Supplementary Information

Valorization of Cash Crop Waste into a decomposable Nanocellulose adsorbent matrix through Bacterial Cell Factories for the Management of Agricultural Runoff Contaminants

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Experimental

Text S1. Chemicals and Bacterial Strain

All reagents were of analytical grade and obtained from Hi Media, India. The Hestrin-Schramm (HS) medium consisted of yeast extract (5 g/L), peptone (5 g/L), disodium hydrogen phosphate (2.7 g/L), and citric acid (1.15 g/L). The 2,4-D compound was obtained from Hi Media, India, while TC was acquired from Tokyo Chemical Industry (TCI), India. *Komagataeibacter saccharivorans* NUWB1, previously isolated from decomposed apple, was used in this study.

Text S2. Adsorption Capacity and Removal Efficiency Calculations

Batch adsorption experiments assessed the uptake of 2,4-D and TC. BNC was added to Erlenmeyer flasks containing solutions of defined initial concentrations. Mixtures were agitated at controlled temperatures for specific durations. Parameters such as adsorbent dosage, initial pollutant concentration, contact time, pH, and temperature were systematically varied.

Concentrations of 2,4-D and TC before and after adsorption were measured using a UV–visible spectrophotometer (Shimadzu, Japan) at wavelengths of 282 nm (2,4-D) and 357 nm (TC). Experiments were conducted in triplicate. The adsorption capacity (qe) and removal efficiency (R%) were calculated as follows:

$$Removal\ efficiency(\%) = \frac{C_0 - C_e}{C_0} * 100$$ Eqn. (1)

$$q_e = \frac{(C_0 - C_e)}{m} *\ V$$ Eqn. (2)

where C_0 is the concentration of the initial pollutant (mg L^{-1}), C_e is the equilibrium concentration of the pollutant, m represents the mass (g) of the adsorbent, and V(L) represents the volume of the adsorbate solution.

Text S3. Biodegradation % Calculation

$$\text{Biodegradation \%} = \frac{W1\ - W2}{W1} \times 100$$ Eqn. (3)

W_1 signifies the initial weight of the film, and W_2 signifies its final weight of post-degradation.

Results and Discussion

Text S4. Adsorption Kinetics Models

The Lagergren pseudo-first-order (PFO) model, originally proposed by Lagergren. is represented by Eqn (4):

$$\text{Log}\ (q_e - q_t) = log q_e - k_1 t$$ Eqn. (4)

The variable k_1 represents the rate constant for the pseudo-first-order, and t indicates the duration of contact time. The Ho pseudo-second-order (PSO) kinetic model is shown in Eqn (5):

$$\frac{1}{q_t} = \frac{1}{k_2 q_e} + \frac{t}{q_e}$$ Eqn. (5)

where k_2 is the pseudo-second-order adsorption rate constant

Text S5. Adsorption Isotherm Models

The Langmuir isotherm is given by Eqn (6):

Langmuir isotherm: $$\frac{C_e}{q_e} = \frac{1}{Q_{max} K_L} + \frac{1}{Q_{max}} C_e$$ Eqn. (6)

C_e (mg L^{-1}) represents the concentration of the adsorbate in the solution at equilibrium, q_e(mg/g) represents the amount of adsorbate adsorbed at equilibrium, q_{max} (mg/g) represents the monolayer adsorption capacity, K_L represents the equilibrium adsorption constant

The Freundlich isotherm is given by Eqn (7):

Freundlich isotherm: $$log q_e = \frac{1}{n_F}(log C_e) + log K_F$$ Eqn. (7)

here k_F represents the Freundlich constant, n represents the intensity of the adsorption.

The Temkin isotherm is given by Eqn (8):

Temkin isotherm: $$q_e = \frac{RT}{B_T} Ln(A_T C_e)$$ Eqn. (8)

Here, the variation in adsorption energy is Q (J mol^{-1}), and the Temkin constant is KT (L mg^{-1}).

