SELF-ASSEMBLING HYALURONIC ACID-LIPID CONJUGATES AS PLATFORM FOR TARGETED DRUG DELIVERY

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BACKGROUND

Hyaluronic acid (HA) is a glycosaminoglycan associated with inflammatory processes and tumor development, known to selectively interact with the CD44 receptor, which is highly expressed in multiple cancer types [1,2]. Previous investigations showed that HA oligosaccharide-polyethylene glycol (PEG)-phospholipid conjugates characterized by different degrees of polymerization (DP) promoted increased internalization in CD44-positive (CD44+) lung cancer cells [3].

AIM

The conjugation of an azide-functionalized HA octasaccharide (DP8) with 1,2-dipalmitoyl-sn-glycero-3-phosphoethanolamine-N-dibenzocyclooctyl (PE) was designed to promote the formation of self-assembling nanocarriers.

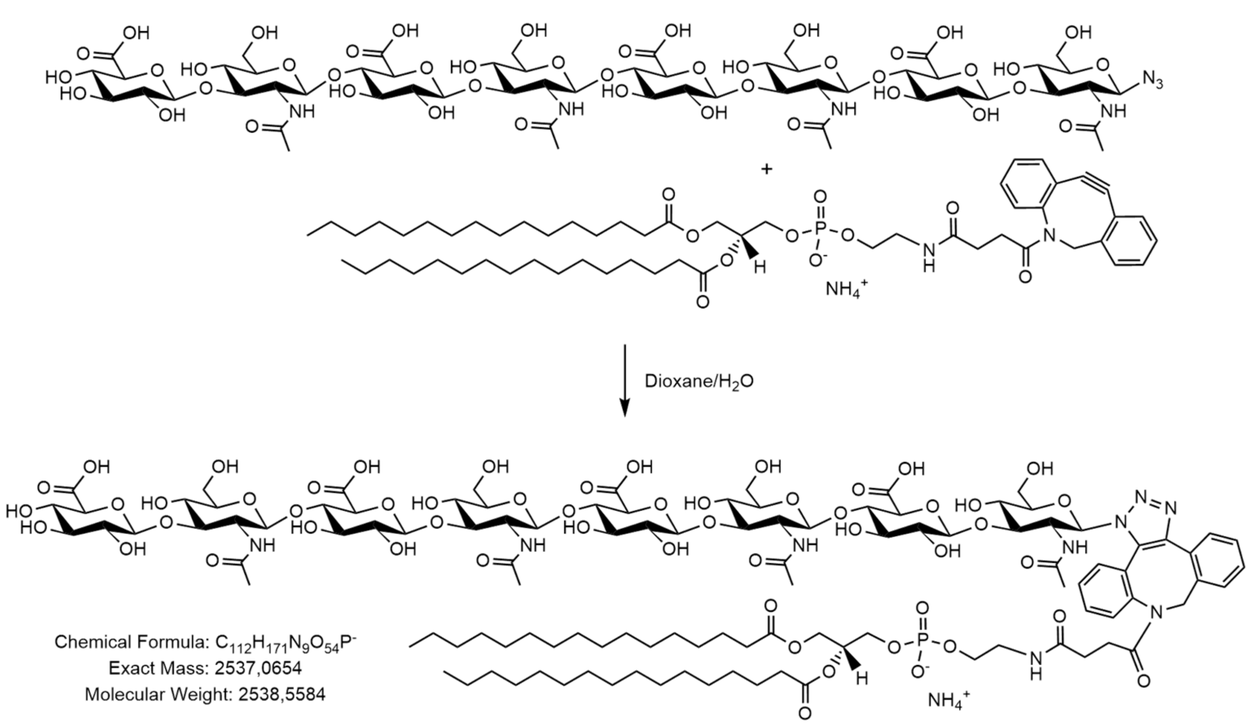

Figure 1. Synthesis of the HA-DP8-PE conjugate.

