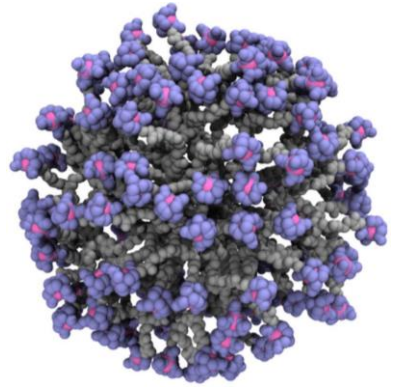
Complexation properties of glycodendrimers towards nucleotides



COST ACTION CA 17140

NANO2CLINIC

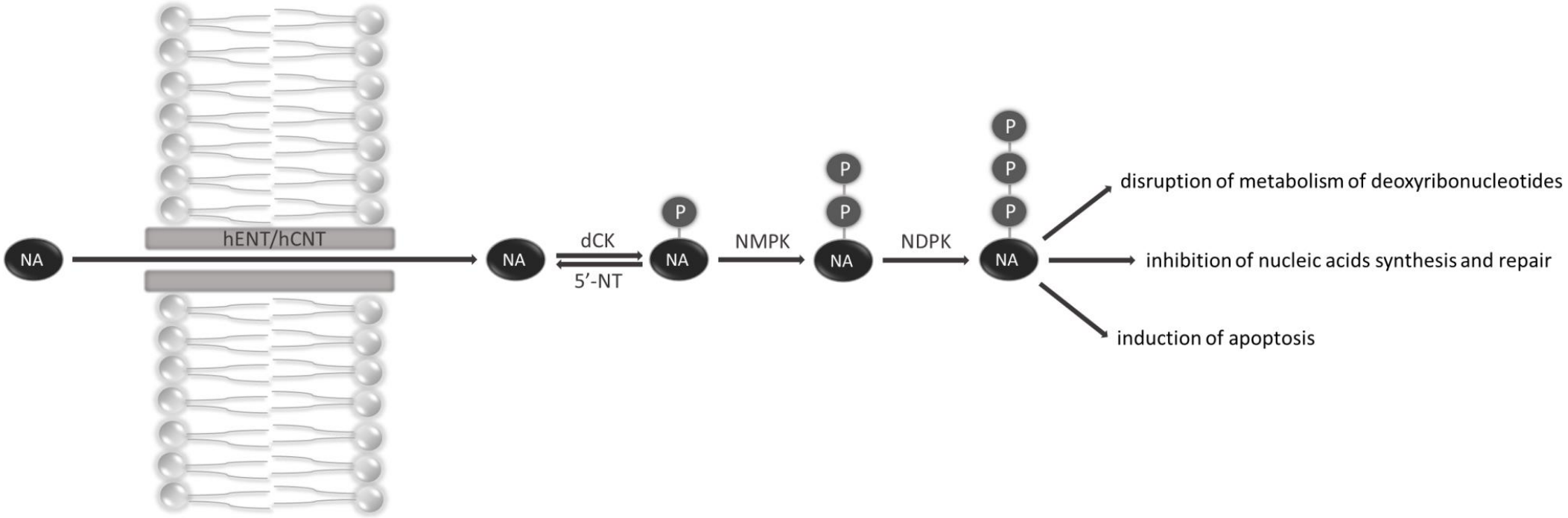
CANCER NANOMEDICINE - FROM THE BENCH TO THE BEDSIDE

Michał Gorzkiewicz, Ph.D

Department of General Biophysics, Faculty of Biology and Environmental Protection, University of Lodz, Lodz, Poland

FACULTY OF BIOLOGY AND ENVIRONMENTAL PROTECTION
University of Lodz

Nucleoside analogues – mechanism of action



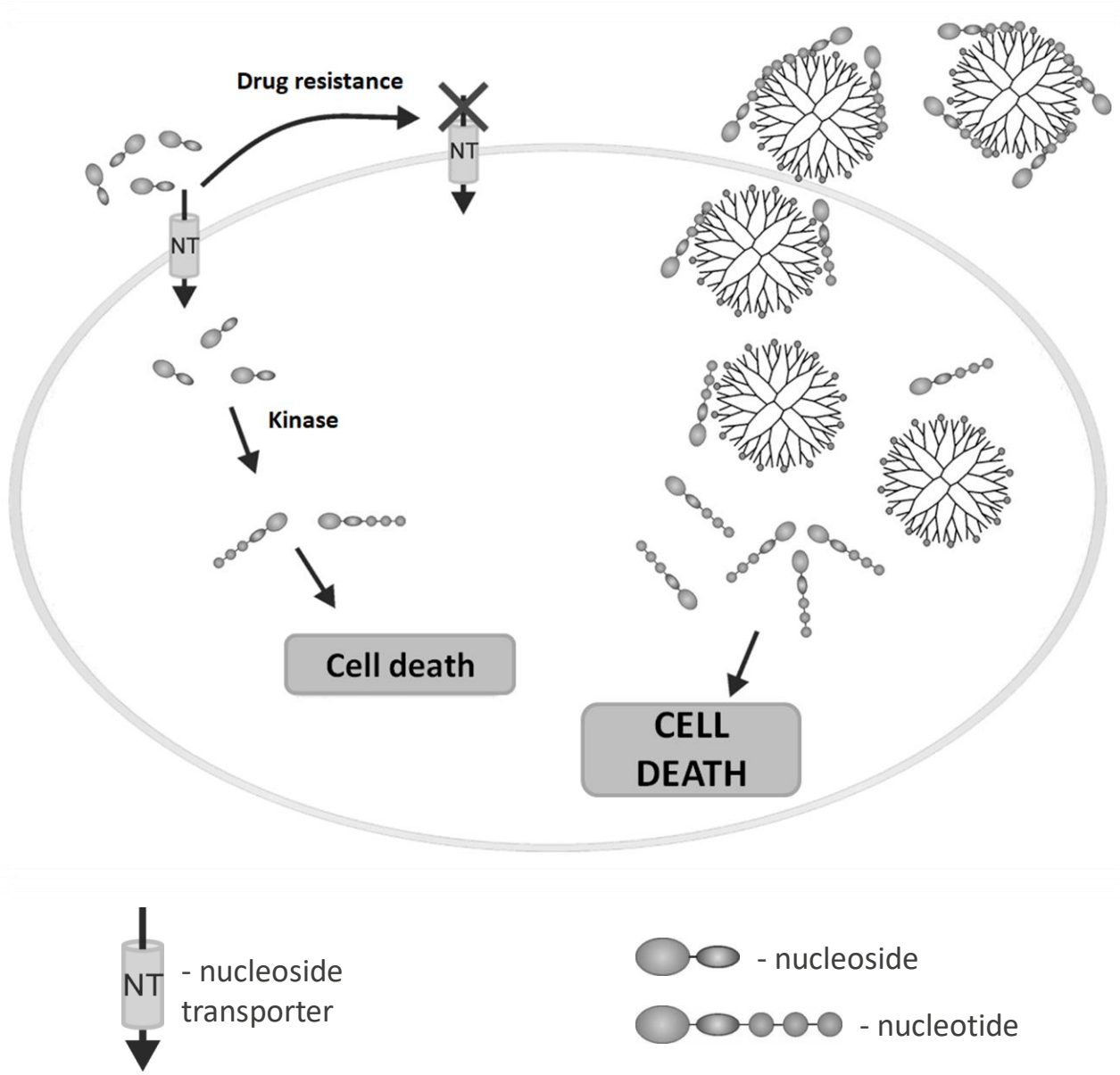
Metabolism and mechanisms of action of nucleoside analogues

