

Supplemental Information

Antibacterial Agents Adsorbed on Active Carbon: A New Approach for Wound Healing Treatment

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The standard curve for the sulfamethoxazole (S)

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Analytical part

The standard curve for the gentamicin-ninhydrin complex

Accurately 100 µg of gentamycin sulfate powder (Sigma-Aldrich CAS: 1405-41-0, Burlington, MA, United States) was weighed and dissolved with 1 mL of phosphate-buffered saline (PBS) solution (pH 7.4) to get the stock solution with the concentration of 100 µg/mL. Then dilutions of gentamycin sulfate were prepared in the range from 50 µg/mL to 1.6 µg/mL. In the next step, 50 mg of ninhydrin (Sigma-Aldrich CAS: 485-47-2, Burlington, MA, United States) was dissolved in 10 mL of PBS at pH = 7.4 to give a slightly yellow solution whose concentration was 5 mg/mL. 50 ml of a 5 mg/ml ninhydrin solution was added to 150 ml of gentamycin sulfate solutions to give final concentrations of gentamycin sulfate in the range of 37.50 to 1.20 µg/ml. The prepared samples were vortexed and heated at 95°C for 5 min.

After the elapsed time, the solutions were transferred to an ice bath for 1 min. and immediately examined spectrophotometrically at 418 nm. All measurements were performed in duplicate.

The standard curve for the sulfamethoxazole (S)

11 mg sulfamethoxazole (Sigma-Aldrich, CAS: 723-46-6, Burlington, MA, United States) was accurately reconstituted, which was then dissolved in 10 mL PBS (pH = 7.4) to give a final concentration of 1.1mg/mL. The sulfamethoxazole dilutions in the range of 0.0042-1.1 mg/mL were prepared from the stock solution and analyzed *via* high-performance liquid chromatography using a Waters 2707 instrument (Waters 1525 Binary HPLC Pump) with a vacuum degasser. A detector with a diode array (Waters 2489 UV/Visible Detector) was used to uncover the signal of substance. The reverse-phase column Ascentis® C8 Cat#: 581425-U (25 mm × 4.6 mm, 5 µm) was used for separation. HPLC-grade solvents were employed throughout the analysis: acetonitrile (Merck, UK) with 0.05% trifluoroacetic acid and water with 0.05% trifluoroacetic acid. The sample injection volume was 15 µl, the flow rate was 1 ml/min, and the wavelength for detection was 280 and 254 nm. The retention time for sulfamethoxazole under the described conditions was an average of 13.38 minutes. All measurements were performed in duplicate.

Supplemental figures:

