

Supplementary materials

Article

Nanocomposite Hydrogel Films Based on Sequential Interpenetrating Polymeric Networks as Drug Delivery Platforms

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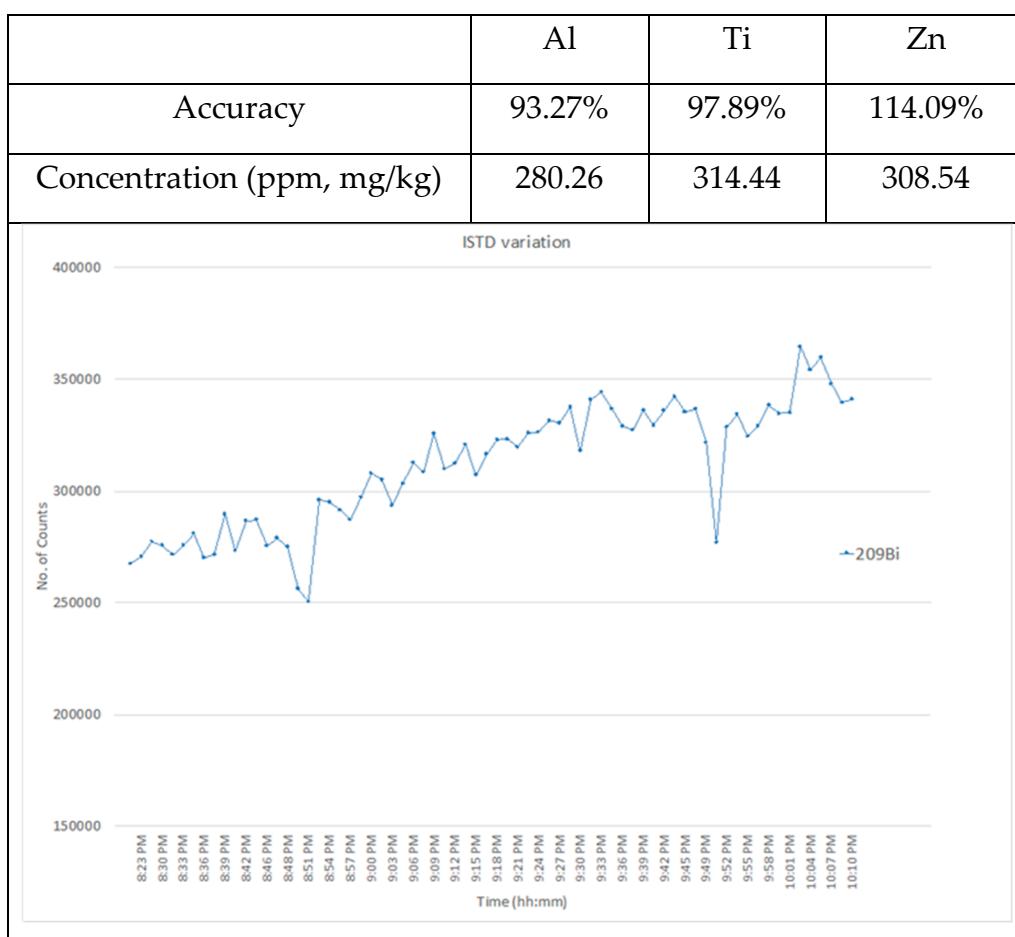
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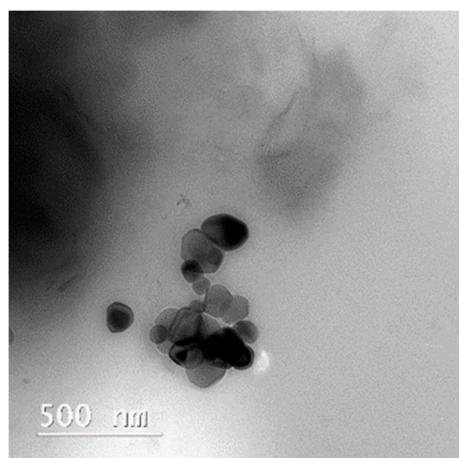
Characterization

ICP – MS

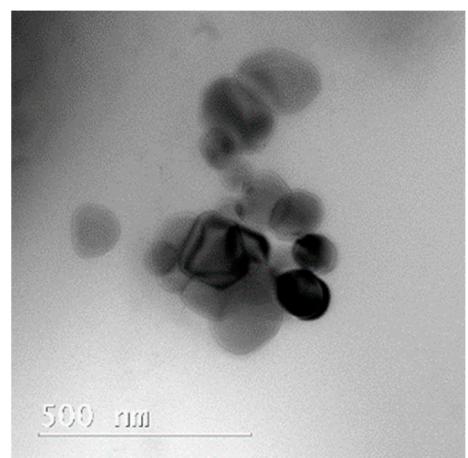
The method's accuracy is between 93% and 114%, and the acceptance limit is (70-150%) with precision under 14% and the acceptance limit 20%. The internal standard used for normalization was ^{209}Bi since it is not present in the measured samples and has a variation of 8.9%.

