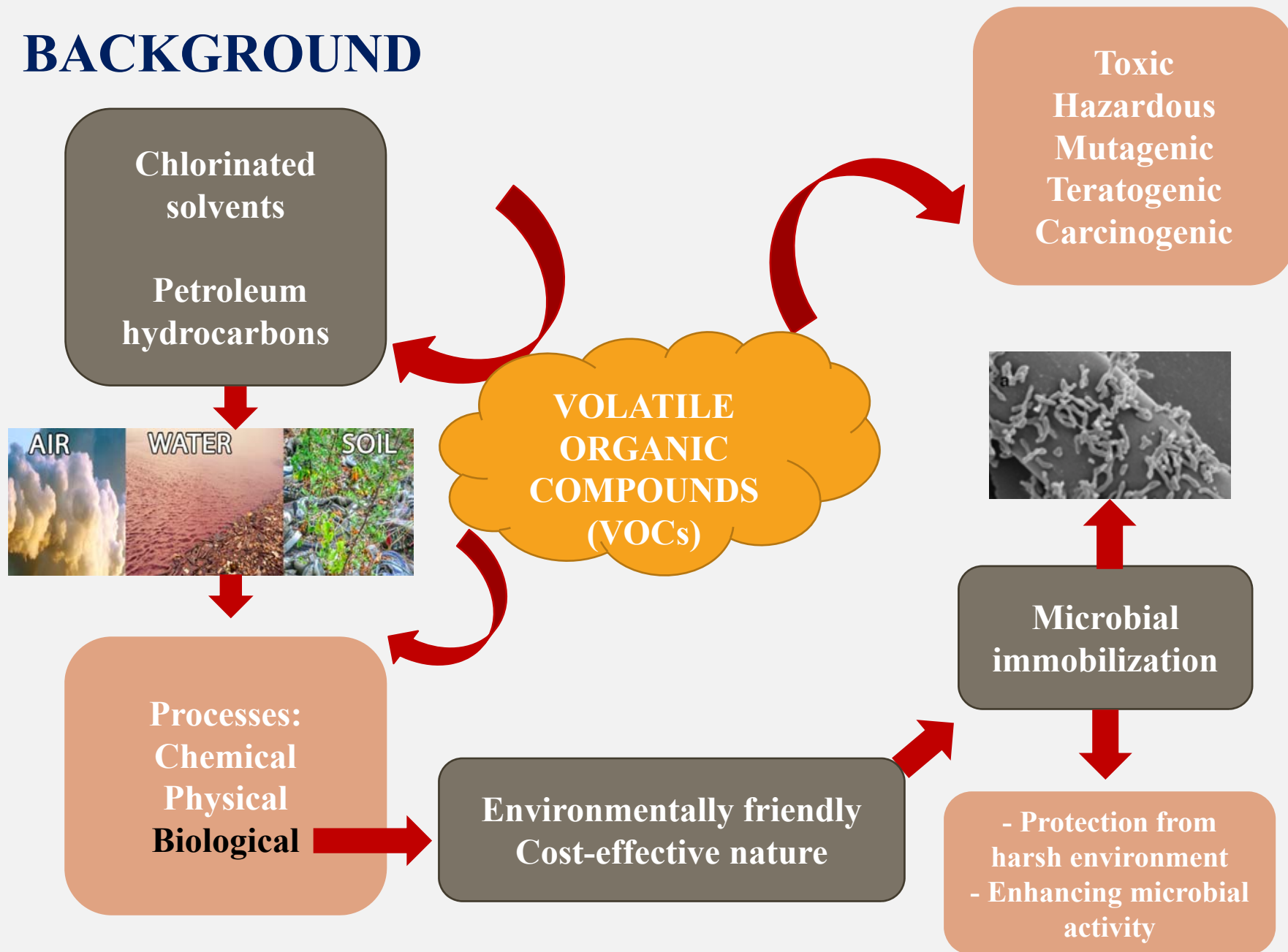
R.A. de Toledo¹, T.T. Shen^{1,2}, H. Shim¹

¹ Department of Civil and Environmental Engineering, Faculty of Science and Technology, University of Macau, Macau SAR, P.R. China

