Supplemental Fig. 1 DNA strands characterization and TDN assembly

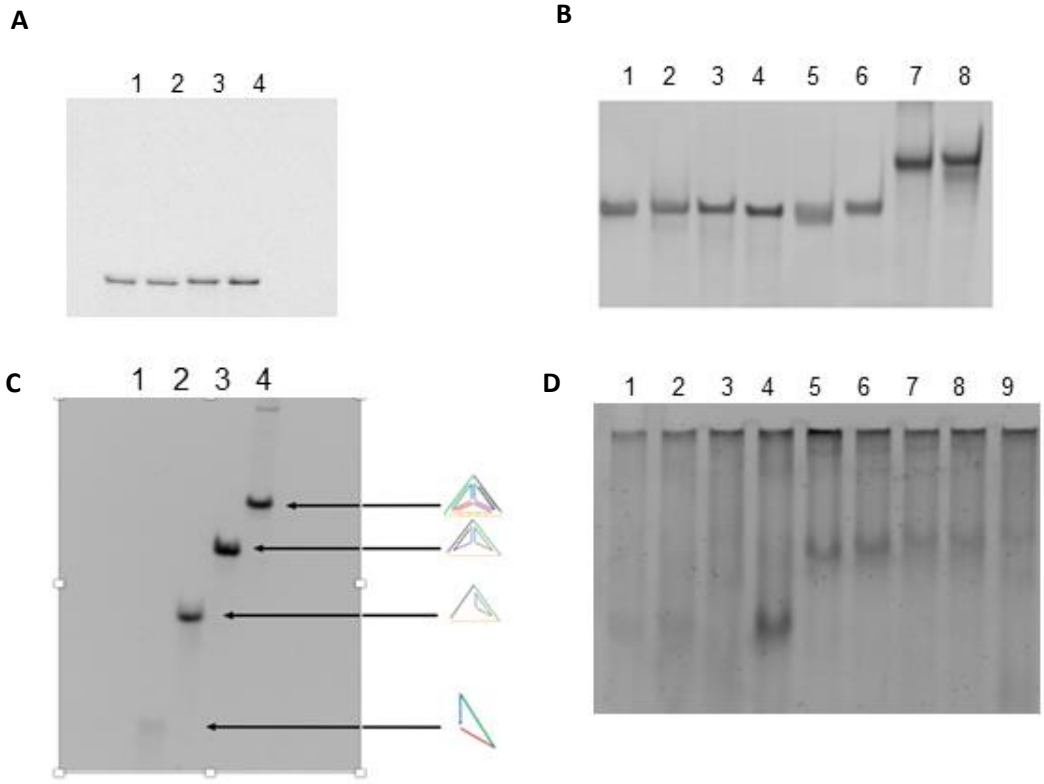

Fig. S1 - All DNA strands were characterized by denaturing PAGE to confirm proper synthesis and purity. Denaturing PAGE experiments were carried out on a 20 x 20 cm vertical Hoefer 600 electrophoresis unit (constant voltage of 300V for 1.5 hours). Gels were stained using the GelRed® or GelGreen®) and images were collected using a Gene Genius Bioimaging system (SYNGENE). **Fig.1.a TDN DNA strands characterization.** 8% denaturing PAGE of unmodified tetrahedron strands (0.02 nmol/lane): Lane 1- **S1**, lane 2- **S2**, lane 3- **S3** and lane 4- **S4**. **Fig.1.b Functionalized TDN DNA strands characterization.** 8% denaturing PAGE of unmodified tetrahedron strands (0.02 nmol/lane): Lane 1- **S1-C12**, lane 2- **S2-C12**, lane 3- **S3-C12**, lane 4- **S4-C12**, lane 5-**Cy5-S1**, lane 6- **Cy5-S1-C12**, lane 7- **A-S4** and lane 8- **A-S4-C12**. DNA strands were assembled into a DNA tetrahedron as previously described. Briefly, 0.02 nmol of each strand **S1-4** were annealed (105-4ºC) in TAEMg for 16 hours. Native PAGE experiments were carried out on a 20 x 20 cm vertical Hoefer 600 electrophoresis unit (constant voltage of 80V for 16 hours). Gels were stained using the GelRed® or GelGreen®) and images were collected using a Gene Genius Bioimaging system (SYNGENE). **Fig.1.c Unmodified TDN assembly characterization**. 8% native PAGE of unmodified TDN assembly (0.02 nmol/lane): Lane 1- **S1**, lane 2- **S1+S2**, lane 3- **S1+S2+S3** and lane 4- **S1+S2+S3+S4**. To confirm the loaded TDN integrity a native 8% PAGE experiment was perfored to confirm