Table S1- ICP-MS accuracy and spike concentrations

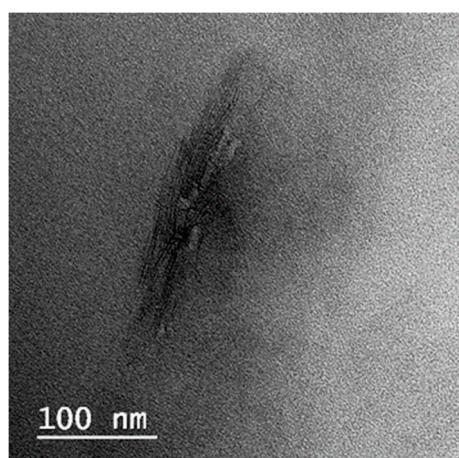




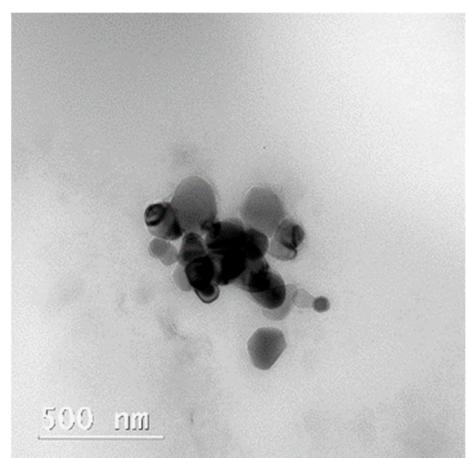
A



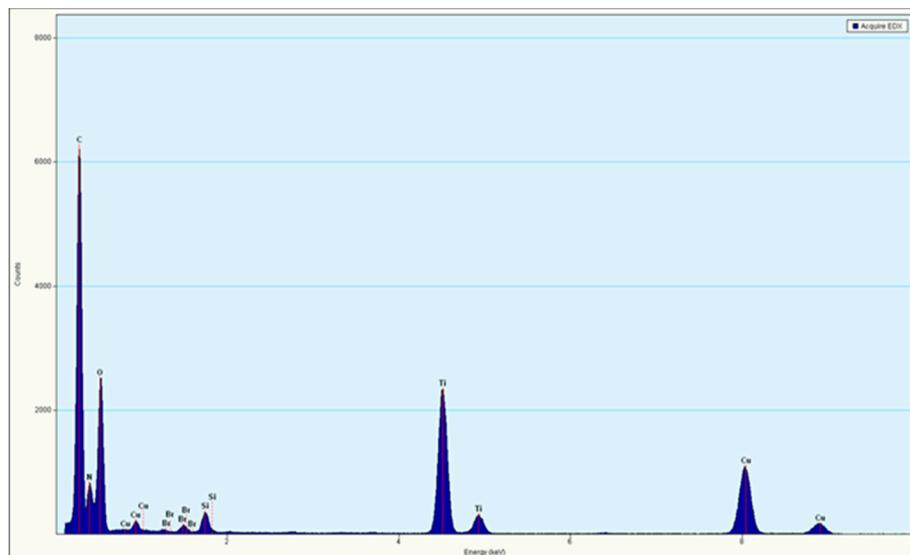
B



C

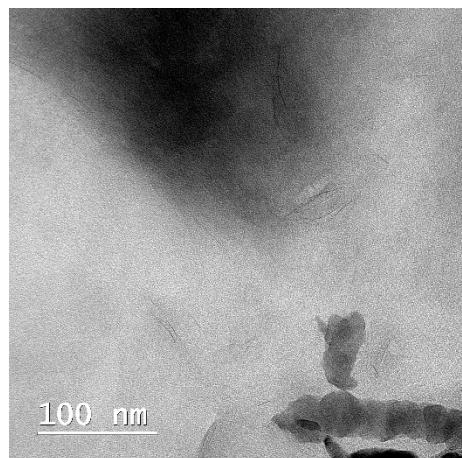


D

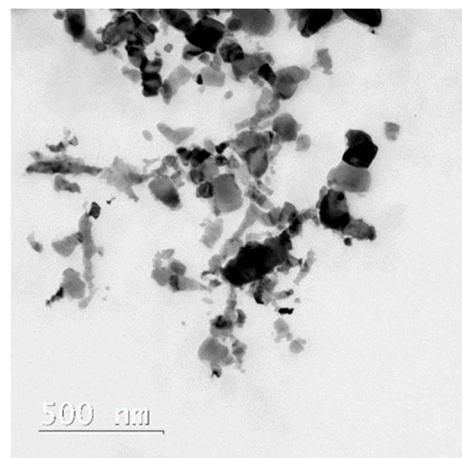


E

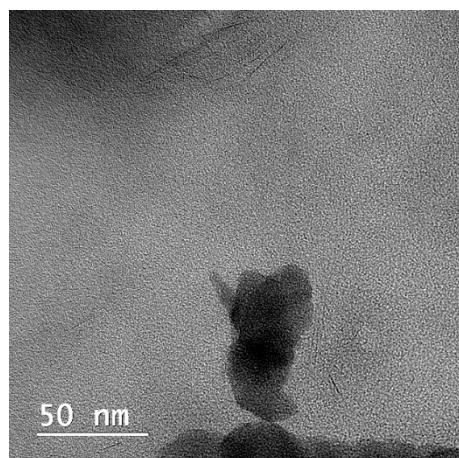
*Figure S1 - TEM images of bentonite layers (A, C) and TiO₂ nanoparticles (A, B, D)
and EDX spectra (E) of the hydrogel film IV.1*