Figure S1

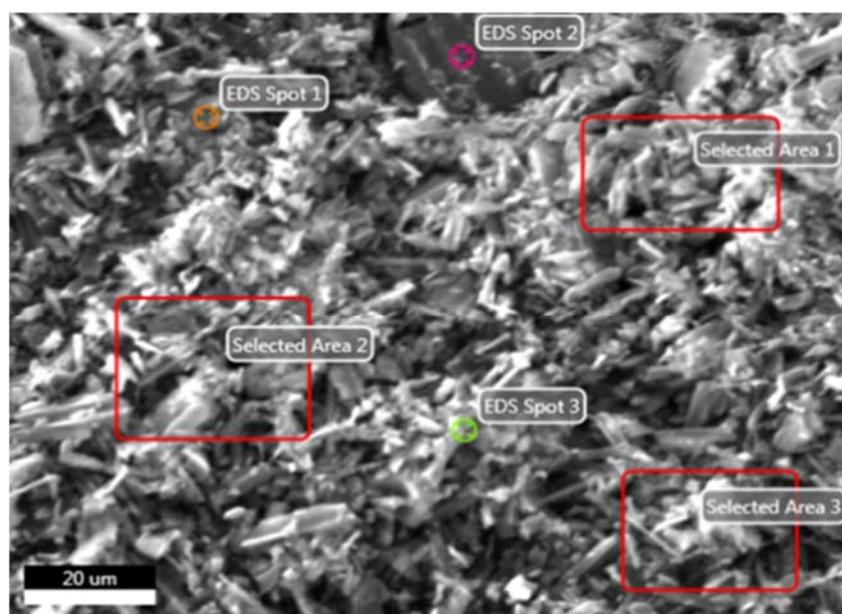
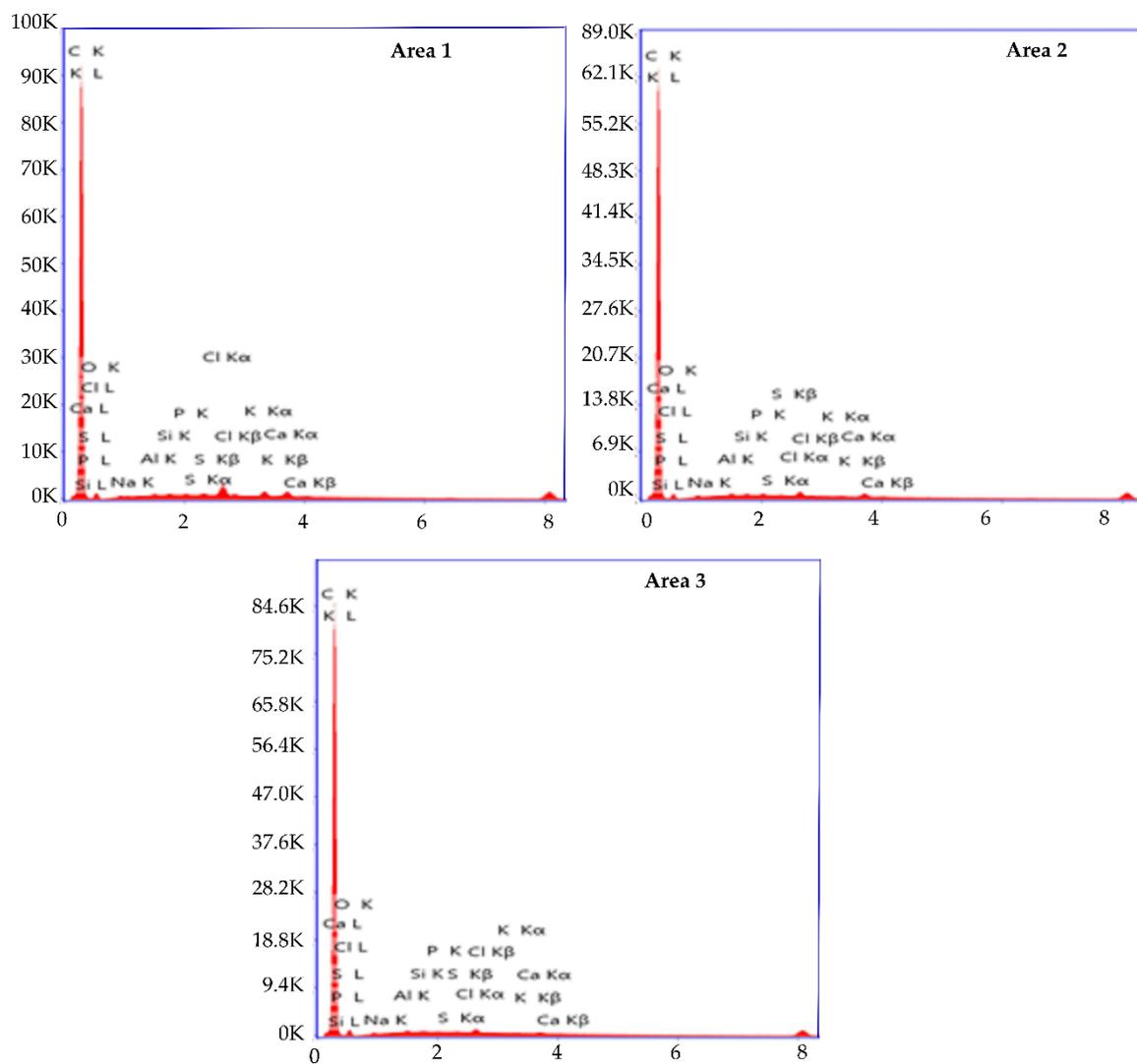


Figure S1. The microscopy picture of the area on PAC surface selected to elemental content analysis. The red windows reveal an area 1-3 on PAC surface. Orange, pink, and green spots show the EDS of 1, 2, and 3, respectively. The areas 1-3 and spots 1-3 of PAC surface were selected to elemental content determination.

