Supplementary materials for the article:

Blood-Nanoparticle Interactions Create a Brain Delivery Superhighway for Doxorubicin

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Table S1. Summary of the formulations used for modeling the distribution and elimination of free doxorubicin and nanodelivery systems.

Formulation	Dose	Study details	Ref.
Doxorubicin solution	Aqueous solution ($5\ mg \cdot kg^{-1}$)	Healthy male Wistar rats Bodyweight 180-200 g Plasma pharmacokinetics (*n* = 5-8) Brain/plasma distribution (*n* = 5-8)	[1]
Doxorubicin solution	Aqueous solution ($6\ mg \cdot kg^{-1}$)	Female Fisher rats (methylcholantrene-induced histiocytoma) Bodyweight 180–200 g Plasma Pharmacokinetics (*n* = 4)	[2]
PBCA nanoparticle	270 nm ($5\ mg \cdot kg^{-1}$)	Healthy male Wistar rats Bodyweight 180-200 g Plasma pharmacokinetics (*n* = 5-8) Kidney/brain distribution (*n* = 5-8)	[1]
Doxil®	91.8 nm ($6\ mg \cdot kg^{-1}$)	Female Fisher rats (methylcholantrene-induced histiocytoma) Bodyweight 180–200g (*n* = 4)	[2]

Table S2. Parameter estimates used for the *in-silico* analysis of the nanodelivery systems with the biodistribution model

Study design	Parameter	Range	Ref.
Doxil® (6 mg·kg^{-1}) in female Fisher rats	V_{DC} (L)	6.57-8.94	[3]
	V_{DF} (L)	27.94-31.18	[4]
	k_{T_in} (h^{-1})	0.000001-0.1	Estimation
	k_{T_out} (h^{-1})	0.000001-0.1	Estimation
	k_{Peri_in} (h^{-1})	8.78-9.6	[4]
	k_{Peri_out} (h^{-1})	0.035-0.042	[4]
	k_{B_in} (h^{-1})	0.000001-0.1	Estimation
	k_{B_out} (h^{-1})	0.000001-0.1	Estimation
	Body weight (g)	180-200	[4]
	Tumor weight (g)	0.015*	[4]
	f_{brain} (%)	1.239-1.323	NBRP Koyoto
PBCA nanoparticles (5 mg·kg^{-1}) in healthy male Wistar rats	V_{DC} (L)	6.624-9.52	[3]
	V_{DF} (L)	1.64-224.54	[5]
	k_{Peri_in} (h^{-1})	0.0206-1.4	[5]
	k_{Peri_out} (h^{-1})	0.0756-0.380	[5]
	k_{B_in} (h^{-1})	0.000001-0.1	Estimation
	k_{B_out} (h^{-1})	0.000001-0.1	Estimation
	Body weight (g)	180-200	[5]
	f_{brain} (%)	0.963-1.245	NBRP Koyoto

The asterisk indicates that a fixed value was set.

