

SUPPLEMENTARY INFORMATION

## Enhancers of amyloid aggregation: novel ferrocene-based compounds selective toward amyloid models

*Sara La Manna*<sup>a,\*</sup>, *Concetta Di Natale*<sup>b</sup>, *Valeria Panzetta*<sup>b,c</sup>, *Paolo Antonio Netti*<sup>b,c</sup>, *Antonello Merlino*<sup>d</sup>, *Konrad Kowalski*<sup>e</sup> and *Daniela Marasco*<sup>a,\*</sup>

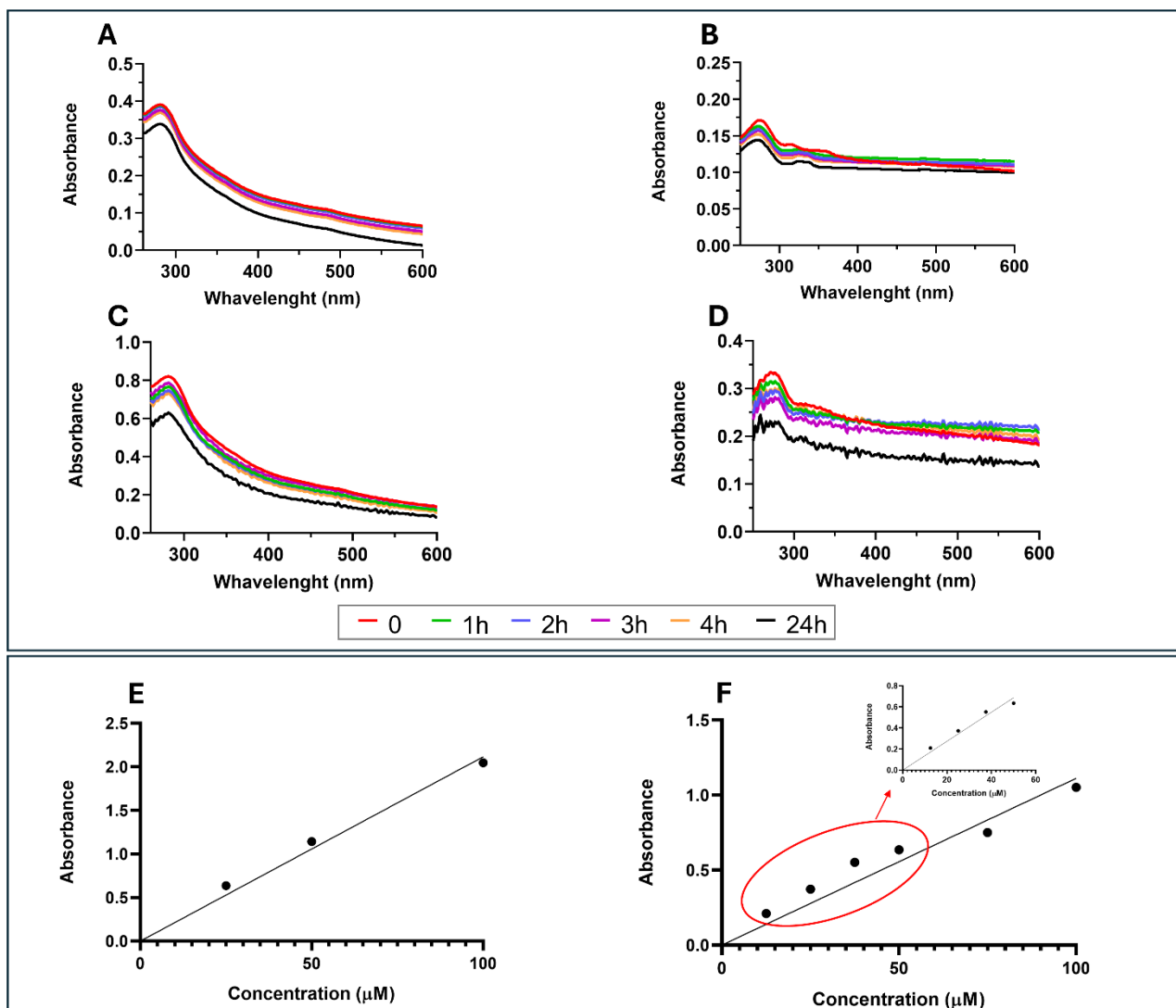
<sup>a</sup>Department of Pharmacy, University of Naples “Federico II”, 80131, Naples, Italy.