Figure S2



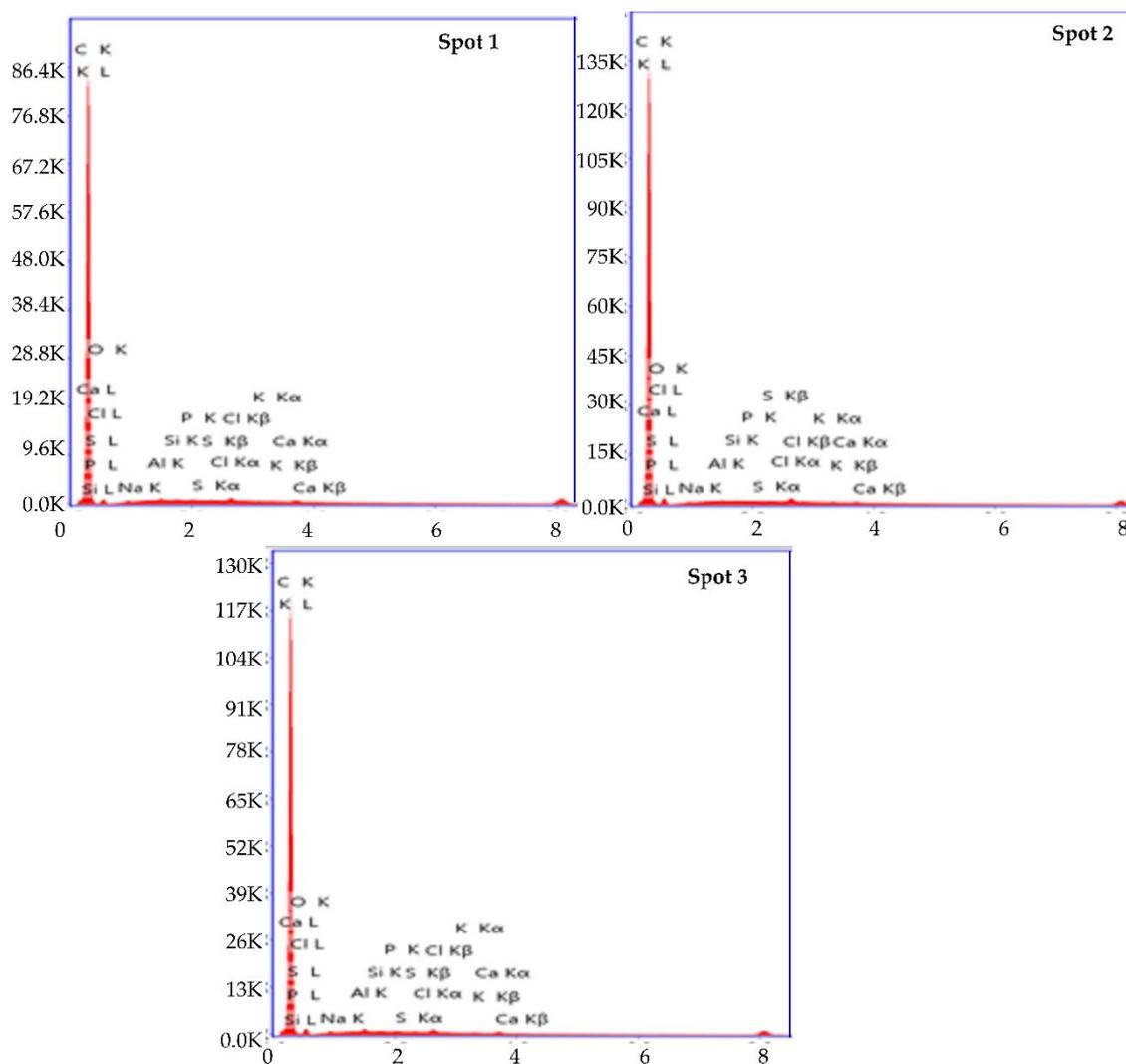


Figure S2. The results of the content of elements for individual selected areas 1-3 and EDS spots 1-3

Figure S3

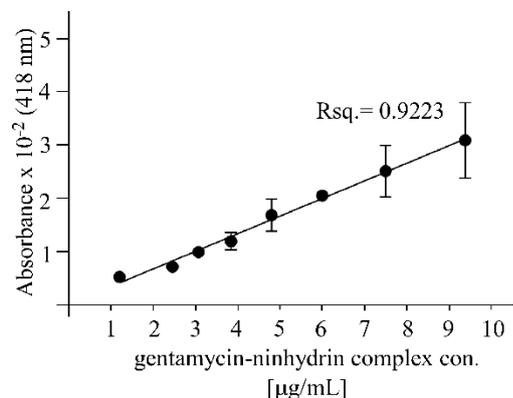


Figure S3. The standard curve for the gentamicin (G)-ninhydrin complex

Figure S4

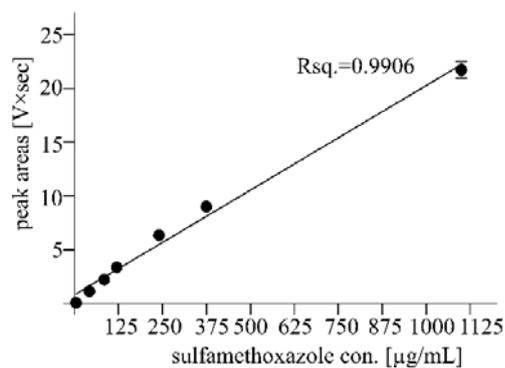


Figure S4 The standard curve for the sulfamethoxazole (S)