Text S6. Thermodynamic Equations for Adsorption

The values of ΔH◦ and ΔS◦ were derived from the slope and intercept of the van’t Hoff plot of ln Kd versus 1/T for the highest initial concentration of TC and 2,4-D using the expression given in Eqn (9).

$$In K_d = \frac{\Delta S°}{R} - \frac{\Delta H°}{RT}$$ Eqn. (9)

where Kd denotes the dimensionless equilibrium distribution coefficient, R (8.314 J/mol K) denotes the universal gas constant and T (K) denotes the absolute temperature. The values of ΔG◦ were calculated from Eqn (10).

$$\Delta G° = \Delta H° - T\Delta S°$$ Eqn. (10)

Using equations 10 and 11, K_d and $\Delta G°$ were determined.

$$K_d = \frac{q_e}{C_e}$$ Eqn. (11)

$$\Delta G° = \Delta H° - T\Delta S°$$ Eqn. (12)

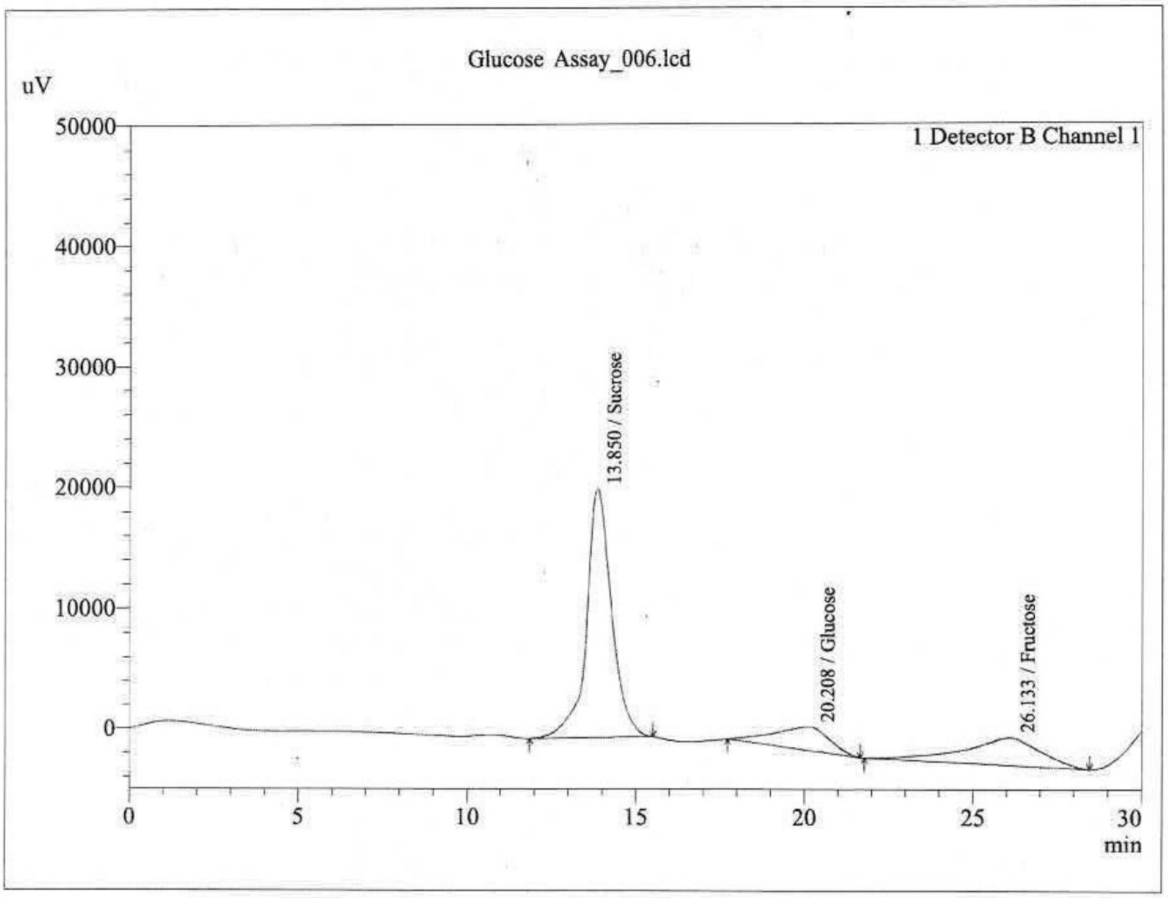
K_d represents the distribution coefficient, q_e is the amount of adsorbate retained by the adsorbent per litre of solution at equilibrium, C_e is the equilibrium concentration of the adsorbate in the solution, R denotes gas constant (8.314 JK^-

