Nano Innovation 2020 Conference & Exhibition — Rome, 15-18 September

Discoid Nanoparticles: reaction environment-dependent Size Response

Iannotta D[1,2], Celia C[1], Wolfram J[3], Papa A[4], Di Marzio L[1]

[1]University "Gabriele d'Annunzio" Chieti-Pescara; [2]University of Teramo (TE), IT; [3]Mayo Clinic, FL, USA; [4]ALFATESTLab S.r.l

INTRODUCTION - In recent years, the develop of new drug delivery system (DDS) was necessary to optimize the treatment efficiency, thus overcoming the limits of traditional therapy such as targeting and drug half life. Tumor targeting plays a pivotal role in anticancer therapy. Tumor targeting can be carried out by conjugating specific targeting agents on the surface of nanoparticles or by modifying their shape and size. Several DDS was developed such as liposomes, niosomes and other type of nanoparticles, but they can have some limits like the recognition and elimination of the immune system or the tumor targeting. To improve the selective targeting and accumulate therapeutic agents inside the tumor tissues, we designed new Discoidal Nanoparticles (DNs). DNs have non-spherical shape that allows better interact and cross through the fenestrated endothelium of tumor tissue compared to spherical particles. DNs can be obtained from liposomes incubated with Styrene-Maleic Acid copolymer (SMA). SMA is a copolymer formed by reversible addition-fragmentation chain transfer polymerization (RAFT) between Styrene and Maleic Anhydride that is able to modify the native liposome size from about 150 nm to ~13 nm. Molecular ratio between styrene and maleic anhydride, temperature and pH of environment reaction can affect the synthesis of DNs [1]. The aim of this work is to study the effect of pH into the DNs production. Dimyristoylphosphatidylcholine (DMPC) liposomes were synthesized by Thin Layer Evaporation, freeze and thaw cycles and extrusion methods. The DNs are products by incubating liposome in Hepes 10 mM with different SMA molecular ratio (2:1 and 4:1) at different pHs (range of pH from 3.5 to 11.5)) and temperatures (4°C, 25°C, 37°C, 65°C). Physicochemical characterized of Liposomes and DNs was carried out using Dynamic Light Scattering (DLS). Liposome were used as experimental control.

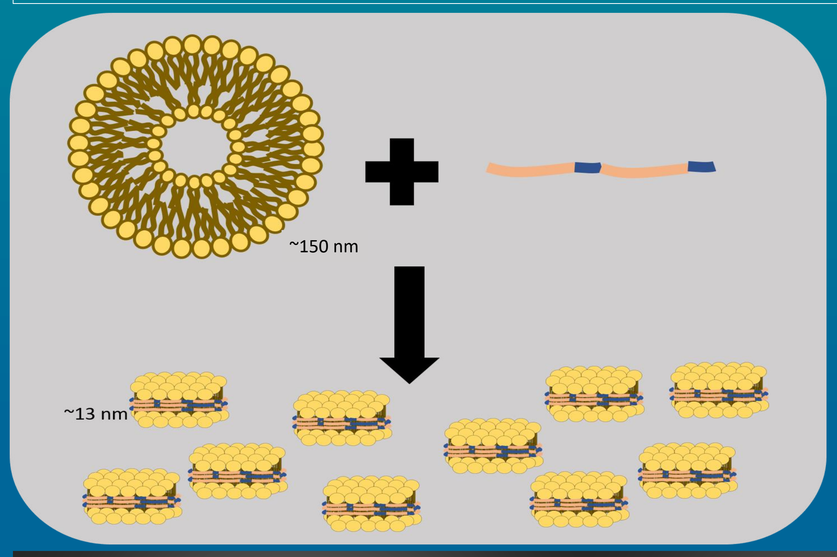

Figure 1: Schematic representation of DNs realization.

T (°C)	SMA 2:1 pH	SMA 2:1 Size (nm)	SMA 4:1 pH	SMA 4:1 Size (nm)
DMPCL	7.4	147.98 ± 10.22	7.4	125.33 ± 0.58
4°C	3.5	188.00 ± 59.18	3.5	100.45 ± 10.68
	5.5	15.20 ± 0.02	5.5	88.35 ± 27.80
	6.5	22.21 ± 2.37	6.5	48.77 ± 44.00
	6.8	11.25 ± 0.03	6.8	134.00 ± 14.50
	7.4	11.69 ± 0.64	7.4	94.00 ± 20.59
	8.5	10.05 ± 0.57	8.5	20.78 ± 4.41
	9.5	10.44 ± 1.12	9.5	26.00 ± 2.75
	11.5	12.23 ± 1.41	11.5	9.04 ± 0.86
25°C	3.5	14.40 ± 8.63	3.5	95.73 ± 28.56
	5.5	13.02 ± 8.70	5.5	65.75 ± 9.12
	6.5	20.20 ± 7.82	6.5	28.17 ± 15.31
	6.8	12.94 ± 4.18	6.8	107.56 ± 98.66
	7.4	13.94 ± 6.46	7.4	33.27 ± 1.69
	8.5	11.32 ± 1.70	8.5	14.18 ± 1.41
	9.5	9.20 ± 0.78	9.5	10.68 ± 3.36
	11.5	14.47 ± 1.22	11.5	16.08 ± 6.84
37°C	3.5	92.93 ± 0.06	3.5	77.38 ± 3.54
	5.5	14.12 ± 1.91	5.5	61.57 ± 3.54
	6.5	19.66 ± 2.83	6.5	240.30 ± 99.56
	6.8	12.55 ± 2.28	6.8	190.30 ± 28.85
	7.4	12.16 ± 0.92	7.4	253.20 ± 19.80
	8.5	11.84 ± 1.06	8.5	26.42 ± 8.32
	9.5	12.02 ± 2.85	9.5	12.19 ± 1.35
	11.5	10.39 ± 1.05	11.5	11.65 ± 0.59
65°C	3.5	21.63 ± 5.61	3.5	94.04 ± 1.63
	5.5	10.53 ± 0.99	5.5	16.42 ± 1.75
	6.5	16.96 ± 0.99	6.5	26.37 ± 15.83
	6.8	13.21 ± 0.32	6.8	232.4 ± 49.21
	7.4	10.91 ± 0.46	7.4	19.73 ± 1.13
	8.5	8.40 ± 0.14	8.5	34.93 ± 3.72
	9.5	12.50 ± 1.80	9.5	12.16 ± 3.54
	11.5	16.33 ± 1.89	11.5	10.00 ± 2.40