the integrity of TDN after DOX intercalation. In this case, we used GelGreen® to stain the DNA, since GelRed® stains the DNA trough intercalation. **Fig. S1.d TDN+DOX assembly characterization.** 8% native PAGE of TDN assemblies loaded with DOX (0.02 nmol/lane): Lane 1- **S1+S2+S3+S4 + DOX**, lane 2- **Cy5-S1+S2+S3+S4 + DOX**, lane 3- **S1+S2+S3+A-S4 + DOX**, lane 4- **S1-C12+S2 +S3 +S4**, lane 5- **S1-C12+S2-C12+S3+S4 + DOX**, lane 6- **S1-C12+S2-C12+S3-C12+S4 + DOX,** lane 7- **S1-C12+S2-C12+S3-C12+S4-C12 + DOX,** lane 8- **Cy5-S1-C12+S2-C12+S3-C12+S4-C12 + DOX** and lane 9- **S1-C12+S2-C12+S3-C12+A-S4-C12 + DOX.**

Supplemental Figure 2 - TDN Circular Dichroism

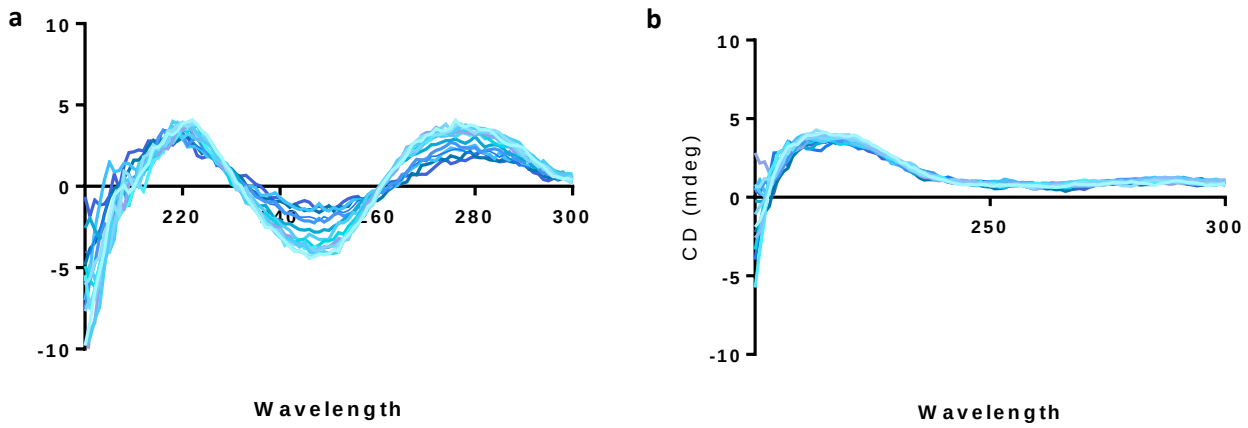

Fig. S2 - To monitor the conformational polymorphism of our nanostructure we performed circular dichroism spectroscopy in the TDN at a 10 µM concentration in a TAEMg buffer solution. [2] **A– Functionalized TDN** thermal melt plots as a function of temperature. Overlaid CD spectra were obtained at different temperatures ranging from 30°C to 95°C. The bottom line (light blue) is for the spectrum obtained at 30°C; the series of blue-coloured lines are plots obtained at successive 5°C intervals and the darkest blue is for the spectrum obtained at 95°C. **B- Functionalized TDN loaded with DOX and PTX** thermal melt plots as a function of temperature. Overlaid CD spectra were obtained at different temperatures ranging from 30°C to 95°C. The bottom line (light blue) is for the spectrum obtained at 30°C; the series of blue-coloured lines are plots obtained at successive 5°C intervals and the darkest blue is for the spectrum obtained at 95°C.