$^{1}mol^{-1}$), and T is the absolute temperature. ΔH° and ΔS° were computed using the graph that plotted ln (K_d) against 1/T.

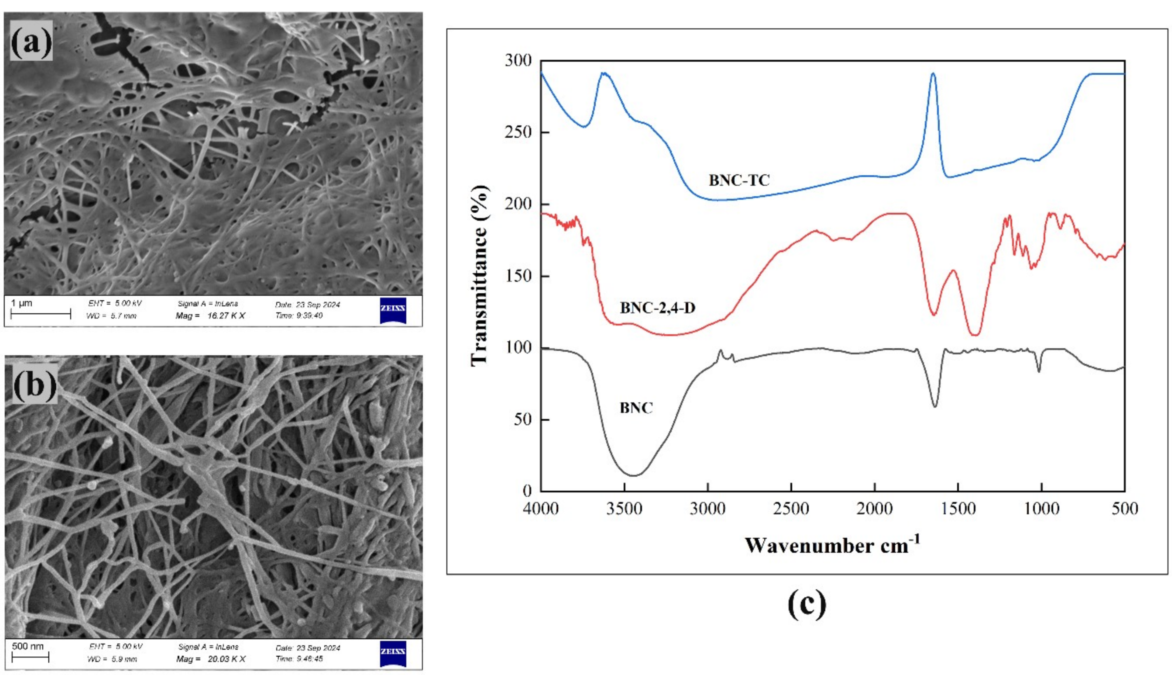
Supplementary Figures

- **Figure S1. Waste Sugarcane Molasses.**

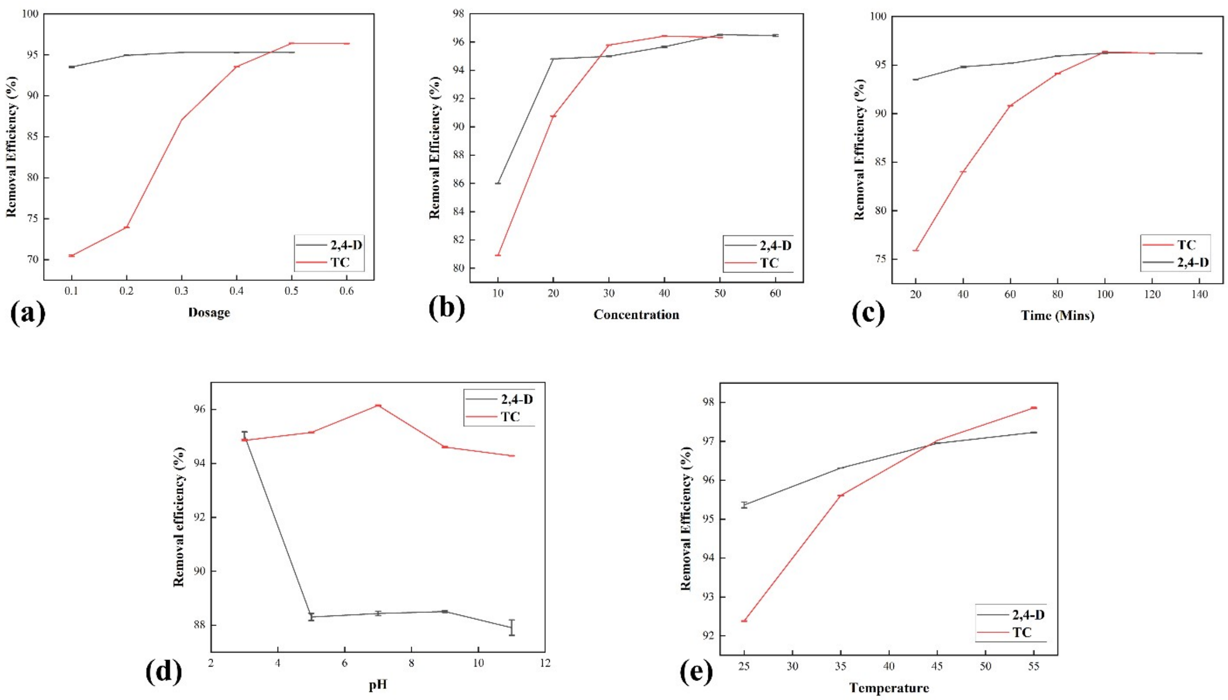