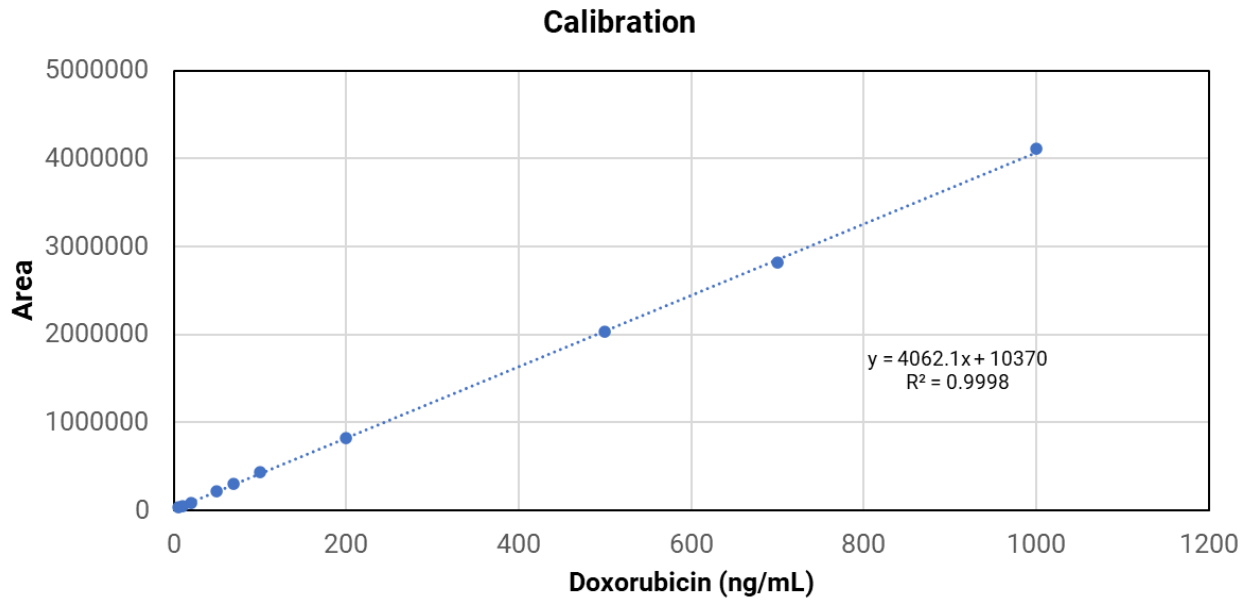

Figure S1. The calibration curve of doxorubicin measured by the HPLC method.

