AFM Study of DNA Complexes with Biologically Active Compounds

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Biologically Active Compounds

- Metal Ions
- Coordination compounds
- Polyamines
- Polycations

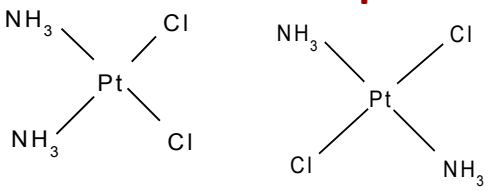
Trivalent:

- Fe3+ ($FeCl_{3)}$
- La3+ ($LaCl_3$)
- Al3+

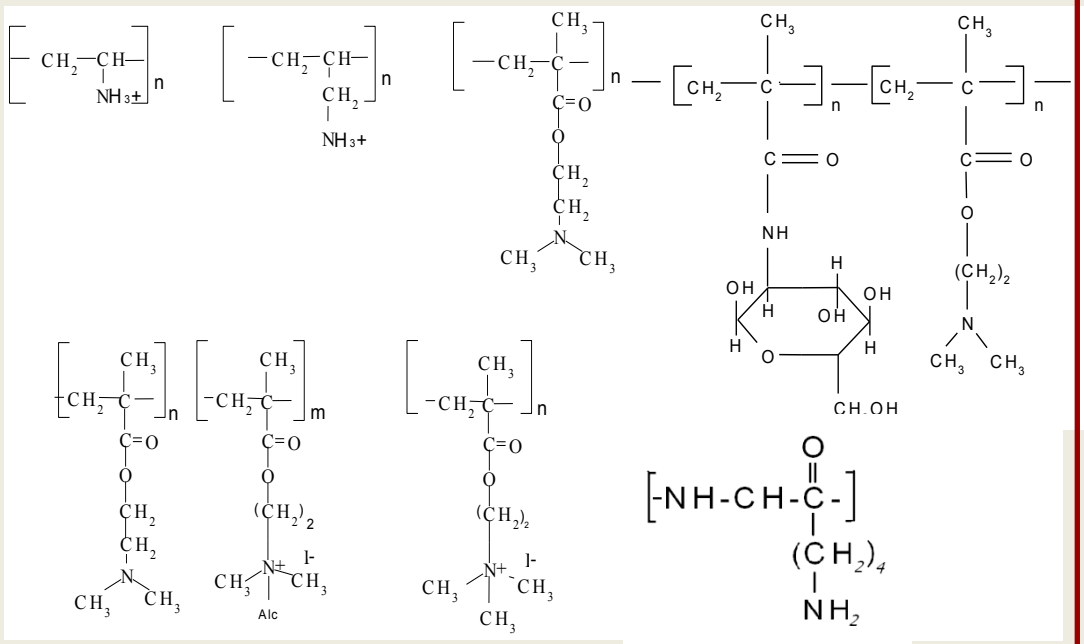
Divalent:

- Mg2+, Ba2+, Ca2+
- Mn2+, Ni2+, Cu2+

Coordination compounds



Platinum
Palladium
Cobalt
Ruthenium



Spermidine

$NH_2 – (CH_2)_3 – NH –(CH_2)_4 – NH_2$

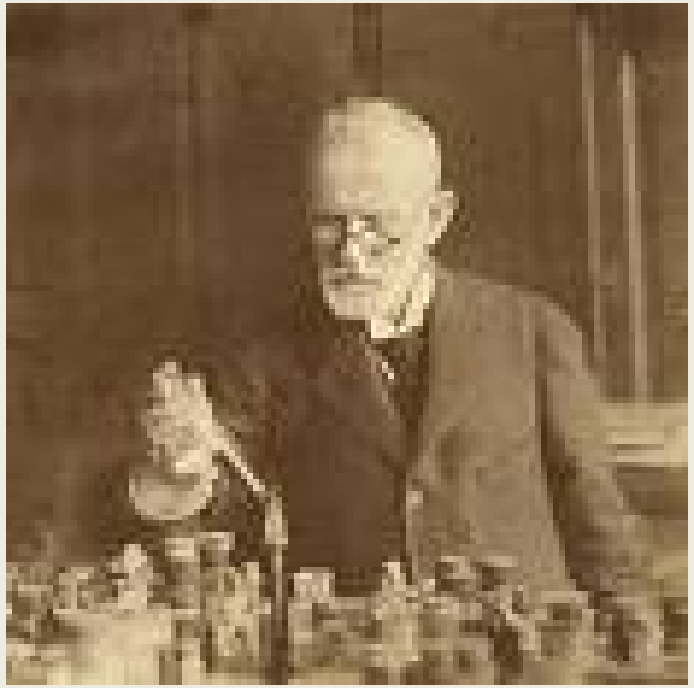
Spermine

$NH_2 – (CH_2)_3 – NH –(CH_2)_4 – NH –(CH_2)_2\text{-}NH_2$

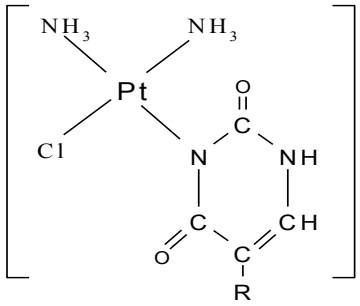
Chemotherapy based on toxic metals

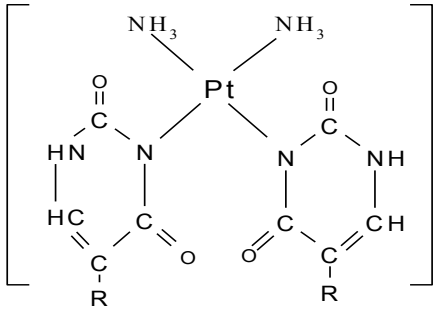
It is over 100 years since Paul Ehrlich envisioned the development of a "**Magic Bullet**" a dye carrying a toxic heavy metal which would target disease causing agents while leaving healthy tissue unharmed

In his Nobel Lecture on December 11th, 1908 Paul Ehrlich layed out the foundation of chemotherapy and the use of drugs to cure patients.

