Supplementary Material (ESI) for Chemical Communications
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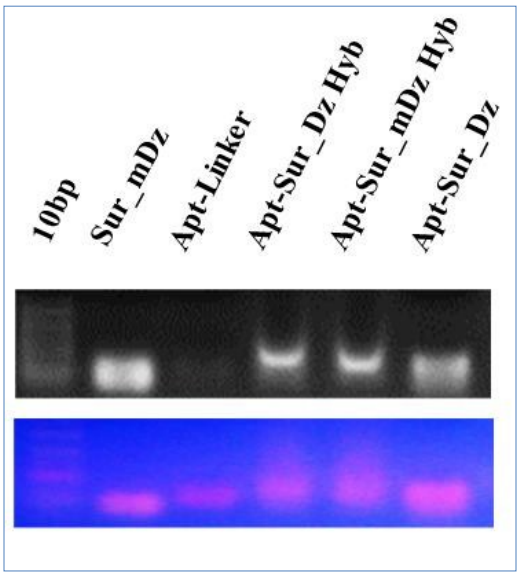
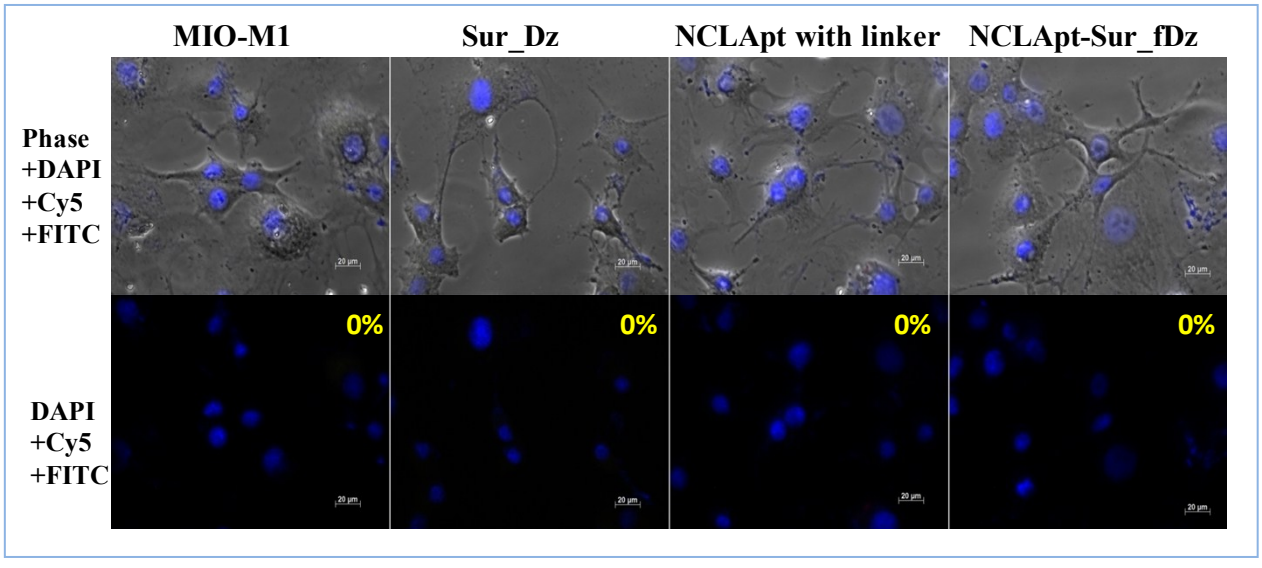
Supplementary figure 1



Supplementary figure 1. **Agarose gel electrophoresis** of the conjugates. **Synthesis of conjugate by annealing.** Survivin mutant DNAzyme, nucleolin aptamer with linker and conjugates prepared in hybridization buffer was electrophoresed on 4% agarose gel.

Supplementary figure 2



Supplementary figure 2**. Cellular uptake of NCL-APT-Dz conjugates in MIO-M1 cells.** Microscopic images showing the cellular uptake of Sur_fDz, NCL-APT-linker and NCL-APT-Sur_fDz in MIO-M1 cells after 2hr of addition and imaged under 40X objective. Percentage positive are represented in the right top corner of representative sample.