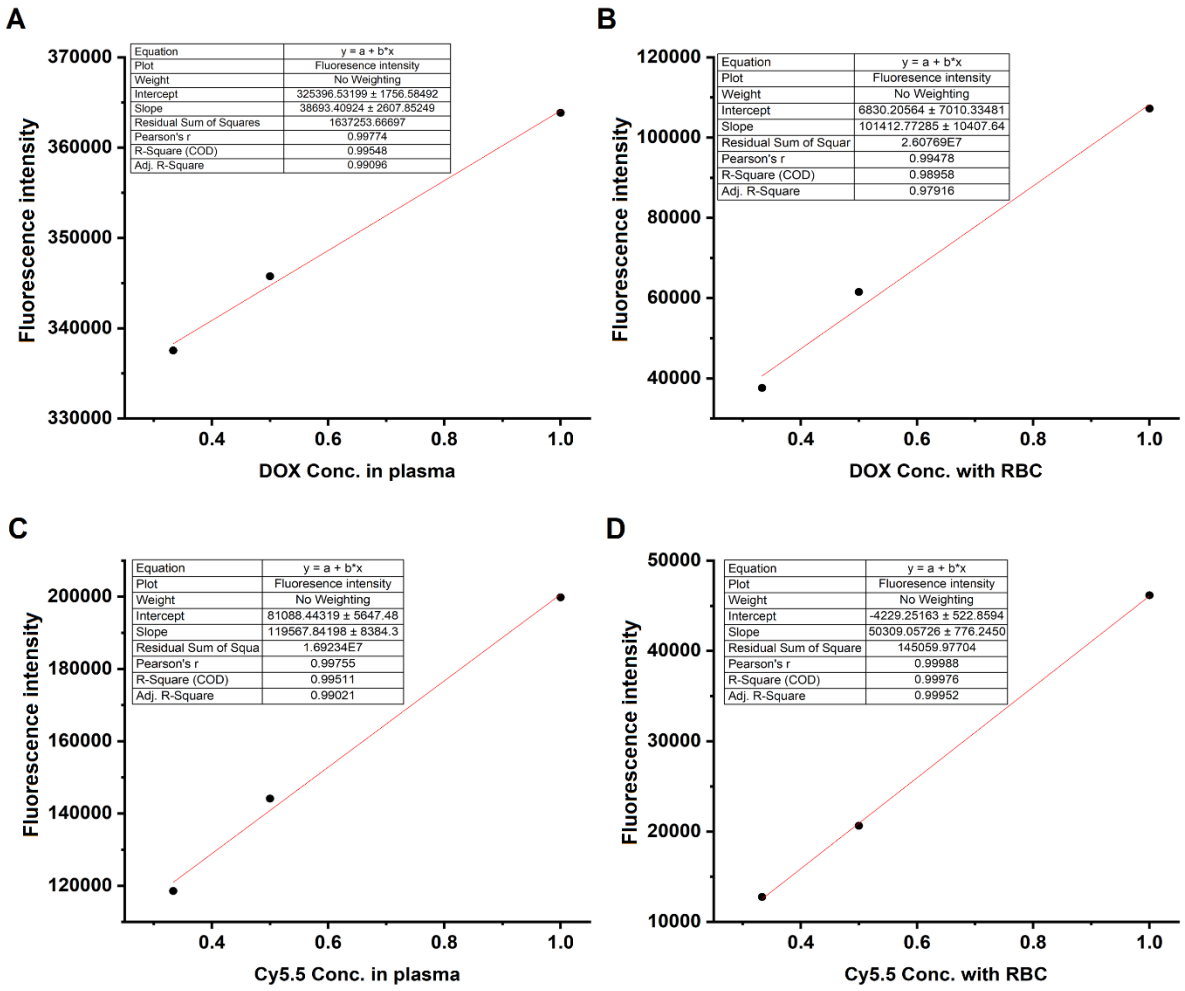

Figure S2. Calibration curves of fluorescence intensity of doxorubicin in the presence of plasma (A) and red blood cells (B), and Cy5.5 in the presence of plasma (C) and red blood cells (D).

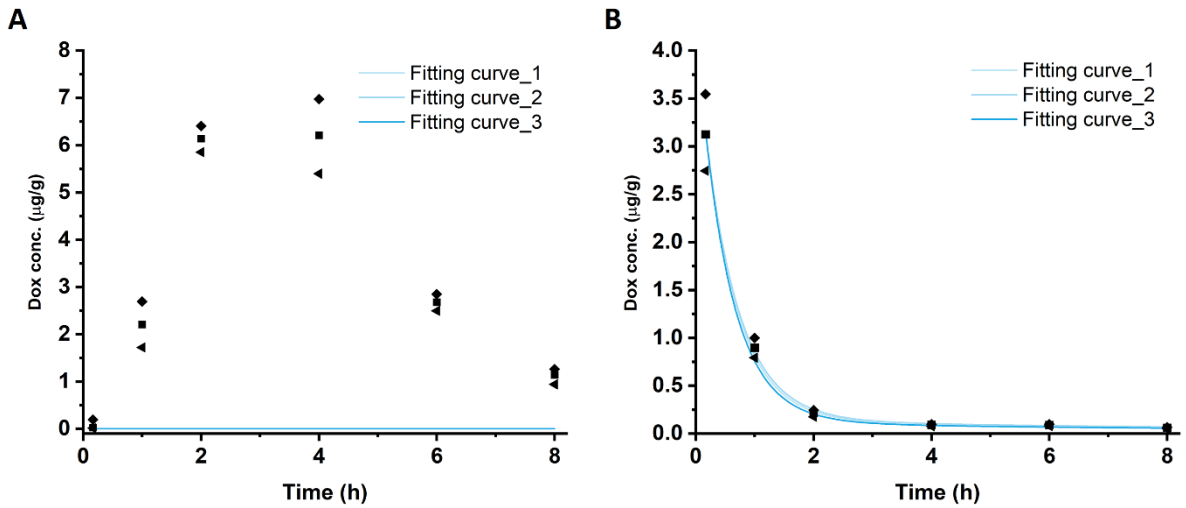

Figure S3. Simulated doxorubicin concentration-time profiles following intravenous administration of PBCA nanoparticles. Overlay between the predicted and the observed concentrations in (A) plasma and (B) brain for the pharmacokinetics observed by Gulyaev et al.. The observed upper (◆), mean (■) and lower (◀) values were fitted, respectively.