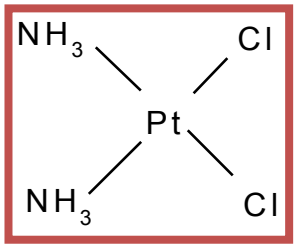


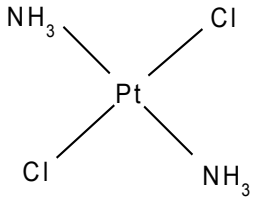
Paul Ehrlich
1854–1915

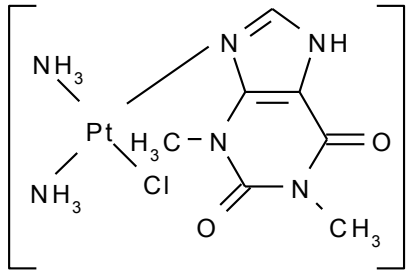


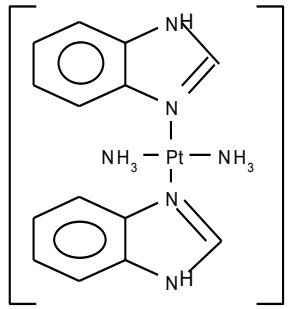


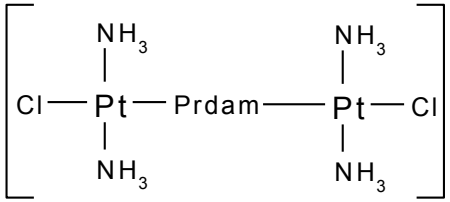
R=Br,H,NO$_2$

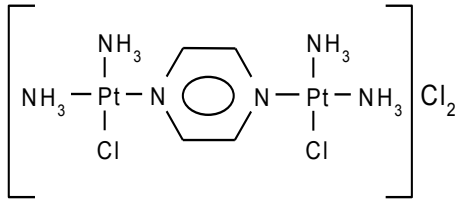


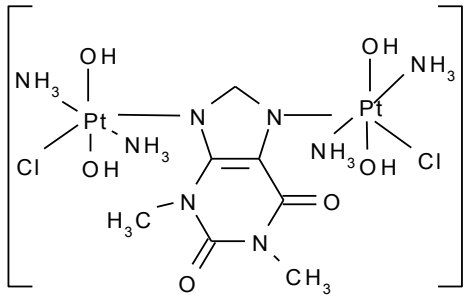


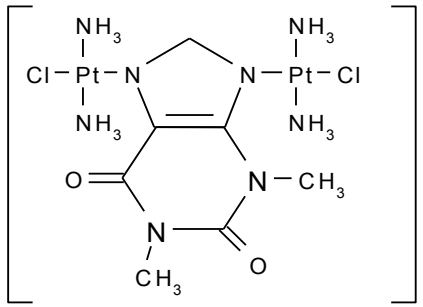


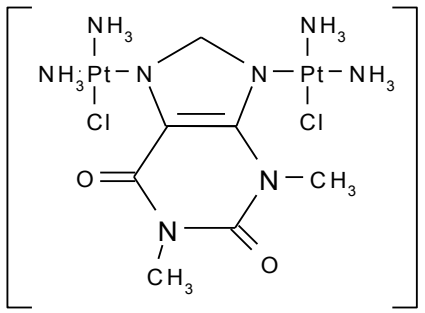


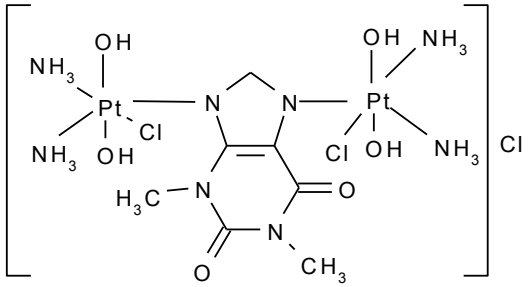




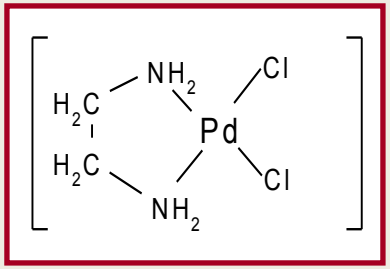


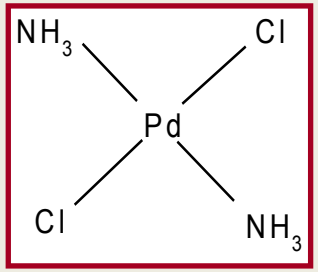


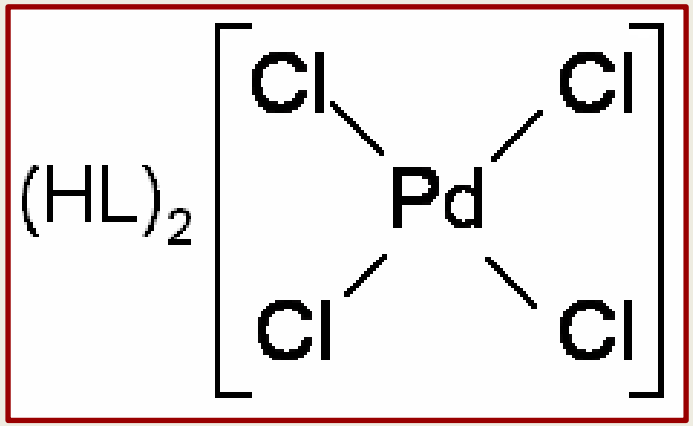


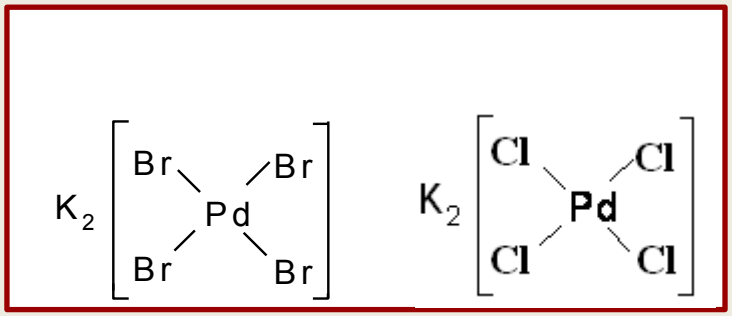


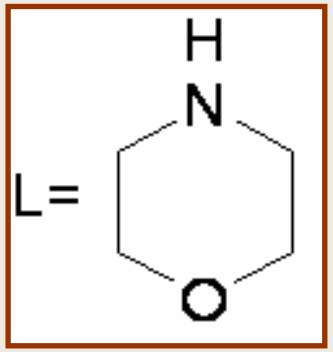
Palladium Coordination Compounds

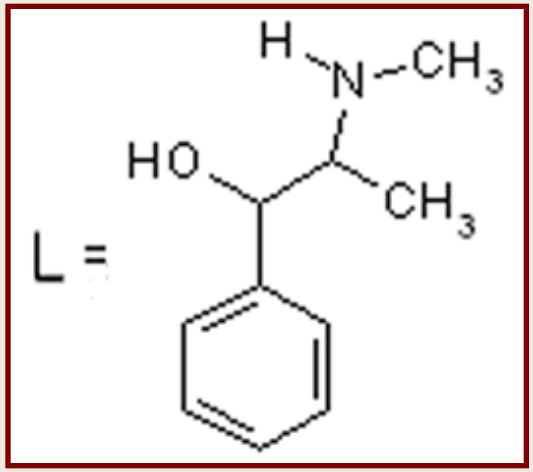


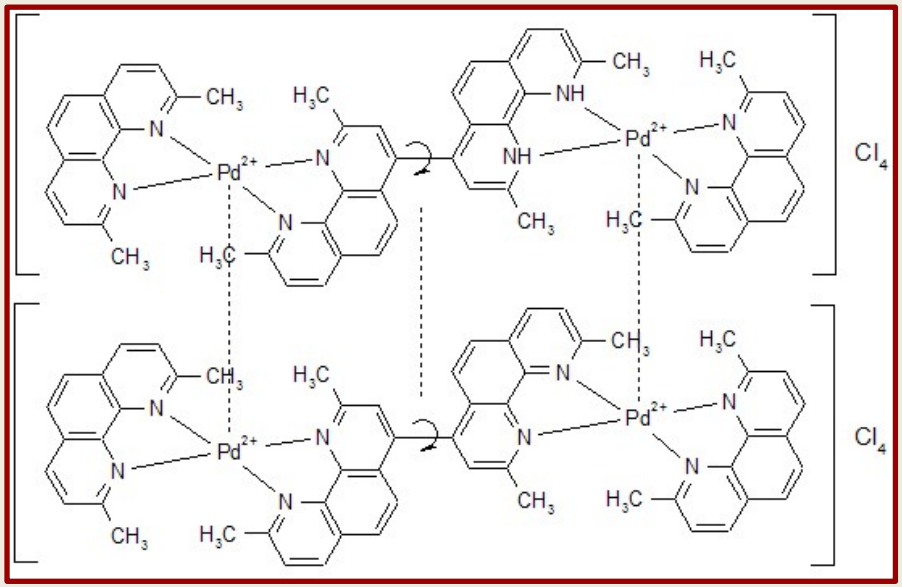




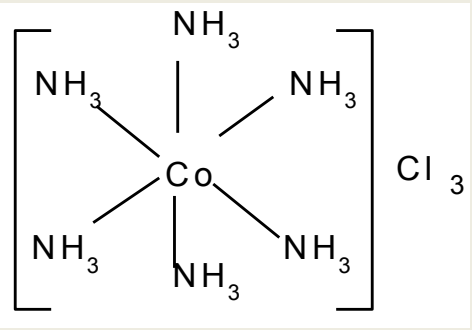


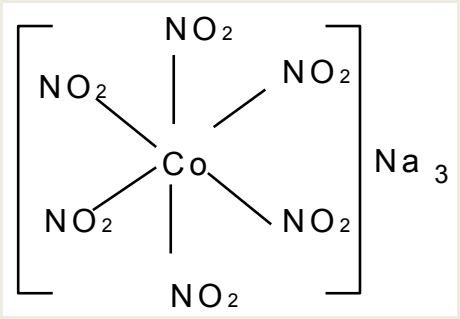


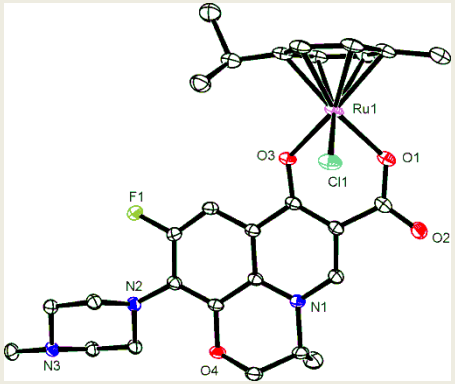


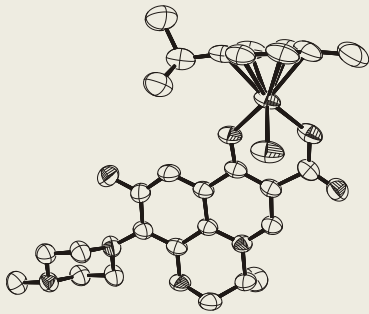


Coordination Compounds of Cobalt and Ruthenium





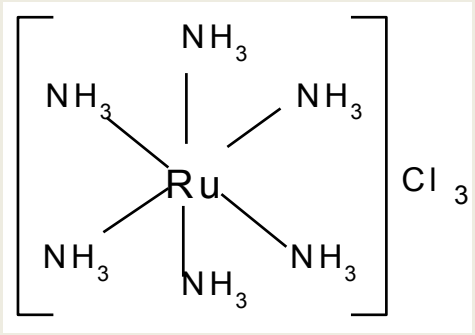


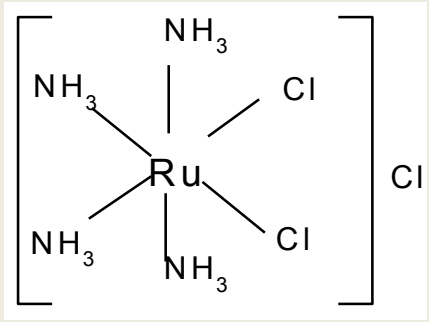


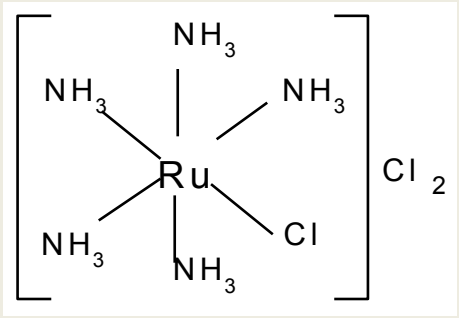
$[Co(NH_3)_6]$ 3+

$[Co(NO_2)_6]$ 3- -> $[Co(NO_2)(H_2O)_5]$ 2+ -> $[Co(H_2O)_6]$ 3+

Iztok Turel University of Ljubljana, Faculty of Chemistry and Chemical Technology







$[Ru(NH_3)_6]$ 3+

$[Ru(NH_3)_4(Cl)_2]$ +

$[Ru(NH_3)_5(Cl)]$ 2+

Experimental methods

- ATOMIC FORCE MICROSCOPY

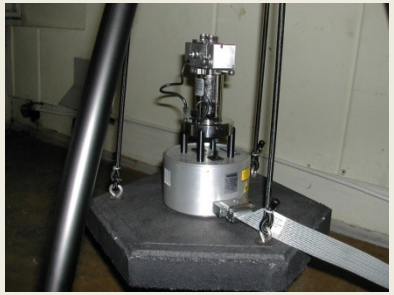
  NanoScope IV, Veeco
- DYNAMIC LIGHT SCATTERING

  PhotoCor, Russia
- CIRCULAR DICHROISM

  Mark IV, Jobin Ivon
- LOW GRADIENT VISCOMETRY

  Zimm-Crozers type viscometer
- FLOW BIREFRIGENCE

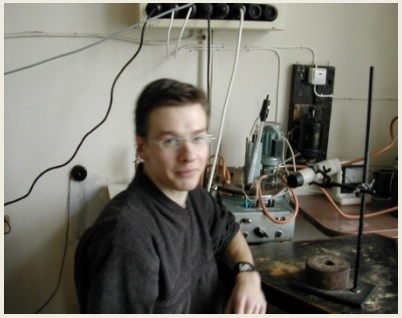
  Original technique
- UV SPECTROSCOPY
- SF 56, Russia
- ELECTROPHORESIS



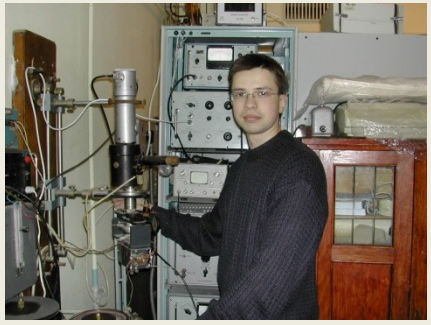
NanoScope IV, Veeco

Dichrograph Mark IV, France

UV-VIS SP-56, Russia

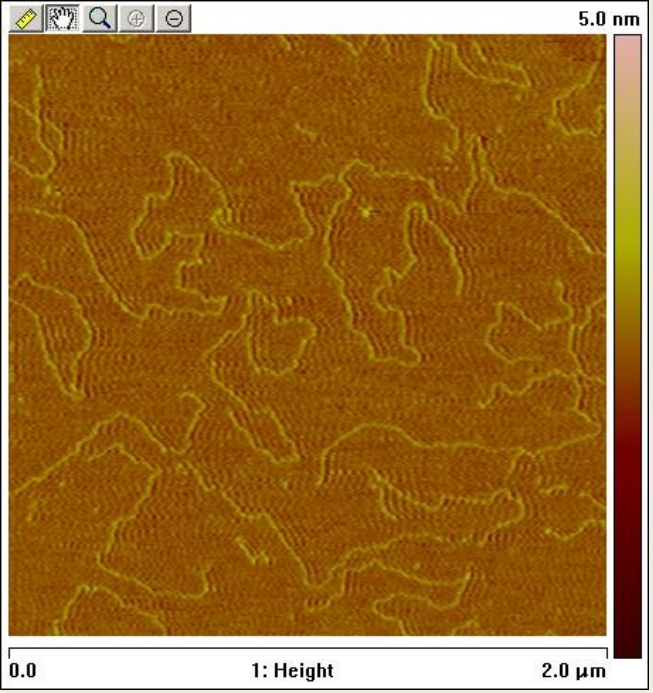


Low gradient Rotation viscometer



Flow Birefrigence

DNA

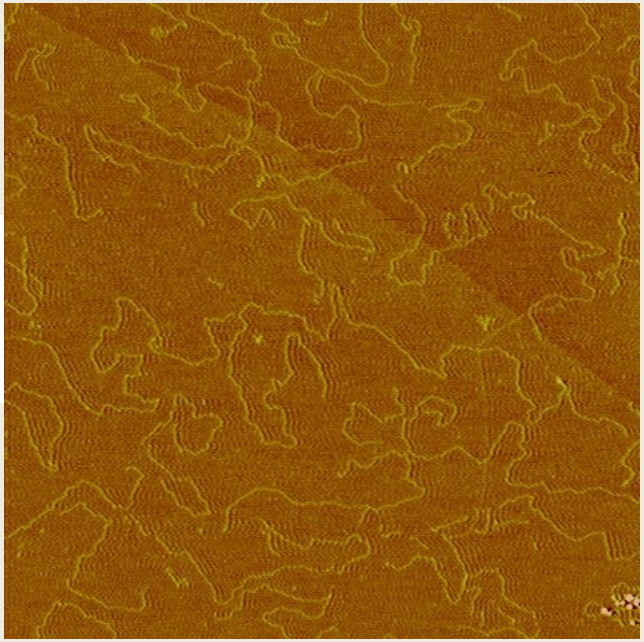


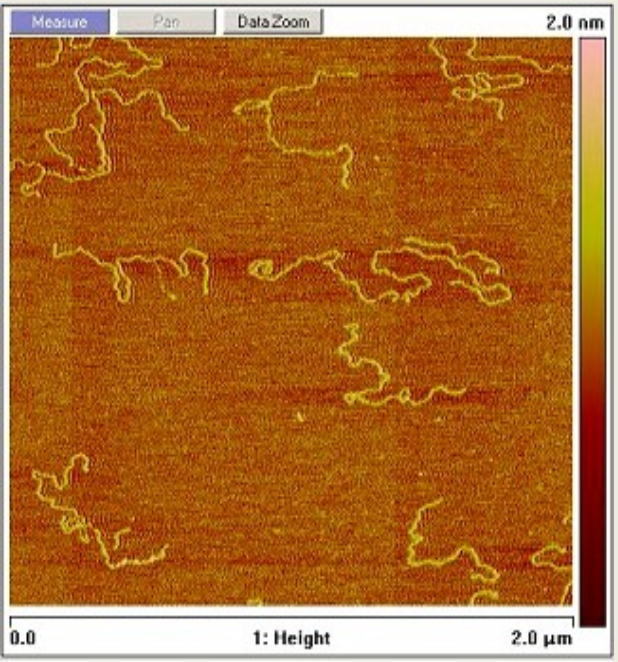
Calf Thymus DNA (Sigma)

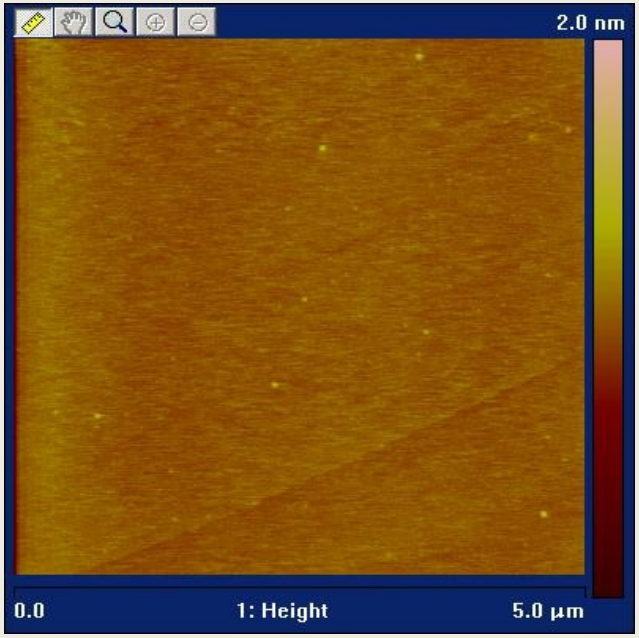
DNA pFL 44/EcoI (4,4 kbp)

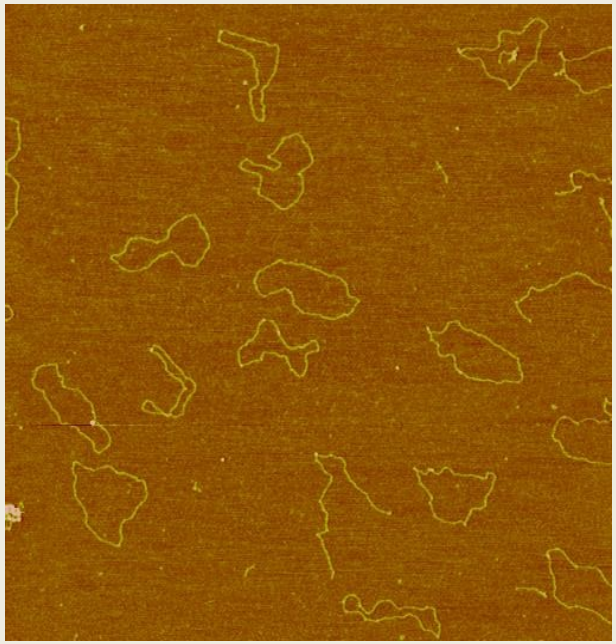
0.005 M NaCl

$5x10^{-4}$ M $MgCl_2$









1 M NaCl

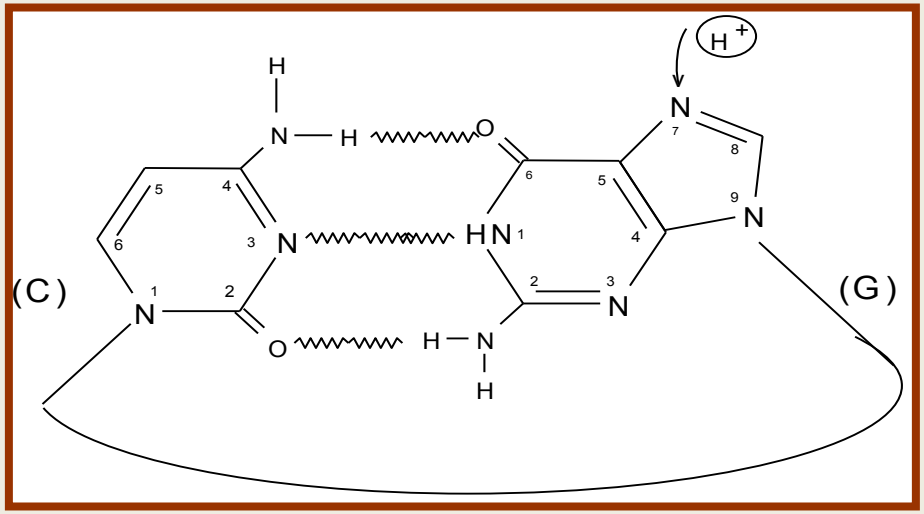
DNA is a highly charged polyion

DNA interaction with ligands in a solutions is greatly depended on electrostatic interaction:

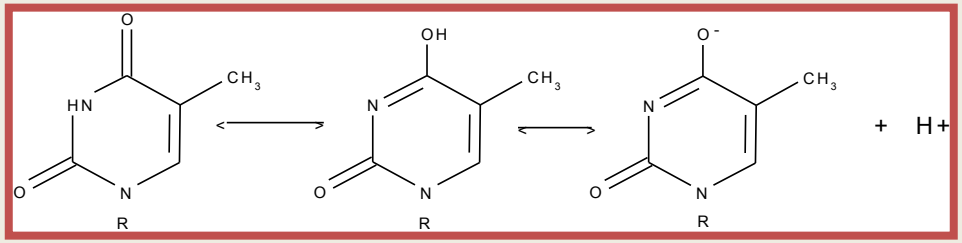
- Long-range electrostatic interactions (responsible for polyelectrolyte swelling)
- Short-rang electrostatic interactions (determine the electrostatic contribution to DNA persistent length)
- Non-electrostatic interaction also play an important role. Water-salt solution is a good solvent for native DNA (for single-strand it isn't as good as for double strand)

Variation in DNA charge density with pH

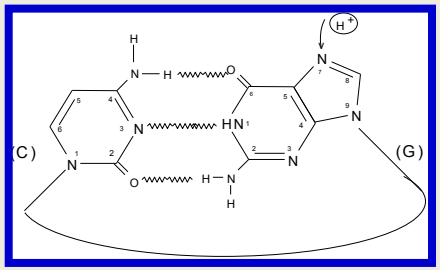
- **negative phosphates** (at pH>1,5)
- **additional positive or negative charges** on nitrogen bases at **acid** or **alkaline pH**

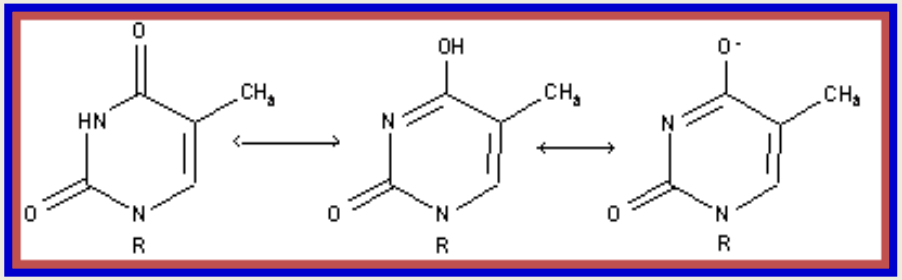


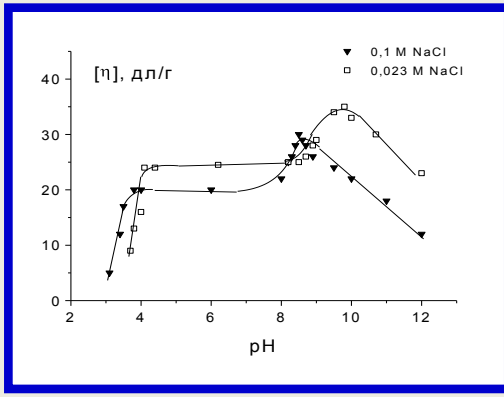
Guanene N7 is the main group for double stranded **DNA protonation**

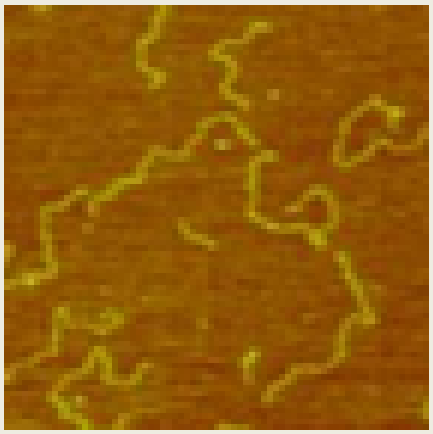


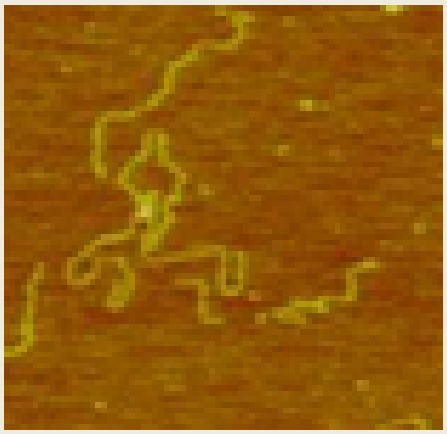
Timine and **Guanine** can get a **negative charge** in alkaline area

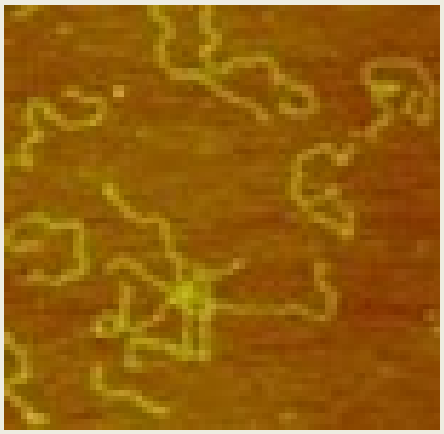




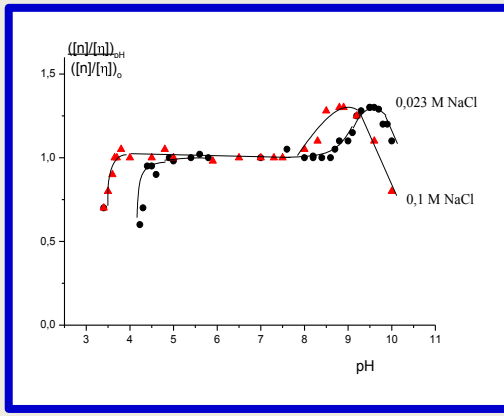


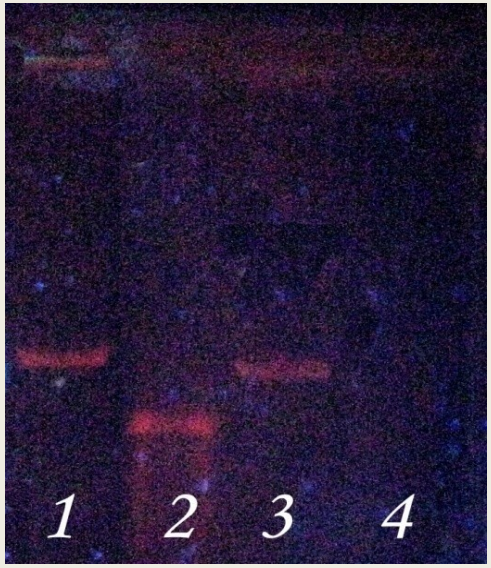






(1μm) **pH=6.2** **pH=11.3** **pH=9.5**



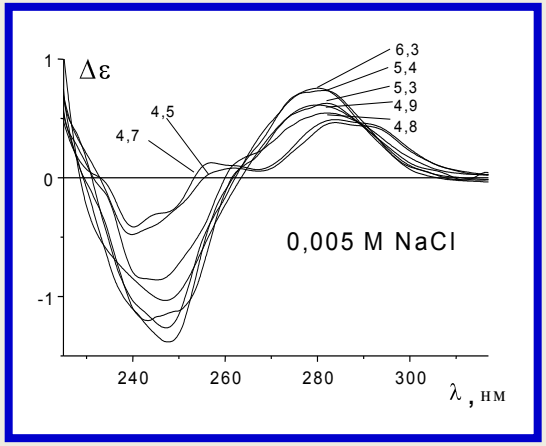


1 – pH=6,2
2 – pH=11,3
3 – pH=9,5
4 – pH=2,55

0,15 M
pK=3,1

0,023
pK=4,0

0,005
pK=4,7



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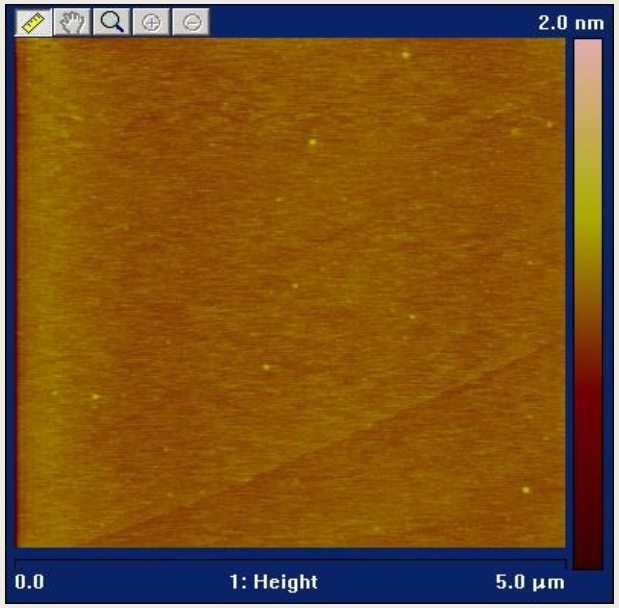
$$\frac{(\Delta n_i/g)_{g\to 0}}{(\eta_r - 1)\eta_0} = \frac{[\eta]}{\eta} = \frac{4\pi}{45kT}\cdot\frac{(\varepsilon_s^2+2)}{n_s}\cdot(\alpha_1-\alpha_2)$$

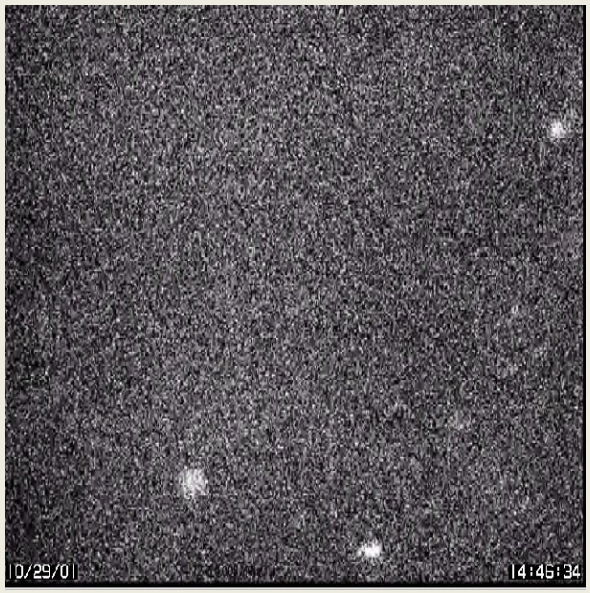
$$\frac{[\eta]}{\eta} \approx S(\beta_{\parallel}-\beta_{\perp})$$

$$(\alpha_1-\alpha_2) = S(\beta_{II}-\beta_{\perp})$$

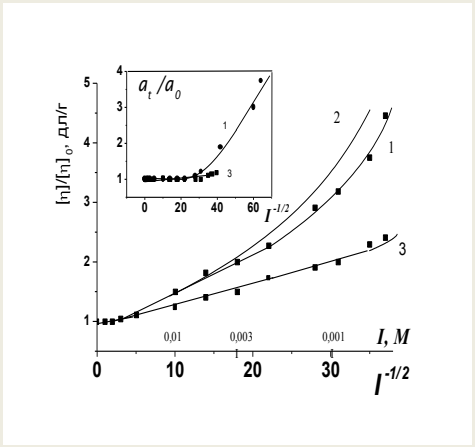
$$[\eta] = \Phi\left(\frac{\langle A^2\rangle}{M}\right)^{3/2}\cdot\chi^3$$

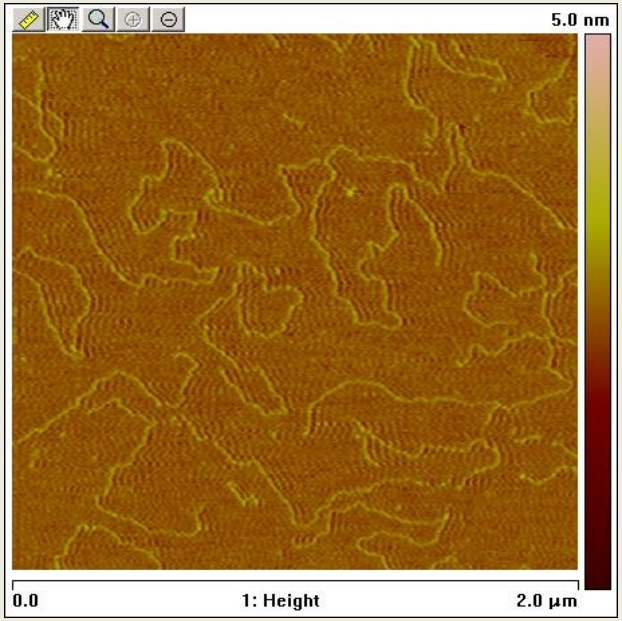


1M NaCl

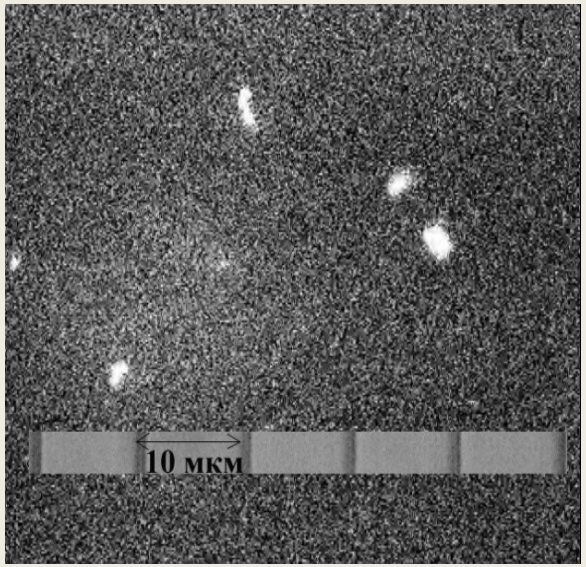


0,1 M NaCl





Calf thymus DNA 0,005 M NaCl

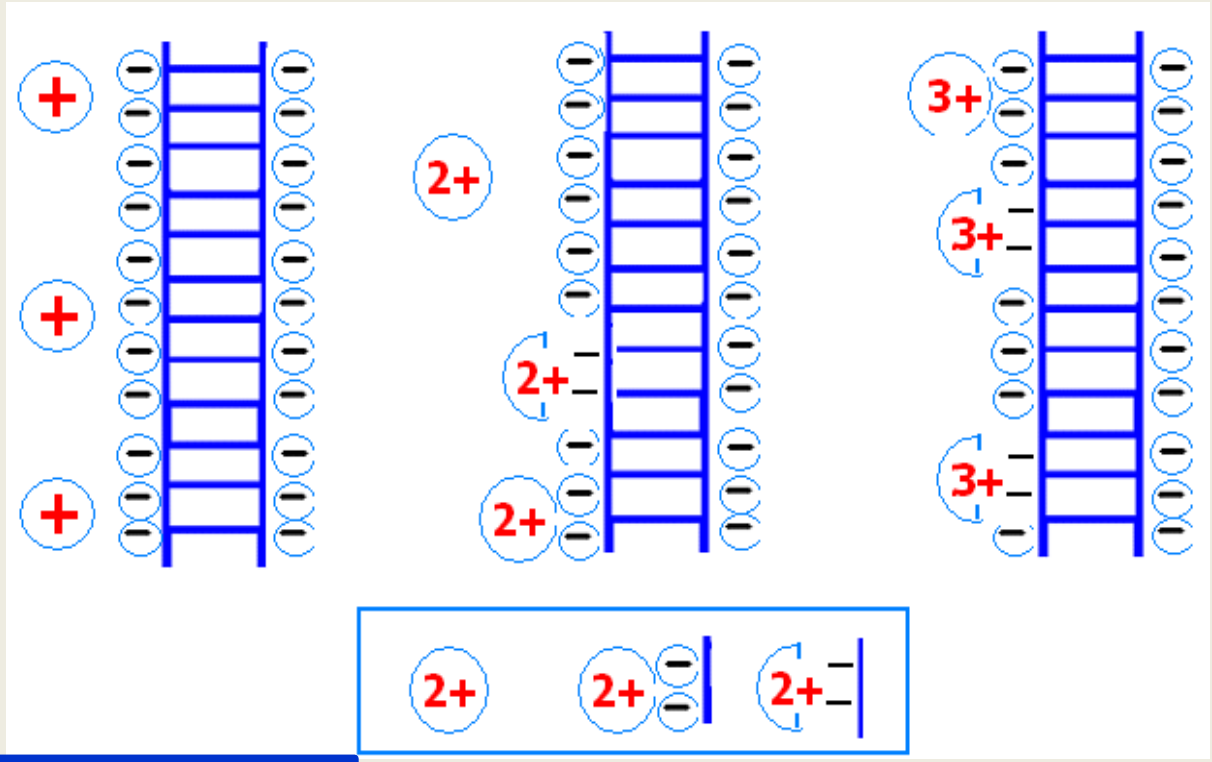


0,005 M NaCl (α ≈ 1.6)