- **Figure S2. HPLC chromatogram showing glucose, fructose, and sucrose in WSM.**



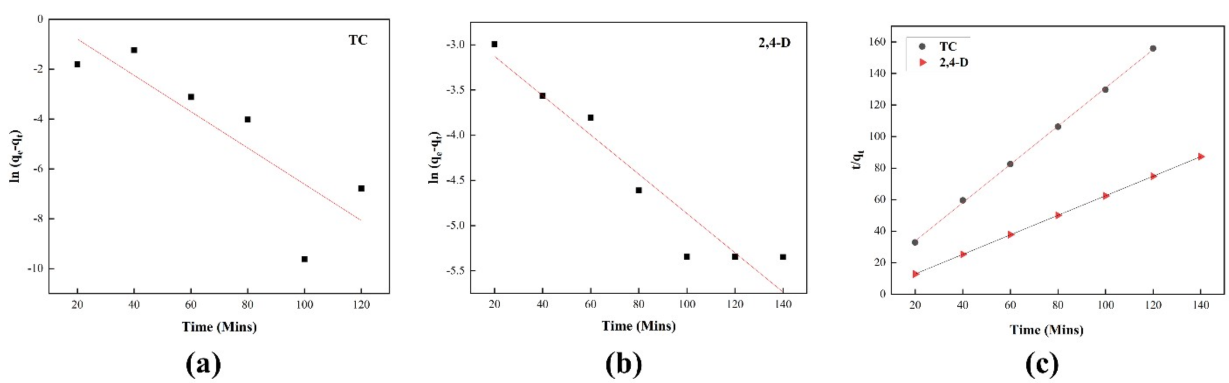
- **Figure S3. FE-SEM images of BNC post-adsorption of (a) TC, (b) 2,4-D, and (c) FTIR spectra pre- and post-adsorption.**