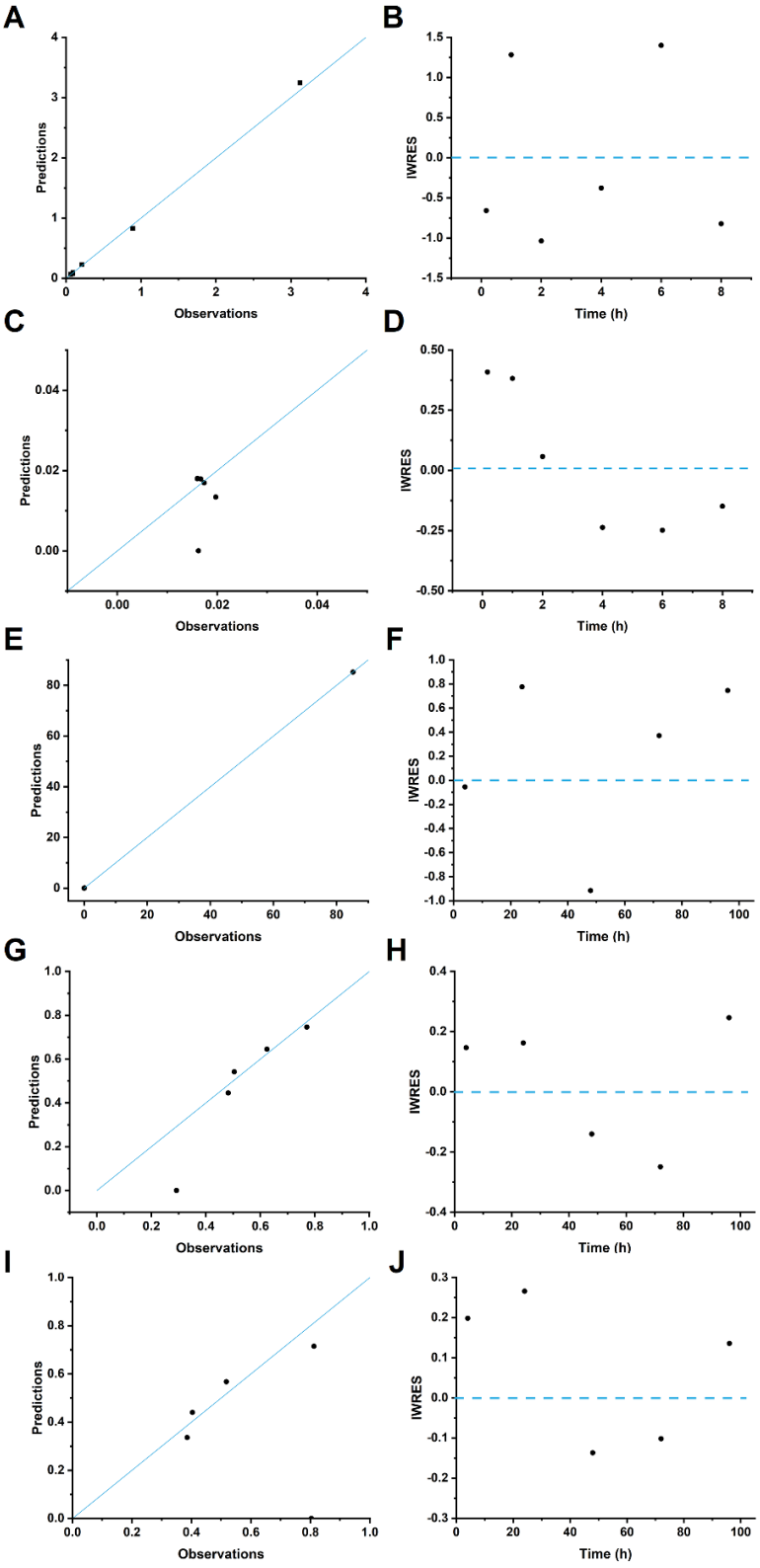

Figure S4. Diagnostic plots for compartmental models simulating doxorubicin concentrations following administration of free doxorubicin. Observed doxorubicin concentrations in (A) plasma and (C) brain obtained by Gulyaev et al., and (E) plasma, (G) brain, and (I) tumor obtained by Siegal et al. versus the individual predicted concentrations (with y = x line as the reference line). Individual weighted residual (IWRES) versus time for (B) plasma and (D) brain obtained by Gulyaev et al., and (F) plasma, (H) brain, and (J) tumor obtained by Siegal et al.