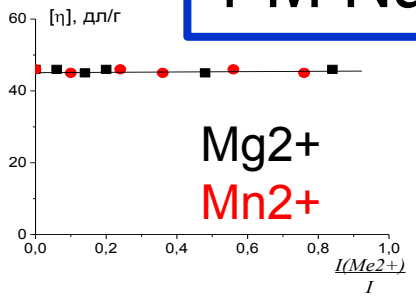
T2 DNA in NaCl solution
with Yo-Yo dye
Axiolab (Carl Zeis)

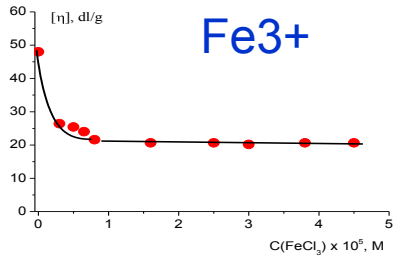
Abramchuk S.S. MSU

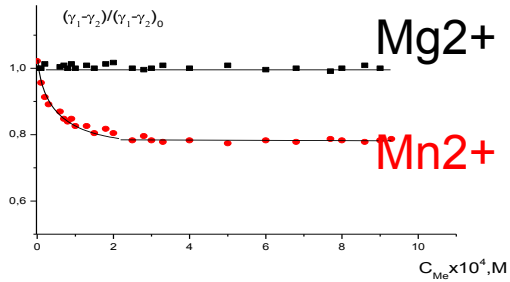
DNA Complexes with Metal Ions



1 M NaCl







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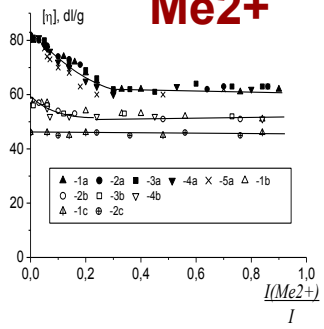
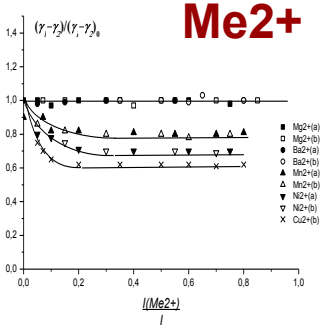
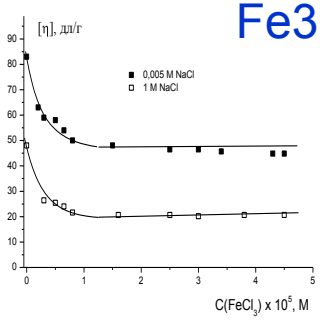
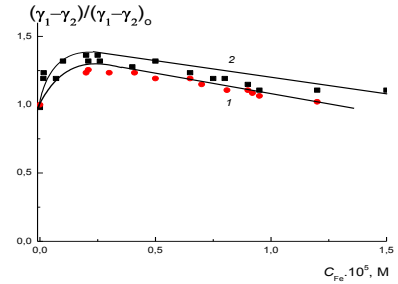
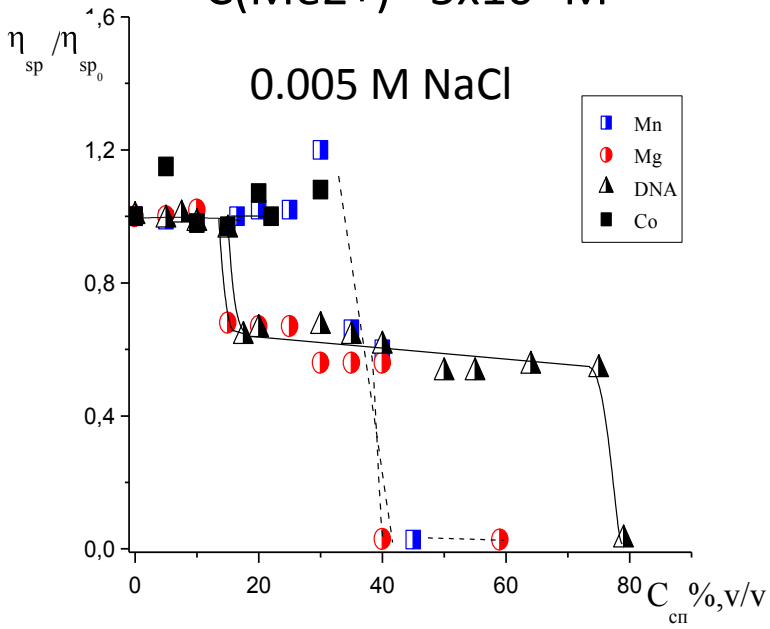
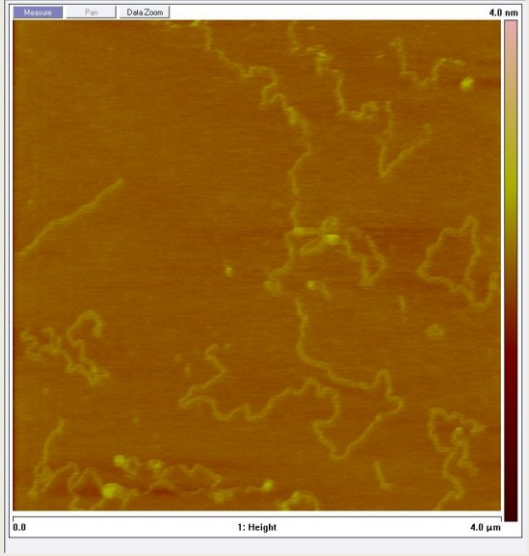