NA: nucleoside analogue; P: phosphate group; hENT/hCNT: human equilibrative/concentrative nucleoside transporter; dCK: deoxycytidine kinase; NMPK: monophosphate kinase; NDPK: diphosphate kinase; 5'-NT: 5'-nucleotidase

Purpose of work and research hypothesis

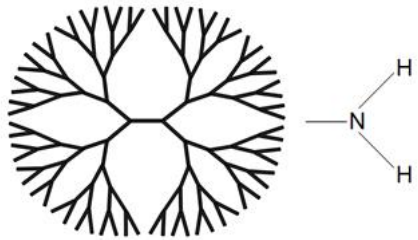
Goal: characterization of complexes of adenosine nucleotides with sugar-modified poly(propyleneimine) (PPI) dendrimers of the 4th generation and evaluation of the possibility their application as carriers of anticancer nucleoside analogues.

Research hypothesis: PPI dendrimers can effectively interact with therapeutic nucleotides and transport active, triphosphate forms of drugs directly into the cells, thus increasing their effectiveness and enabling the overcoming of resistance mechanisms.

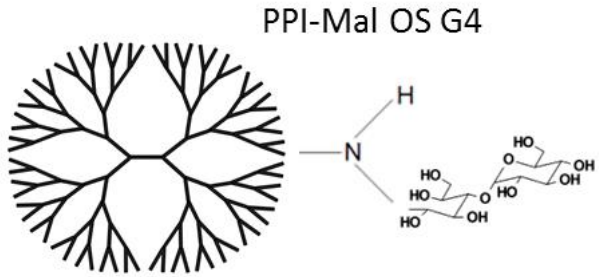


Complex formation – ITC and zeta potential measurements

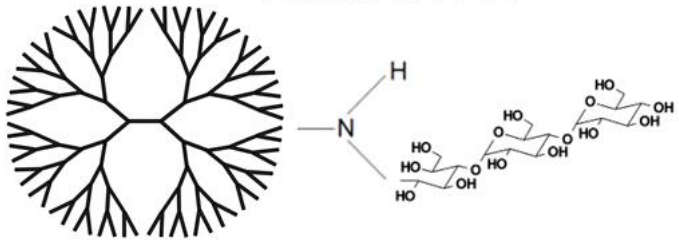
PPI G4



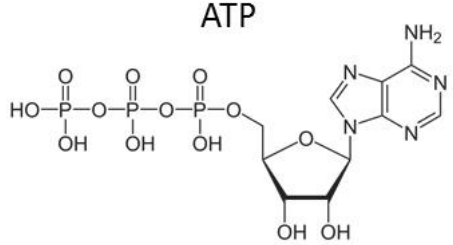
PPI-Mal OS G4

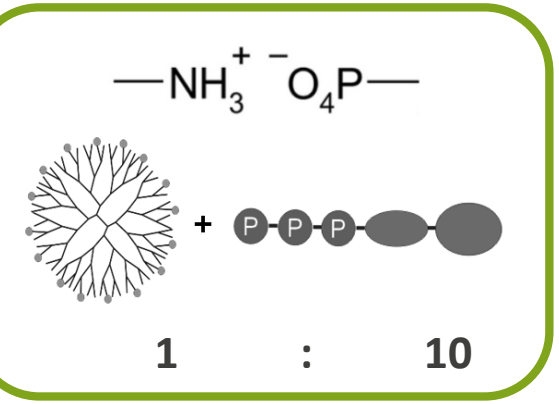


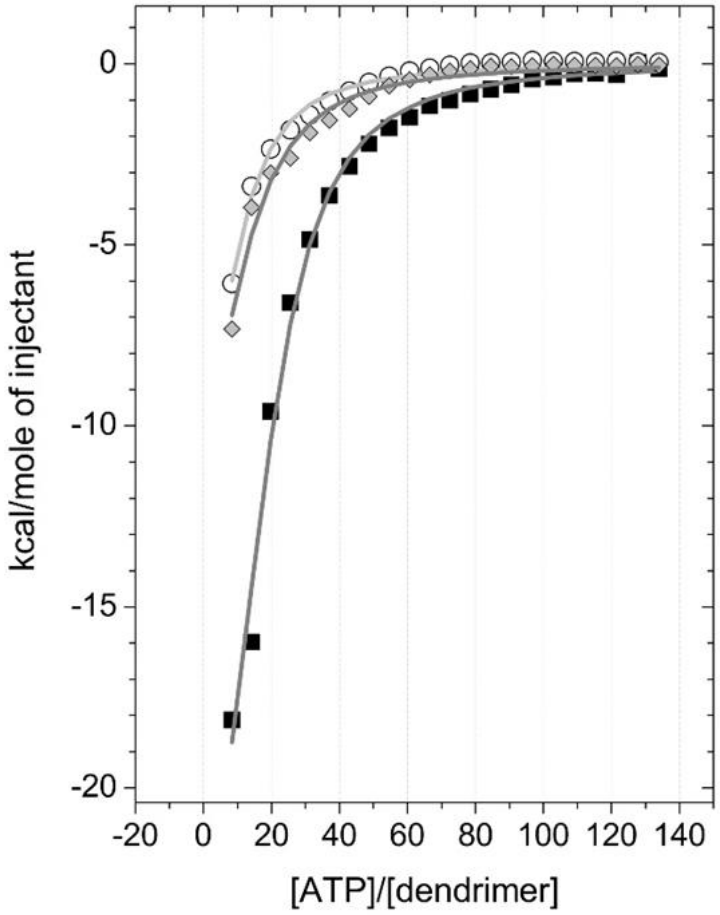
PPI-Mal III OS G4



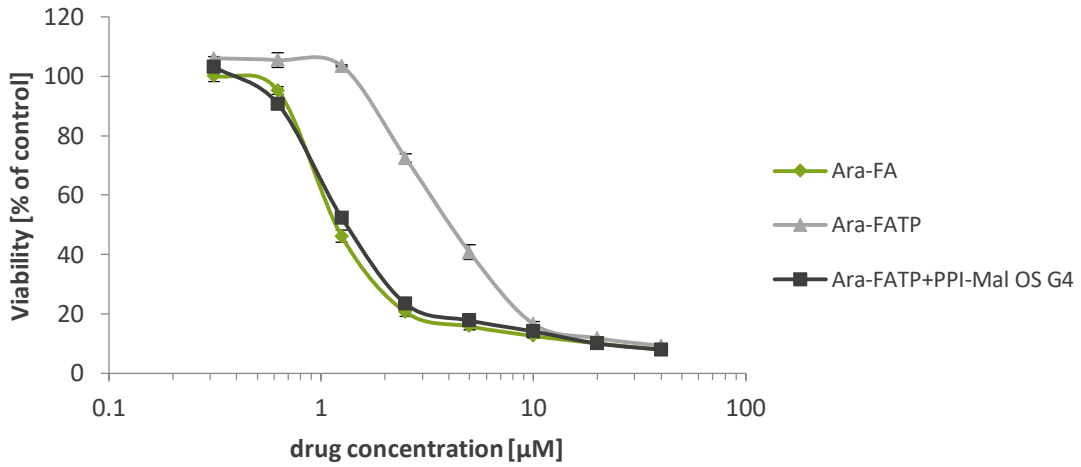
ATP



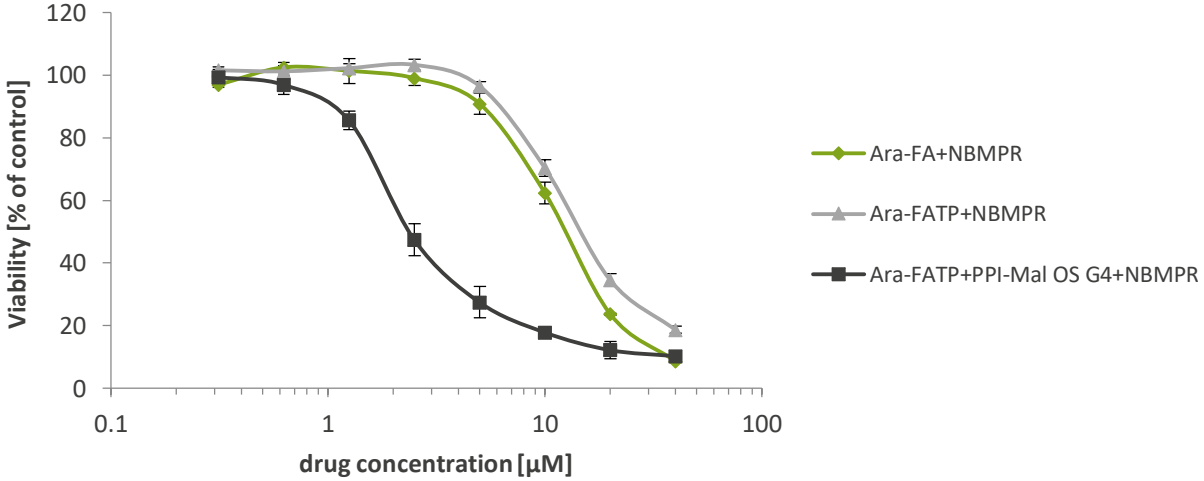




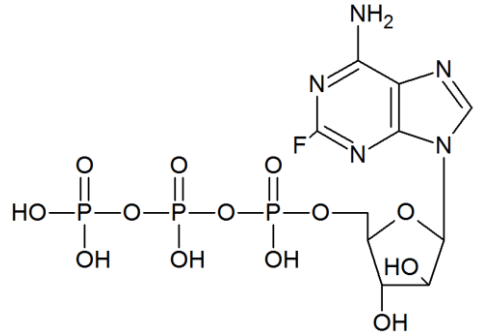
PPI-Mal OS G4 dendrimer as nanocarrier for Ara-FATP



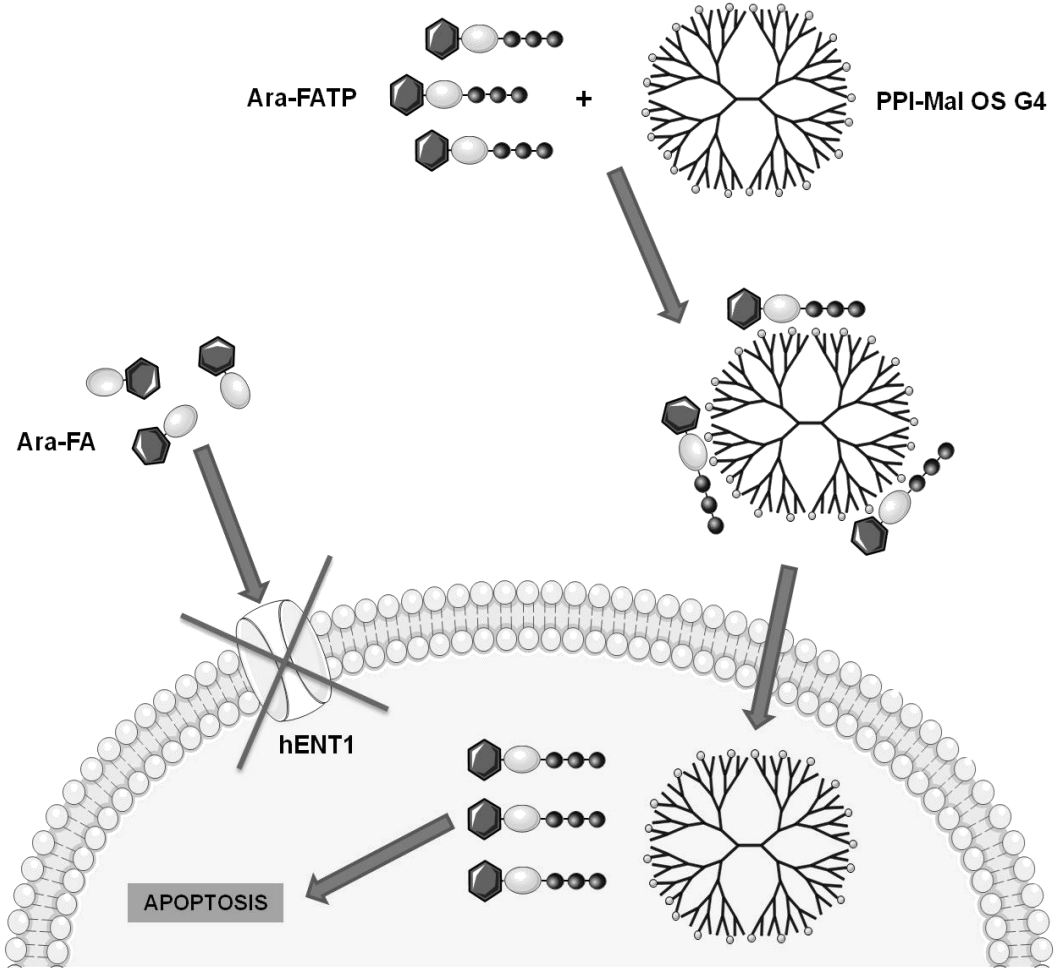
Cytotoxic effect of Ara-FA, Ara-FATP, and Ara-FATP complexed with PPI-Mal OS G4 dendrimer on U937 cells (resazurin assay, average ± S.E.M., n = 6)