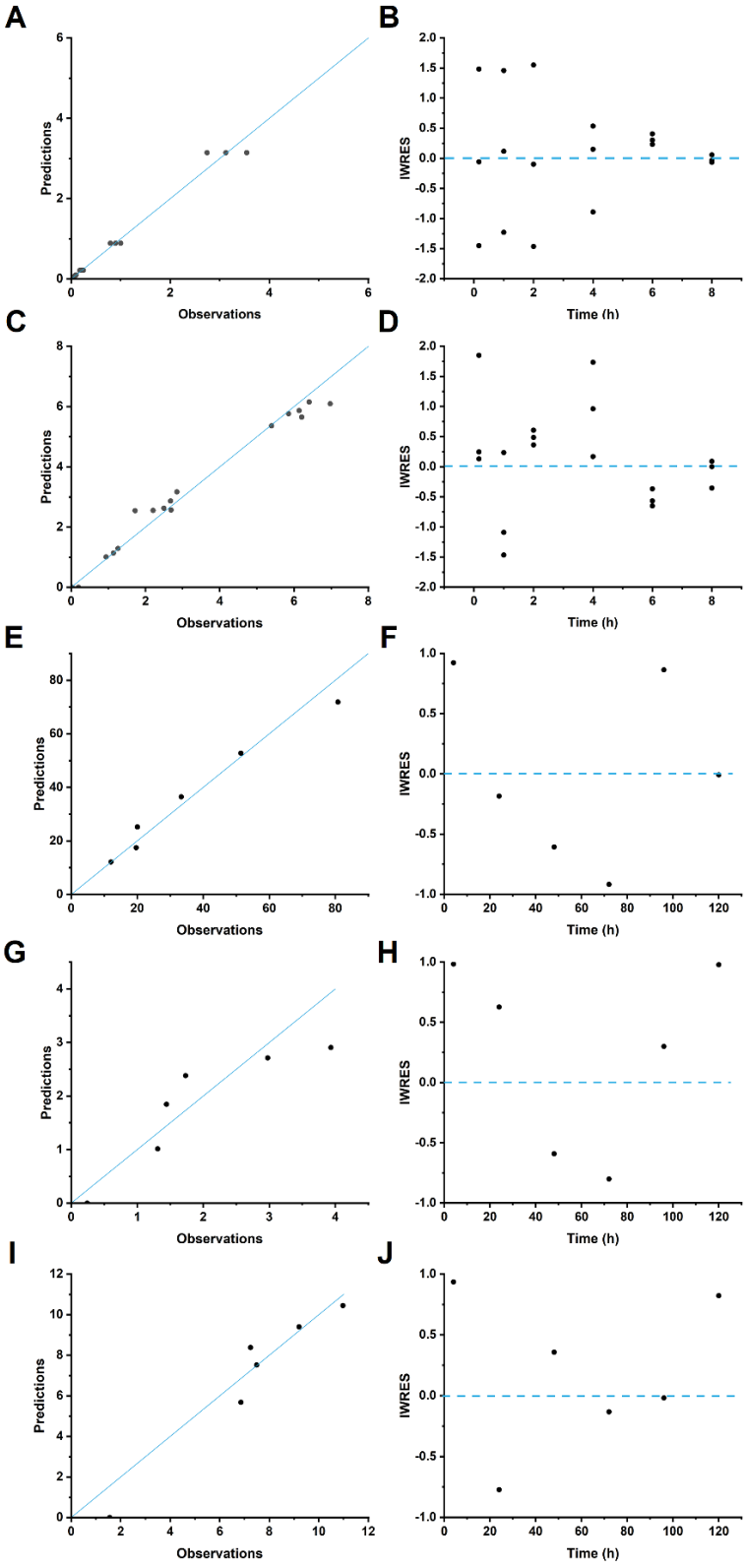

Figure S5. Diagnostic plots for the physiologically-based nanocarrier biopharmaceutics models simulating doxorubicin concentrations following administration of nanodelivery systems. Observed doxorubicin concentrations in (A) plasma and (C) brain obtained by Gulyaev et al., and (E) plasma, (G) brain, and (I) tumor obtained by Siegal et al. versus the individual predicted concentrations (with y = x line as the reference line). Individual weighted residual (IWRES) versus time for (B) plasma and (D) brain obtained by Gulyaev et al., and (F) plasma, (H) brain, and (J) tumor obtained by Siegal et al.