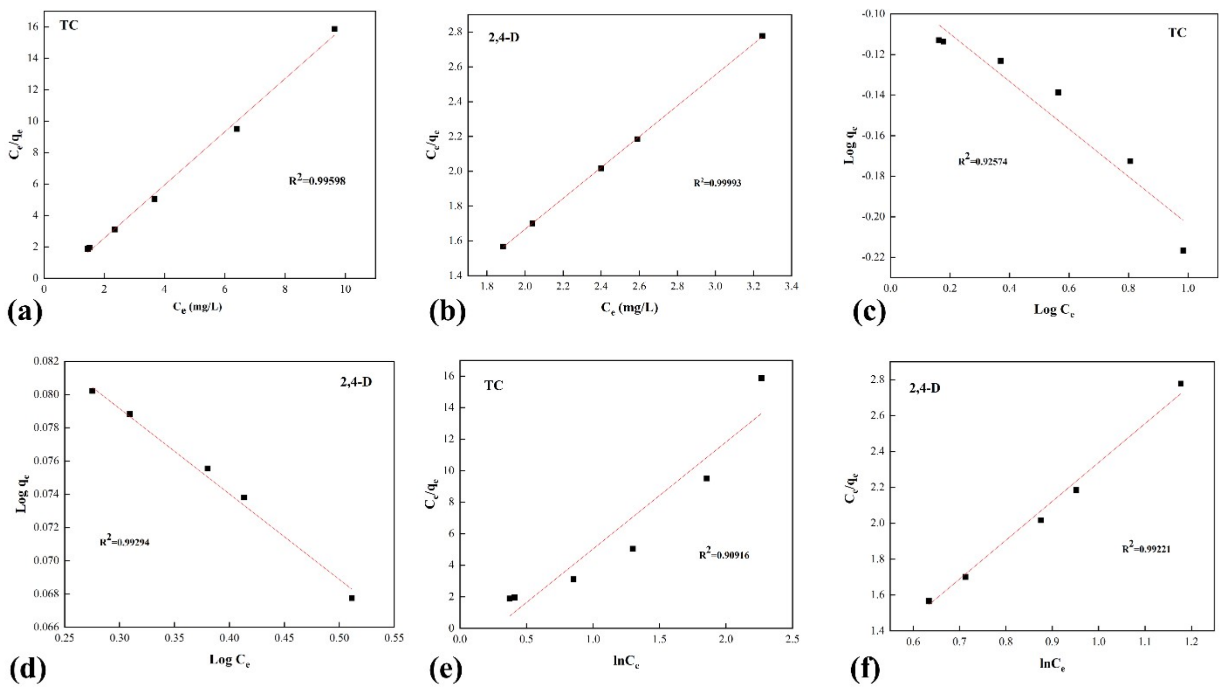
- **Figure S4. Effects of (a) dosage, (b) initial concentration, (c) time, (d) pH, and (e) temperature on adsorption of TC and 2,4-D by BNC**.