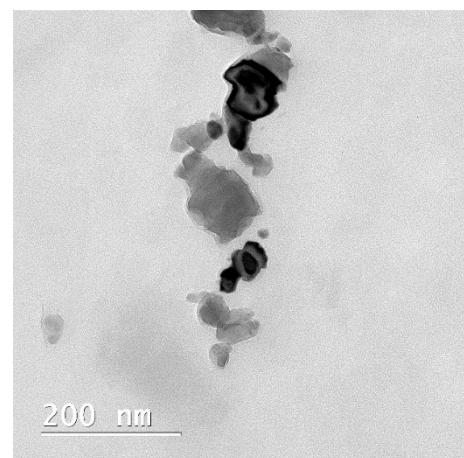
A



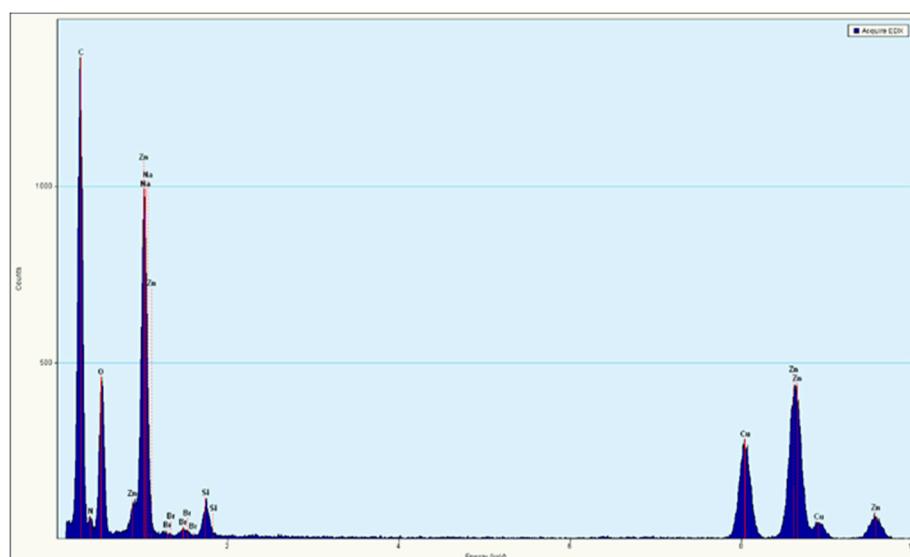
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C

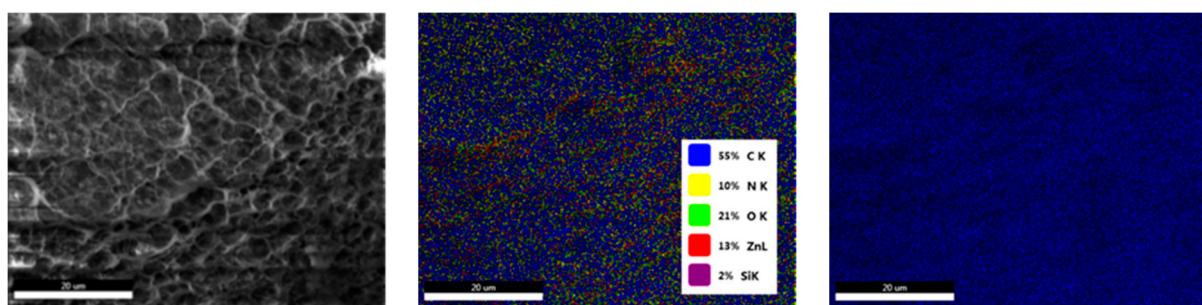


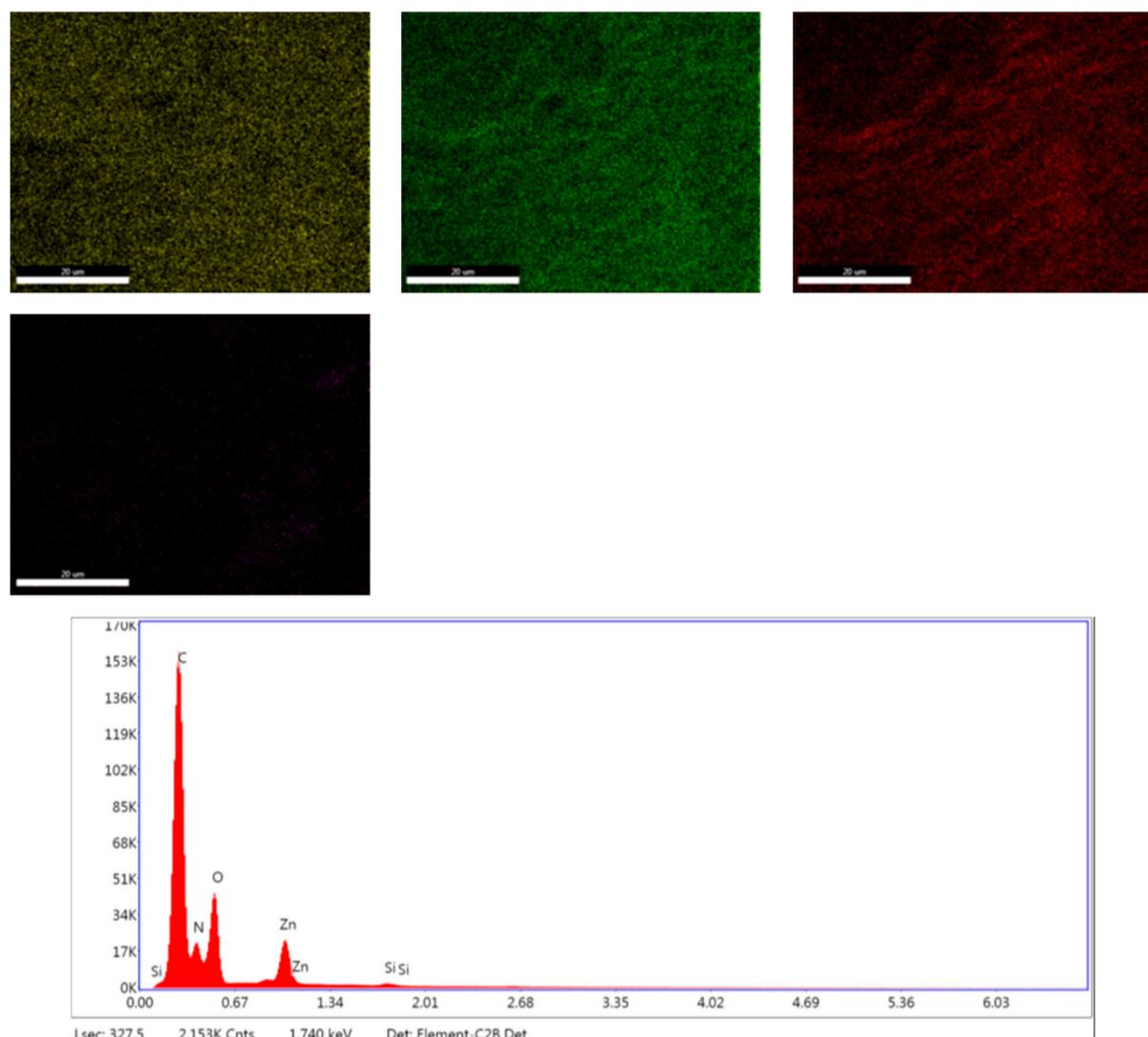
D



E

Figure S2 - TEM images of bentonite layers (A,C) and ZnO nanoparticles (A, B, C, D) and EDX spectra (E) of the hydrogel film IV.2





