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# **APPROACH: Sensitive Detection of Exosomal Biomarkers by Aptamer-Mediated Proximity Ligation Assay and Time-Resolved Förster Resonance Energy Transfer**

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Table S1. Sequences of oligonucleotides used in this work.

Name	Sequence (5'-3')
Apt-CD63 <sup>1</sup>	ATATACACCCACCTCGCTCCCGTGACACTAATGCTATTTTTTTTTT TTTTT <b>GACGCTAATAGTTAAGACGCT</b>
Cho primer <sup>1</sup>	Cholesterol- GACCCTAAGCATAACATGCTCACTGACGCTAGGTTTTTTTTTTT TTTT <b>ATATGACAGAACTAGACACTCTT</b>
Apt-HER2	TCTAAAAGGATTCTTCCCAAGGGGATCCAATTCAAACAGCTTTTT TTTTTTTTTT <b>GACGCTAATAGTTAAGACGCT</b>
Apt-PD-L1	ACGGGCCACATCAACTCATTGATAGACAATGCGTCCACTGCCCCG TTTTTTTTTTTTTT <b>TGACGCTAATAGTTAAGACGCT</b>
Backbone <sup>1</sup>	Phosphate- <b>CTATTAGCGTCCAGTGAATTATACCCGGTCGCTTCTTTATGCC</b> <b>GTCAAGAGTGTCTA</b>
Connector <sup>1</sup>	Phosphate- <b>GTTCTGTCATAITTAAGCGTCTTAA</b>
Tb probe	Tb- <b>TGAATTATACCCGGT</b> (conjugation ratio: 0.87 Tb per DNA)
Cy5 probe	Cy5- <b>CGCTTCTTTATGCCG</b> (conjugation ratio: 0.92 Cy5 per DNA)
Cho primer- Cy5	Cholesterol-.GACCCTAAGCATAACATGCTCACTGACGCTAGGTTTTT TTTTTTTTTTATATGACAGAACTAGACACTCTT-Cy5 (conjugation ratio: 1.5 Cy5 per DNA)
Apt-CD63-Cy5	ATATACACCCACCTCGCTCCCGTGACACTAATGCTATTTTTTTTTT TTTTTGACGCTAATAGTTAAGACGCT-Cy5 (conjugation ratio: 1.5 Cy5 per DNA)
MU-Cy5	Cy5-CTTTTCGGGATTTATGC (conjugation ratio: 1 Cy5 per DNA)

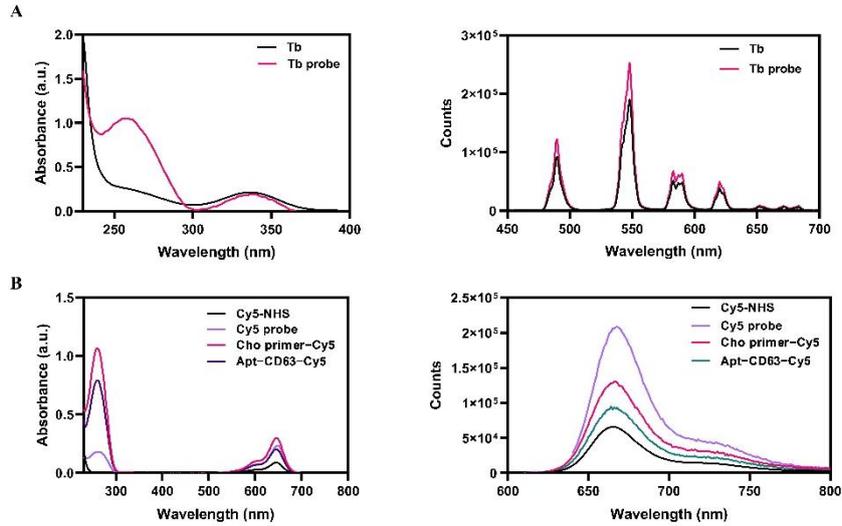


Figure S1. Spectral characterization of Tb probe, Cy5 probe, Cho primer-Cy5 and Apt-CD63-Cy5. The concentrations and labeling ratios (dye/oligonucleotides) were determined through absorption spectroscopy. The absorbance and emission spectra of Tb and Cy5 probes were presented in (A) and (B), respectively.

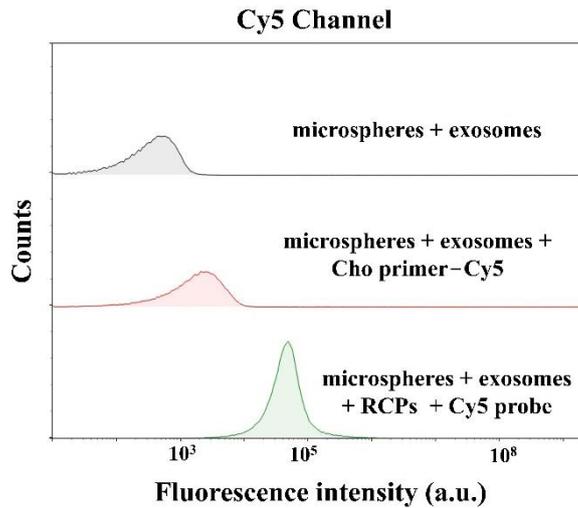


Figure S2. Flow cytometry analysis of RCA products (RCPs).

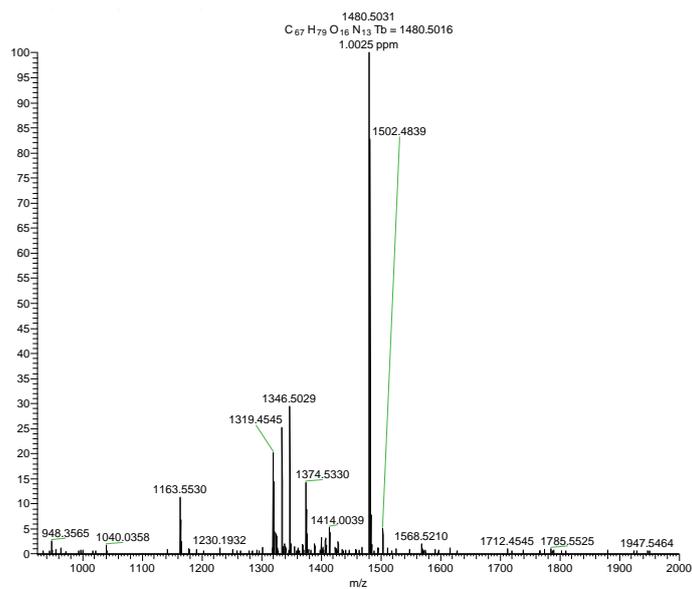


Figure S3. The HRMS spectrum of [CoraFluor-like-NHS].

References:

1. Zhao, X.; Luo, C.; Mei, Q.; Zhang, H.; Zhang, W.; Su, D.; Fu, W.; Luo, Y., Aptamer-Cholesterol-Mediated Proximity Ligation Assay for Accurate Identification of Exosomes. *Anal. Chem.* 2020, 92 (7), 5411-5418, DOI:10.1021/acs.analchem.0c00141.