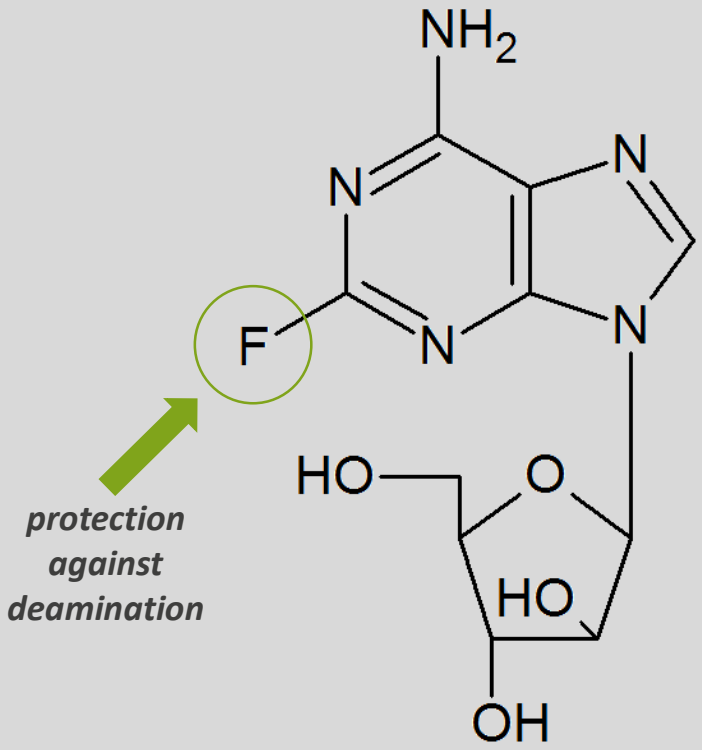
Cytotoxic effect of Ara-FA, Ara-FATP, and Ara-FATP complexed with PPI-Mal OS G4 dendrimer in the presence of hENT1 inhibitor (NBMPR) on U937 cells (resazurin assay, average ± S.E.M., n = 6)



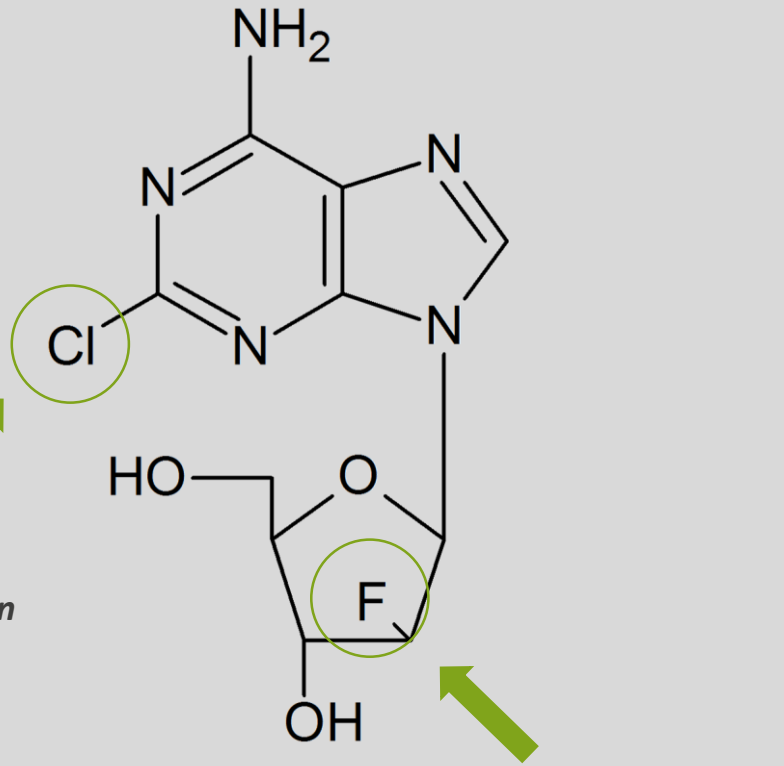
Chemical structure of fludarabine triphosphate (Ara-FATP)