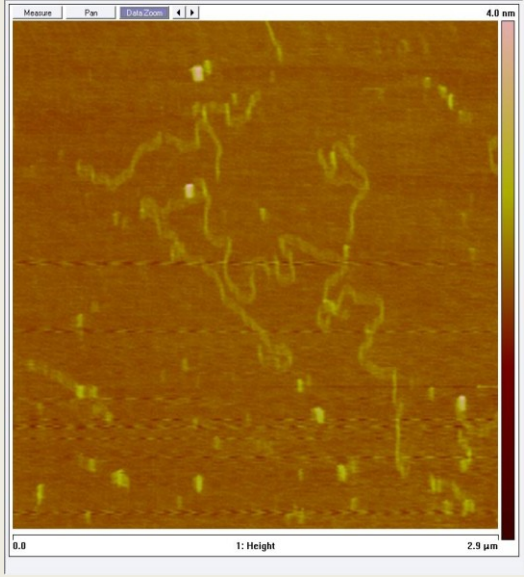




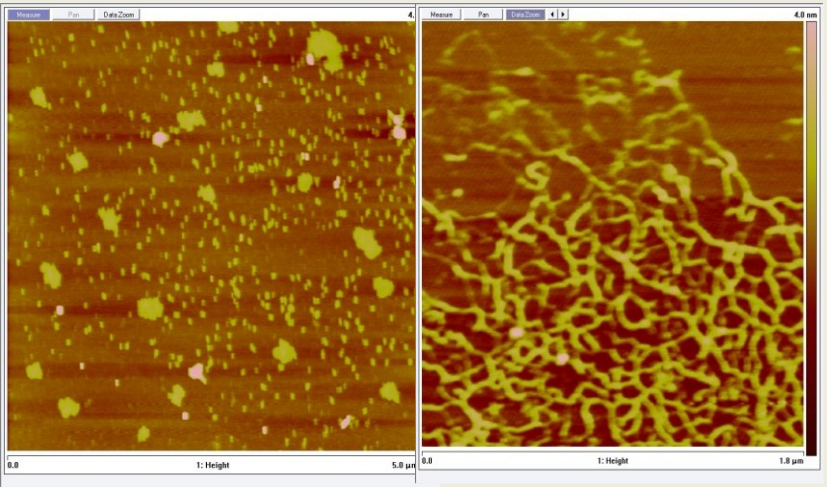




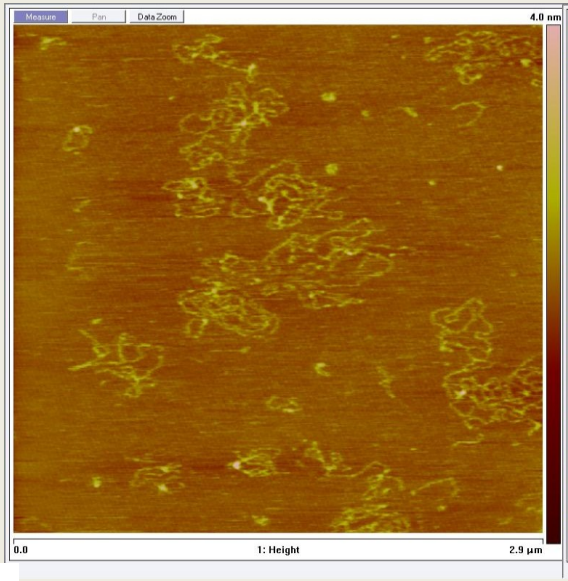


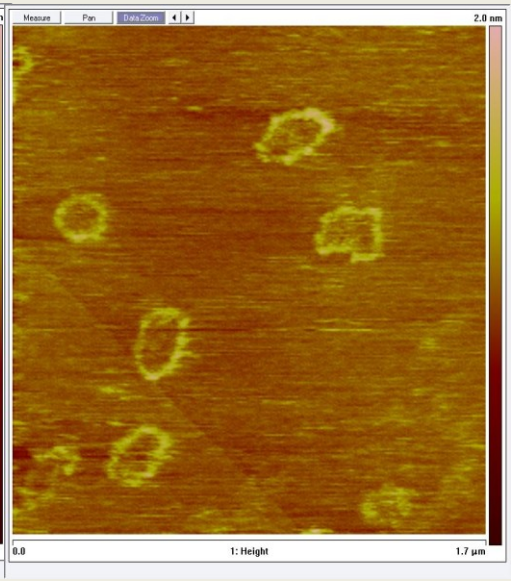


Mn2+ C(alc)=0 % **Mn2+** C(alc)=20 %

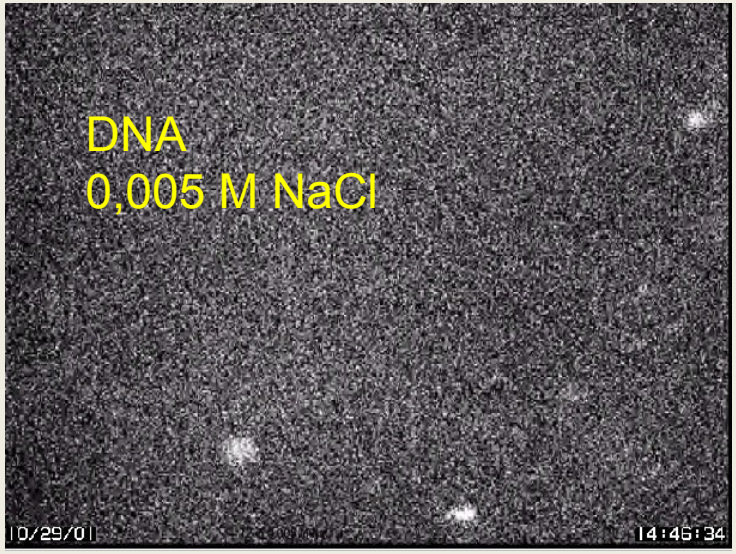


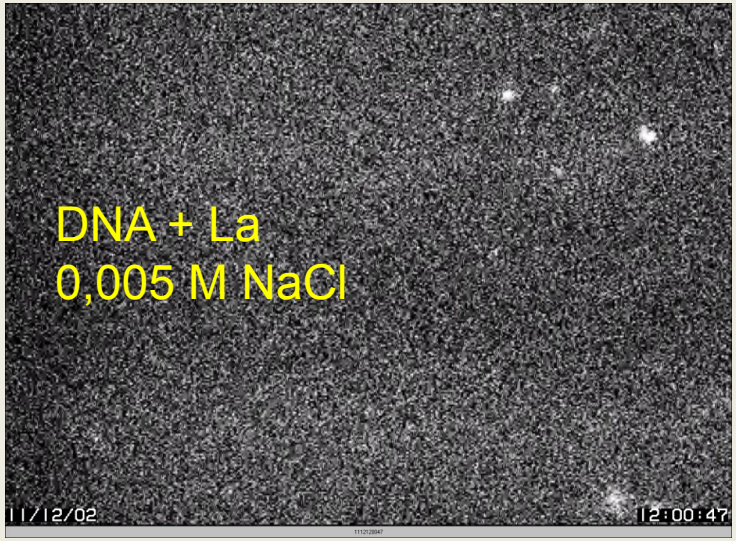
Mn2+ C(alc)=35 % **Mg2+** C(alc)=55 %





Mg2+ C(al)=20 % **Mg2+** C(al)=75 %





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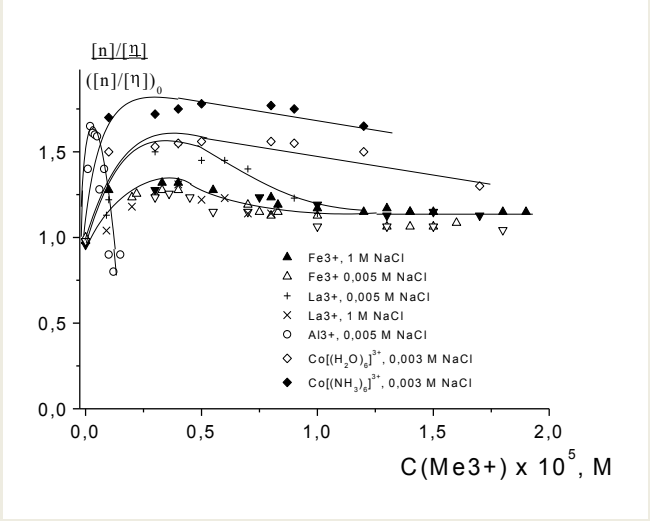
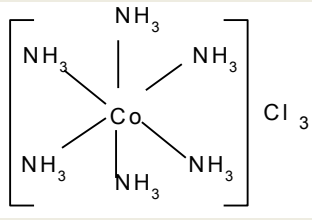
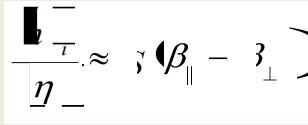
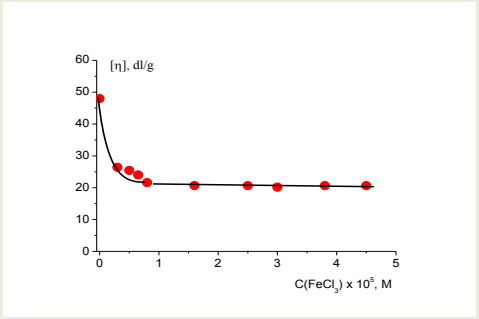
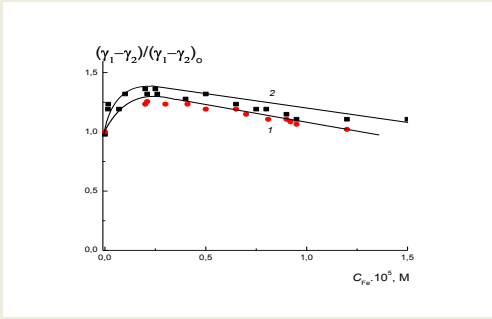
Abramchuk S.S. MSU

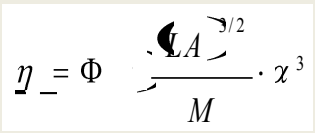


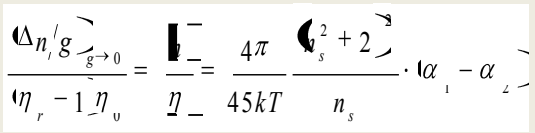


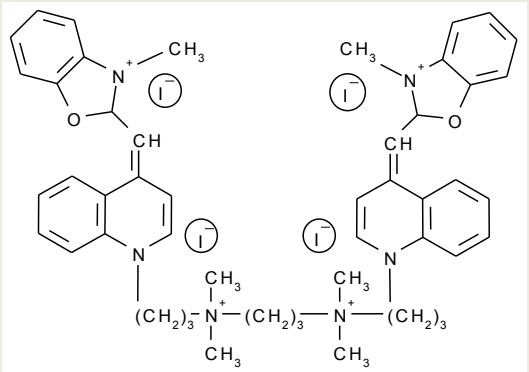


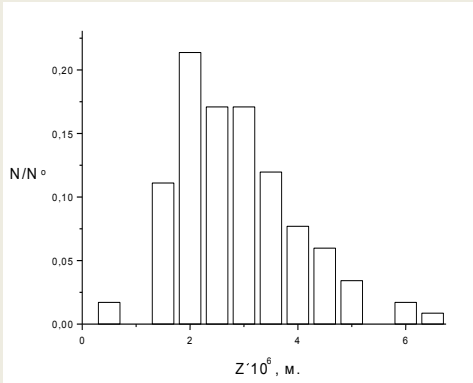


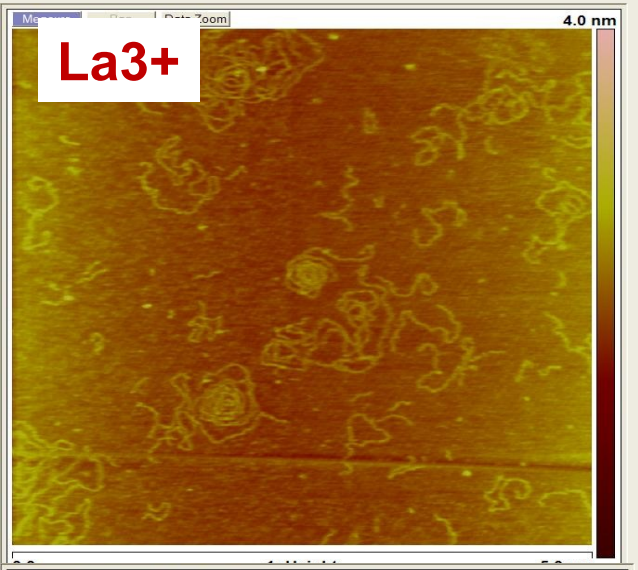


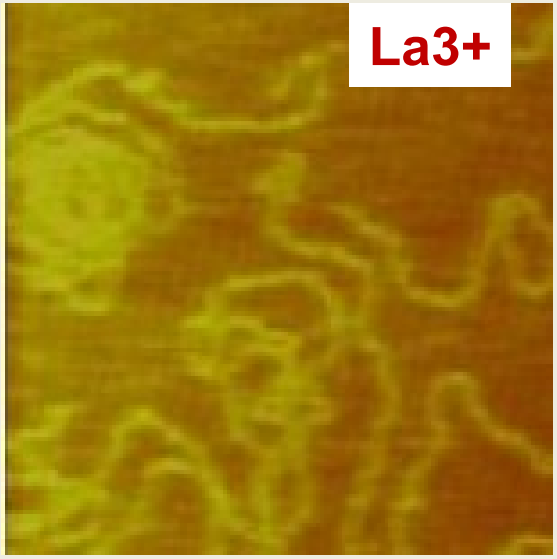


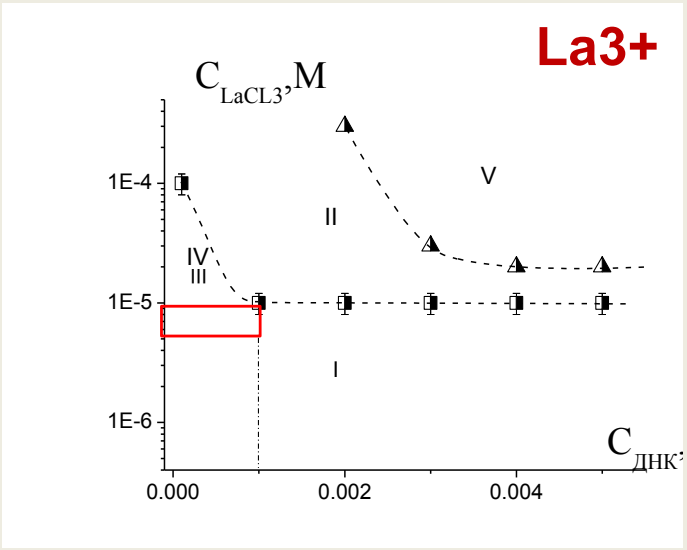


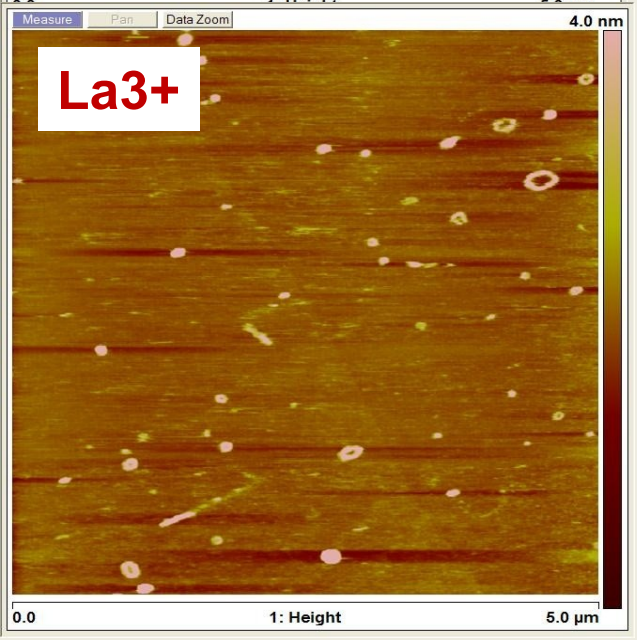


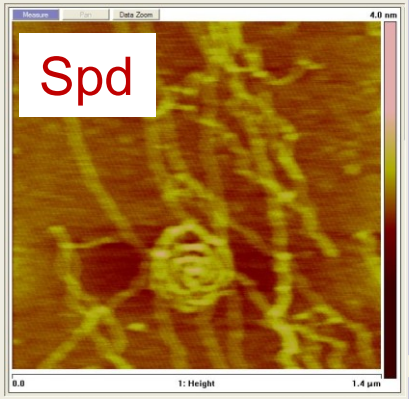


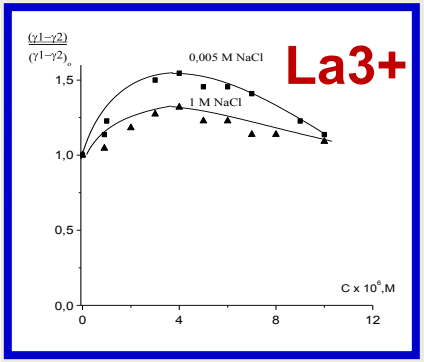


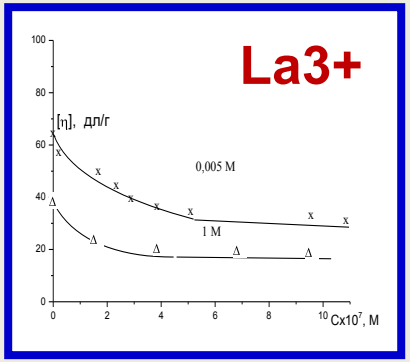


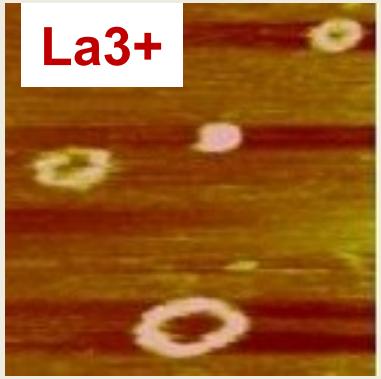


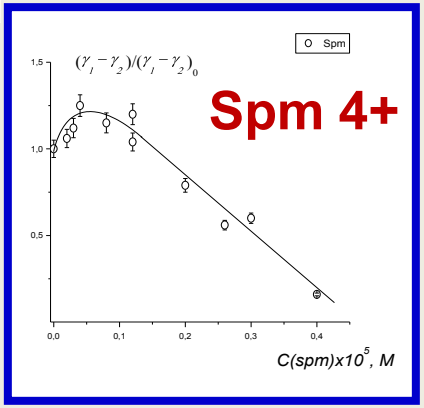


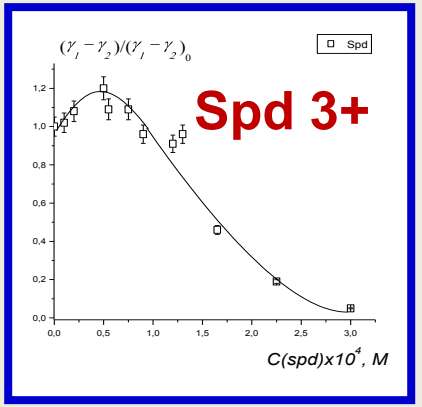






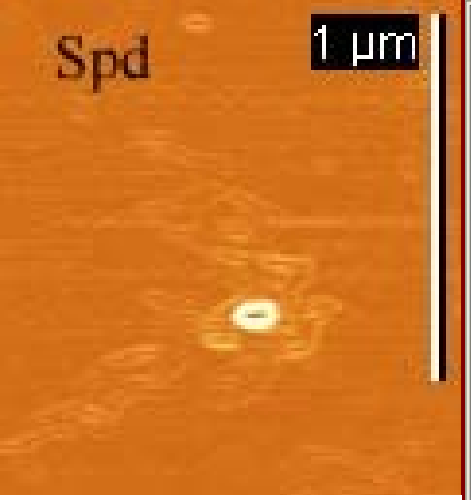




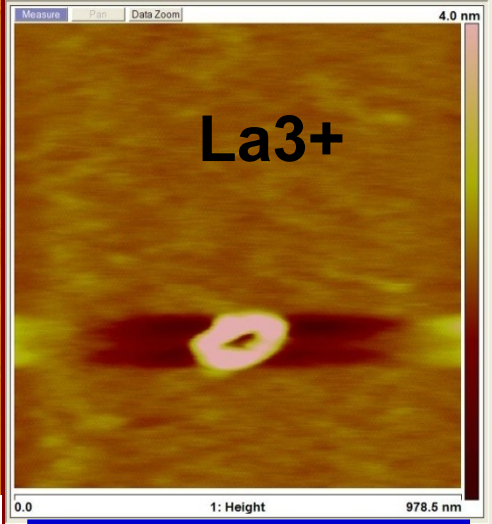


D= (110) nm

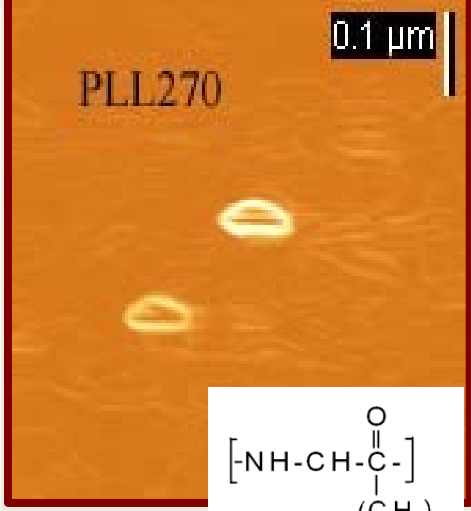
ДНК/$LaCl_3$ в 0.005 M NaCl
$C_{ДНК}$=0.0001%, C_{LaCl3} = 4*10^{-6} M.