eZAF Smart Quant Results

Element	Weight %	Atomic %	Net Int.	Error %	Kratio	Z	R	A	F
C K	46.93	56.84	3604.60	5.49	0.3154	1.0555	0.9761	0.6367	1.0000
N K	19.42	20.18	479.20	9.60	0.0500	1.0240	0.9862	0.2513	1.0000
O K	22.30	20.28	940.40	8.68	0.0737	0.9970	0.9950	0.3315	1.0000
ZnL	10.75	2.39	373.70	2.78	0.0720	0.7135	1.1632	0.9390	0.9991
SiK	0.60	0.31	42.40	4.86	0.0049	0.8844	1.0329	0.9158	1.0043

Figure S3 – SEM-EDX mapping of sample IV

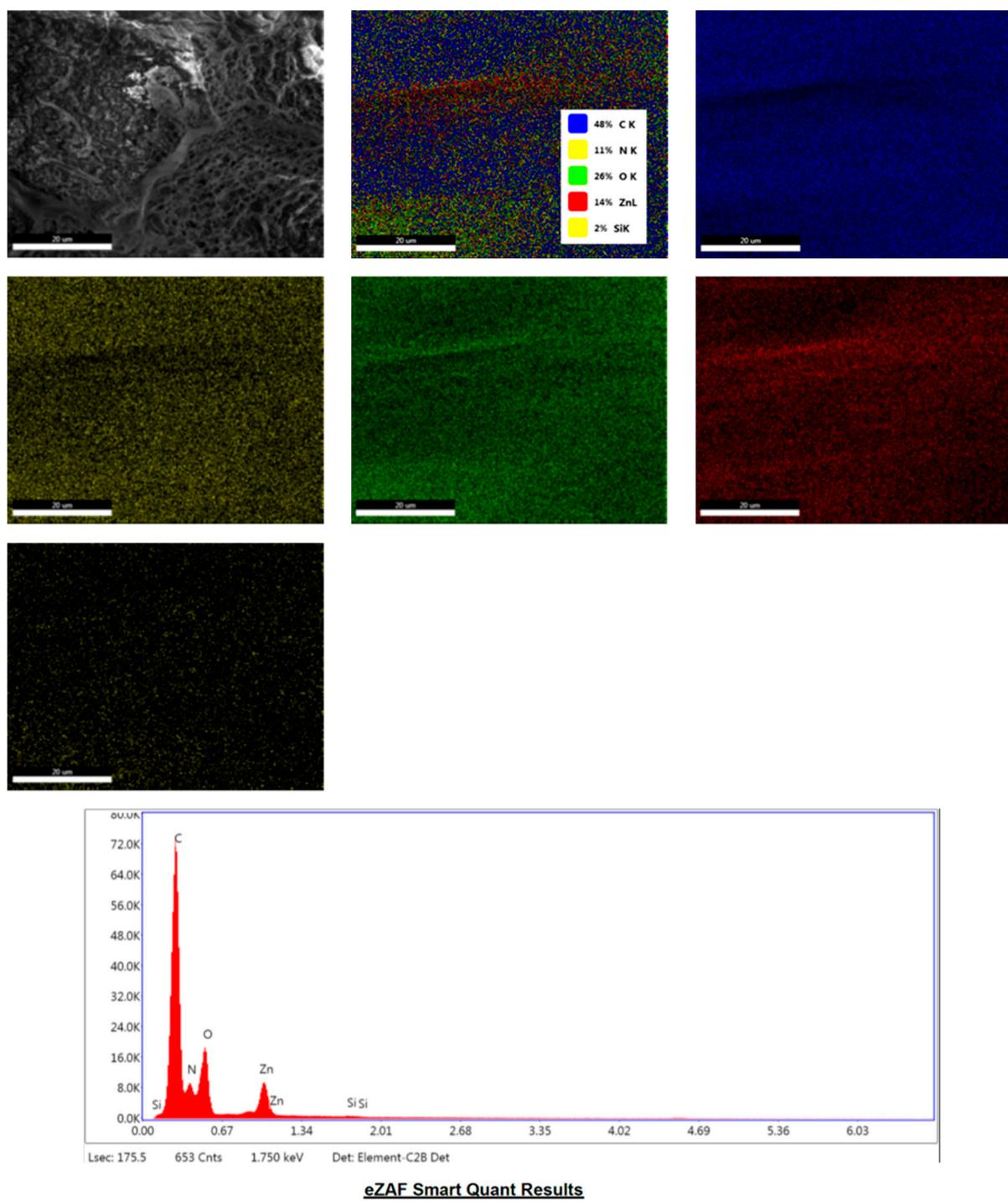
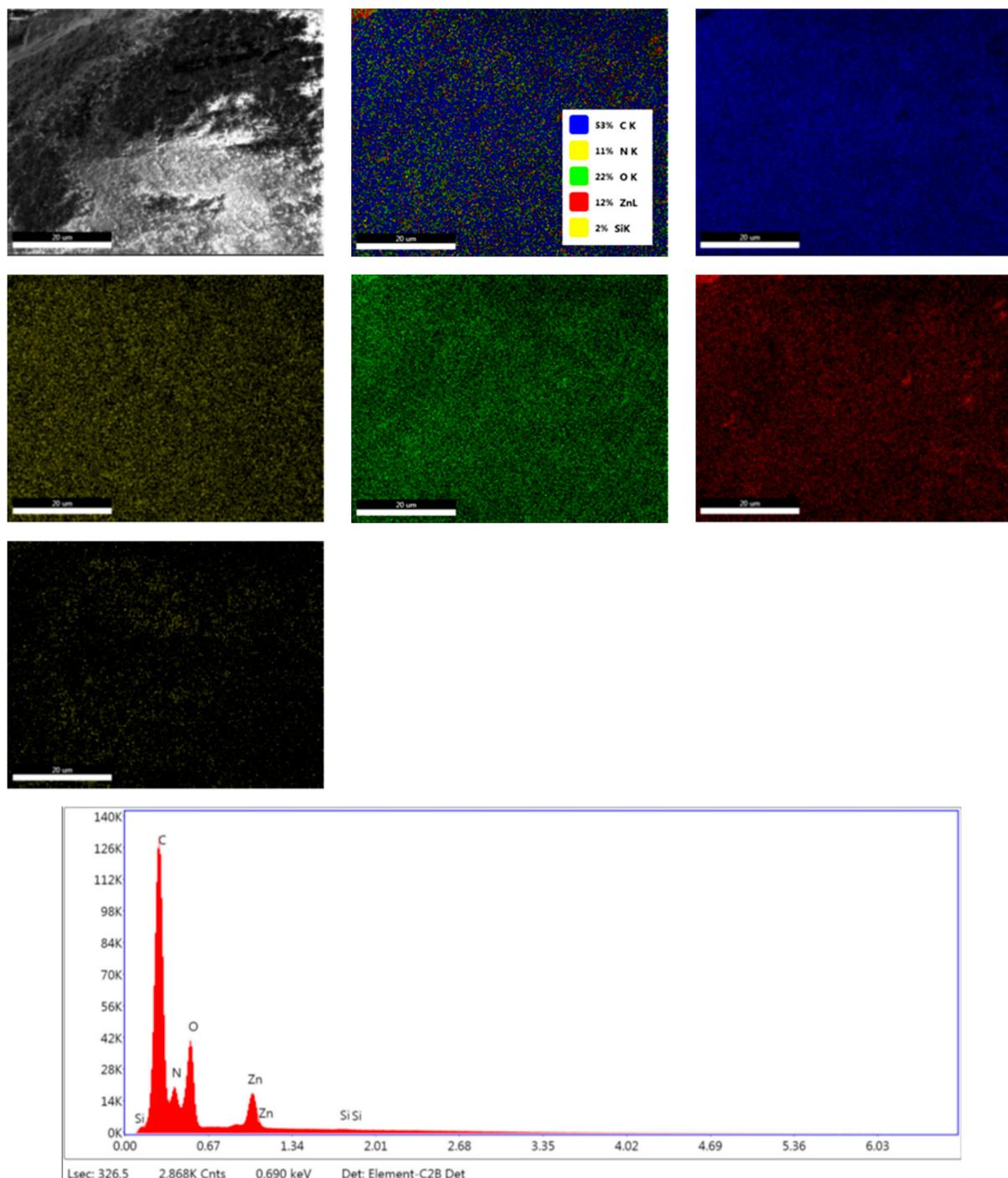