Fludarabine vs. clofarabine



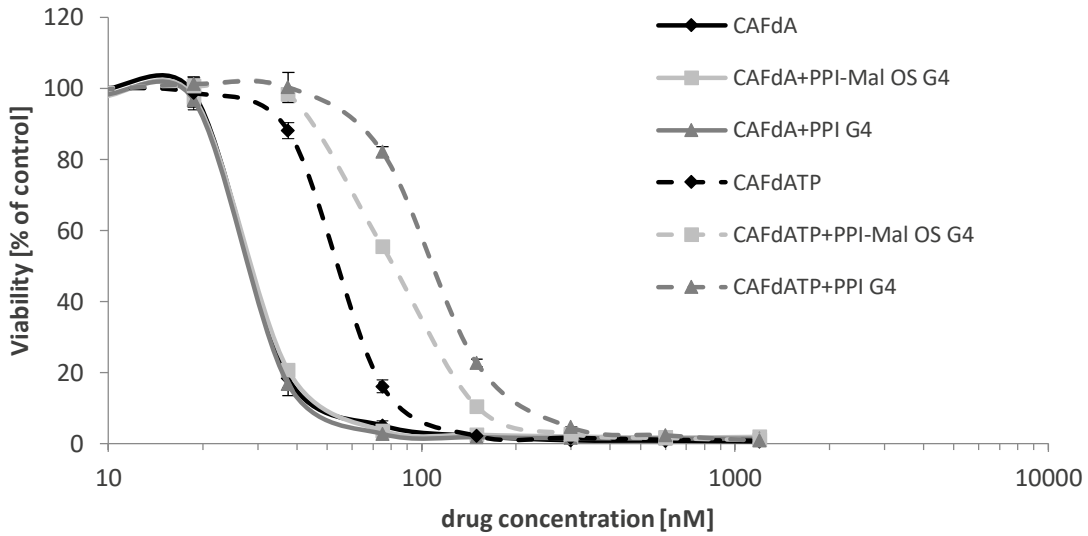
protection against deamination



protection against deamination

enhanced stability of glycosidic bond in acidic pH

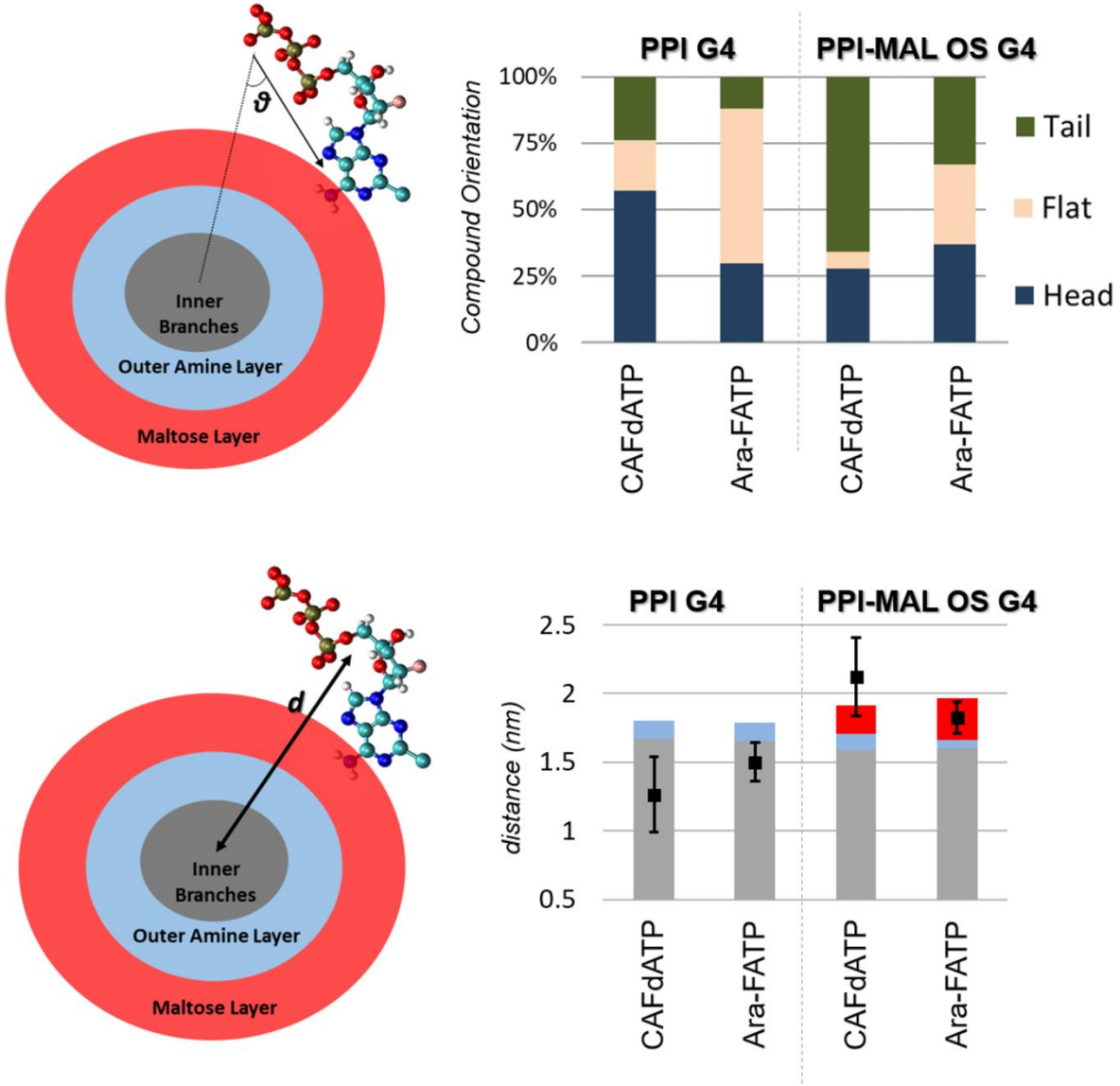
Fludarabine vs. clofarabine



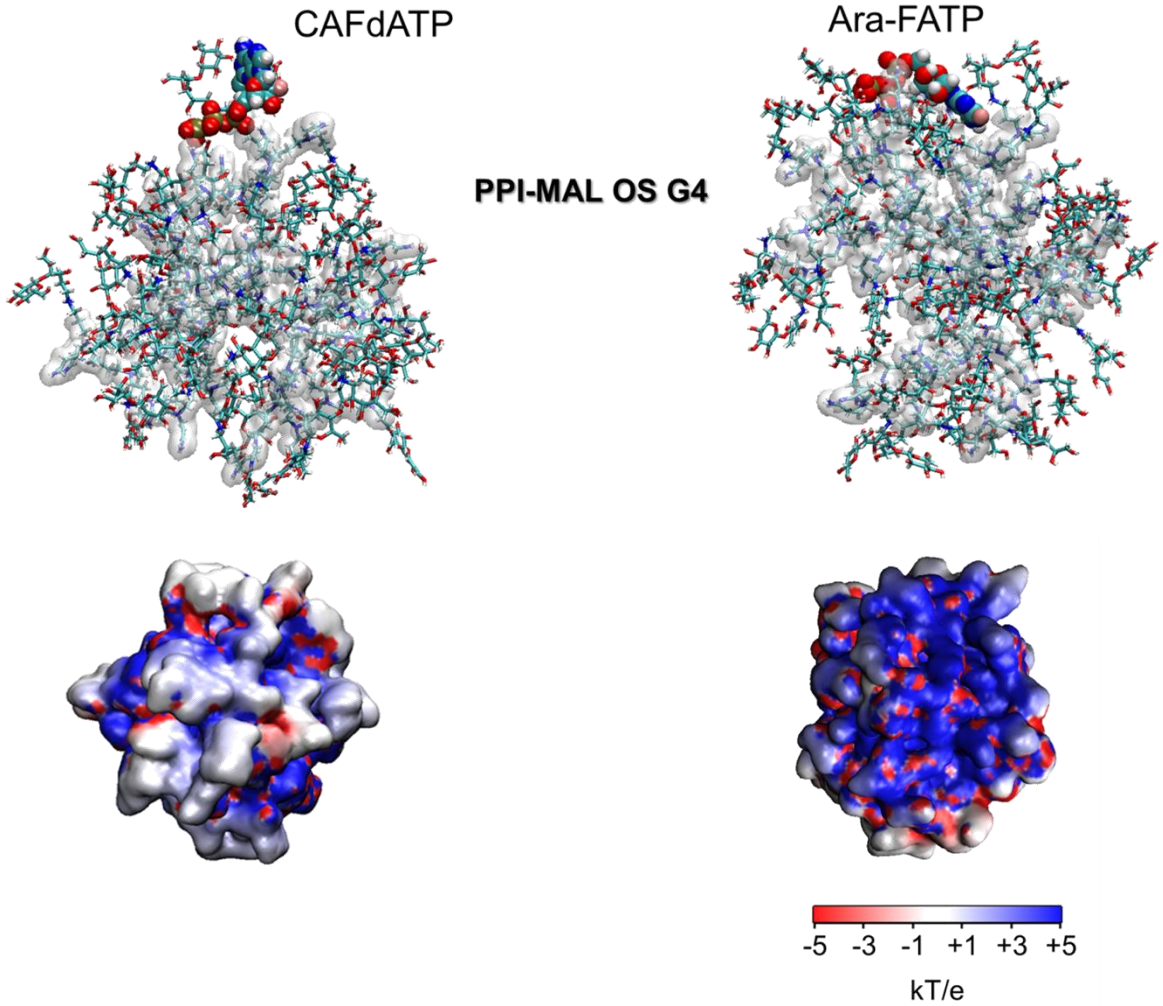
Cytotoxic effect of CAFdA, CAFdATP and their complexes with PPI G4 and PPI-Mal OS G4 dendrimers on U937 cells (resazurin assay, average ± S.E.M., n = 3)

	IC50 ± S.E.M. [nM]
CAFdA	31.05±2.66
CAFdA+PPI-Mal OS G4	32.81±2.50
CAFdA+PPI G4	30.69±1.67
CAFdATP	54.45±1.48
CAFdATP+PPI-Mal OS G4	78.70±1.39
CAFdATP+PPI G4	107.40±4.74

Fludarabine vs. clofarabine



Scheme of the nucleotide-dendrimer interaction mode with focus on nucleotide orientation and nucleotide distance from the dendrimer core



