Shaping bio-inspired nanotechnologies to target thrombosis for dual opticalmagnetic resonance imaging

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Fig. S1. Bioconjugation reaction schemes for conjugation of contrast agents along with either PEG or targeting ligands to CPMV and TMV. (A) Synthesis of particles for targeting with CREKA. (B) Particle conjugation for targeting with GPRPP. (C) Additional conjugation of Gd(DOTA) for MR imaging before dye and GPRPP attachment.



Fig. S2. Fluorescence intensity measurements of particles. Fluorescence intensity was measured at a concentration of 0.25 mg/mL, with much higher fluorescence observed for CPMV particles compared to TMV particles. This can be explained by presentation of the fluorophores in varying molecular environments and with varying inter-dye distances given by the elongated rod *versus* the icosahedral shaped carrier system. The data were normalized based on the relative fluorescence intensities.



Fig. S3. MALDI-TOF MS spectra. (A) Spectra of native TMV, TMV-PEG, and TMV-CREKA particles used for CREKA studies. (B) Spectra of TMV_{Lys} , TMV-PEG, and TMV-GPRPP particles used for GPRPP studies. Tabulated masses compared to predicted masses can be found in **Table S3**. Note that this is not a quantitative method as different ionization probabilities (and thus different peak intensities) can result from slight changes in structure.¹

Table S1. Measured and theoretical mass for peaks o	obtained b	v MALD	I-TOF MS.
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Sample	Measured Mass	Predicted Mass
TMV	17,558	17,534
TMV-PEG	17,668	17,534
	18,230	18,217
TMV-CREKA	17,564	17,534
	18,877	18,804
TMV _{Lys}	17,609	17,561
TMV-PEG	17,621	17,561
	18,206	18,136
TMV-GPRPP	17,598	17,561
	18,796	18,761



Fig. S4. Relaxivity measurements. Determination of relaxivity of Gd(DOTA)-loaded TMV (**A**) and CPMV (**B**) was based on a multi-slice saturation recovery Look-Locker sequence to measure R_1 *versus* Gd concentration.



Fig. S5. Pharmacokinetics of particles. A647-TMV-GPRPP and A647-TMV-PEG were injected intravenously and fluorescent measurements of serum samples taken at each time point (n = 3). Data were analyzed using Prism software. The half-lives of the particles were 6.9 and 8.3 min, respectively.

References:

1. S. Weidner, G. Kühn and U. Just, *Rapid Commun. Mass Spectrom.*, 1995, 9, 697-702.