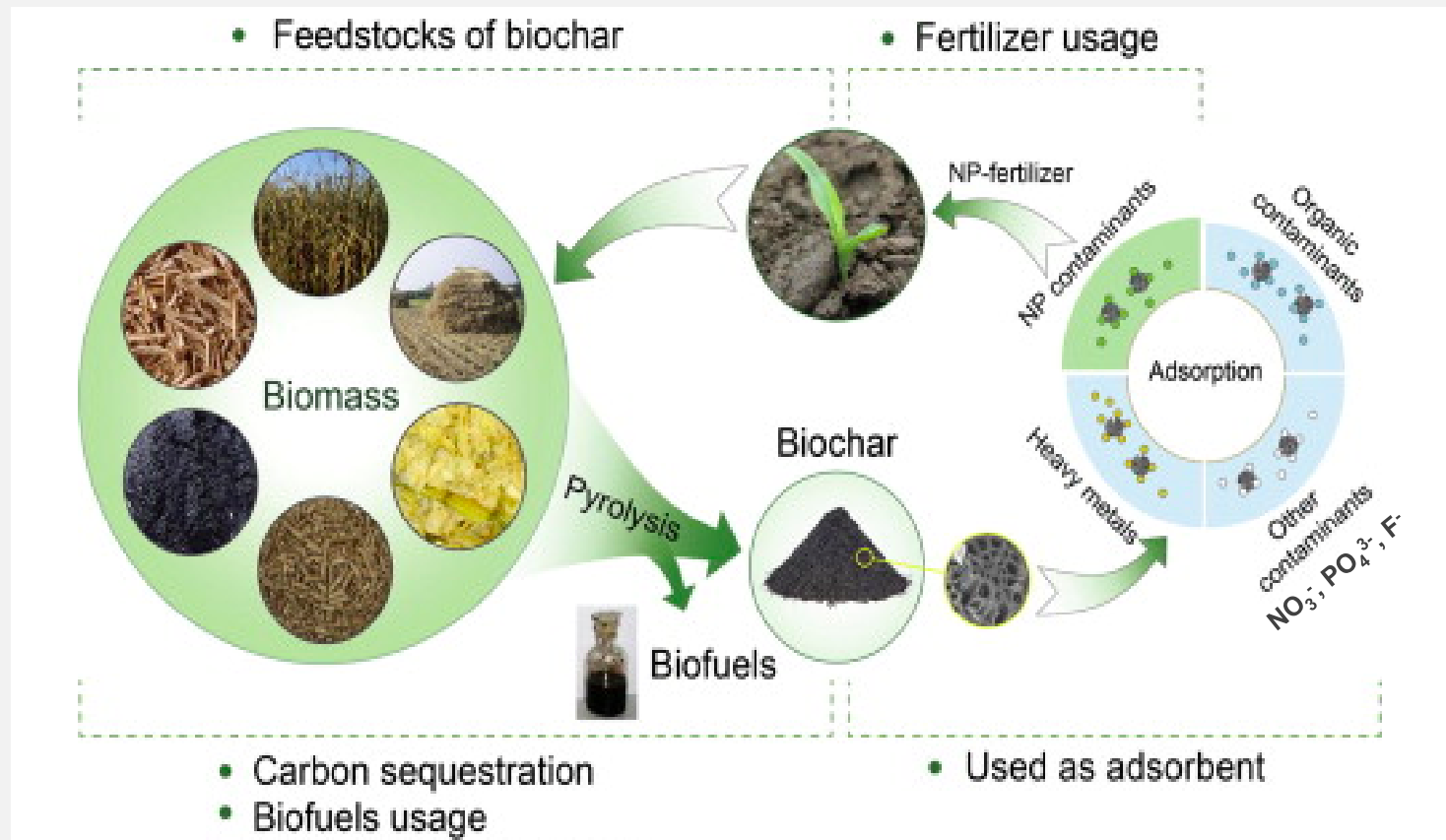
² School of Environmental Science and Engineering, Southern University of Science and Technology, Shenzhen 518055, P.R. China