Figure S4 – SEM-EDX mapping of sample IV.1



eZAF Smart Quant Results

Element	Weight %	Atomic %	Net Int.	Error %	Kratio	Z	R	A	F
C K	49.51	58.99	2790.10	5.14	0.3505	1.0500	0.9786	0.6742	1.0000
N K	17.46	17.84	292.60	9.84	0.0439	1.0186	0.9886	0.2471	1.0000
O K	23.59	21.10	704.30	8.68	0.0796	0.9917	0.9973	0.3401	1.0000
ZnL	9.43	2.06	226.90	3.15	0.0629	0.7095	1.1655	0.9401	0.9991
SiK	0.00	0.00	0.10	95.38	0.0000	0.8794	1.0346	0.9223	1.0045

Figure S5 – SEM-EDX mapping of sample IV.2

Table S2 – Concentrations of the representative elements in the nanocomposite hydrogels

Sample code	Al ppm, mg/kg	Ti ppm, mg/kg	Zn ppm, mg/kg
IV	$559.60 \pm 5.9\%$	$12.44 \pm 2.9\%$	$61449.57 \pm 0.3\%$
IV.1	$646.00 \pm 2.9\%$	$337.12 \pm 3.9\%$	$49524.46 \pm 7.2\%$
IV.2	$587.03 \pm 10.9\%$	$13.37 \pm 11.8\%$	$50787.42 \pm 14.0\%$

