Supplementary Information

Repurposing Mesalamine *via* Peptide-Functionalized Zeolitic Imidazolate Framework-8 (ZIF-8) Nanoparticles for Selective Breast Cancer Targeting

Nur Airin Syahira Johari[1], Mostafa Yousefzadeh Borzehandani[2], Gayaatri Silvaraju[1], Mohd Akmal Azhar[1], Norazalina Saad[3], Rosniza Razali[4], Nurul Akmarina Mohd Abdul Kamal[1]*

[1]PharmTech Cluster, Faculty of Chemical and Process Engineering Technology, Universiti Malaysia Pahang Al-Sultan Abdullah (UMPSA), 26300 Gambang, Pahang, Malaysia
[2]Department of Chemistry and Key Laboratory for Preparation and Application of Ordered Structural Materials of Guangdong Province, Shantou University, 515063 Guangdong, China
[3]UPM-MAKNA Cancer Laboratory, Institute of Bioscience, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia
[4]Medical Technology Division, Malaysian Nuclear Agency, 43000 Kajang, Selangor, Malaysia

*Corresponding author: nurulakmarina@umpsa.edu.my

Table S1. BET surface area, pore volume and diameter of nZIF-8, MES@nZIF-8, and MES-RGD@nZIF-8 determined from N_2 adsorption–desorption isotherms.

Nanoparticles	BET Surface area ($m^2 g^{-1}$)	Pore volume ($cm^3 g^{-1}$)		Pore diameter (nm)	
		Adsorption	Desorption	Adsorption	Desorption
nZIF-8	1,174.44	0.4372	0.5407	1.4889	1.8416
MES@nZIF-8	871.49	0.3204	0.3984	1.4708	1.8288
MES-RGD@nZIF-8	206.29	0.0711	0.0940	1.3776	1.8232