BACKGROUND



Biochar application



Biochar applied as an effective adsorbent for wastewater treatment

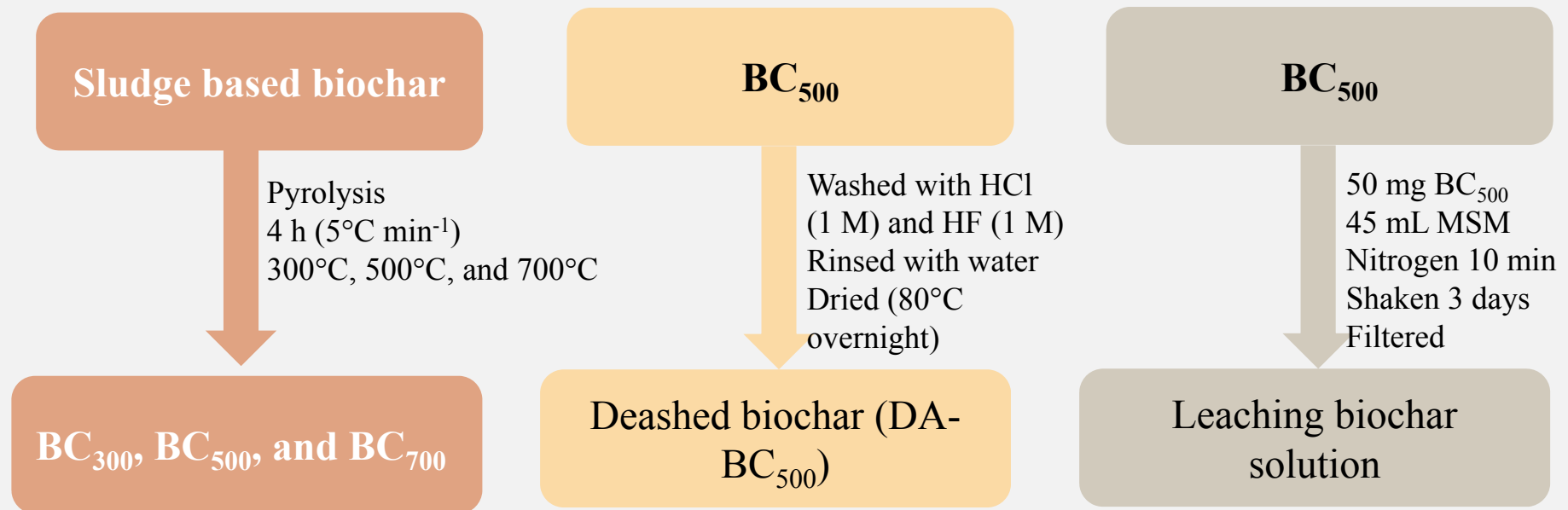
Tan, X., Liu, Y., Zeng, G., Wang, X., Hu, X., Gu, Y., Yang, Z. 2015. Application of biochar for the removal of pollutants from aqueous solutions. *Chemosphere* 125, 70-85.

OBJECTIVES

- ✓ To evaluate the role of sewage sludge-based biochar, generated at different pyrolysis temperatures (300°C, 500°C, and 700°C), on the biological removal of toluene, a model VOC, by *Pseudomonas plecoglossicida*.
- ✓ To further evaluate the effects of deashed biochar and leaching biochar solution on toluene bioremoval performance, whether accelerating toluene mineralization.

EXPERIMENTAL

- ✓ Sewage sludge: From local wastewater treatment plant; Dried (80°C overnight), crushed, and sieved.



- ✓ *Pseudomonas plecoglossicida* previously isolated from a petroleum contaminated site in Xiamen (China) and subcultured in MSM (pH 7) containing (g/L): KH₂PO₄ 1.0; K₂HPO₄ 1.0; NH₄NO₃ 1.0; MgSO₄·7H₂O 0.2; Fe₂(SO₄)₃ 0.05; CaCl₂ 0.02; and toluene (150 mg/L)

Microcosms:

Serum bottles: MSM (mineral salts medium) solution (45 mL)

Toluene (250 mg/L)

Inoculum (5 mL)

Biochar (BC₃₀₀, BC₅₀₀, BC₇₀₀, DA-BC₅₀₀; 50 mg)