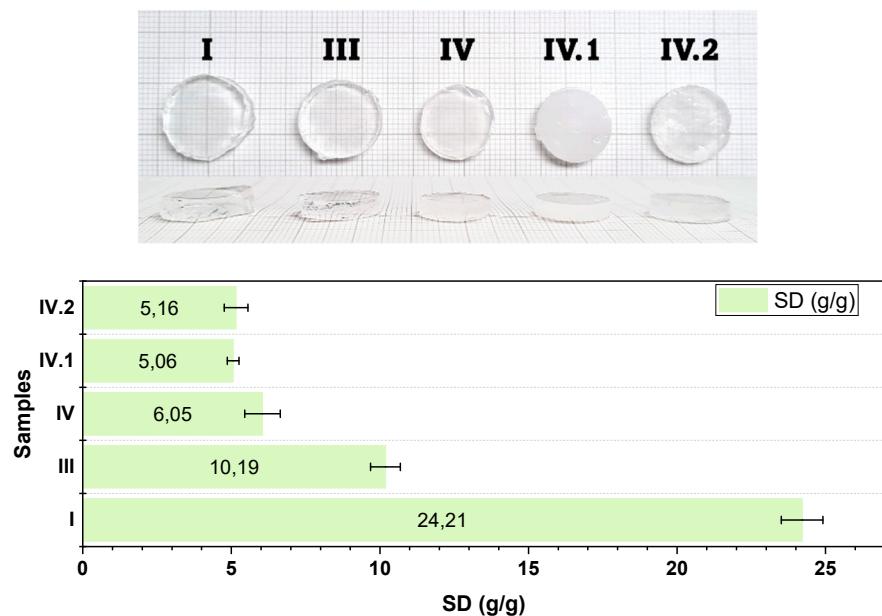


Figure S6 – Swelling degree at equilibrium

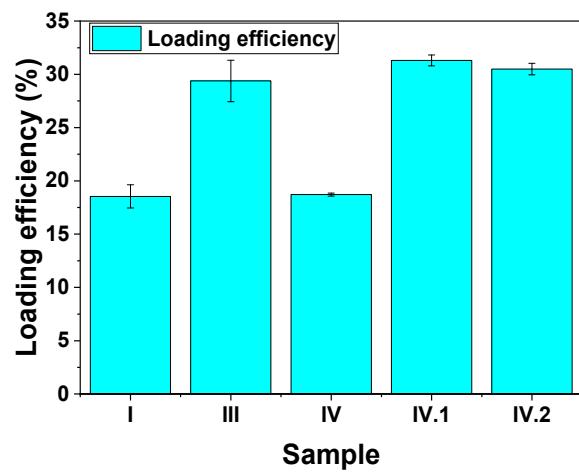
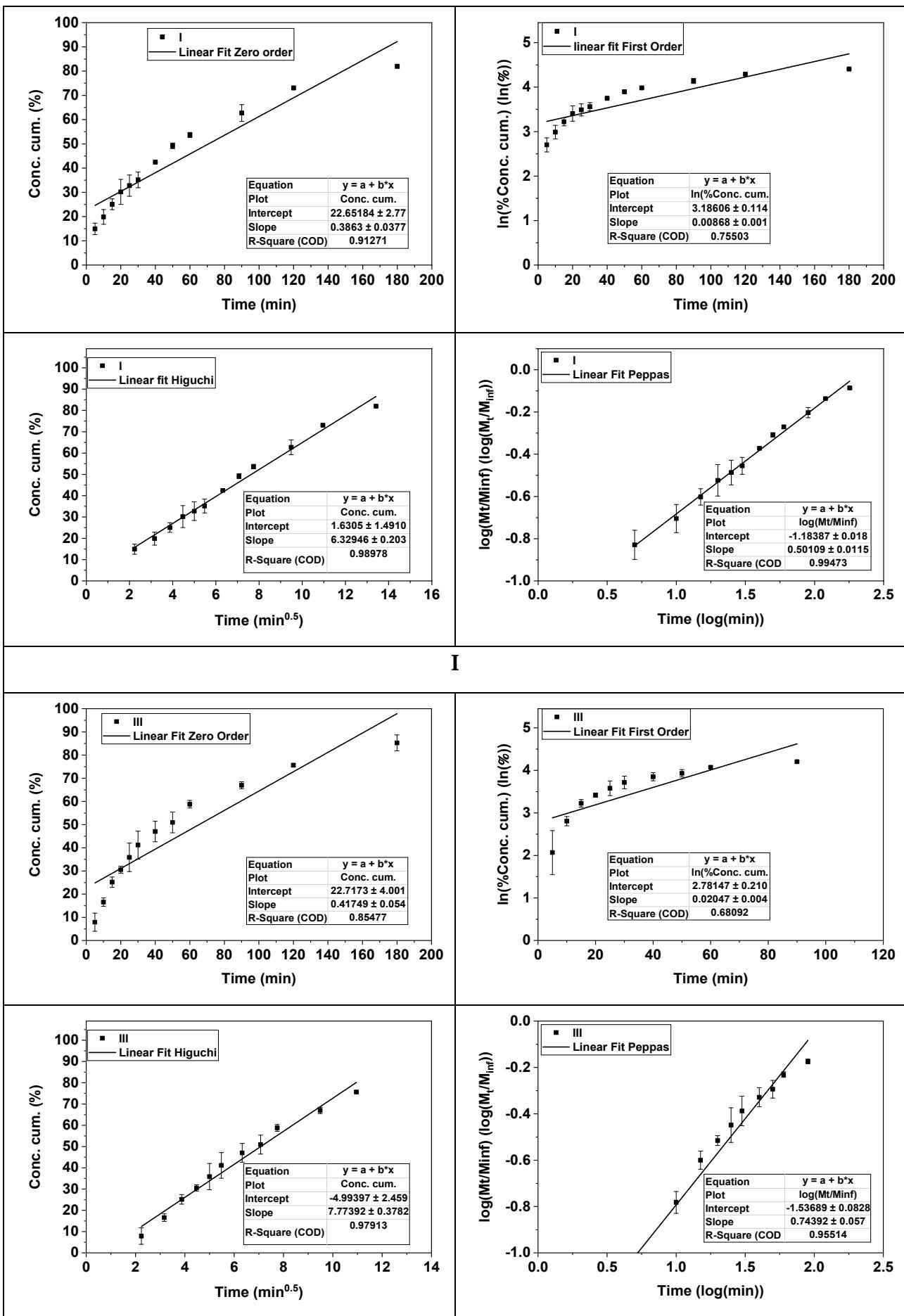
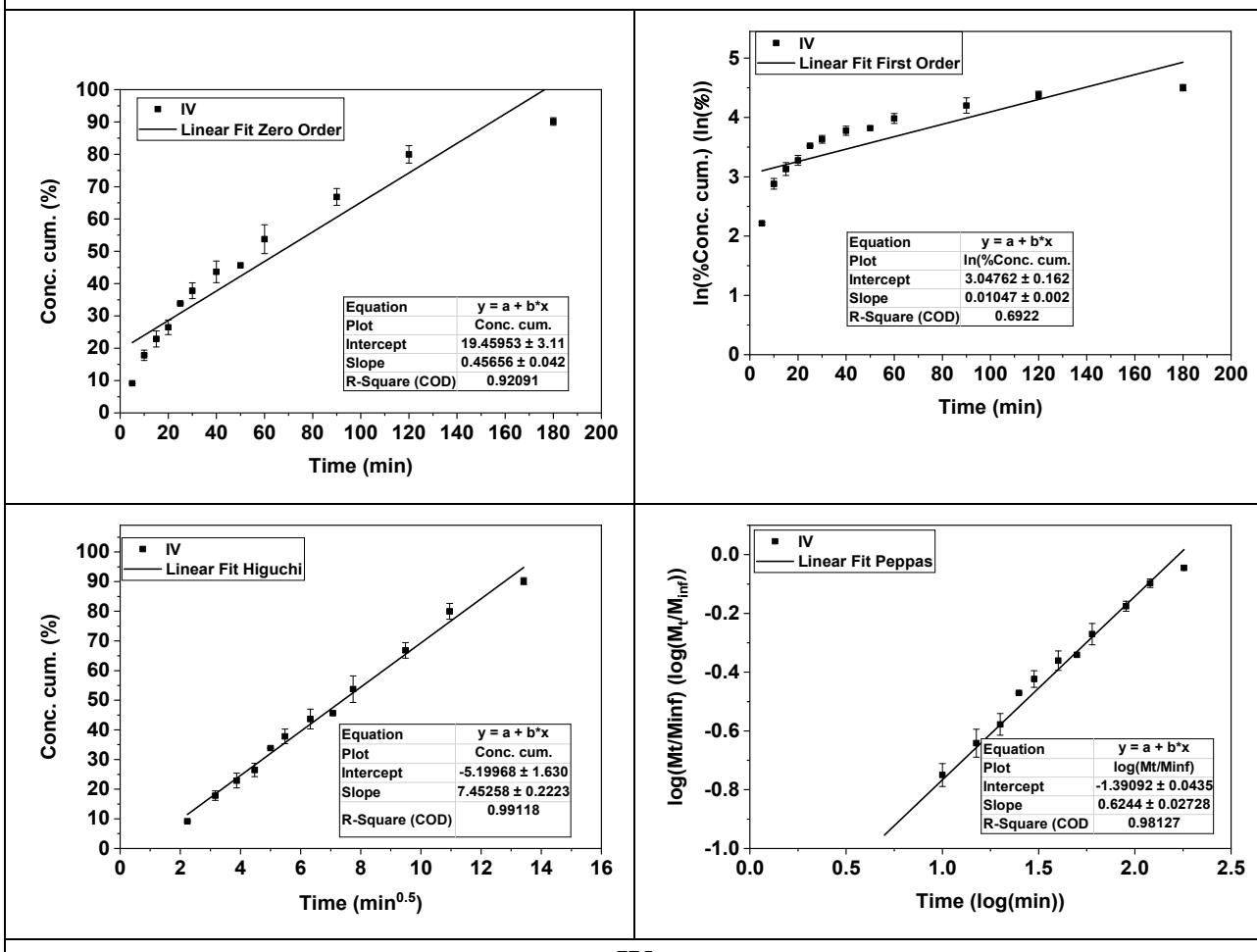