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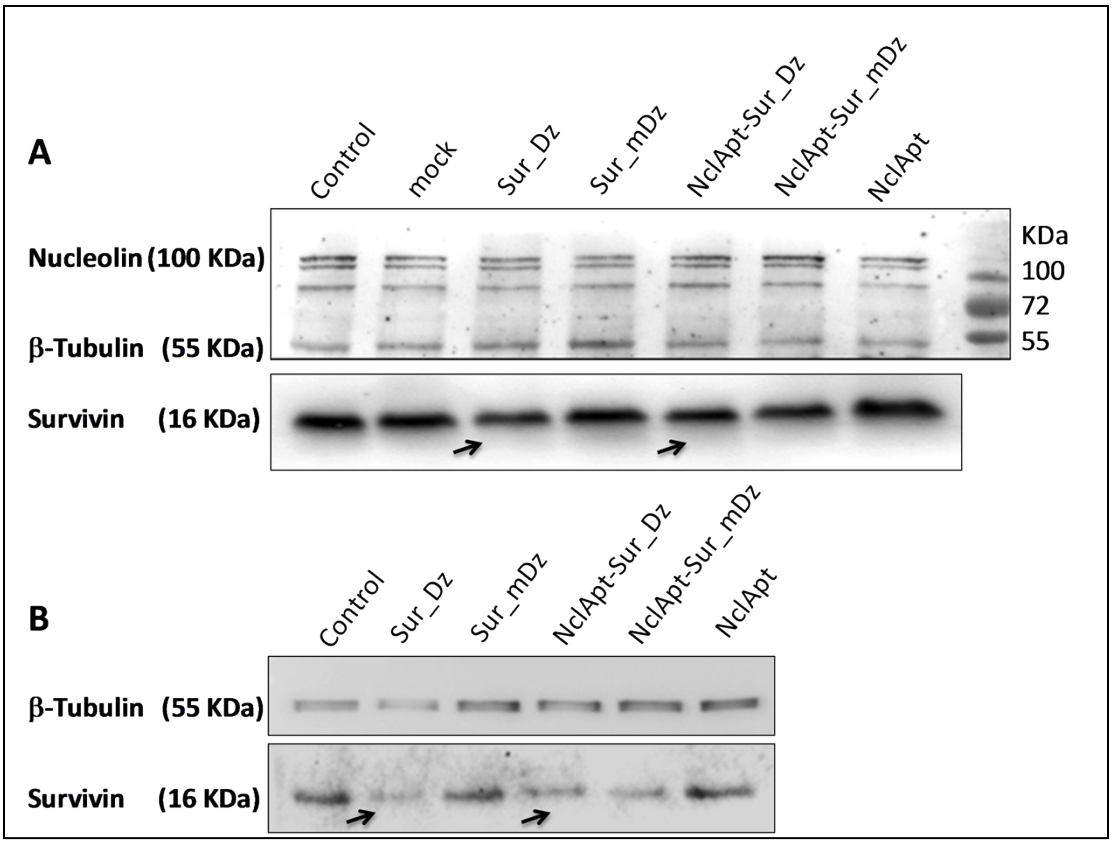
Supplementary figure 3



Supplementary figure 3. **Western blotting.** A. WERI-Rb1 cells and Y79 (B) were treated with the Dzs, conjugates and NclApt alone for 48h and analyzed for the survivin protein levels by western blotting. β-tubulin was used as housekeeping control to normalize the survivin expression. Additionally nucleolin was checked as the housekeeping proteins showed minor variation.

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Supplementary methods

Materials

RB cell lines, Y79 and WERI-Rb1 were procured from RIKEN BioResource center and noncancerous Muller glial cell line, MIO-M1 derived from retina was obtained as a gift from Dr. G.A. Limb UCL Institute of Ophthalmology were used in the study. Ncl-Apt aptamer with linker labeled with Cy5 (Apt-Linker) was synthesized from Integrated DNA technologies (Coralville, IA, USA), survivin DNAzyme (Sur_Dz), mutant survivin DNAzyme (Sur_mDz) were synthesized from VBC biotech (Wien, Austria) and fluorescein labeled survivin DNAzyme (Sur_fDz) was obtained from Sigma Aldrich (Bangalore, India) and the sequences were presented in table 1. Survivin antibody was procured from Santa Cruz biotech (USA). The SYBR green qPCR (Thermoscientific, India) was performed to quantify the fold change in survivin mRNA levels, normalized to β-2-microgloubulin using untreated samples.