Incubation: 150 rpm, 30°C, pH 7, 3 days



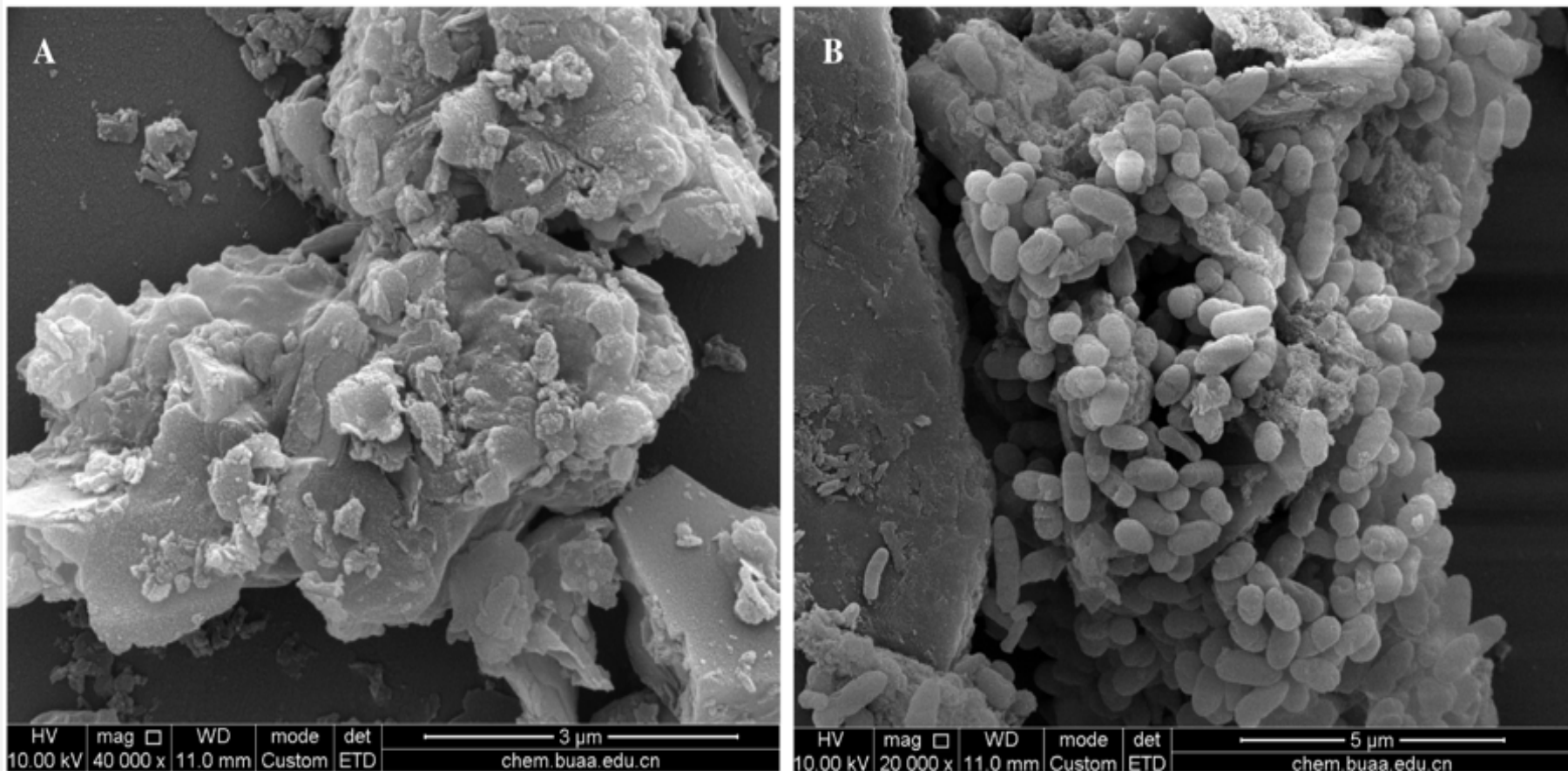
- ✓ Liquid samples collected daily to analyze toluene residual concentration (GC-FID)
- ✓ Microcosms without inoculum set as control (abiotic losses)
- ✓ Toluene adsorption capacity (q_e) on different biochars assessed at different times (5, 24, 48, and 72 h)
- ✓ Langmuir and Freundlich isotherm models for toluene (20, 50, 100, 200, and 300 mg/L) on 50 mg biochar (BC₅₀₀) in 50 mL MSM solution