<sup>b</sup>Department of Chemical, Materials, and Industrial Production Engineering (DICMaPI), University of Naples Federico II, 80125 Naples, Italy.

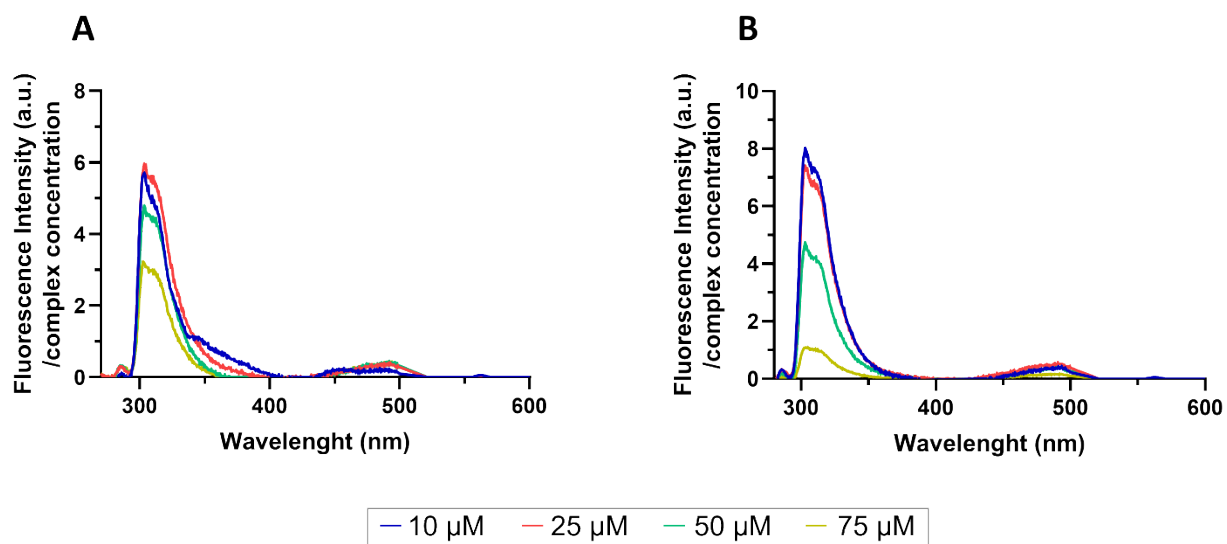
<sup>c</sup>Interdisciplinary Research Centre on Biomaterials (CRIB), University of Naples Federico II, Istituto Italiano di Tecnologia, 80125, Naples, Italy.

<sup>d</sup>Department of Chemical Sciences, University of Naples “Federico II”, 80126, Naples, Italy

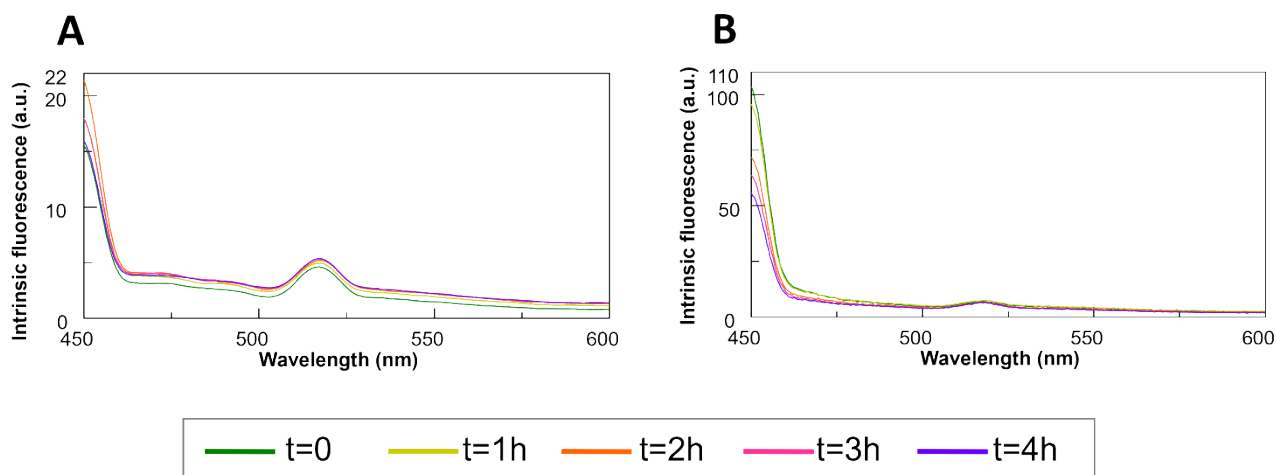
<sup>e</sup>University of Łódź, Faculty of Chemistry, Department of Organic Chemistry, Tamka 12, 91-403 Łódź, Poland.