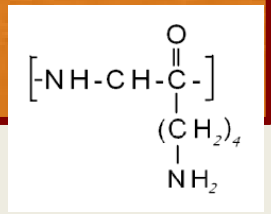


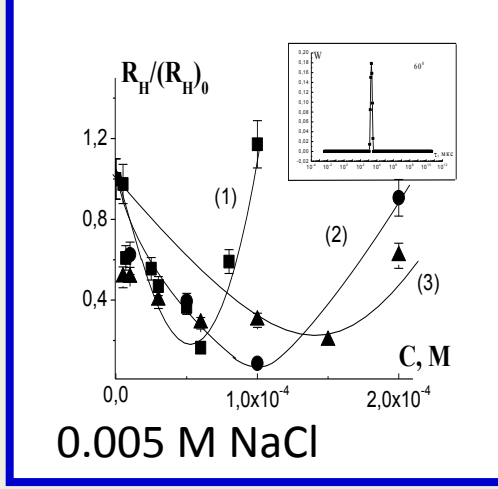
NH2 – (CH2)3 – NH –(CH2)4 –NH2



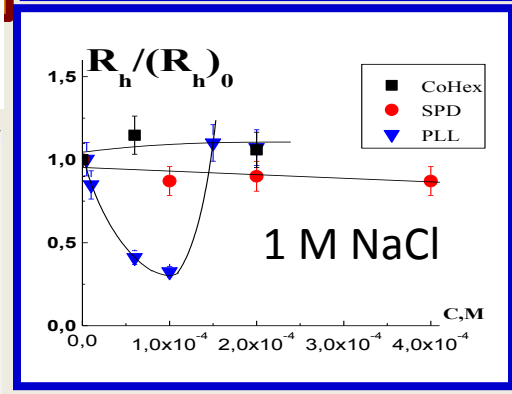
C(La)=4*10^{-6}M



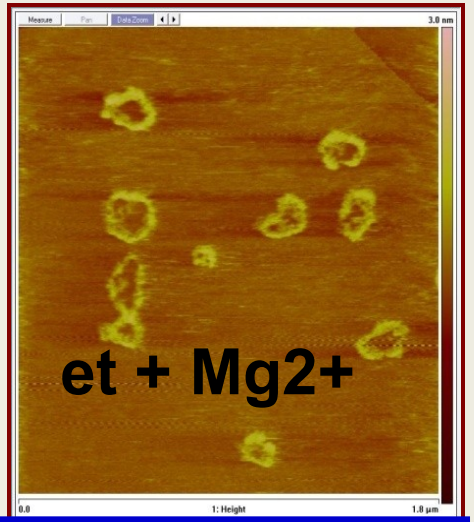




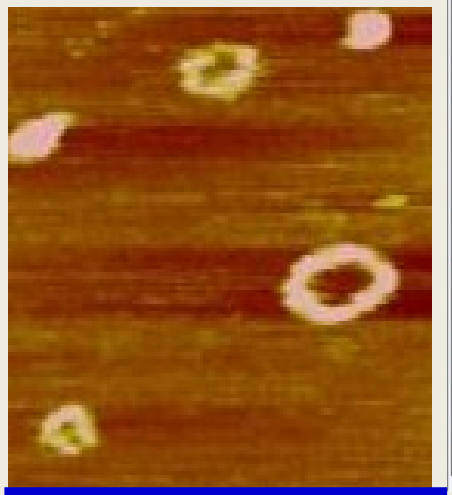
0.005 M NaCl



1 M NaCl

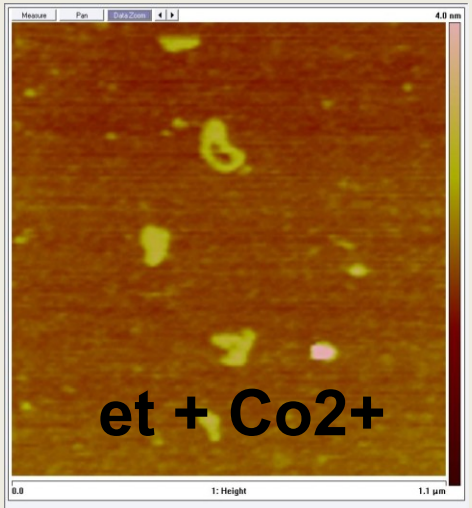


C(et)=75%
C(MgCl2)=5 10^{-4}M

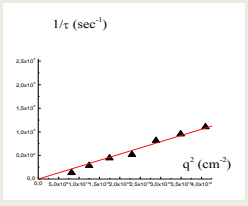


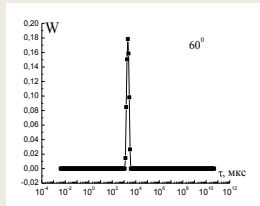
C(La)=4*10^{-6}M

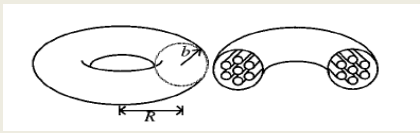
R=(65±10) nm



C(et)=65%
C(CoCl2)=5 10-4M







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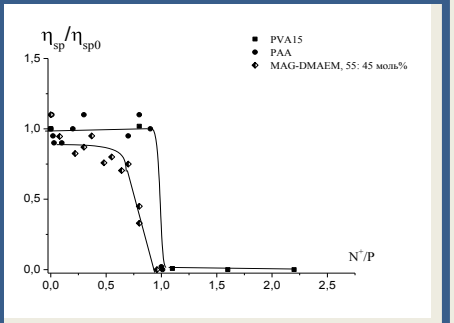
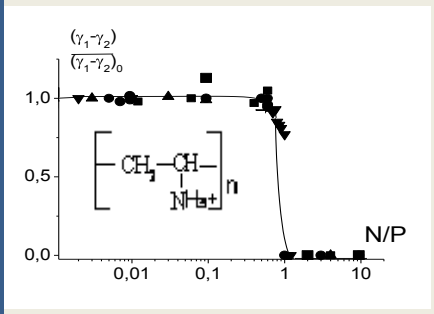
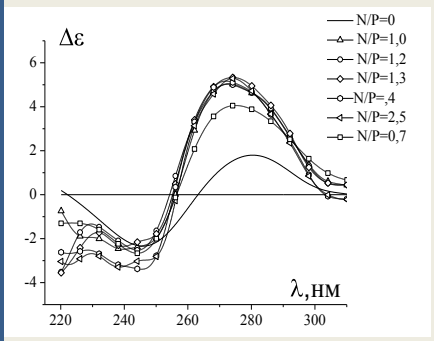
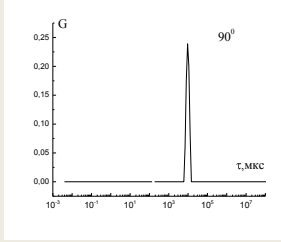
$$A_0 = \frac{D\eta_0([\eta]M)^{1/3}}{T} = 3{,}4\times 0^{-7}\ Дж\cdot град^{-}\ моль^{-/3}$$

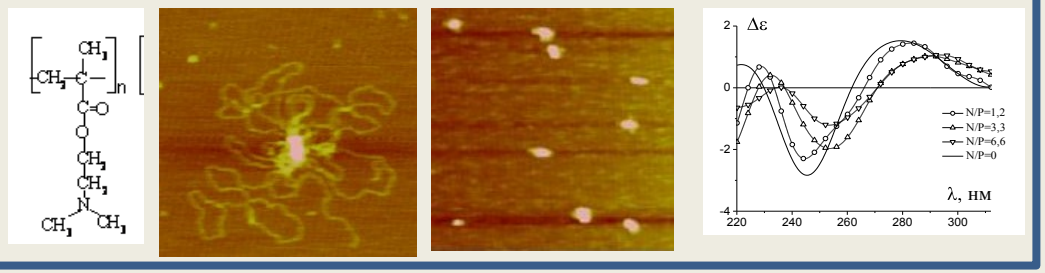
DNA complexes with polycations

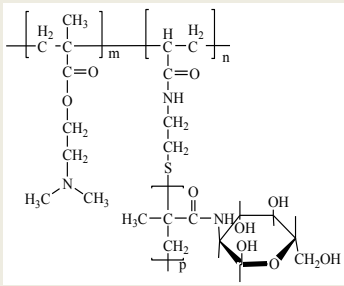


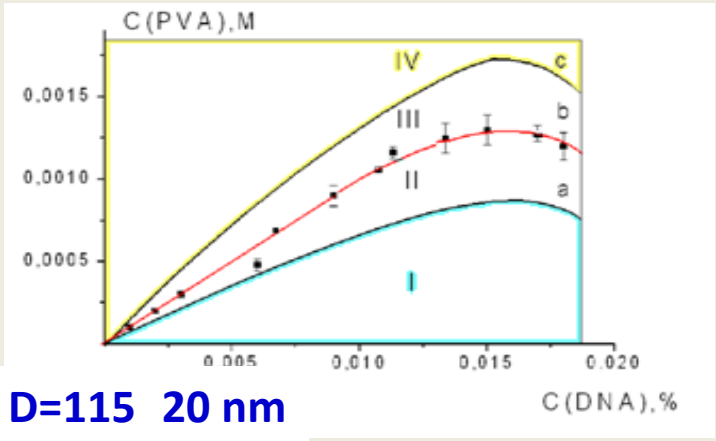




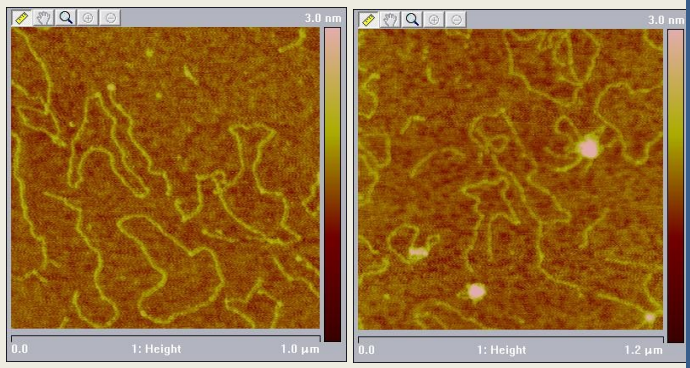




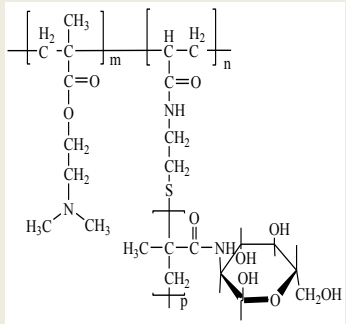


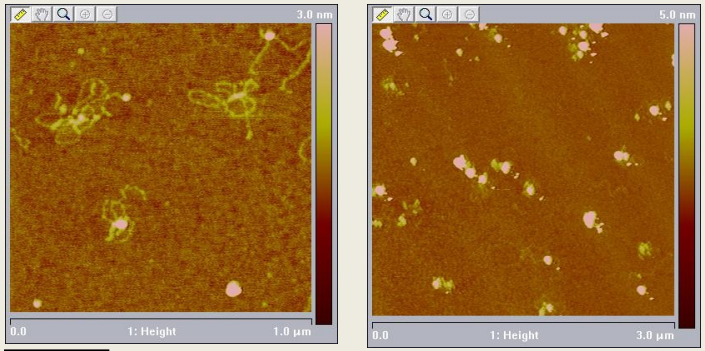


D=115 20 nm

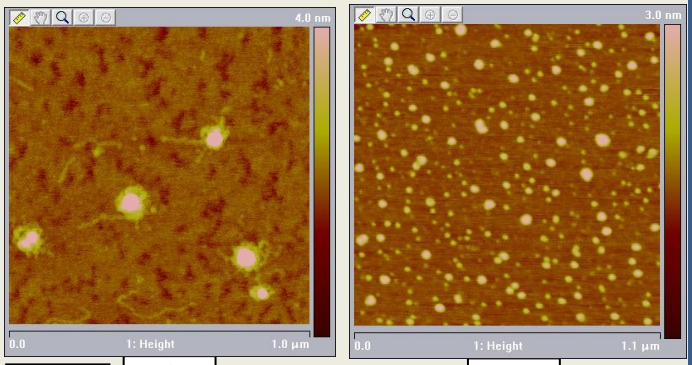


N^+/P 0 DNA pFL 44 0,7





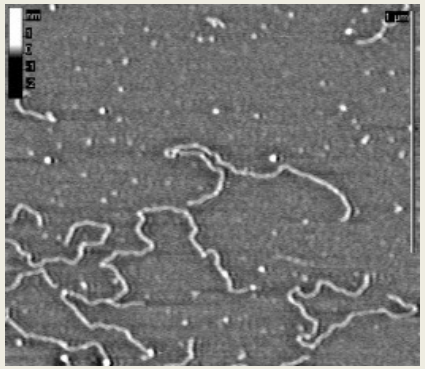
N^+/P 1 2



N^+/P 1 1,4

DNA

1)



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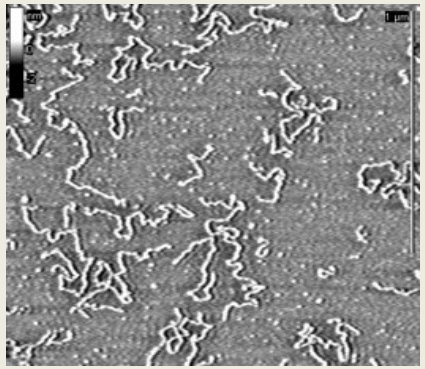
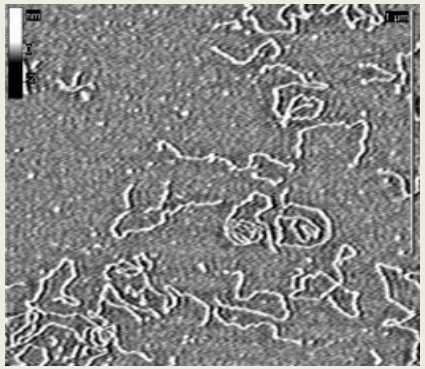


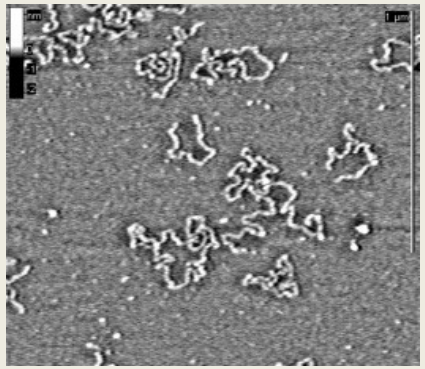
DNA+cis-DDP

DNA+trans-DDP

3)



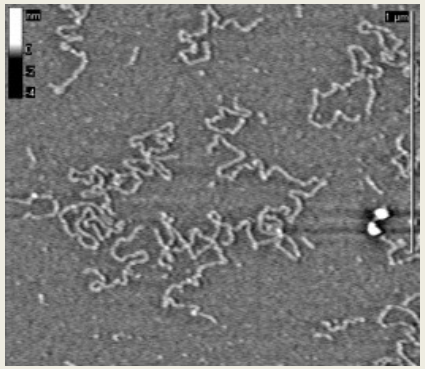
4)