Representative snapshots of nucleotide-dendrimer configurations (1:1 ratio, upper panel) and electrostatic maps for nucleotide-dendrimer complexes (10:1 ratio, lower panel)

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Zeta potential of PPI dendrimers and nucleotide-dendrimer complexes, presented as mean ± SD, n = 9.
[a, b, c] Statistically significant difference between samples at p <0.05

	Zeta potential [mV] ± SD
PPI G4	35.7±5.7
Ara-FATP-PPI G4 (10:1)	26.8±1.8 [a]
Ara-FATP-PPI G4 (20:1)	13.0±0.3 [b]
CAFdATP-PPI G4 (10:1)	10.5±0.9 [a]
CAFdATP-PPI G4 (20:1)	-1.4±0.8 [b]
PPI-Mal OS G4	28.3±3.3
Ara-FATP-PPI-Mal OS G4 (10:1)	13.2±1.6 [c]
CAFdATP-PPI-Mal OS G4 (10:1)	4.0±0.3 [c]

Fludarabine vs. clofarabine

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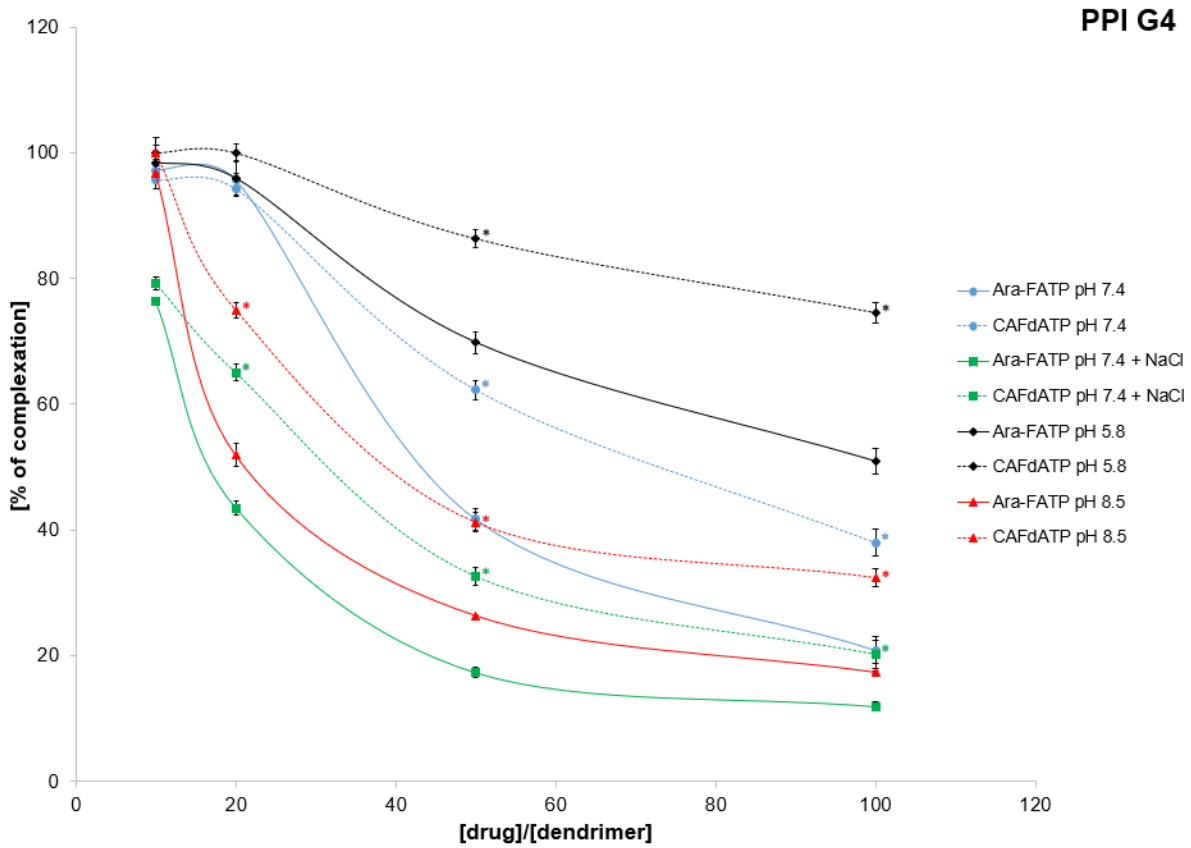