Figure S5

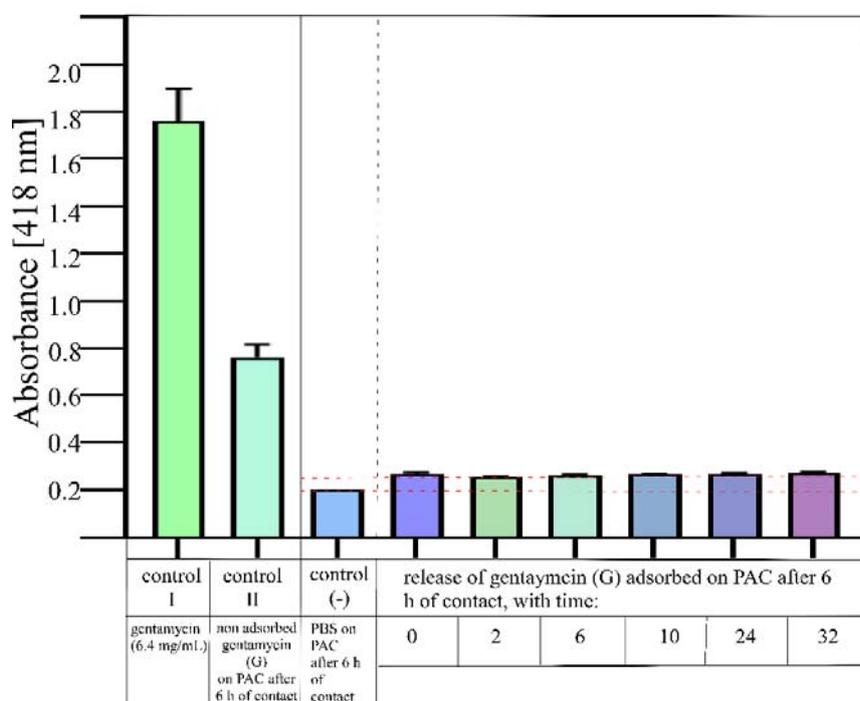


Figure S5 Release of gentamycin sulfate from PAC under aqueous conditions.

Table S1 The results of intelligent eZAF calculations of the content of elements for individually selected areas 1-3 and EDS spots 1-3

Element	Weight %						Atomic %					
	A1	A2	A3	S1	S2	S3	A1	A2	A3	S1	S2	S3
C	92.77	94.04	93.31	94.87	93.2	93.42	95.07	95.84	95.19	96.34	94.99	95.2
O	5.65	4.87	5.84	4.44	6.27	5.9	4.35	3.73	4.47	3.39	4.8	4.52
Na	0.17	0.2	0.13	0.11	0.08	0.21	0.09	0.11	0.07	0.06	0.05	0.11
O	0.05	0.12	0.11	0.09	0.06	0.13	0.02	0.05	0.05	0.04	0.03	0.06
Si	0.07	0.09	0.08	0.06	0.05	0.03	0.03	0.04	0.03	0.03	0.02	0.02

P	0.09	0.12	0.09	0.08	0.05	0.05	0.04	0.05	0.03	0.03	0.02	0.02
S	0.11	0.08	0.09	0.06	0.05	0.03	0.04	0.03	0.03	0.02	0.02	0.01
Cl	0.53	0.21	0.18	0.14	0.16	0.12	0.18	0.07	0.06	0.05	0.05	0.04
K	0.28	0.08	0.08	0.05	0.04	0.03	0.09	0.02	0.02	0.02	0.01	0.01
Ca	0.29	0.2	0.09	0.09	0.05	0.07	0.09	0.06	0.03	0.03	0.01	0.02

A1-3 MEANS AREAS 1-3, S1-3 MEANS SPOTS 1-3

Table S2 Filtration effectiveness of control samples: bacterial cultures *E. coli* and *S. aureus* and activated carbon PAC with LB medium.

CON	A 600 nm	
	Before filtration	After filtration
PAC in LB	0.044	0.045
<i>S. aureus</i> cultured	0.976	0.994
<i>E. coli</i> cultured	0.672	0.616

CON: control samples; A 600 nm: absorbance measured in 600 nm