RESULTS

Some physicochemical parameters of biochars

Biochar	pH	Dv (50) μm	Extractable TP (mg/g BC)	Extractable COD (mg/g BC)	Extractable TN (mg/g BC)
BC ₃₀₀	7.2	77.3	0.43	2.13	0.29
BC ₅₀₀	9.5	61.4	0.27	0.07	0.02
BC ₇₀₀	10.8	59.2	0.07	0.10	0.007
DA-BC ₅₀₀	3.2	72.9	0.13	0.17	0.16

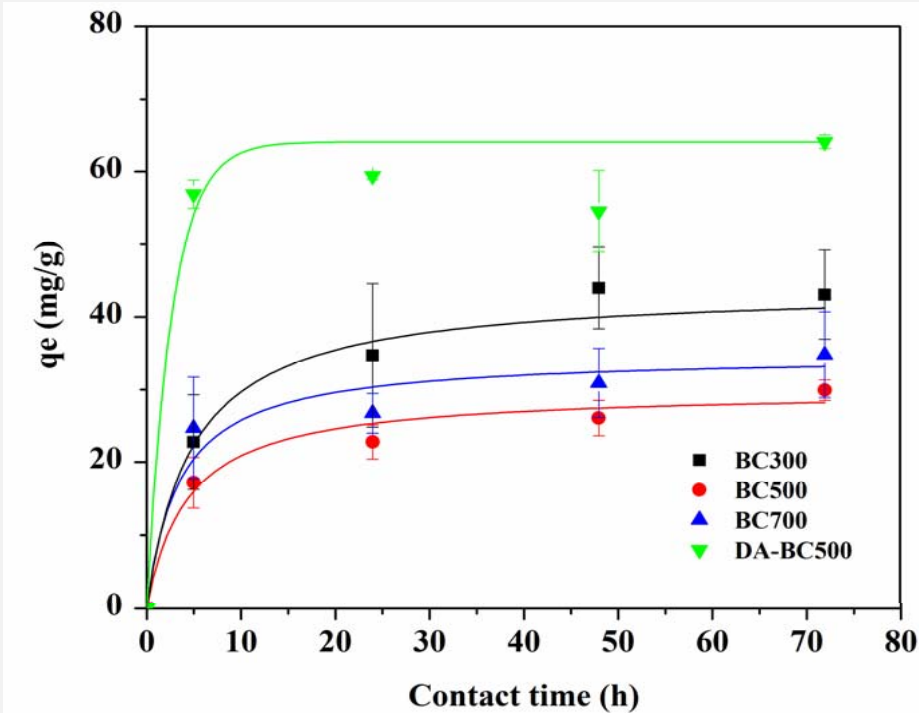
- ✓ pH significantly increased when higher pyrolysis temperatures applied ($p < 0.05$)
- ✓ Deashing treatment with a significant impact on the biochar fine portion, compared to the thermal treatment only; Fine particle portion of biochar decreasing with increasing temperatures
- ✓ Extractable TP, COD, and TN higher for the biochar produced at lower pyrolysis temperature (300°C)



SEM micrographs for (A) biochar (BC₅₀₀) (x40,000) and (B) biochar (BC₅₀₀) with *Pseudomonas* sp. attached on its surface (x20,000)

- ✓ **Surface morphology coarse and heterogeneous**
- ✓ **Rough aggregated micrometric structures with irregular size and orientations**
- ✓ ***Pseudomonas* sp. colonized the biochar surface efficiently**