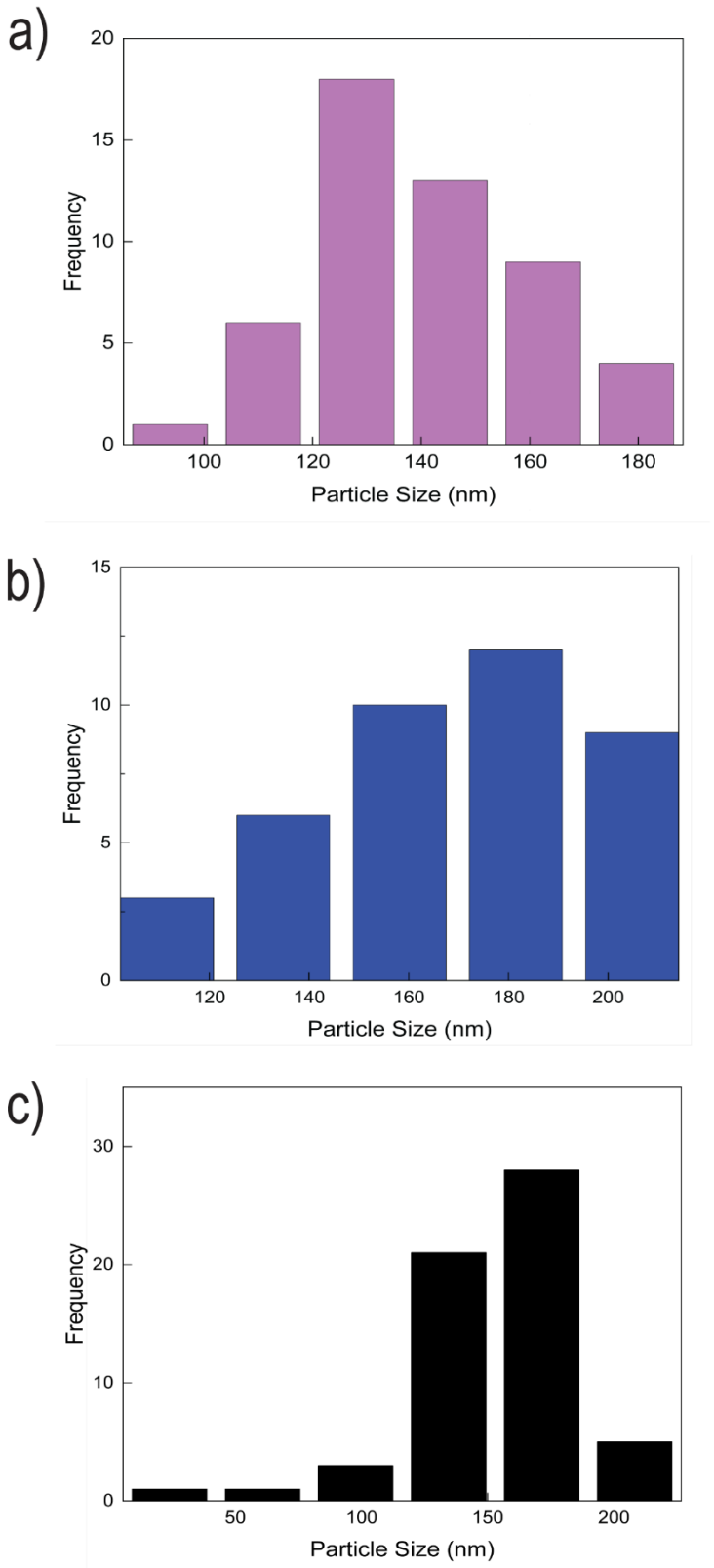


Figure S1. Particle size distribution of synthesized nanoparticles determined from FE-SEM images, which a) nZIF-8, b) MES@nZIF-8, and c) MES-RGD@nZIF-8.

UHPLC Method and Quantification of Mesalamine (MES)

UHPLC Operating Conditions

UHPLC analysis was performed using an Agilent 1290 Infinity II system equipped with a UV detector. Separation was achieved on a reversed-phase C18 column (Accucore, 150 × 4.6 mm, 2.6 µm) maintained at 25 °C. The mobile phase consisted of acetonitrile and ultrapure water (ACN: UPW, 40:60, v/v) delivered at a flow rate of 0.2 mL min^{-1}. The injection volume was 3 µL, and MES was detected at 330 nm. The total run time was 7 min per injection, with MES eluting at approximately 5.18 min in DMSO and 5.85 min in PBS under these conditions (Figure S2).

Calibration Curve

Two independent calibration curves for MES (Figure S3) were constructed to ensure accurate quantification under different experimental conditions. For encapsulation efficiency and loading capacity determination, MES standard solutions were prepared in dimethyl sulfoxide (DMSO) over the concentration range of 0–200 µg mL^{-1} to ensure complete solubilization of the extracted drug. The DMSO-based calibration curve exhibited good linearity, with a regression equation of $y = 56.928x + 207.1$ and a correlation coefficient of $R^2 = 0.9962$. For in vitro drug release studies, MES standards were prepared in phosphate-buffered saline (PBS, pH 5.0) over the same concentration range (0–200 µg mL^{-1}) to match the release medium. The PBS-based calibration curve showed excellent linearity, with a regression equation of $y = 63.458x + 148.83$ and a correlation coefficient of $R^2 = 0.9993$ (Figure S4). These calibration curves were used for quantitative determination of MES in post-centrifugation supernatants for encapsulation efficiency, loading capacity, and in vitro release studies.

