Optimization of Pegylated Iron Oxide Nanoplatforms for Antibody Coupling and Bio-targeting.

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Supporting Information. Hydrodynamic diameter (D_H) and zeta potential as a function of pH for bare NPs, after coating with the various PEG ratio x values , and zeta potential curve as function of pH for γ Fe₂O₃ NPs surface functionalized with PO-PEG-CH₃, x=0 (Fig. S1), EDX analysis (Fig S.2) Absorption spectrum (Fig. S3), Fluorescence emission spectra (Fig. S.4), hydrodynamic diameter distribution in volume and intensity measured in water for the nanoplatforms γ Fe₂O₃@PO-PEG₈₅ , with 1 or 2 antibodies mAb-AF488 (Fig. S5), Variation of the average number of PO-PEG per nanoparticle considering a preferential interaction of 1200-PO-PEG-COOH (Table S1), Average number of Ab/NP deduced with fluorescence and OPA titration (Table S2)

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The isoelectric point (IEP) of bare IO NP is around pH 7 and they precipitate at physiological pH, being positively and negatively charged at acid and basic pH, respectively (Fig. S1A). After coating with the various PEG ratio x values (Fig. S1B), it can be observed that the hydrodynamic size is independent of the pH and clearly show that NP diameter is stable for each x ratio.



Fig. S1 Hydrodynamic diameter (D_H in volume, blue line) and zeta potential (red line) as a function of pH for bare NPs (A) and after coating with the various PEG ratio x values (B), zeta potential curve as function of pH for γ Fe₂O₃NPs surface fonctionalized with PO-PEG-CH₃, x=0 (C)



Fig. S2 EDX spectra for γFe_2O_3 @PO-PEG₁₀₀ NPs



Fig. S3 Absorption spectrum of iron oxide NPs ([Np] = 45nM)



Fig. S4 Fluorescence emission spectra of the γ Fe₂O₃@PO-PEG₈₅-Ab-AF88 using two initial ratio R= n Ab-AF488/n NP =2 (blue) or 4 (red). [NP] = 15 nM



Fig. S5 hydrodynamic diameter distribution in number (A) and intensity (B) measured in water (pH 7 and [Fe]=0.25 mM) for the nanoplatforms γ Fe₂O₃@PO-PEG₈₅ without (black) or with 1 (blue) or 2 (red) antibodies mAb-AF488

Table S1. Calculation considering a preferential interaction of 1200-PO-PEG-COOH with NP surface

А	В	С	D	E	F	G
Initial x (%)	Average number of PEG (EDX results)	Theoritical PEG-COOH number (X*515)	Theoritical PEG-CH3 number (X*677)	Number of PEG COOH considering preferential grafting (x*515)	Number of PEGCH3 onto NP surface (B-E)	effective experimental x ratio (100*E/(E+F))
0	677	0	677	0	677	0
15	565	77.25	575.45	77.25	487.75	13.67
50	480	257.5	338.5	257.5	222.5	53.64
85	532	437.75	101.55	437.75	94.25	82.28
100	515	515	0	515	0	100

 Table S2. Average number of Ab/NP deduced with fluorescence and OPA titration

	F	energiane a		
Ri	Fluorescence titration (supernatant)	OPA titration	Quenching Factor	
2	0.9 ± 0.4	1.2 ± 0.3	3.2	
4	2.1 ± 0.2	1.9 ± 0.4	2.9	