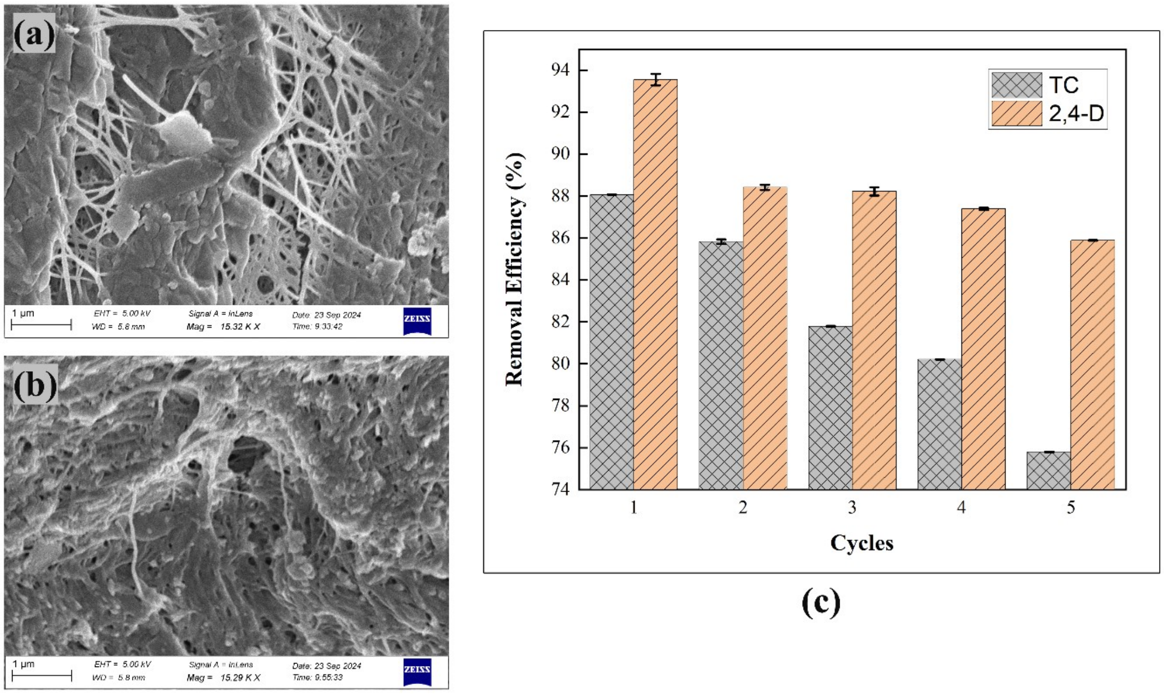
- **Figure S5. (a) and (b) Pseudo-first-order for TC and 2,4-D and (c) pseudo-second-order kinetic plots for TC and 2,4-D.**



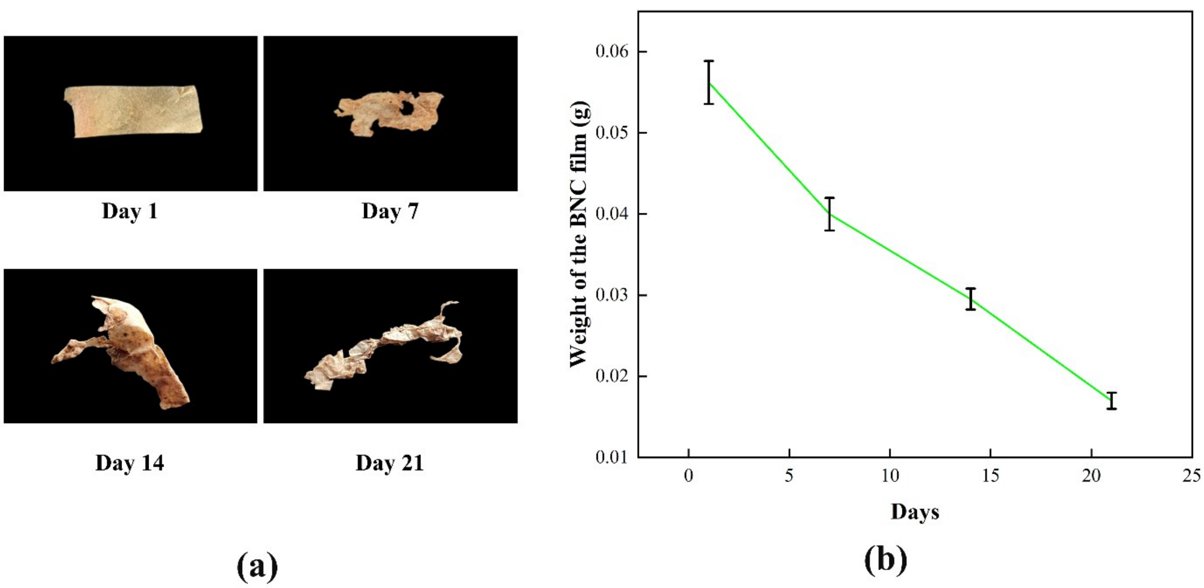
- **Figure S6. Langmuir, Freundlich, and Temkin isotherms for TC and 2,4-D adsorption.**