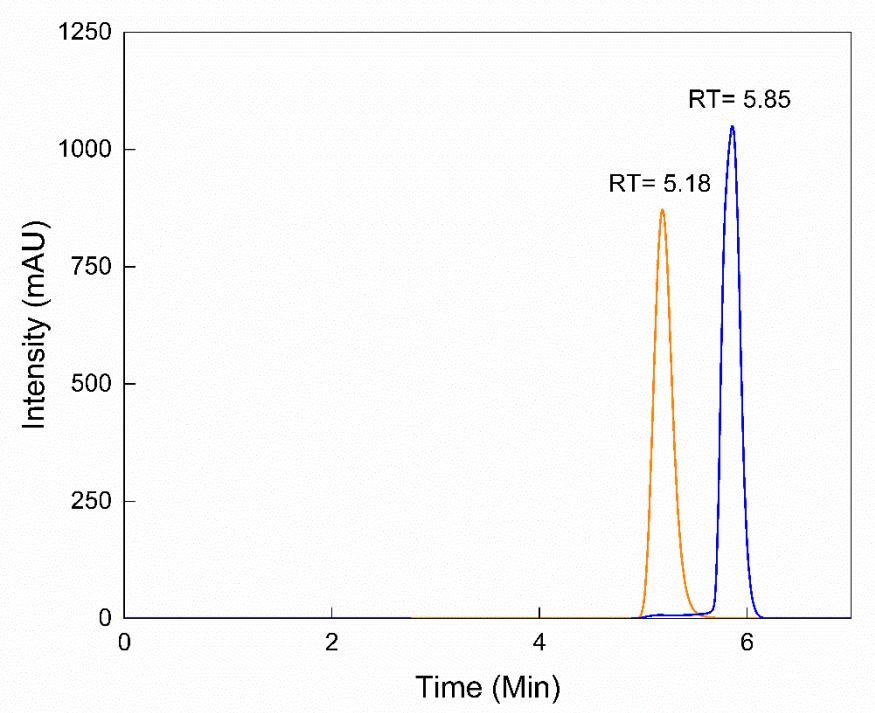
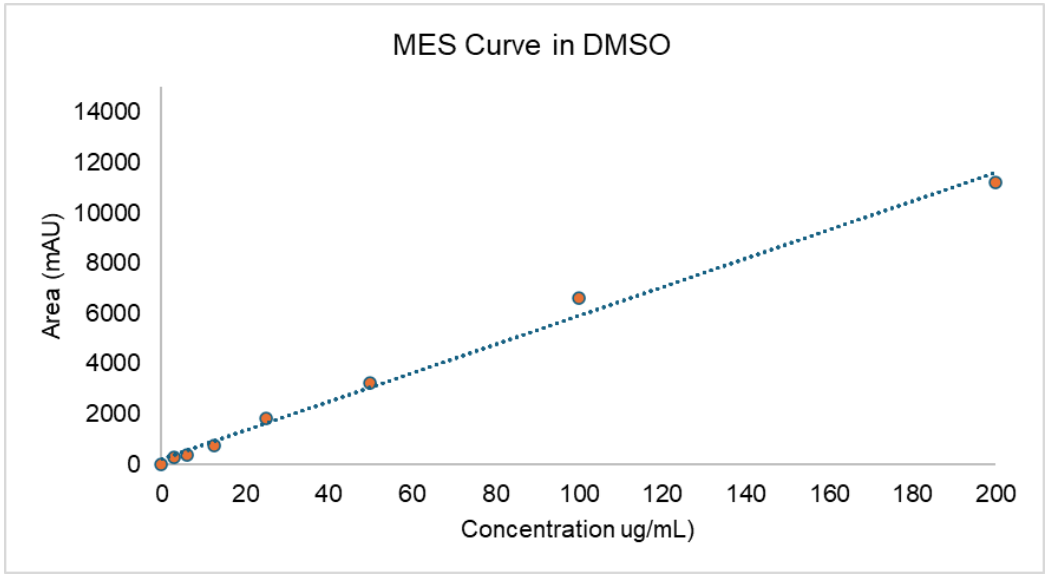


Figure S2. UHPLC chromatograms of mesalamine (MES) standards showing retention times of 5.18 min (DMSO) and 5.85 min (PBS).

a)



b)

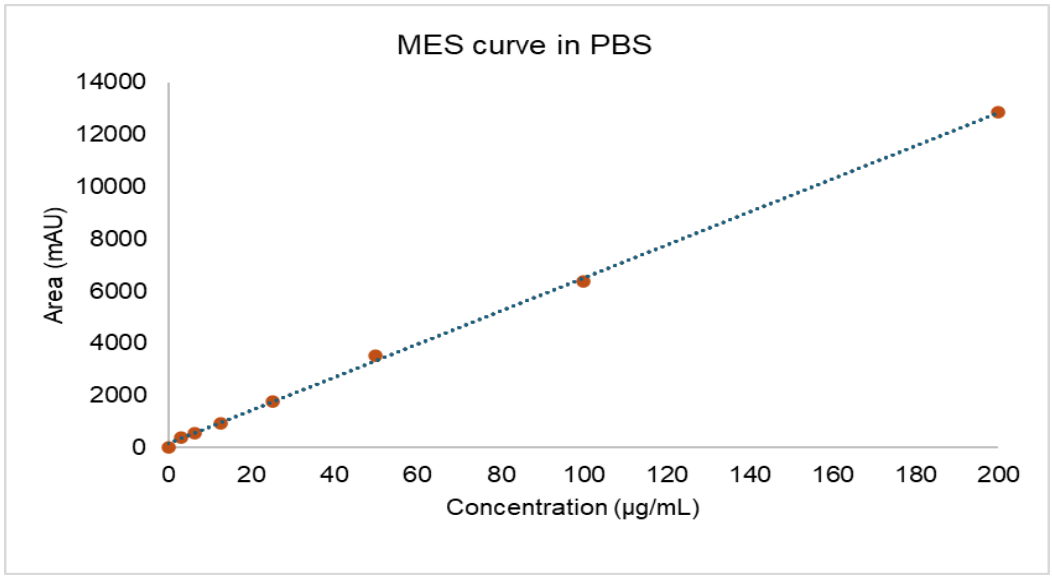


Figure S3. MES calibration curves prepared in a) DMSO and b) PBS showing linear relationships between peak area and concentration under identical UHPLC conditions.