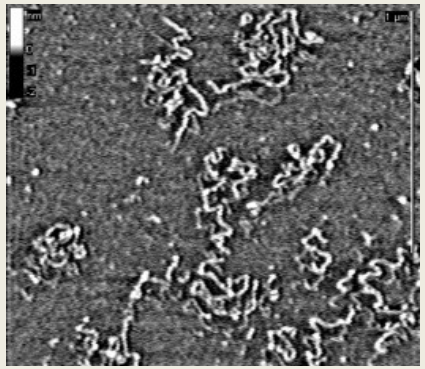
(DNA+cis)+trans

(DNA+trans)+cis

5)



6)

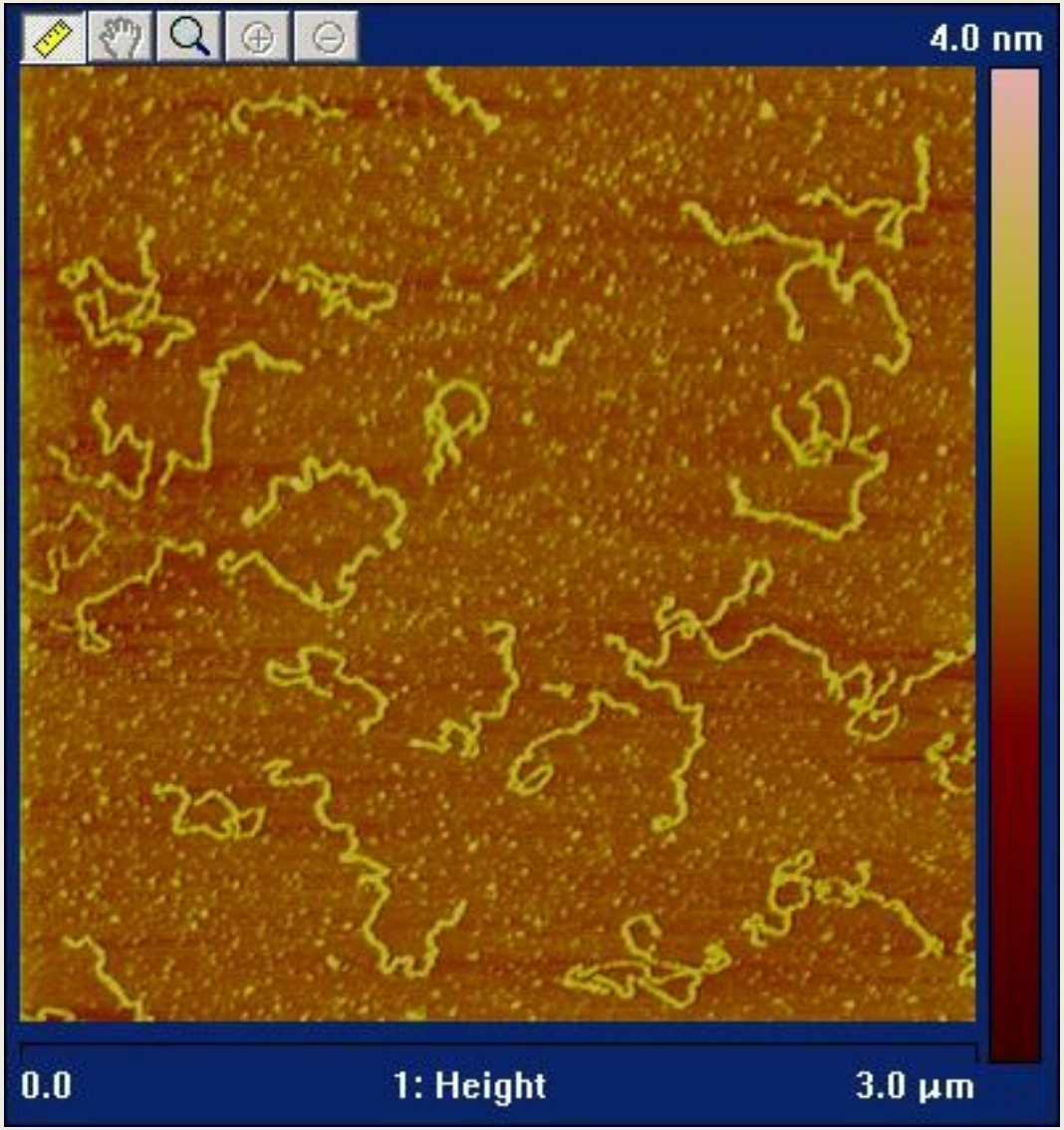


DNA+cis+trans

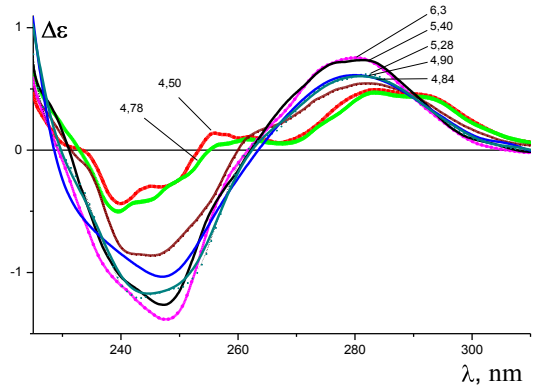
Рисунок 1. АСМ-изображение ДНК pFL44s/EcoRI и ее комплексов с цисДДП и трансДДП. 1- ДНК; 2- ДНК+цисДДП; 3- ДНК+транс-ДДП; 4-(ДНК+цис-ДДП)+транс-ДДП; 5- (ДНК+транс-ДДП)+цис-ДДП; 6- ДНК+цис-ДДП+транс-ДДП.

Alexei Bogdanov (poster)

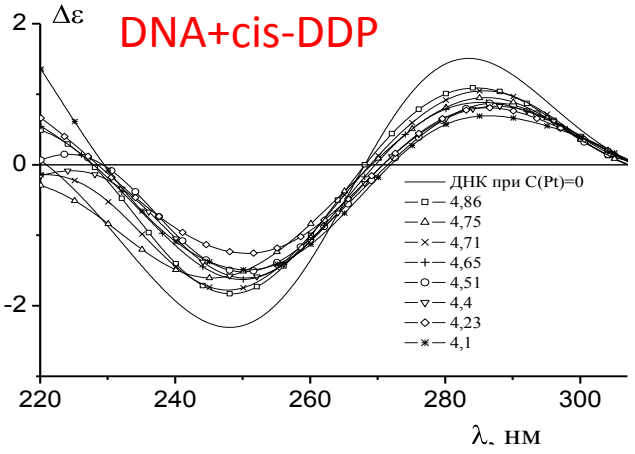
DNA with cis-DDP



DNA



DNA+cis-DDP



DNA + La

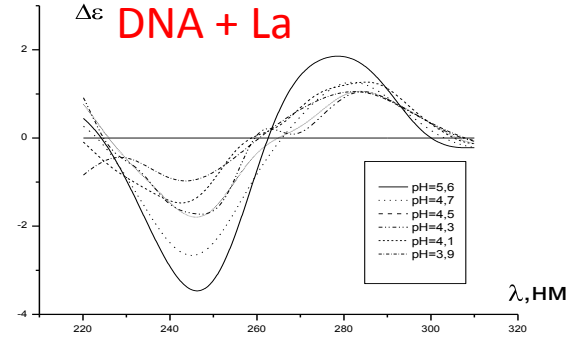
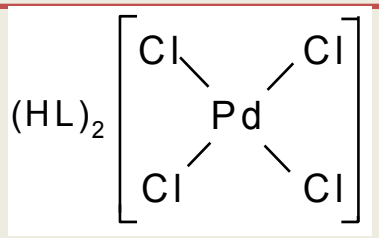
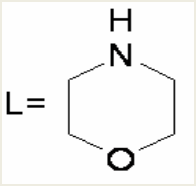
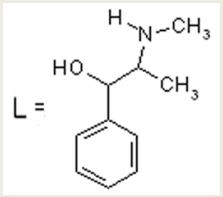


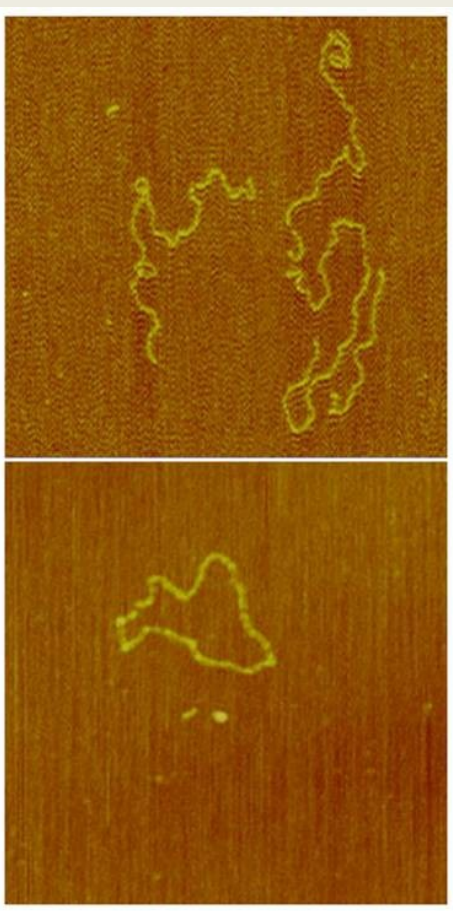
Рис. 4.44. Спектры КД ДНК в комплексе с La^{3+}($C=10^{-3}$M) в 0,005M NaCl при разных pH