NANOCARRIERS

The resulting nanoassemblies (NAs) were loaded with a nitric oxide-releasing derivative of doxorubicin, namely DR6, which has demonstrated activity against doxorubicin-resistant cells [4].

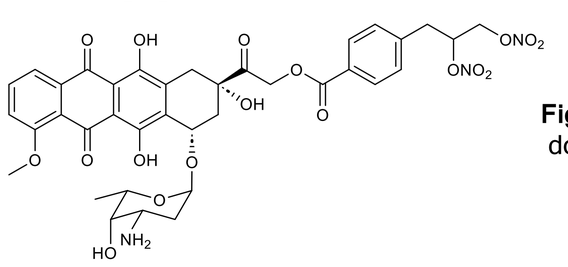

Figure 2. Chemical structure of NO-donor doxorubicin derivative (DR6).

Table 1. Physico-chemical characteristics of HA-DP8-PE NAs

	Mean diameter (nm ± SD)	PdI	ζ-potential (mV ± SD)	EE (%)	DL (%)
HA-DP8-PE NAs	134 ± 55	0.431	-24 ± 3	/	/
DR6-HA-DP8-PE NAs	108 ± 28	0.366	-31 ± 6	86 ± 10	10 ± 1

IN VITRO

UPTAKE

Time-dependent uptake of the NAs in CD44+ cancer cell lines compared to CD44- counterparts.

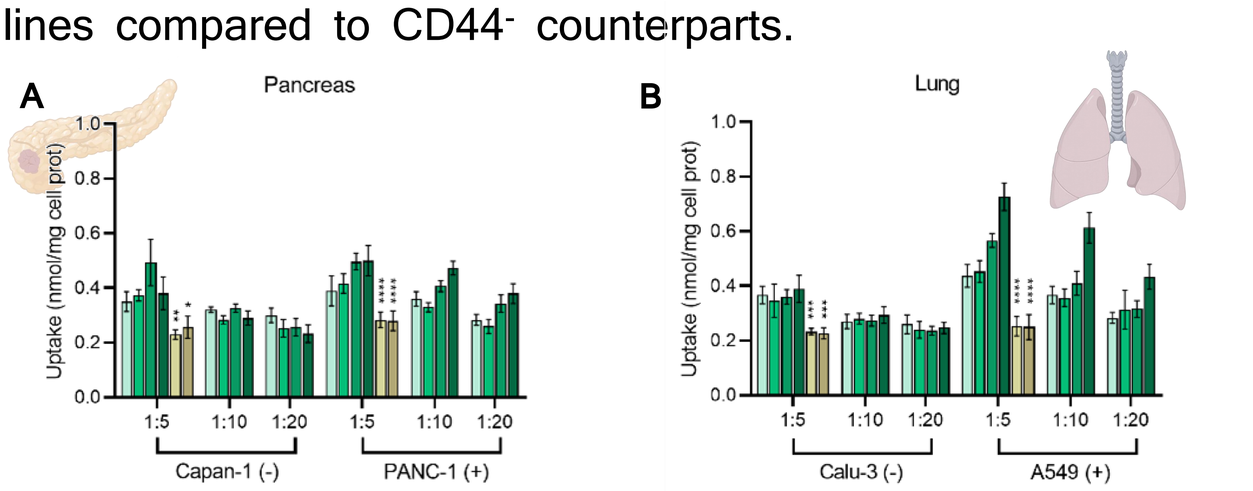

Figure 3. Cellular uptake by pancreatic (Capan-1 and PANC-1), lung (Calu-3 and A549) and breast (MCF-7 and MDA-MB-231) cancer cells of NAs, diluted 1:5, 1:10 and 1:20, labelled with Nile Red (NR), after 1, 3, 6 or 24 h. All cell lines were treated with NAs diluted 1:5 for 24 h, in the presence of HA (100 μM) and anti-CD44 antibody diluted 1/100 (Ab). ⁎p<0.05, ⁎⁎p<0.01, ⁎⁎⁎p<0.001, ⁎⁎⁎⁎p<0.0001 vs NAs, diluted 1:5 for 24 h.