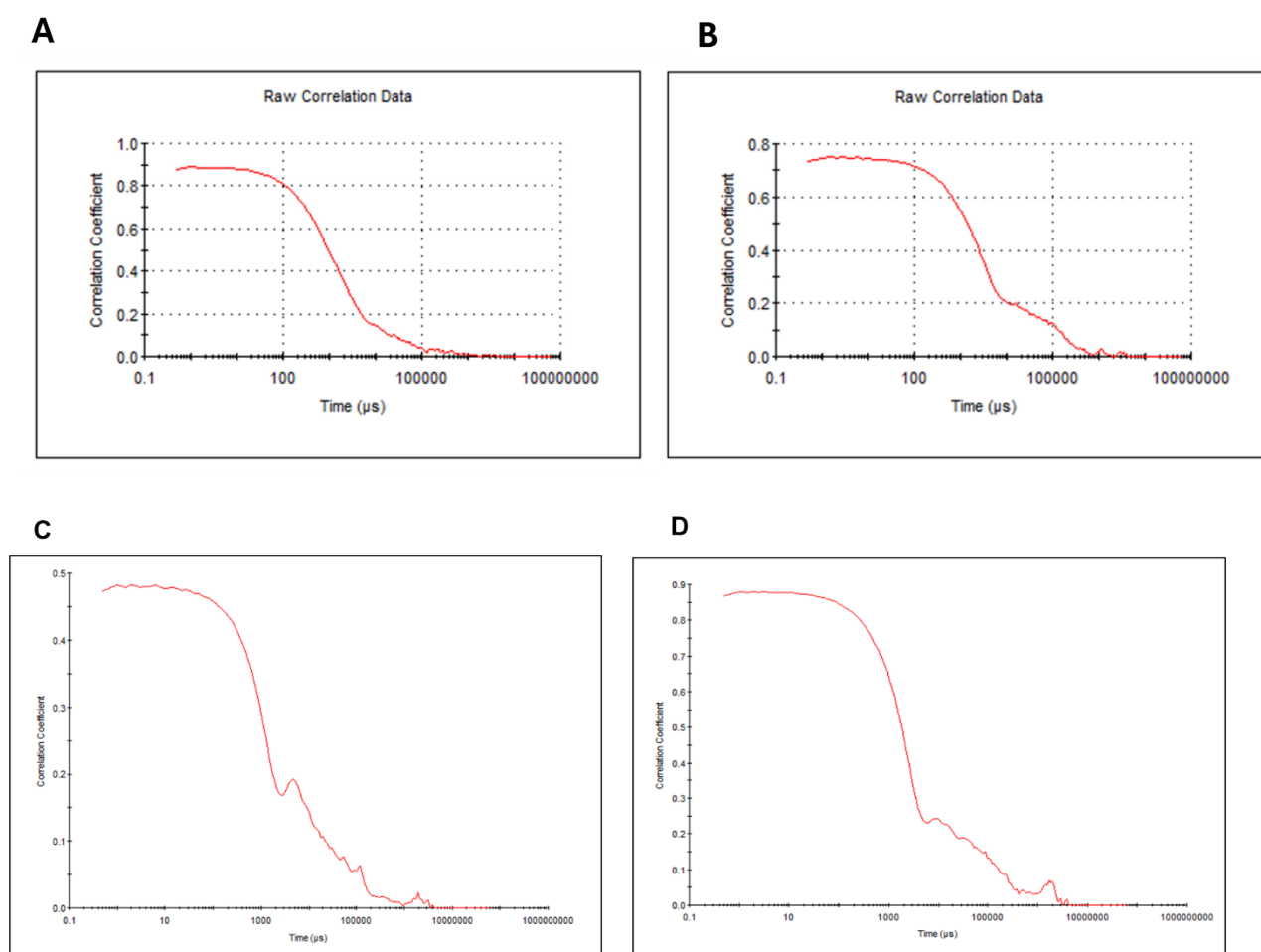
**Figure S1.** (Upper panel) UV-Vis spectra overtime of **mono-T\_Fc** (A, C) and **di-T\_Fc** (B, D): 25  $\mu\text{M}$  (A, B) and 75  $\mu\text{M}$  (C, D) in 10 mM phosphate buffer at pH 7.4 (0.2% DMSO, v/v). (Lower panel) Solubility assays of **mono-T\_Fc** (E) and **di-T\_Fc** (F) in 10 mM phosphate buffer at pH 7.4 (0.2% DMSO, v/v). As inset of panel F, magnification of the region from 0 to 60  $\mu\text{M}$ . The values shown in panels E and F are the average of two separate measurements.