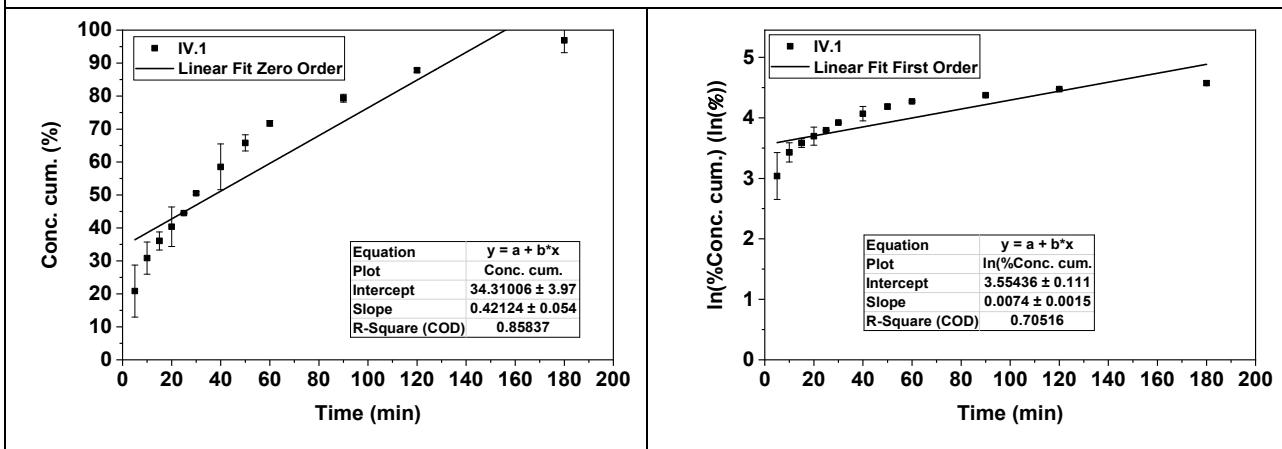
Figure S7 – Nafcillin loading efficiency at pH 7.4