Adsorption kinetics



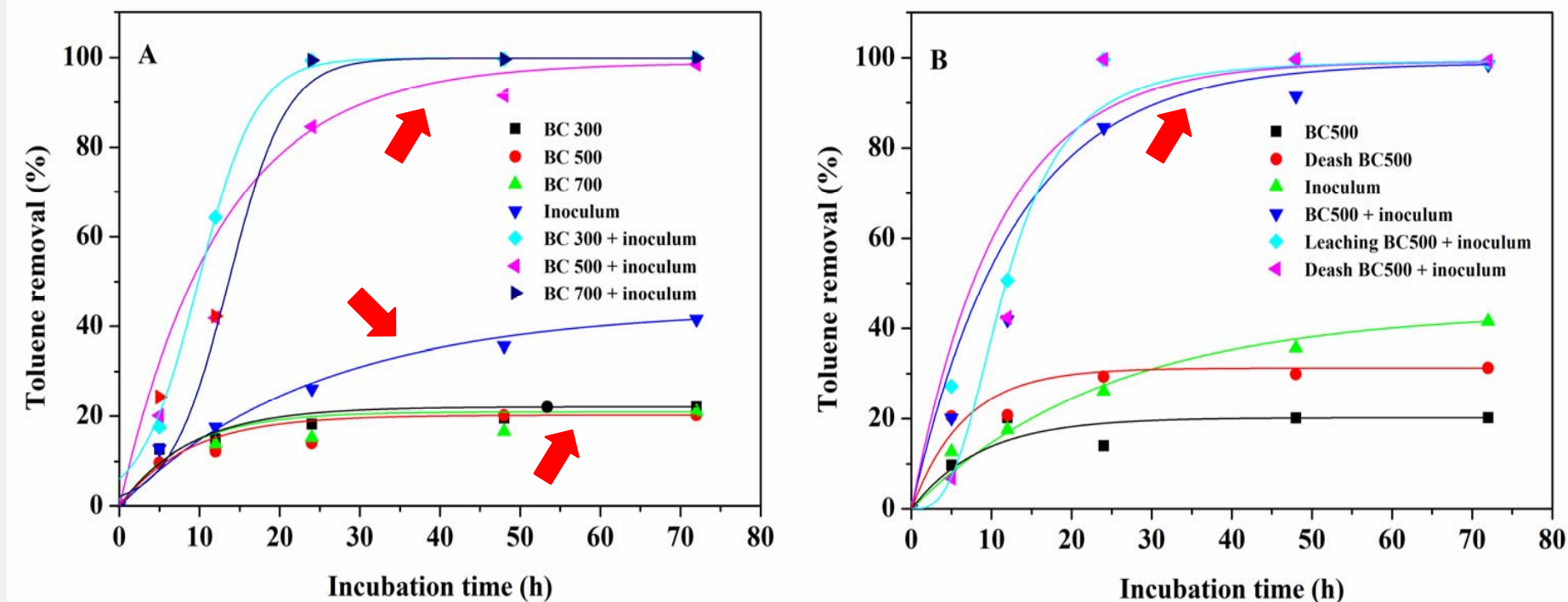
- ✓ Highest adsorption capacity on deashed biochar (64.1 ± 0.9 mg/g) ($p < 0.05$)
- ✓ Removal of the inorganic fraction (ash) might have created additional sorption sites on its surface
- ✓ Main purpose of biochar addition, to enhance toluene bioremoval (through biosorption)
- ✓ BC500: Showing the lowest toluene adsorption capacity, used to generate deashed biochar and leaching biochar solution

Toluene (250 mg/L) adsorption capacity (q_e) on different (BC_{300} , BC_{500} , BC_{700} , $DA-BC_{500}$) biochars (50 mg)

Adsorbent	Langmuir			Freundlich		
	C_m (mg/g)	K_L (L/mg)	r^2	K_f (mg/g) (L/mg) ^{1/n}	1/n	r^2
Biochar (BC_{500})	3.28	0.005	0.60	0.002	1.80	0.99