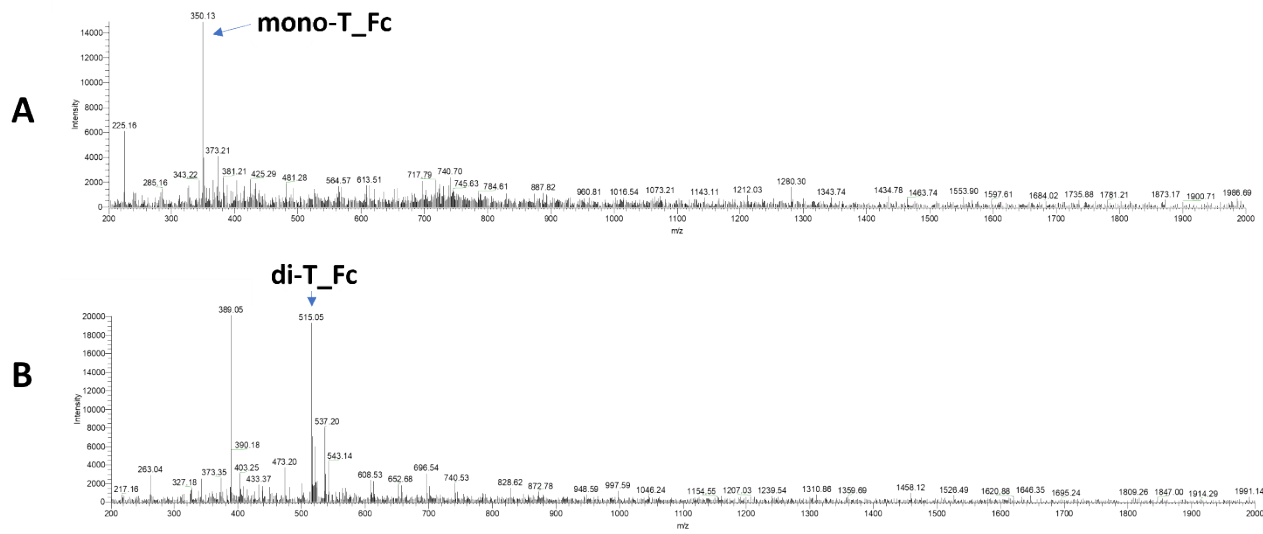
**Figure S2.** Overlay of fluorescence emission spectra normalized for complexes concentration, (A) **mono-T\_Fc** and (B) **di-T\_Fc**.



**Figure S3.** Overlay of fluorescence emission spectra of **mono-T<sub>2</sub>Fc** (A) and **di-T<sub>2</sub>Fc** (B) at 25 $\mu$ M ( $\lambda_{exc}$  = 440 nm).

