Supplementary

A Chelate-Free Nano-platform for Incorporation of Diagnostic and Therapeutic Isotopes

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Figure S1 Experimental set-up for [90 Y]Y-FH and [177 Lu]Lu-FH radiolabeling: **a,b**) [90 Y]YCl₃ source and the prepared reaction mixtures; **c,d**) heating vortex system for heating and stirring reaction mixtures; The heating vortex technique was developed by integrating an orbital shaker with a silicon oil bath heater. The vials containing the reaction mixtures were placed inside the tube holder in the silicon oil bath and an applied force pressed the vials into the bottom of the oil bath so the motion is transmitted to the reaction mixture and a vortex is created. The heating vortex technique was used for heating, cooling and quenching the [90 Y]Y-FH, [177 Lu]Lu-FH and [64 Cu]Cu-FH reaction mixtures; **e,f**) the [177 Lu]LuCl₃ source vial and prepared reaction mixtures.

Abbreviations: FH, Feraheme.



Figure S2 SEC analyses for [⁹⁰Y]Y-FH, [¹⁷⁷Lu]Lu-FH and [⁶⁴Cu]Cu-FH by PD-10: **a)** PD-10 column with loaded [⁹⁰Y]Y-FH reaction mixture and separation of [⁹⁰Y]Y-DFO complex from [⁹⁰Y]Y-FH; similar procedure was applied to separate the [¹⁷⁷Lu]Lu-DFO and [⁶⁴Cu]Cu-DFO complexes; **b,c,d)** collected volume fractions (e.g. 200 μ L) from [^{9b}Y]Y-FH, [¹⁷⁷Lu]Lu-FH and [⁶⁴Cu]Cu-FH elution, respectively (excluding the dead volume fractions). The arrows indicates the direction of collection.

Abbreviations: SEC, size exclusion chromatography; DFO, deferoxamine mesylate salt; FH, Feraheme.



Figure S3 PD-10 purification and radio-FH product fraction collection: **a,b**) SEC separation of $[^{90}Y]Y$ -FH from $[^{90}Y]Y$ -DFO complex by PD-10 column eluted by saline ; **c,d**) concentrating and collecting the final product using Amicon 30 kDa MC centrifugal filter; **e**) Amicon centrifugal filter after collecting the product showing ignorable iron and activity loss. Similar method was applied for collecting the final $[^{177}Lu]Lu$ -FH and $[^{54}Cu]Cu$ -FH products.

Abbreviations: SEC, size exclusion chromatography; DFO, deferoxamine mesylate salt; FH, Feraheme.



Figure S4 Radio-TLC for [⁹⁰Y]Y-FH reaction mixtures and final product: a) prepared radio-TLCs with \approx 1 µL [⁹⁰Y]Y-FH reaction mixture spotted at origins; b) Developed radio-TLC elutions with chelexed water; separating [⁹⁰Y]Y-FH NPs from [⁹⁰Y]Y-DFO complex. Similar method was applied for [⁹⁰Y]Y-FH final product, [⁶⁴Cu]Cu-FH and [¹⁷⁷Lu]Lu-FH reaction mixture and final product samples.

Abbreviations: radio-thin layer chromatography, radio-TLC; DFO, deferoxamine mesylate salt; FH, Feraheme.



Figure S5 Attachment of FH nanoparticles to the magnetic stirrer bars post HIR. It could be recovered by washing with loading buffers to maximize RCY.

Abbreviations: FH, Feraheme; RCY, radiochemical yield.

Example for the use of equation 1.

Reaction rate calculation for HIR:

Considering the HIR for the copper-64 isotope with $t_{1/2} = 12.7$ hr, reaction time T = 2 hr and an initial activity of $A_0 = 17.6$ (≈ 18) MBq (e.g. $17.60 \times 10^6 \frac{disintegration}{second}$, however, since all of the variables are in hr this needs to be converted into disintegration per hr: $17.60 \times 10^6 \times 3600 \frac{disintegration}{hr}$) the reaction rate can be calculated using Eq.1:

$$\dot{R}(t) = \frac{\tilde{A}\int_{0}^{T} e^{-\lambda t} dt}{T} = \frac{\frac{A_{0}}{\ln(0.5)} \times t_{1/2} \int_{0}^{T} e^{-\ln(0.5)\frac{t}{t_{1/2}}} dt}{T}$$
$$\frac{\frac{17.6 \times 10^{6} \times 3600}{\ln(0.5)} \times 12.7 \times \frac{e^{-\ln(0.5)\frac{2}{12.7}} - 1}{\frac{\ln(0.5)}{12.7}}}{2}$$

$$\dot{R}(t) = 1.22 \times 10^{12} hr^{-1}$$

Reaction rate calculation for HRR-HIR:

We can similarly calculate the $\dot{R}(t)$ for HRR-HIR with T = 1 hr, and $A_0 = 22.2 ~(\approx 22)$ MBq (e.g. $22.2 \times 10^6 \frac{disintegration}{second}$ or $22.2 \times 10^6 \times 3600 \frac{disintegration}{hr}$)

$$\dot{R}(t) = \frac{\tilde{A} \int_{0}^{T} e^{-\lambda t} dt}{T} = \frac{\frac{A_{0}}{\ln(0.5)} \times t_{1/2} \int_{0}^{T} e^{-\ln(0.5) \frac{t}{t_{1/2}}} dt}{T}$$
$$\frac{\frac{22.2 \times 10^{6} \times 3600}{\ln(0.5)} \times 12.7 \times \frac{e^{-\ln(0.5) \frac{1}{12.7} - 1}}{\frac{\ln(0.5)}{12.7}}}{1}$$
$$\dot{R}(t) = \frac{1.50 \times 10^{12} hr^{-1}}{1}$$

Based on the above calculations, we can see the reaction rate for HRR-HIR is increase by $\approx 19\%$.