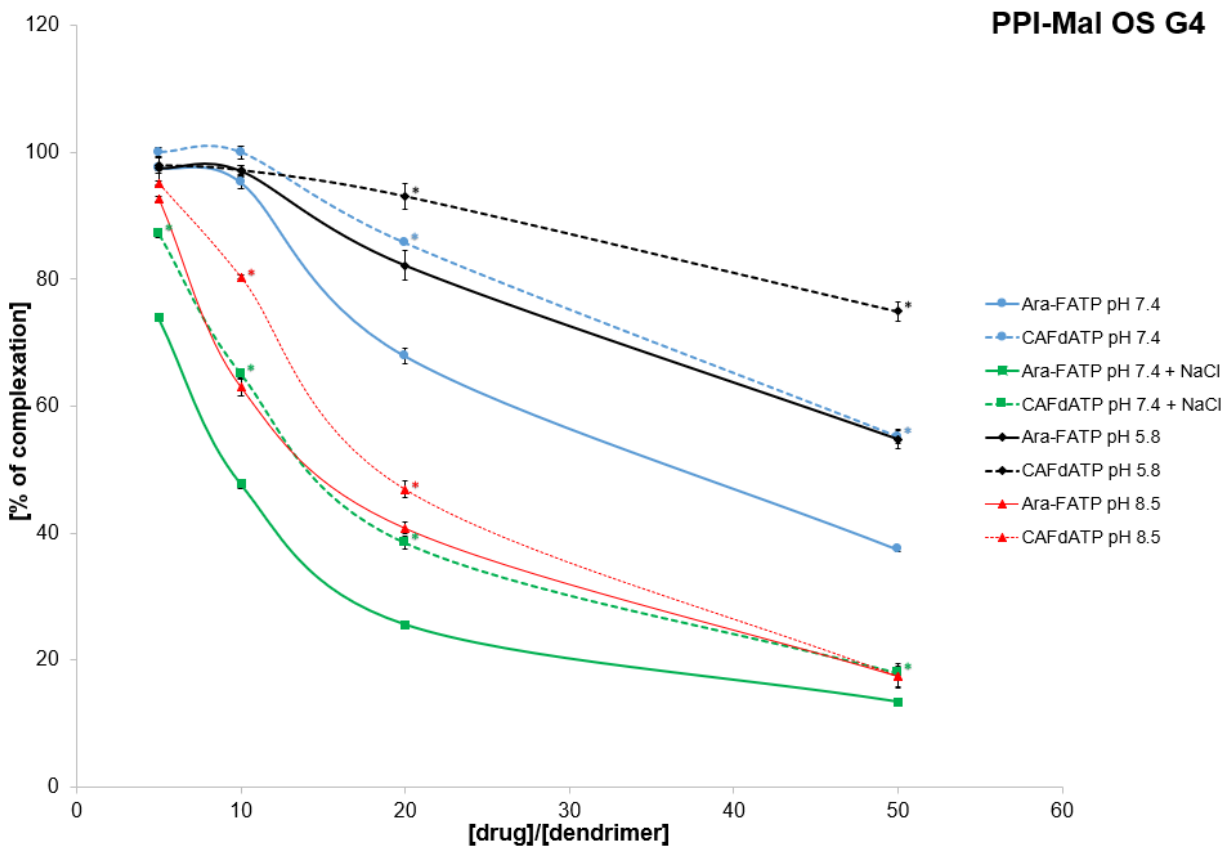

Analysis of the stability of the complexes using ultrafiltration. Results are presented as percentage of drug complexation efficiency, mean ± SD, n = 8.
** Statistically significant difference compared to Ara-FATP at p <0.05*