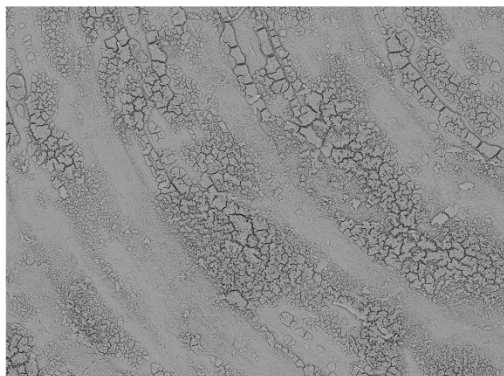


**Figure S4.** Raw correlation by DLS of: (A,C) A $\beta$ <sub>21-40</sub> alone and (B,D) with **mono-T<sub>2</sub>Fc** at 1:0.5 peptide:metal complex molar ratio, recorded after 4 (A-B) and 6 h (C-D) of aggregation.



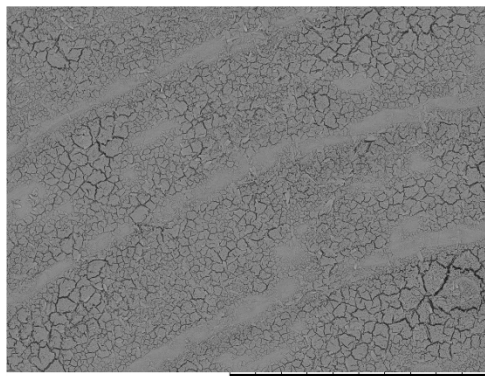
**Figure S5.** ESI-MS spectra of (A) mono-T\_Fc and (B) di-T\_Fc.

A



TM3000 2023/11/17 15:19 HL D8.2 x500 200 um  
DICMAPI - Test Misure

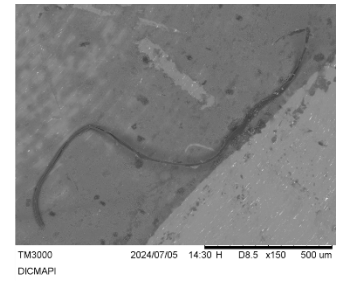
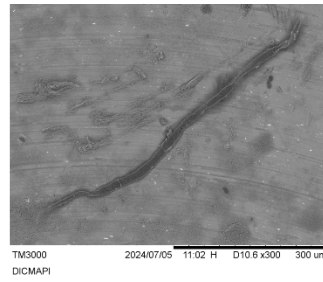
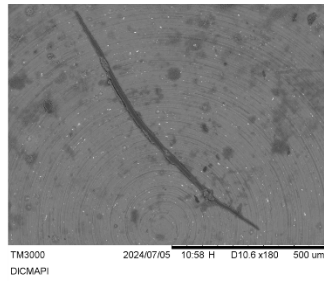
B



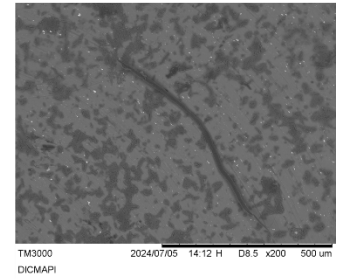
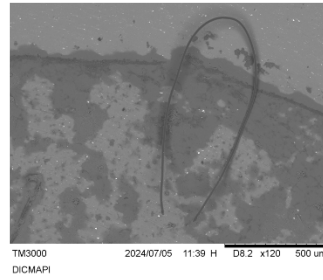
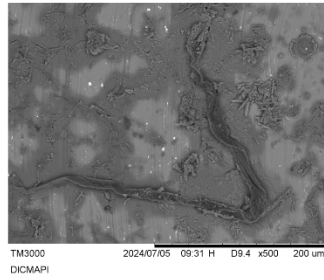
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**Figure S6.** SEM micrographs images of (A) **mono-T\_Fc** and (B) **di-T\_Fc**.