- **Figure S7. FE-SEM images of BNC after desorption of (a) TC, (b) 2,4-D, and (c) BNC regeneration cycles.**



- **Figure S8. (a) Morphological changes in BNC films over 21 days of soil burial, and (b) weight loss during degradation.**



Supplementary Tables

Table S1. Comparison of BNC production in different beaker types and broth volumes

Beaker Volume (mL)	Culture Volume (mL)	Surface Area (cm^2)	Depth (cm)	S/V Ratio (cm^{-1})	BNC Production (g)
500	300	349	6.2	1.6	3.279 ± 0.023
500	200	349	4.1	2.69	3.085 ± 0.09
250	200	220	5.6	1.1	1.621 ± 0.014
250	100	220	3.2	2.2	1.77 ± 0.063

Table S2. d-Spacing and Dominant Allomorph of BNC Synthesized from WSM

Bacterium	Carbon Substrate	d_1 (nm)	d_2 (nm)	d_3 (nm)	Dominant Allomorph
K. saccharivorans NUWB1	WSM	0.601	0.515	0.385	Iα

Table S3(a). Kinetic Parameters for Adsorption of TC and 2,4-D onto BNC

Adsorbate	Model	qe (exp)	qe (cal)	k	R^2
TC	Pseudo-1st Order	0.771	1.51	0.000727	0.72
TC	Pseudo-2nd Order	0.771	0.824	0.07	0.99
2,4-D	Pseudo-1st Order	1.608	0.3713	0.00018	0.91
2,4-D	Pseudo-2nd Order	1.608	1.614	5.1379	0.99

Table S3(b). Adsorption Isotherm Parameters for TC and 2,4-D on BNC

Adsorbates	Langmuir	Freundlich	Temkin
TC	q_{max} (mg g^{-1}) = 1.22	K_F = 2.44	b_T = 6.7869
	K_L = 0.072	1/n = 8.524	K_T = 1.2944
	R^2 = 0.995	R^2 = 0.92	R^2 = 0.90
2,4-D	q_{max} (mg g^{-1}) = 9.3057	K_F = 1.24	b_T = 2.16528
	K_L = 10.488	1/n = 19.447	K_T = 1.08344
	R^2 = 0.99993	R^2 = 0.99294	R^2 = 0.99065

Table S3(c). Thermodynamic Parameters for Adsorption of TC and 2,4-D

Adsorbate	*ΔH°* (kJ/mol)	*ΔS°* (kJ/mol)	*ΔG°* (kJ/mol)			
			298(K)	308(K)	318(K)	328(K)
TC	35.916	0.10917	-32.5246	-33.6163	-34.708	-35.7997
2,4-D	14.681	0.04633	-13.129	-13.5924	-14.0558	-14.5192

Table S4. Weight Loss and Biodegradation of BNC Films

Day	Film Weight (g)	BNC Degradation (%)

1	0.056 ± 0.0026	0
7	0.04 ± 0.003	40.58
14	0.029 ± 0.0012	47.48
21	0.017 ± 0.001	69.76