III



IV



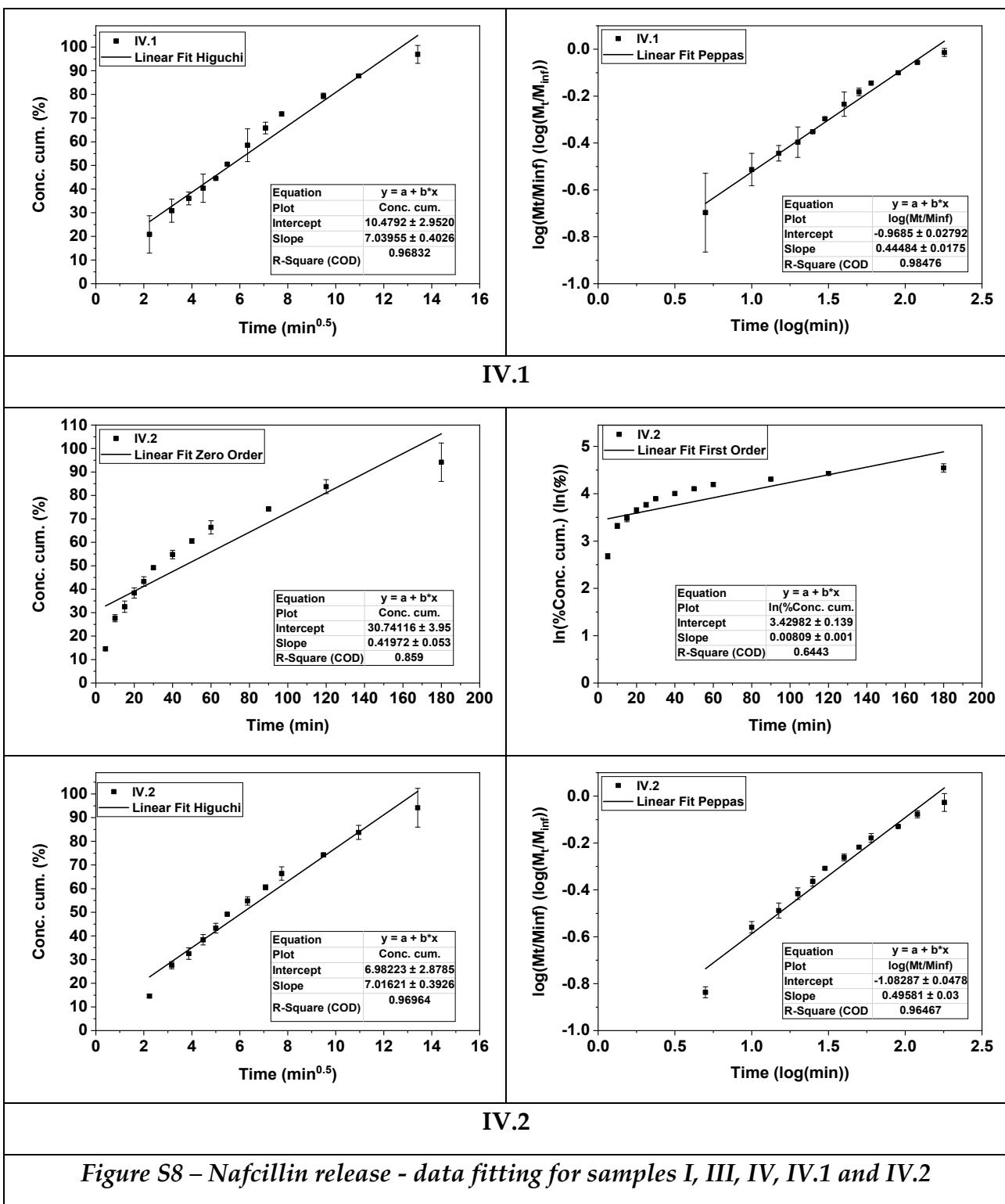


Table S3 - Number of CFU when no drug was loaded in the nanocomposite hydrogel films

Samples	No. of CFU growth after 15 min		No. of CFU growth after 30 min		No. of CFU growth after 1 hour	
	<i>S. aureus</i>	<i>E. coli</i>	<i>S. aureus</i>	<i>E. coli</i>	<i>S. aureus</i>	<i>E. coli</i>
IV	2×10^7	5×10^7	3×10^7	12×10^7	4×10^7	9×10^7
IV.1	10^7	3×10^7	2×10^7	5×10^7	10^7	12×10^7
IV.2	3×10^7	9×10^7	4×10^7	10×10^7	2×10^6	7×10^6
PC <i>S. aureus</i>				5×10^7		
PC <i>E. coli</i>				10×10^7		
PC = control						

Table S4 - Minimal inhibitory concentration (MIC) and Minimal bactericidal concentration (MBC) values

Sample / Microorganism	<i>E. coli</i>		<i>S. aureus</i>	
	MIC (%)	MBC (%)	MIC (%)	MBC (%)
TiO ₂ - NPs powder	> 2	--	> 2	--
ZnO - NPs powder	0.125	--	0.25	--

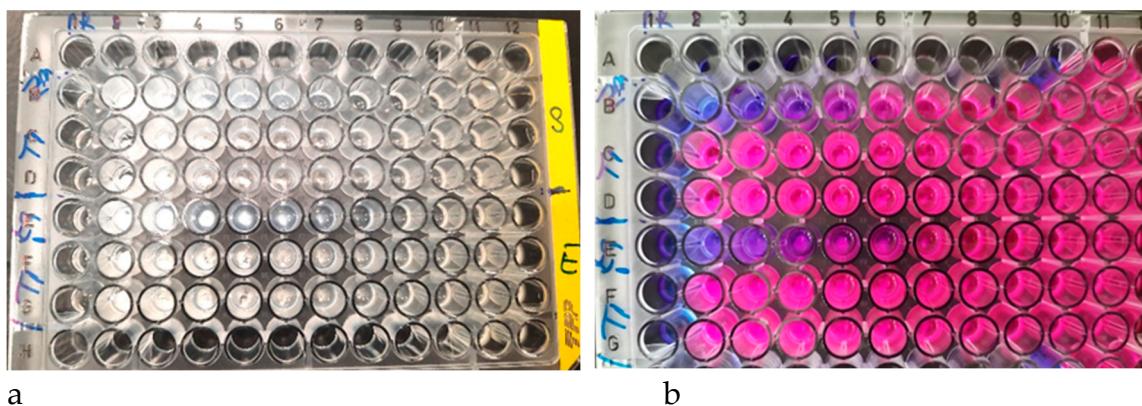


Figure S9. - MIC determination observed from broth microdilution assay using MH broth and resazurin.

S. aureus ATCC 6538 (row B: ZnO; row C: TiO₂; row D: Positive Control *S. aureus*);
E. coli ATCC 8739 (row E: ZnO; row F: TiO₂ and row G: Positive Control *E. coli*);
Bk = first column (a)before resazurin (b)after resazurin