Table 1. DNA sequences

Strand	Sequence (5' to 3')
S1	ATTTATCACCCGCCATAGTAGACGTATCACCAGGCAGTTGAGACGAACATTCCTAAGTCTGAA
S2	ACATGCGAGGGTCCAATACCGACGATTACAGCTTGCTACACGATTCAGACTTAGGAATGTTCG
S3	ACTACTATGGCGGGTGATAAAACGTGTAGCAAGCTGTAATCGACGGGAAGAGCATGCCCATCC
S4	ACGGTATTGGACCCTCGCATGACTCAACTGCCTGGTGATACGAGGATGGGCATGCTCTTCCCG
S1-C12	ATTTATCACCCGCCATAGTAGACGTATCACCAGGCAGTTGAGACGAACATTCCTAAGTCTGAA**[C12]**
S2-C12	ACATGCGAGGGTCCAATACCGACGATTACAGCTTGCTACACGATTCAGACTTAGGAATGTTCG**[C12]**
S3-C12	ACTACTATGGCGGGTGATAAAACGTGTAGCAAGCTGTAATCGACGGGAAGAGCATGCCCATCC**[C12]**
S4-C12	ACGGTATTGGACCCTCGCATGACTCAACTGCCTGGTGATACGAGGATGGGCATGCTCTTCCCG**[C12]**
Cy5-S1	**Cy5**-ATTTATCACCCGCCATAGTAGACGTATCACCAGGCAGTTGAGACGAACATTCCTAAGTCTGAA
Cy5-S1-C12	**Cy5**-ATTTATCACCCGCCATAGTAGACGTATCACCAGGCAGTTGAGACGAACATTCCTAAGTCTGAA**[C12]**
A-S4	GCAGCGGTGTGGGGGCAGCGGTGTGGGGGCAGCGGTGTGGGGTTTTACGGTATTGGACCCTCGCATGACTCA ACTGC CTGGTGATACGAGGATGGGCATGCTCTTCCCG
A-S4-C12	GCAGCGGTGTGGGGGCAGCGGTGTGGGGGCAGCGGTGTGGGGTTTTACGGTATTGGACCCTCGCATGACTCA ACTGC CTGGTGATACGAGGATGGGCATGCTCTTCCCG**[C12]**

Table S1. Unmodified DNA aptamer and tetrahedron sequences were obtained from a previous publication [1]. Strands **S1-4** were purchased from IDT DNA with polyacrylamide gel electrophoresis purification. The functionalized DNA strands were purchased from GeneLink with PAGE. When desired, **Cy5** was attached to the 5'-end of strand **S1**, while an alkyl (**C12)** was attached to the 3'-end of strands **S1-4** on a DNA synthesizer. When needed, a previously published HER2+ aptamer (**A**) was incorporated on the 5'-end of strand **S4**.

Supplemental methods

1. Tris(hydroxymethyl)aminomethane base (ultra pure, TRS001), ethylenediaminetetraacetic acid disodium dihydrate (EDTA, reagent grade, EDT001), formamide (biotechnology grade, min. 99.5%, FOR001), tetramethylethylenediamine (TEMED, electrophoresis grade, TEM001), 40% acrylamide/bis-acrylamide solution 19:1, ACR005) and urea (ultra pure, molecular biology grade, URE-001) were purchase from Bioshop Canada. Boric acid (B6768), ammonium persulfate (≥98%, A3678), doxorubicin (suitable for fluorescence, 98.0-102.0% (HPLC), 44583), paclitaxel (from semisynthetic, ≥97%, T7191), Nile Red (suitable for fluorescence, ≥98.0% (HPLC), 19123) were purchase from Sigma-Aldrich. Magnesium chloride hexahydrate (crystalline, USP/FCC, M35-500) was purchased from Fisher Chemical™. Annexin V FITC apoptosis staining / detection kit (ab14085), cleaved caspase-3 staining kit (ab65613), Phalloidin-iFluor 488 (ab176753) and anti-CY5 mouse monoclonal (ab52061) were purchase from Abcam. Acetic acid glacial (lab grade, 00615-540) from Anachemia. PAGE GelRed® Nucleic Acid Gel Stain (41008) and GelGreen® Nucleic Acid Gel Stain (41005) were purchased from Biotium. Illustra microspin G-25 columns (27-5325-01) were purchased from GE Healthcare. AlamarBlue™ Cell Viability Reagent (DAL1025) was purchased from Invitrogen. Alpha Minimun Essential Medium (098150) and fetal bovine serum (FBS, 310-010-CL) was purchased from Multicell. CellLight™ Plasma Membrane-CFP, BacMam 2.0 (C10606) and pHrodo™ Red Dextran, 10,000 MW, for Endocytosis (P10361) were purchased from Invitrogen.

References:

(1) Liu, Z.; Duan, J.-H.; Song, Y.-M.; Ma, J.; Wang, F.-D.; Lu, X.; Yang, X.-D. *Journal of Translational Medicine* **2012**, *10*, 148.

(2) Agudelo, D.; Bourassa, P.; Bérubé, G.; Tajmir-Riahi, H.-A. *International Journal of Biological Macromolecules* **2014**, *66*, 144.