Table I: Size of DNs at different molar ratio, temperature and pH.

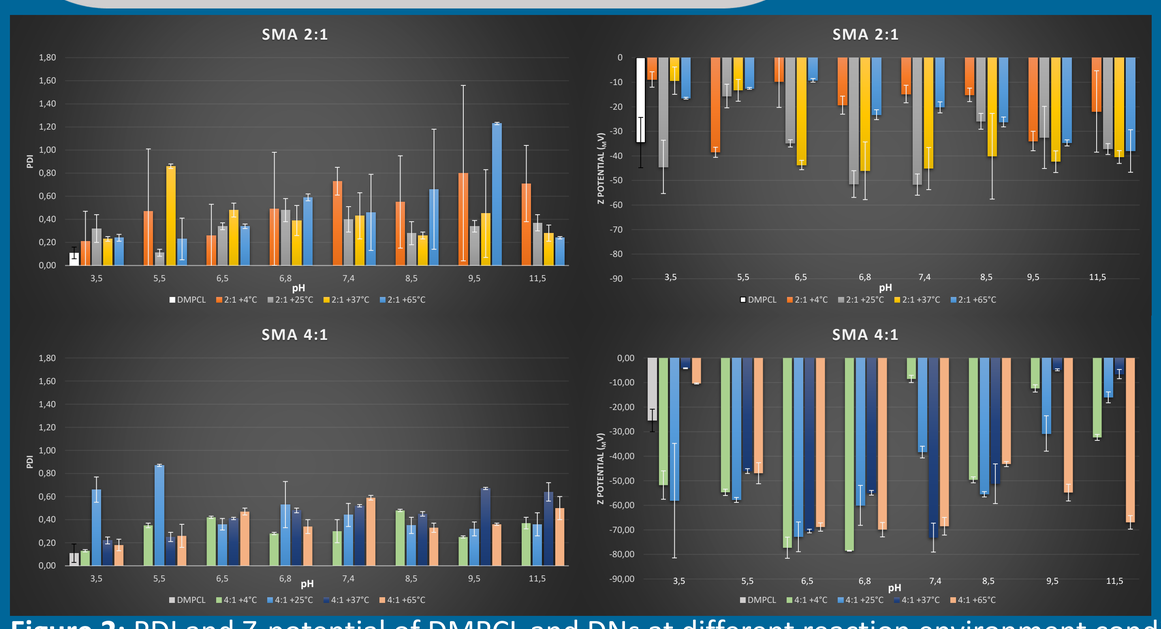

Figure 2: PDI and Z-potential of DMPCL and DNs at different reaction environment conditions.

RESULTS AND DISCUSSION - The physicochemical characterization of DMPC Liposome (DMPCL) was made at all pH (data not show). DMPCL is a homogeneous population with a low PDI, a size around 150 nm and a Z-potential about -40 mV. After incubation with SMA the physicochemical characterization shows a Z-potential variation, an increase of PDI (Figure 2), and size difference (Table I). DNs are obtained only under certain condition (Table I) and results are agree with literature data on SMA behaviour [1]. SMA with a molar ratio 2:1 forms DNs best at pH around neutrality, while the polymer with molar ratio 4:1 at basic pHs. The increase of DNs PDI compared to DMPCL could be due to the presence to bare liposomes and some SMA aggregates (data not show).

CONCLUSION - DNs could be an innovative drug delivery systems for anticancer therapy. Reaction environment of SMA affects the synthesis of DNs as well as drug delivery.

ACKNOWLEDGMENT - This research is funded by the Italian Ministry of Instruction, University, and Research under the national project PON Ricerca e Innovazione 2014-2020.

REFERENCES - "Effect of polymer composition and pH on membrane solubilization by styrene-maleic acid copolymers". Scheidelaar S., Koorangevel M.C., van Walree C.A., Dominguez J.J., Dorr J.M. and Killian J.A., Biophysical Journal, 2016.

UNIONE EUROPEA
Fondo Sociale Europeo

Ministero dell'Istruzione, dell'Università e della Ricerca

PON RICERCA E INNOVAZIONE 2014 - 2020