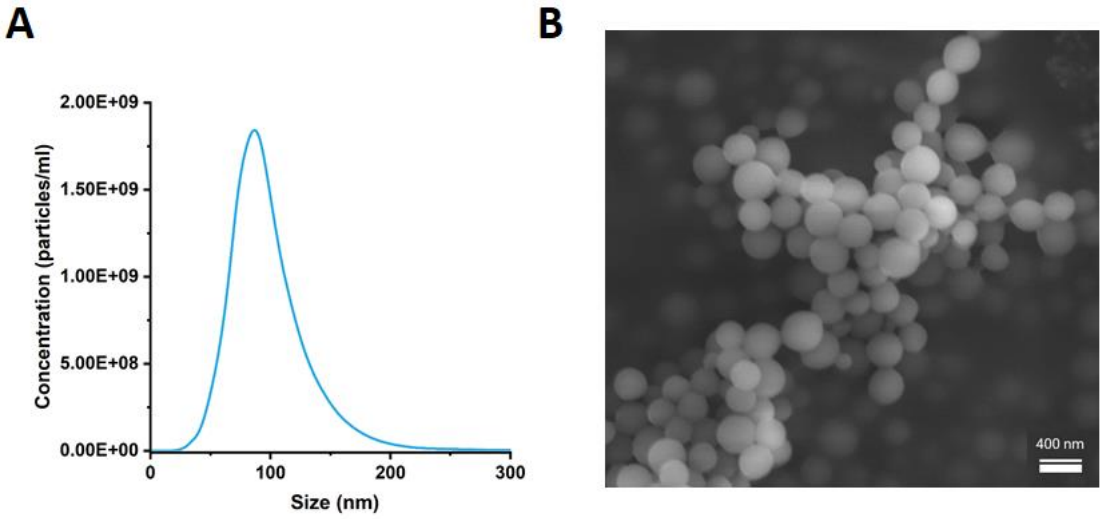

Figure S6. Characteristics of PBCA nanoparticles. (A) Concentration of PBCA nanoparticles in water at 37°C measured by nanoparticle tracking analysis. (B) Scanning electron micrograph of PBCA nanoparticles recorded with a TESCAN MIRA3 at 5 kV at 45,700-fold magnification.

References

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[2] T. Siegal, A. Horowitz, A. Gabizon, Doxorubicin encapsulated in sterically stabilized liposomes for the treatment of a brain tumor model: biodistribution and therapeutic efficacy, J Neurosurg, 83 (1995) 1029-1037.
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