Table S2. Similarity factor (f_2) values comparing the release profiles of MES@nZIF-8 and MES-RGD@nZIF-8 at pH 7.4 and 5.5. f_2 values between 50 and 100 indicate profile similarity.

Medium	Comparison	f_2 values
7.4	MES@nZIF-8 x MES-RGD@nZIF-8	48.8
5.5	MES@nZIF-8 x MES-RGD@nZIF-8	36.4

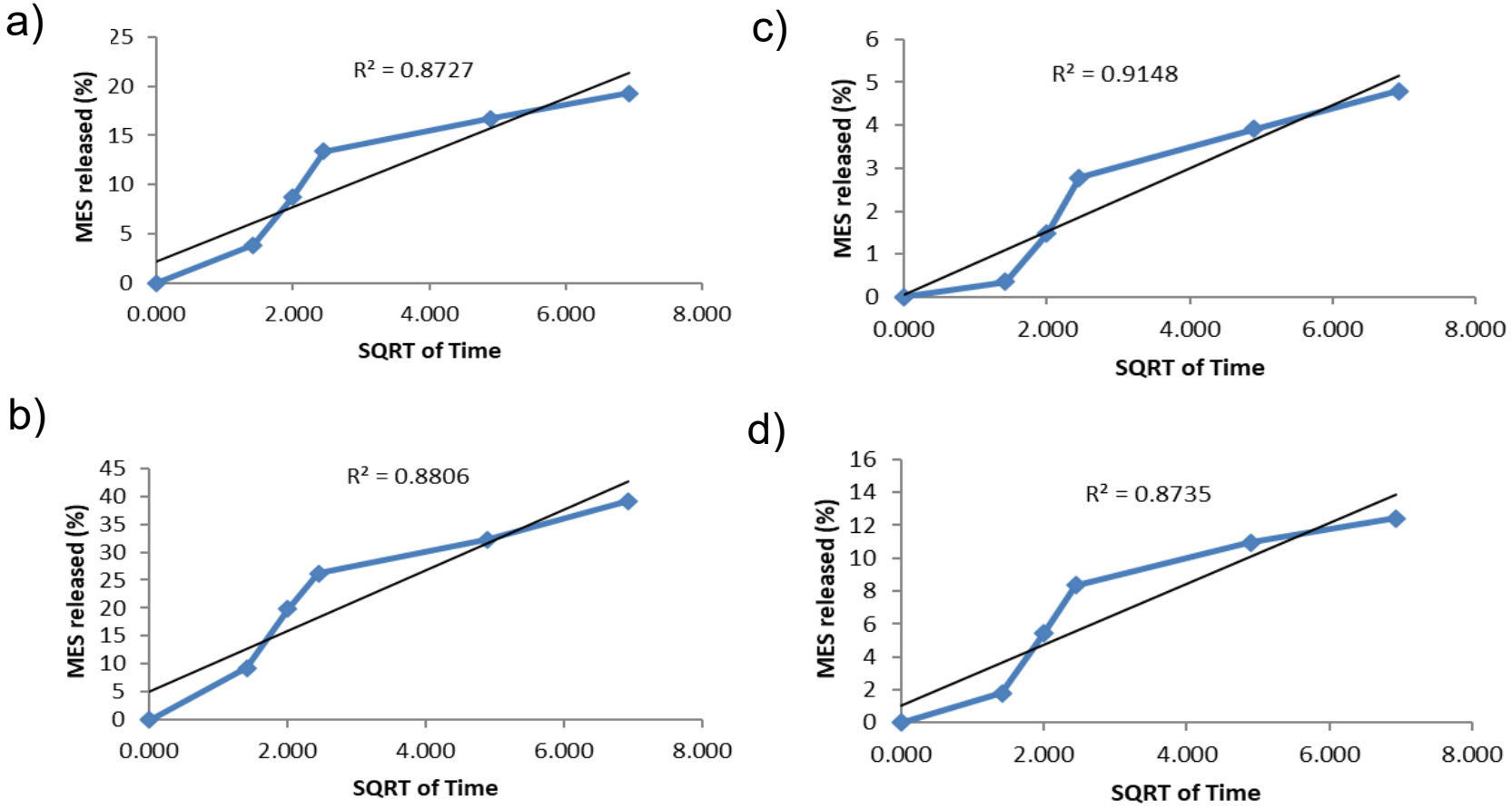


Figure S4. Higuchi kinetic model fitting for MES release from a) MES@nZIF-8 and b) MES-RGD@nZIF-8 at pH 7.4, and c) MES@nZIF-8 and d) MES-RGD@nZIF-8 at pH 5.5, showing diffusion-controlled release behavior.