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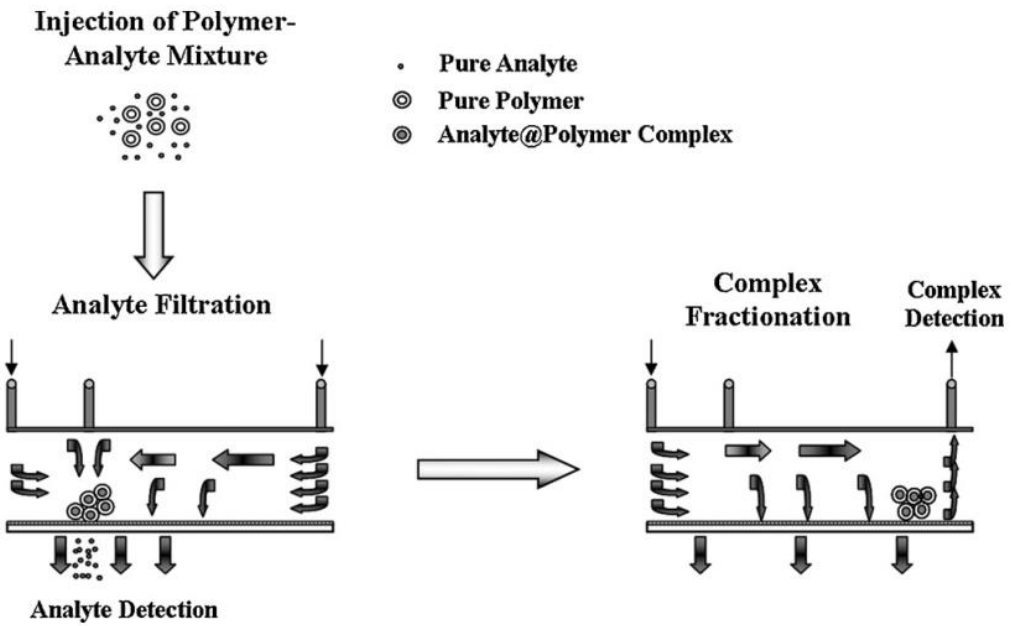
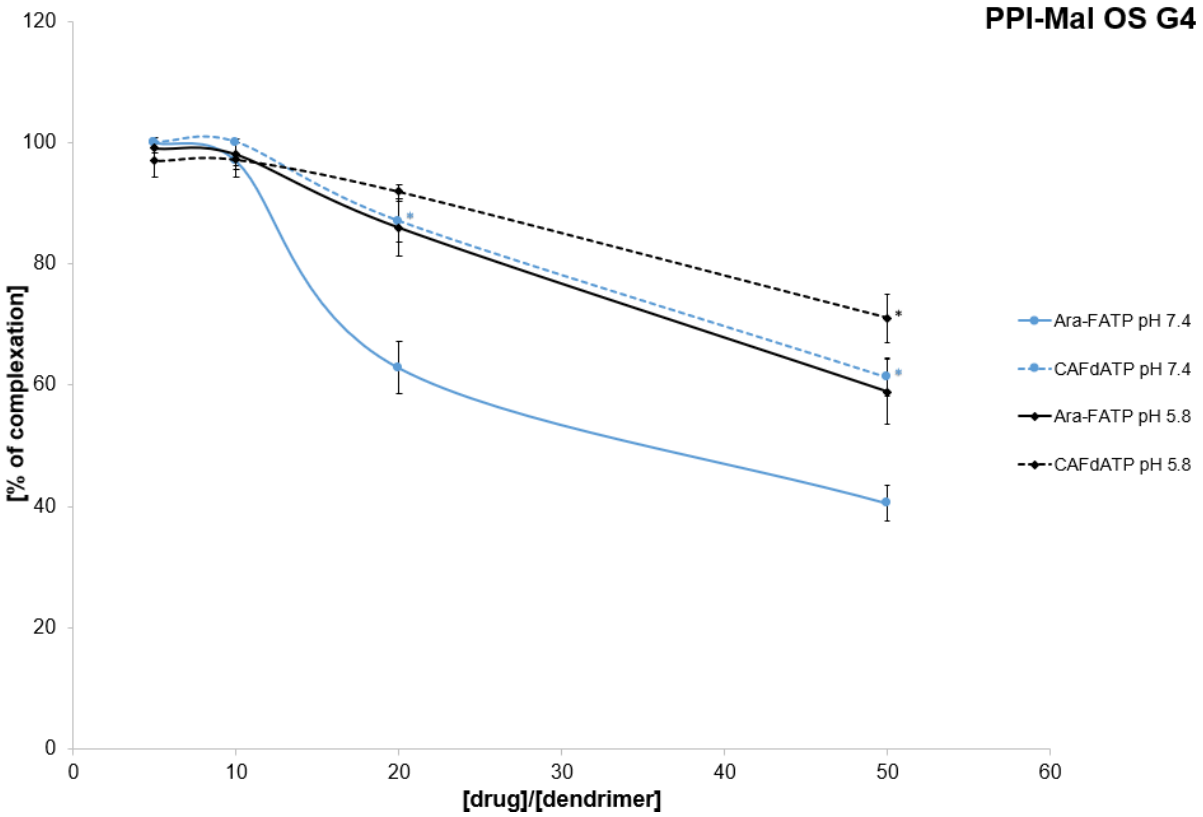
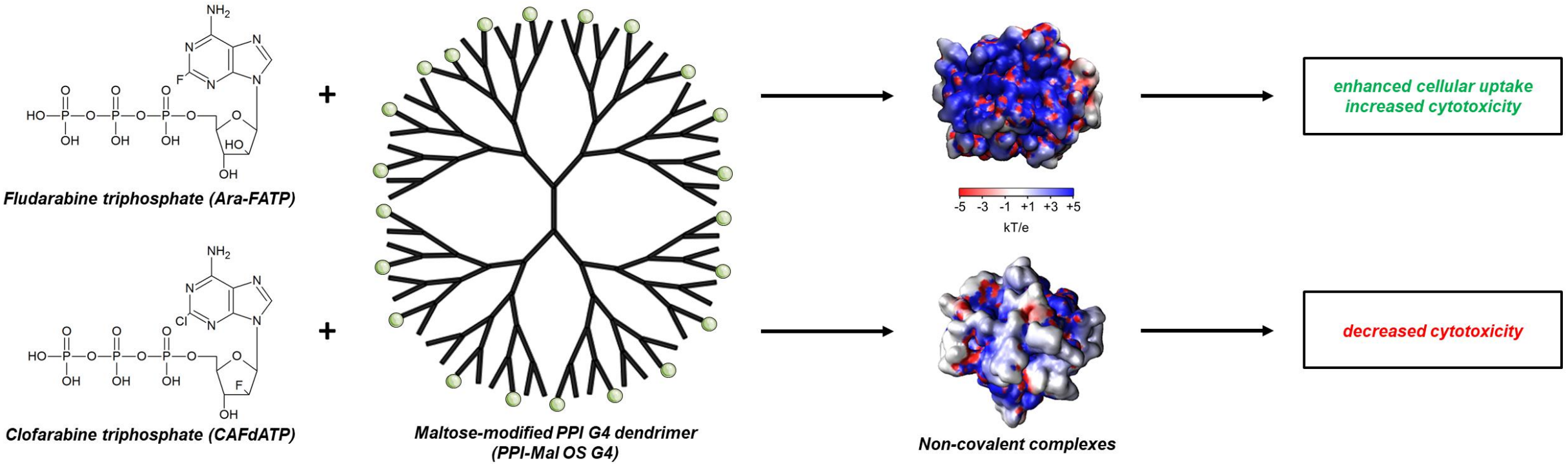


Principle of AF4 method for analyte filtration and analyte@polymer complex fractionation
(Boye et al., J Chromatogr A. 2010, 1217 (29) 4841-4849).



Analysis of the stability of the complexes with the use of AF4. Results are presented as percentage of drug complexation efficiency, mean ± SD, n = 8.
** Statistically significant difference compared to Ara-FATP at p <0.05*

Conclusions



Gorzkiewicz, M., Appelhans, D., Boye, S., Lederer, A., Voit, B., & Klajnert-Maculewicz, B. (2019). Effect of the structure of therapeutic adenosine analogues on stability and surface electrostatic potential of their complexes with poly (propyleneimine) dendrimers. Macromolecular Rapid Communications, 40(15), 1900181.

Thank you for your attention