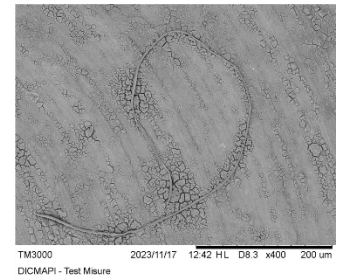
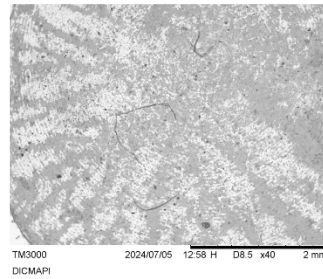
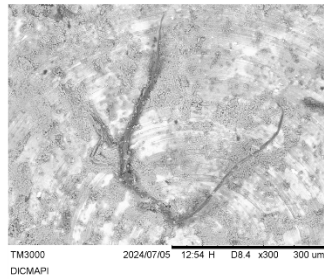
Alone



+ mono-T\_Fc

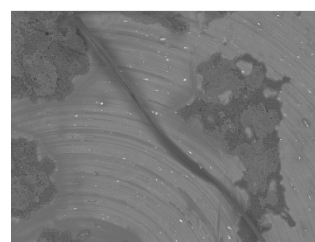
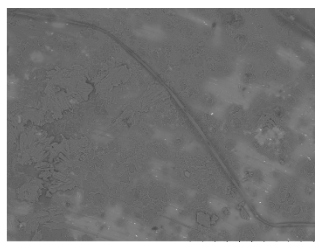
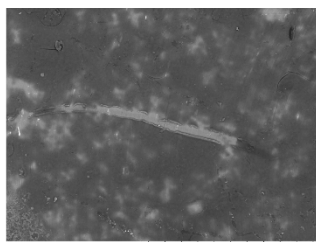


+ di-T\_Fc

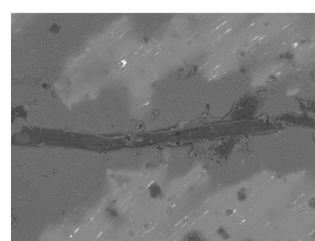
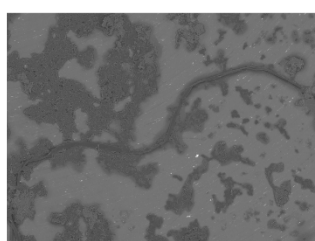
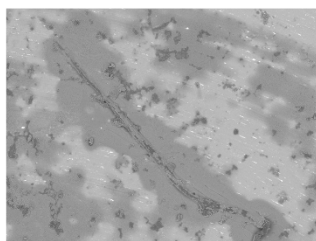


**Figure S7.** SEM micrographs images of  $A\beta_{21-40}$  in the absence and presence of **mono-T\_Fc** or **di-T\_Fc** at 1:0.5 peptide:metal complex molar ratio. For each sample, the three images are derived from 3 independent experiments.

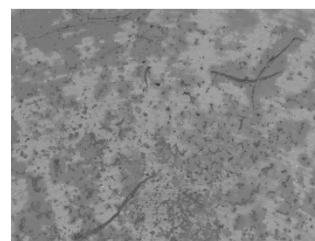
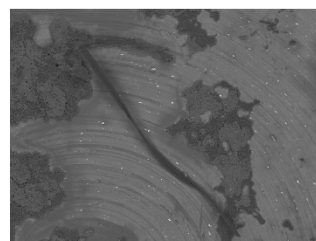
Alone



+ mono-T\_Fc



+ di-T\_Fc



**Figure S8.** SEM micrographs images of NPM1<sub>264-277</sub> in the absence and presence of **mono-T\_Fc** or **di-T\_Fc** at 1:0.5 peptide:metal complex molar ratio. For each sample, the three images are derived from 3 independent experiments



**Table S1:** Time values at which ThT fluorescence emissions reach their maximum value/2 are named  $t_{1/2}$  and maxima intensity values related to ThT experiments.