CELLULAR VIABILITY

DR6-HA-DP8-PE NAs demonstrated increased cytotoxic effects in CD44+ (MB-MDA-231) cells compared to CD44- (MCF-7), following a dose- and time-dependent trend.

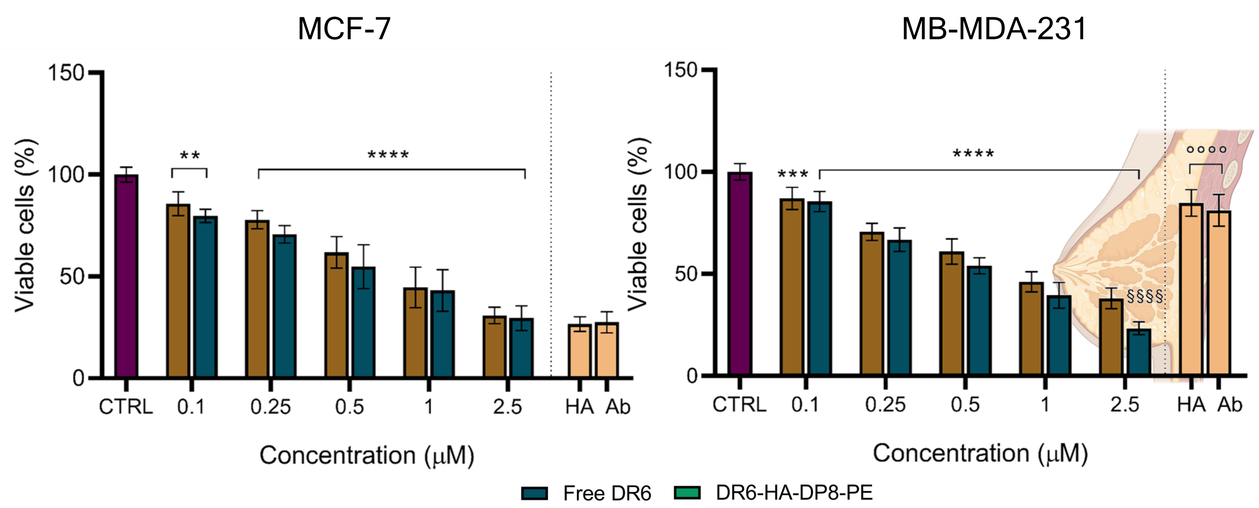

Figure 4. Cell viability of MCF-7 and MDA-MB-231 cells. Cells were incubated with free ND or NA containing different concentration (0.1, 0.25, 0.5, 1, 2.5 μM) of ND, for 72 h. Cell viability of untreated cells was 100 % for all experimental conditions. Cells were grown for in fresh medium or in the presence of ND-HA-DP8-PE NAs containing 2.5 μM ND, in the absence or presence of HA (100 μM) or anti-CD44 antibody diluted 1/100 (Ab), then cell viability was measured. ND/ND-HA-DP8-PE NAs vs untreated cells: ⁎⁎p<0.005, ⁎⁎⁎p<0.001, ⁎⁎⁎⁎p<0.0001; ND vs ND-HA-DP8-PE NAs at the same concentration: §§§§p<0.0001; HA/Ab-treated cells vs ND-HA-DP8-PE NAs (2.5 μM): °°°°p<0.0001.

TAKE HOME MESSAGE

The HA-DP8-PE nanoassemblies exhibit strong colloidal stability and favorable biocompatibility, together with efficient and selective CD44-mediated internalization, reflecting the relevance of HA-based targeting strategies in CD44-overexpressing tumors. Their ability to enhance intracellular delivery of the encapsulated therapeutic payload supports improved drug accumulation in cancer cells, including resistant phenotypes, while maintaining a well-defined and self-assembled nanoscale structure.

References

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