Electronic Supplementary Information (ESI)

In silico prediction of the in vitro behavior of polymeric gene delivery vectors

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Parameters: Bonds, angles and dihedral angles for branched PEIs

Albeit the lack of published parameters, the criteria for replacing unparametrized bonds, angles and dihedral angles always pursued preferring the most similar among all available. In fact, there were only two issues: whether a *missing charge* (a published parameter considering an "S" bead instead of an "Sq"), issue that can be understood when considering such high protonation level of secondary amines according to *in vitro* results (100%), or a *missing bead* (published "s" bead instead of t). The latter situation was infrequent.

Table S1: Bond parameters replacements

BONDS	
Missing charge	
Not published	Replaced with
sq-pq	s-pq
sq-sq	sq-s (same as s-sq)

Table S2: Angle parameters replacements

ANGLES	
Missing charge	
Not published	**Replaced with**
sq-t-sq	sq-t-s
t-t-sq	t-t-s
t-sq-pq	t-s-pq
Nsq-t-t	Ns-t-t
Np-t-sq	Np-t-s
t-sq-sq	t-s-sq
sq-sq-t	sq-s-t

Table S3: Dihedral parameters replacements

DIHEDRALS ANGLES			
Missing charge		**Missing bead**	
Not published	Replaced with	Not published	Replaced with
sq-t-t-p	s-t-t-p	sq-t-s-pq	sq-t-t-pq
Nt-t-sq-pq	Nt-t-s-pq	t-t-sq-sq	t-t-t-s
Nsq-t-t-sq	Ns-t-t-s	sq-sq-t-t	s-t-t-t
t-sq-sq-t	t-sq-s-t		
Nsq-t-t-p	Ns-t-t-p		
sq-sq-t-sq	s-sq-t-s		
sq-t-sq-pq	s-t-s-pq		
t-t-sq-pq	t-t-s-pq		
Np-t-sq-pq	Np-t-s-pq		

Figure S1. Timeline of siRNA-PEIs transfection assays.

t$_{-24h}$ t$_{0h}$ t$_{4h}$ t$_{48h}$

cell
seeding

polyplex
delivery

polyplex
removal and
cell incubation

cytotoxicity (%) and
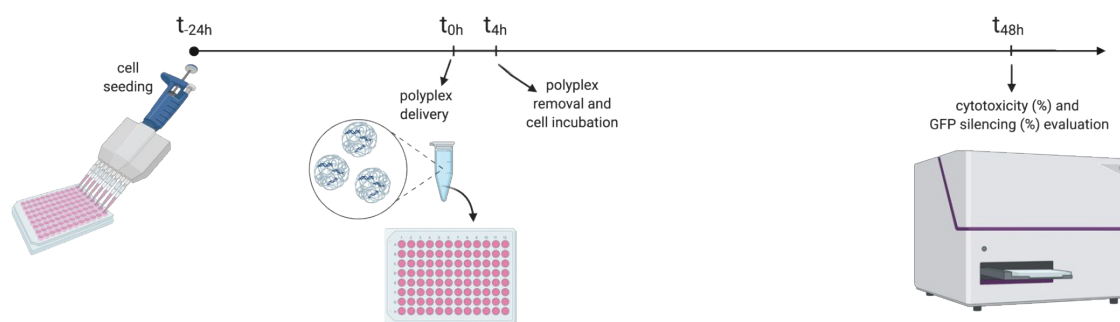GFP silencing (%) evaluation

Figure S2. Cytotoxicity (%) evaluated on GFP-293 cells of siRNA scramble-PEIs complexes (10 kDa *l*PEI = white bars; 10 kDa *b*PEI = black bars) prepared at different N/Ps (5, 10, 15, 30). Results are expressed as mean ± SD (n ≥ 3).
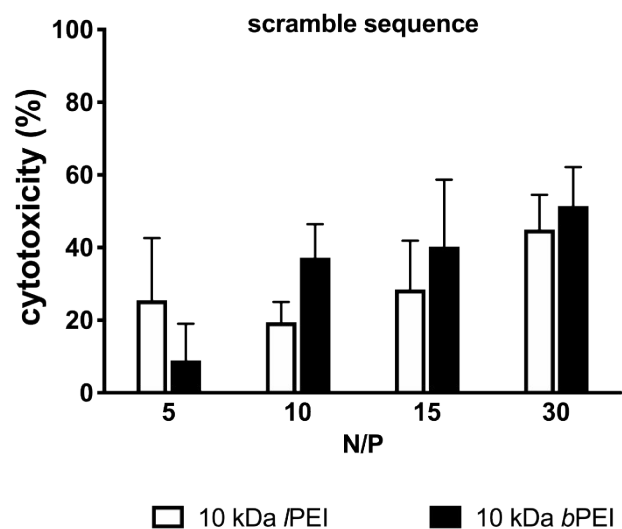
Figure S3. Polydispersity index (PDI) of siRNA-PEIs complexes prepared at N/P 10. Data were obtained while measuring the size (expressed in terms of D_H) using Dynamic Light Scattering (DLS) technique.