	Sample	$t_{1/2}$ (Min)	Maximum Intensity (a.u.)
<b>NPM1<sub>264-277</sub></b>	Alone	8.20	3682.0
	+ mono-T_Fc 1:1.5	8.37	2461.0
	+ mono-T_Fc 1:1	8.80	2434.0
	+ mono-T_Fc 1:0.5	8.50	2203.0
	+ mono-T_Fc 1:0.2	8.30	3321.0
	+ di-T_Fc 1:1.5	7.80	2302.0
	+ di-T_Fc 1:1	8.40	2545.5
	+ di-T_Fc 1:0.5	14.5	2145.0
	+ di-T_Fc 1:0.2	12.0	3818.5
<b>A<math>\beta</math><sub>21-40</sub></b>	Alone	188.6	559.5
	+ mono-T_Fc 1:1.5	219.5	719.1
	+ mono-T_Fc 1:1	220.3	770.5
	+ mono-T_Fc 1:0.5	221.5	892.5
	+ mono-T_Fc 1:0.2	235.0	580.0
	+ di-T_Fc 1:1.5	210.3	1278.6
	+ di-T_Fc 1:1	262.0	2342.5
	+ di-T_Fc 1:0.5	268.4	3768.7
	+ di-T_Fc 1:0.2	205.9	889.2
<b>ASA (A<math>\beta</math><sub>21-40</sub>)</b>	+Seed mono-T_Fc	172.3	3043.0
	+Seed di-T_Fc	145.2	948.4

**Table S2:** Table of main observed ions relative to the species formed by the A $\beta_{21-40}$  alone and mixed with **mono-T\_Fc** or **di-T\_Fc**. Experimental and theoretical mass and charge were reported for each adduct.

Description	m/z (charge)		Theoretical m/z	
	Peptide	+metal complex		
A $\beta_{21-40}$ : <b>mono-T_Fc</b>	A $\beta_{21-40}$	1887.08 (+1) 943.97 (+2)	1887.08 (+1) 943.86 (+2)	1887.22 944.11
	b <sub>16</sub>	1556.73 (+1)	1555.59 (+1)	1555.81
	b <sub>15</sub>	1456.61 (+1)	1457.90 (+1)	1456.75
	b <sub>14</sub>	1325.86 (+1)	1325.77 (+1)	1325.71
	b <sub>12</sub>	1156.63 (+1)	1155.61 (+1)	1155.60
	A $\beta_{21-40}$ + <b>mono-T_Fc</b>	-	1118.68 (+2) 746.7 (+3)	1119.19 (+2) 746.46 (+3)
	A $\beta_{21-40}$ : <b>di-T_Fc</b>	A $\beta_{21-40}$	1887.08 (+1) 943.97 (+2)	1887.08 (+1) 943.89 (+2)
b <sub>16</sub>		1556.73 (+1)	-	1555.81
b <sub>15</sub>		1456.61 (+1)	1457.86 (+1)	1456.75
b <sub>14</sub>		1325.86 (+1)	1325.83 (+1)	1325.71
b <sub>12</sub>		1156.63 (+1)	-	1155.60
A $\beta_{21-40}$ + <b>di-T_Fc</b>		-	1201.17 (+2)	1201.27

**Table S3:** Table of main observed ions relative to the species formed by the NPM1<sub>264-277</sub> peptide alone and mixed with **mono-T\_Fc** or **di-T\_Fc**. Experimental and theoretical mass and charge were reported for each adduct.

	Description	m/z (charge)		Theoretical m/z
		peptide	+metal complex	
NPM1 <sub>264-277</sub> : <b>mono-T_Fc</b>	NPM1 <sub>264-277</sub> Covalent dimer	591.76 (+6)	591.74 (+6)	591.30
	NPM1 <sub>264-277</sub> Covalent dimer	886.85 (+4)	886.81 (+4)	886.45
	NPM1 <sub>264-277</sub> Covalent dimer	1181.50 (+3)	1181.54 (+3)	1180.93
	NPM1 <sub>264-277</sub> Covalent dimer	1772.97 (+2)	1771.96 (+2)	1771.9
	NPM1 <sub>264-277</sub> + <b>mono-T_Fc</b>	-	1060.95 (+2)	1061.45
NPM1 <sub>264-277</sub> : <b>di-T_Fc</b>	NPM1 <sub>264-277</sub> Covalent dimer	591.75 (+6)	591.74 (+6)	591.30
	NPM1 <sub>264-277</sub> Covalent dimer	886.85 (+4)	886.83 (+4)	886.45
	NPM1 <sub>264-277</sub> Covalent dimer	1181.50 (+3)	1181.57 (+3)	1180.93
	NPM1 <sub>264-277</sub> Covalent dimer	1772.9 (+2)	1771.97 (+2)	1771.9
	NPM1 <sub>264-277</sub> + <b>di-T_Fc</b>	-	1143.39 (+2)	1143.95