Table 1. Oligonucleotide sequence of the aptamer and the DNAzyme

S.No.	Oligo sequence	Sequence (* indicates Phosphorothioate)
1	Sur_Dz	5'-C*CTCGGCCA GGC TAG CTA CAA CGA CCGCTCCG*G-3'
2	Sur_mDz	5'-C*CTCGGCCA GGC TAT GTA CAA CGA CCGCTCCG*G-3'
3	NclApt-linker	5' GGTGGTGGTGGTTGTGGTGGTGGTGGT*T*T*T*T*CCGGAGG 3'

Conjugation in different buffers

The conjugation of NclApt-Sur_Dz was carried out by denaturing in water bath for 10 mins and gradual cooling to room temperature. Conjugate formation was analyzed in different buffers such as 1x SSC buffer and hybridization buffer. The chimeric conjugate prepared with hybridization buffer showed noticeable difference in the size of conjugate compared to Sur_Dz and Apt-linker. This ensured the conjugation reaction was carried out perfectly and the optimized Dz concentration had hybridized with NclApt-linker and showed no unbound or free Dz or aptamer.

Binding assay

Cellular binding in Y79, WERI-Rb1 and MIO-M1 were performed for checking the efficiency of the conjugate upon chimerization. 1x10^6cells/ml was subjected to binding assay (protocol published earlier Subramanian et al., 2012) with 500nM concentration of APT-linker alone and APT-Sur_Dz conjugate were used for and incubated at 25˚C for 2 h. The cells were washed twice with PBS to remove unbound conjugatesand subjected for FACS acquisition.

Conjugate internalization

The conjugate binding and uptake in cells and delivery of DNAzyme into cells, internalization was studied following earlier published protocol (Subramanian et al., 2014). 500nM of NCL-APT -Sur_fDZ conjugate, NCL-APT -linker alone and sur_fDZalone were added to Y79, WERI-Rb1 and MIO-M1 cells grown on coverslips. After 2 h of incubation the cells were washed twice with PBS, fixed with 4% paraformaldehyde for 10 min and counterstained with DAPI for ten mins. Then images were captured using Zeiss Axio vision fluorescent microscope (Carl Ziess, Germany).

Functional activity of conjugates

The functional activity of conjugates on cells was studied by either transfecting Sur_Dz and Sur_mDz alone or by adding the conjugates to the cells. 12.5μM of NCL-APT -Sur_Dz and NCL-APT -Sur_mDz conjugates were added to the cells and the same concentration of APT-Linker alone was also added directly to the cells and Sur_Dz, Sur_mDz alone were also transfected as controls. Cells were collected 48h post transfection and subjected for RNA isolation and protein extraction were performed. The primer sequences used are listed below in table 2. The cell proliferation was quantified by measuring the metabolic activity of the cell using MTT assay. qPCR, western blotting and MTT assays were performed following the published protocol (Subramanian et. al., 2013 and Subramanian et. al., 2012).

Table 2. List of primers used for the qPCR.

Primer	sequence (5'-3')
BIR F	GACCACCGCATCTCTACATTC
BIR R	TGCTTTTTATGTTCCTCTATGGG
B-2-M F	TATCCAGCGTACTCCAAAGA
B-2-M R	GACAAGTCTGAATGCTCCAC

REFERENCES:

1. Subramanian N, Raghunathan V, Kanwar JR, et al. (2012) Target-specific delivery of doxorubicin to retinoblastoma using epithelial cell adhesion molecule aptamer. *Molecular vision*18:2783-2795.
2. Subramanian N, Navaneethakrishnan S, Biswas J, et al. (2013) RNAi Mediated Tiam1 Gene Knockdown Inhibits Invasion of Retinoblastoma. *PLoS ONE* 8(8): e70422.
3. Subramanian N, Sreemanthula JB, Balaji B, Kanwar JRet al. (2014) A strain-promoted alkyne-azide cycloaddition (SPAAC) reaction of a novel EpCAM aptamer-fluorescent conjugate for imaging of cancer cells. *Chemical communications* 4,**50**, 11810-11813.