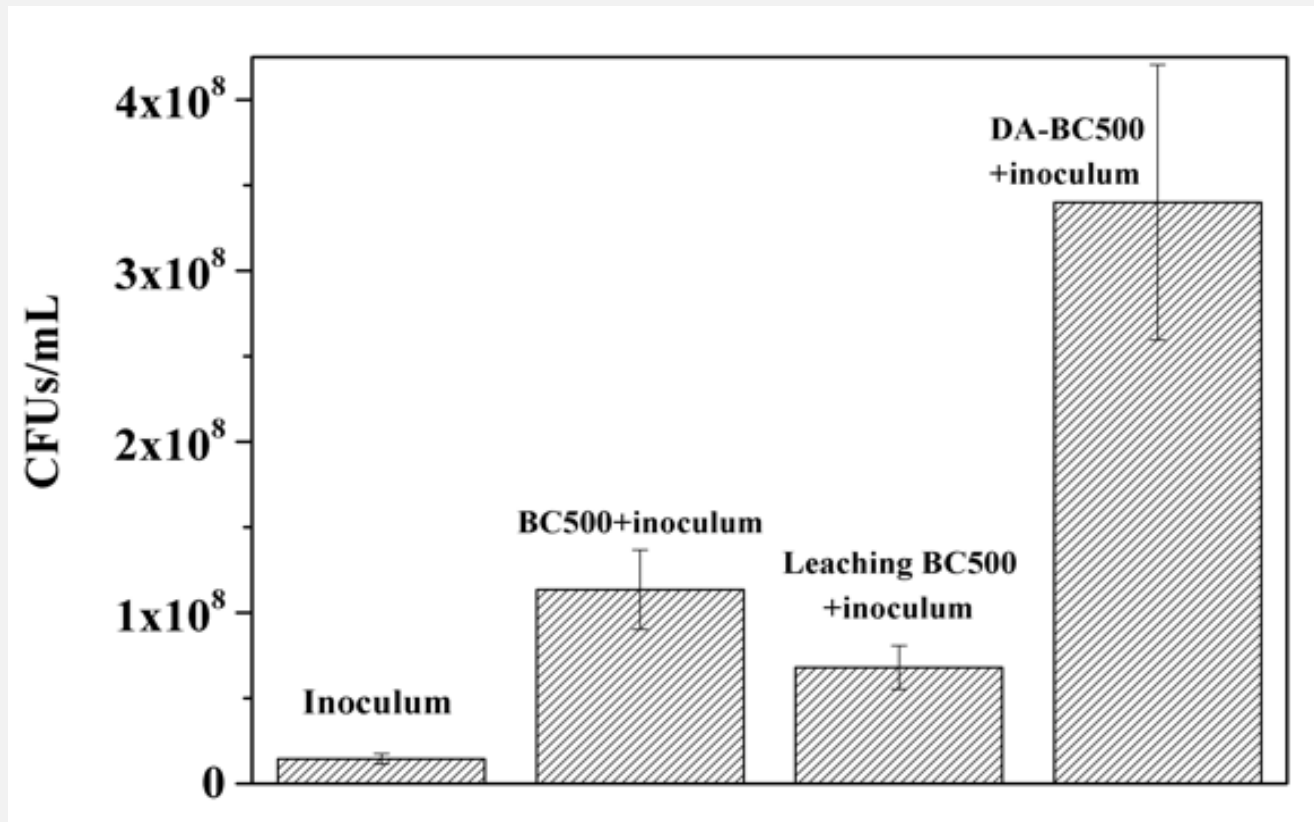
- ✓ Freundlich isotherm showed a better fit ($r^2 = 0.99$) to the adsorption data
- ✓ Biochar surface is heterogeneous with adsorption sites of different affinities

Biological removal of toluene and role of biochar



Toluene removal efficiency of *Pseudomonas* sp.: (A) with biochar produced under different temperatures (BC₃₀₀, BC₅₀₀, and BC₇₀₀) and (B) with BC₅₀₀, deashed BC₅₀₀, BC₅₀₀ leaching solution

Microbial growth in the presence of biochar



Colony forming units (CFUs) for the microcosms with inoculum only and with inoculum + biochars

- ✓ Removal of ash from biochar exposing additional surface area, mainly small-diameter pores, favoring the inoculum colonization and proliferation

CONCLUSIONS

- ✓ The biological and sorption removal of toluene were not that effective when isolate and biochars (BC₃₀₀, BC₅₀₀, and BC₇₀₀) applied alone. However, when biochar applied together with inoculum, toluene was almost completely removed after 2 days (through biosorption).
- ✓ Biochar promoted the microbial immobilization on its surface and substantially enhanced/facilitated the toluene bioremoval compared to the system with the microbial isolate only.
- ✓ Biochar could be considered an efficient electron mediator, and its redox moieties could essentially influence the electron transfer between microbial cells and toluene, with both sorbed on the biochar particles.
- ✓ The developed hybrid (physical/sorption + biological/immobilization) process as a promising technology considering the biochar as ecofriendly nature waste with special redox characteristics.

FUTURE WORKS

- Further studies warranted to further clarify the mechanism of toluene removal in the presence of biochar and what kind of compounds (organics and/or inorganics) stimulating the microbial growth especially when the biochar leachate used to remove toluene efficiently.
- Long-term stability of biochar and its reutilization to be further evaluated, especially in terms of contaminants/nutrients availability and microbial immobilization.
- Application of this hybrid technology can also be extended to other VOCs commonly found in contaminated sites (subsurface environment; soil and groundwater).

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