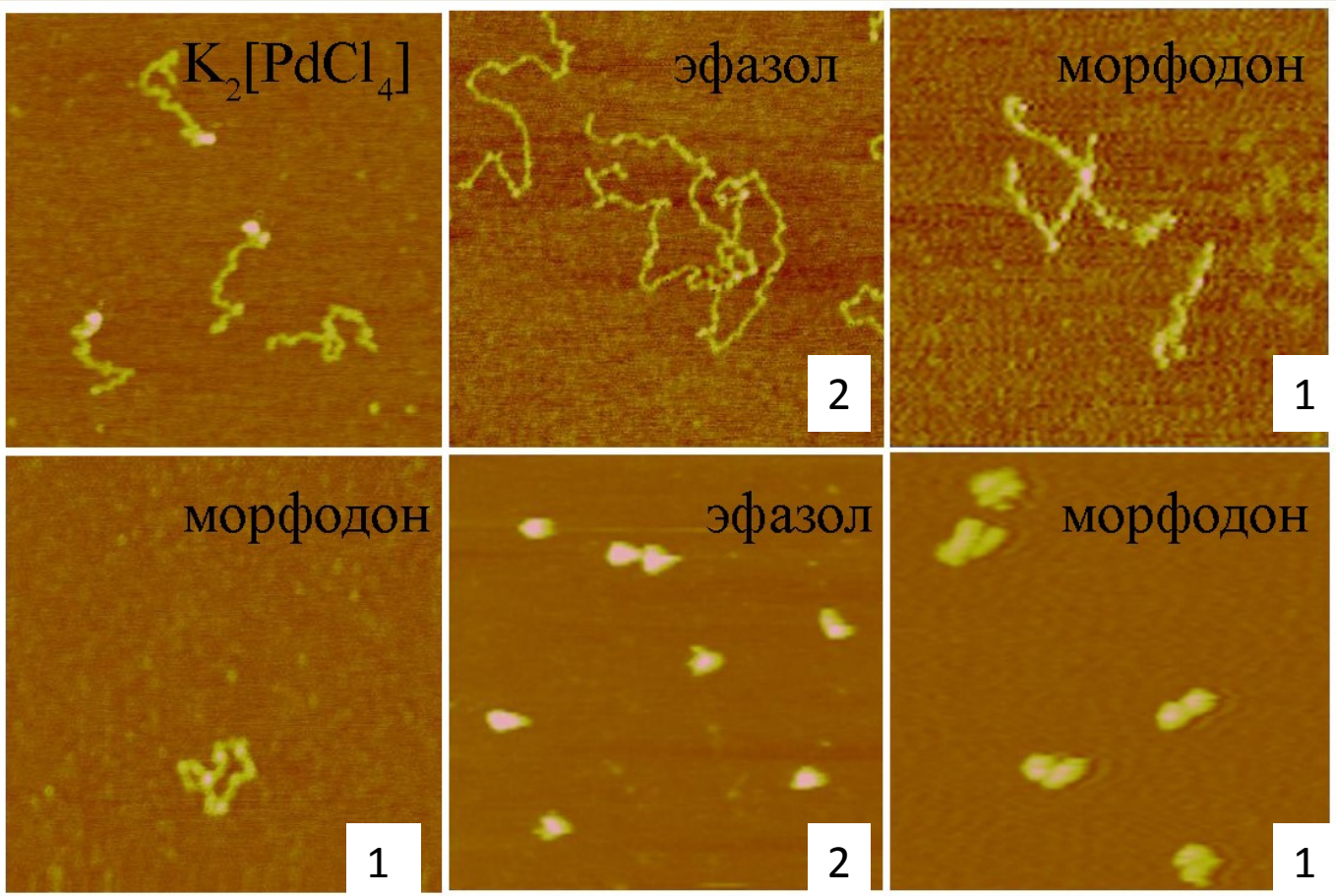
DNA Complexes with Palladium Compounds



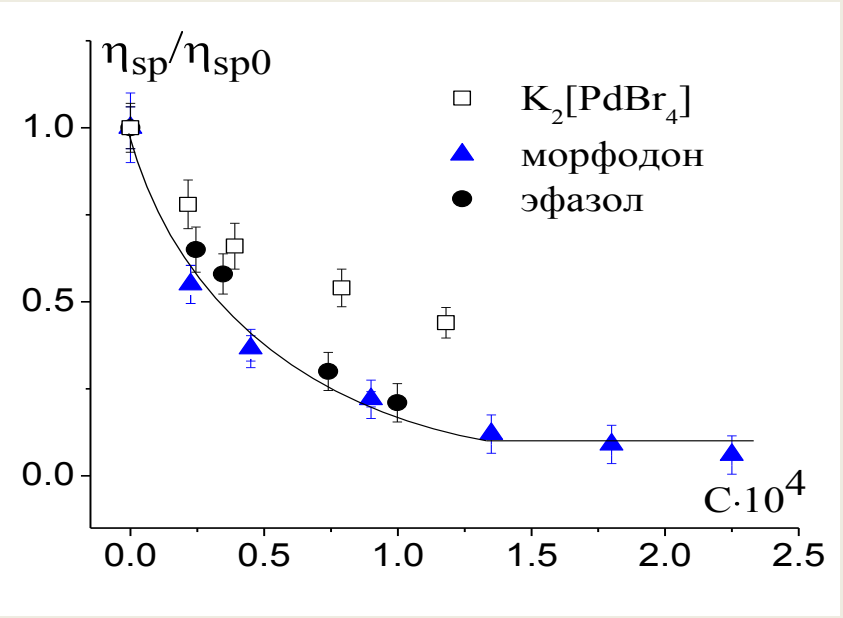
1 - L= 

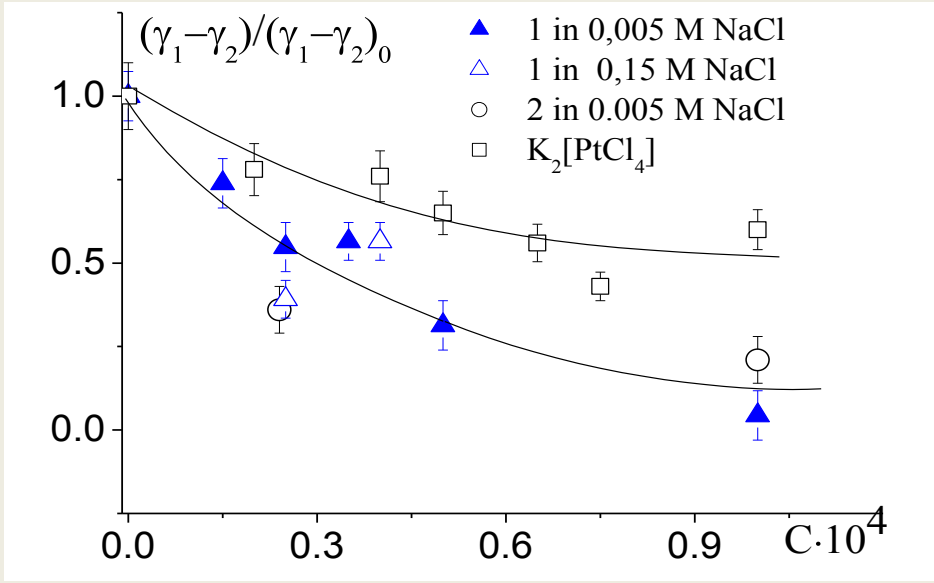
2 - L = 

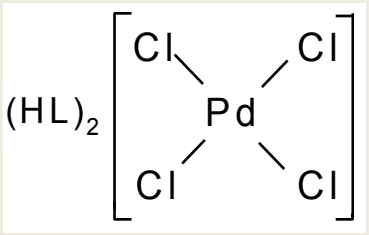


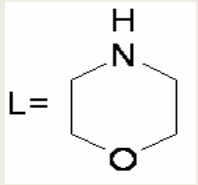


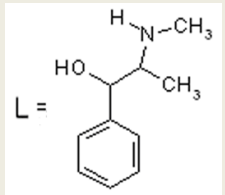
DNA complexes with Palladium compounds

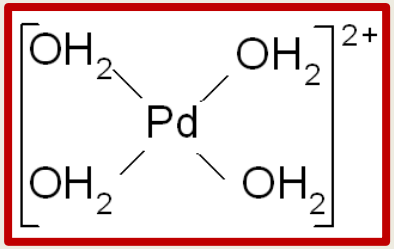


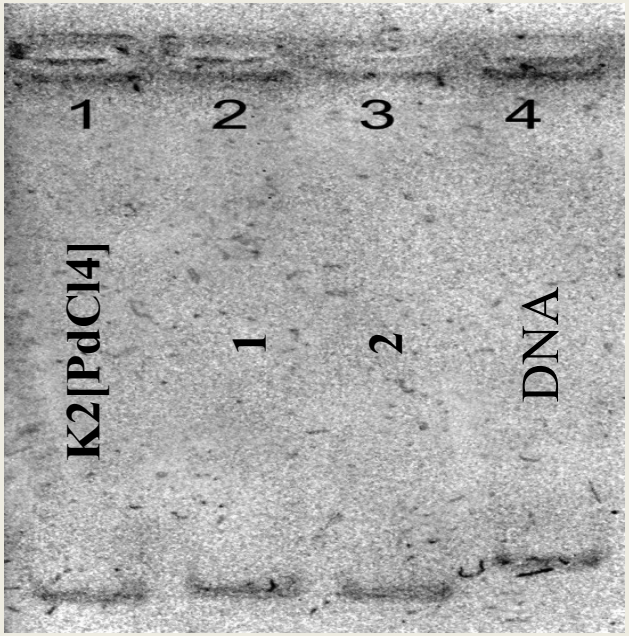


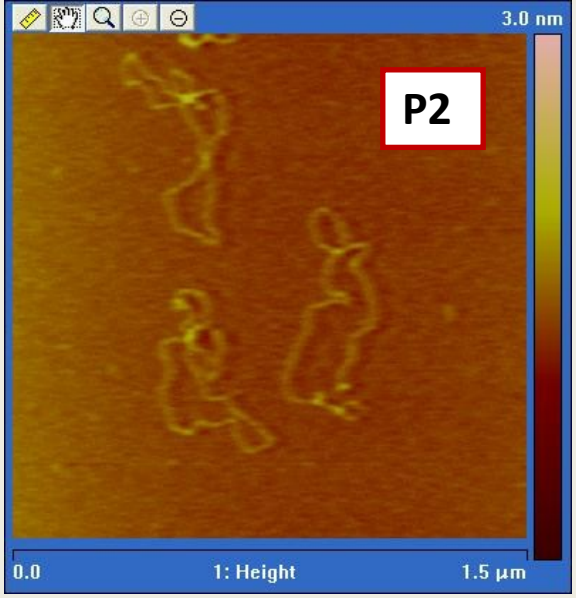


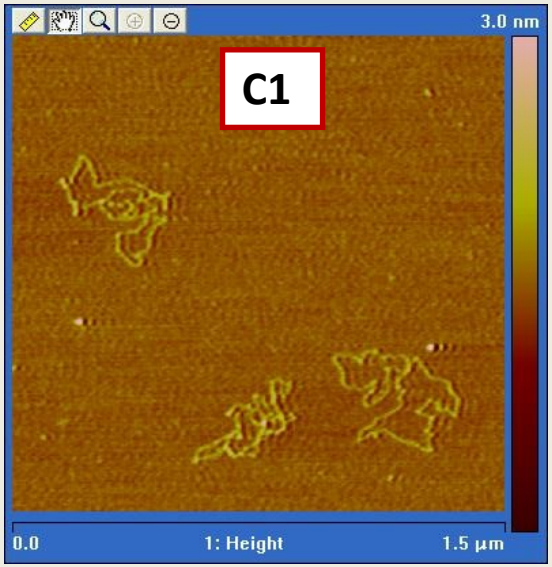
1 - 

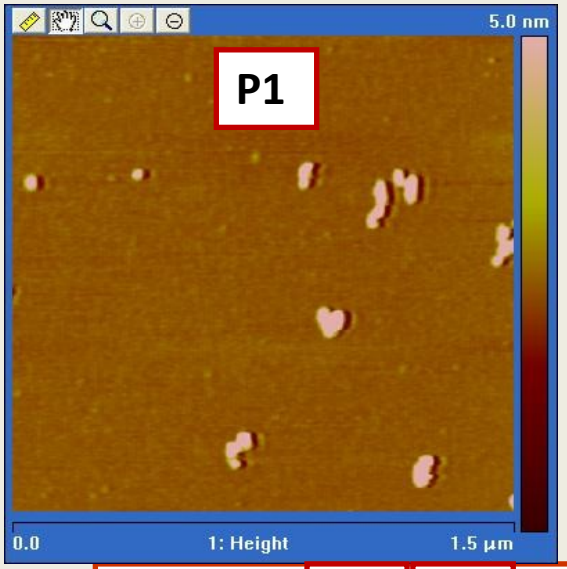
2 - 

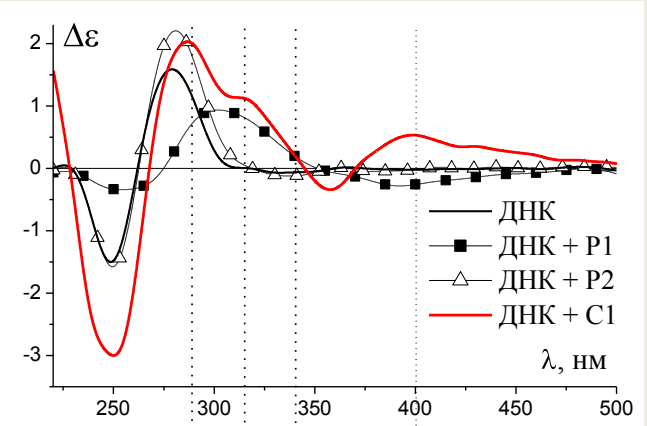


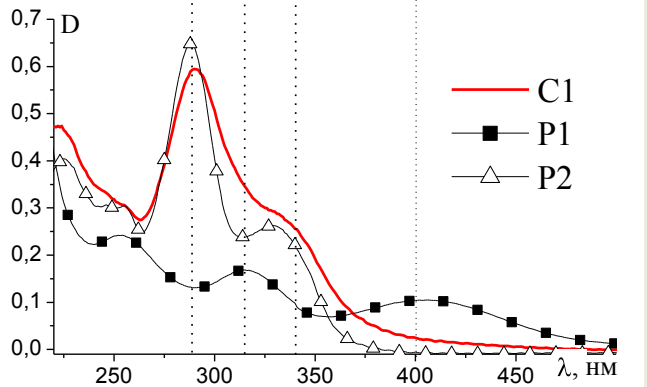


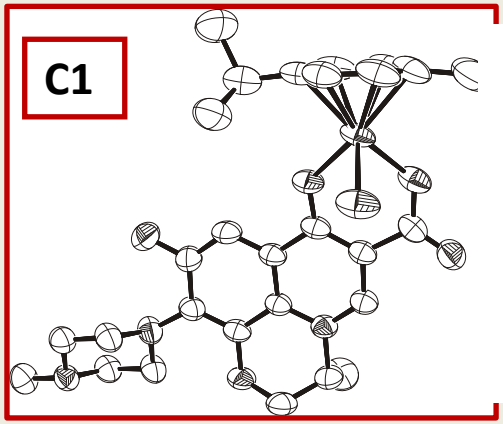

P2
3.0 nm
0.0
1: Height
1.5 µm

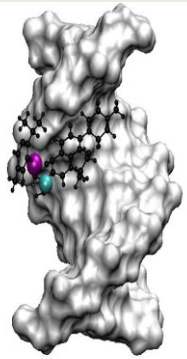

C1
3.0 nm
0.0
1: Height
1.5 µm

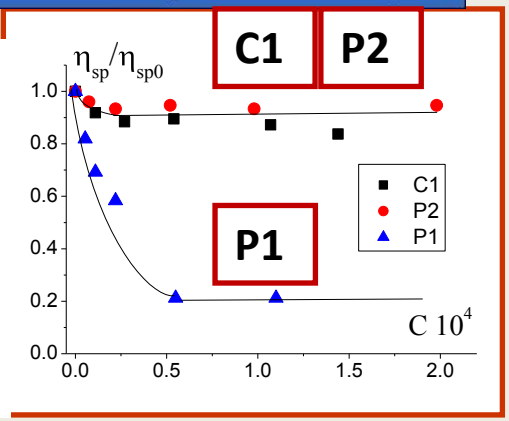

P1
5.0 nm
0.0
1: Height
1.5 µm

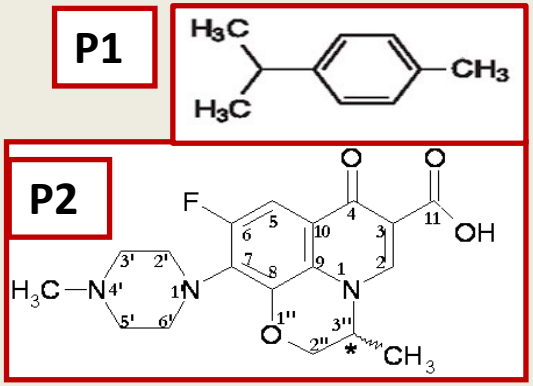

Δε
ДНК
ДНК + P1
ДНК + P2
ДНК + C1
λ, нм

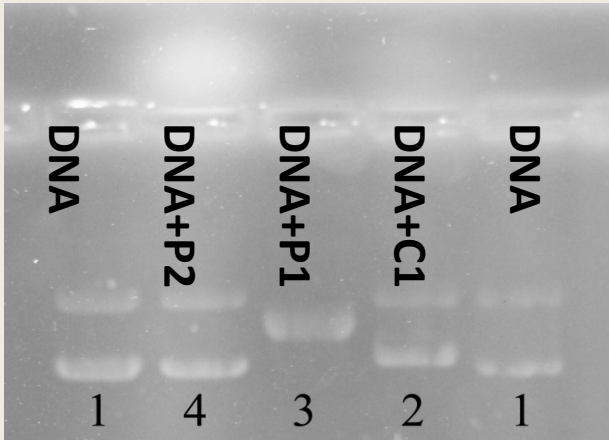

D
C1
P1
P2
λ, нм


C1

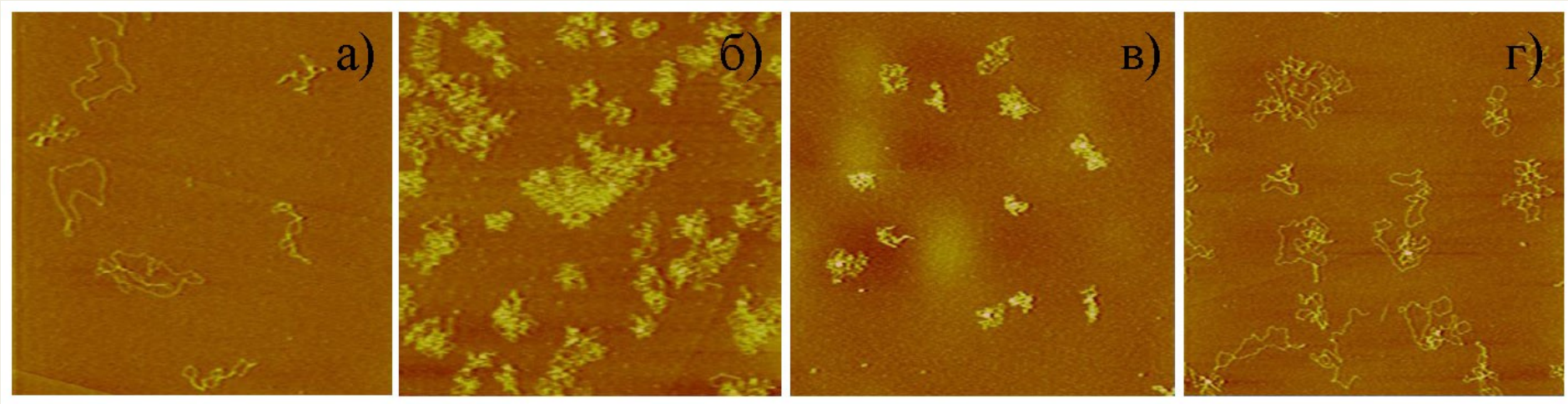



C1
P2
P1
ηsp/ηsp0
C 10^4
C1
P2
P1


P1
P2

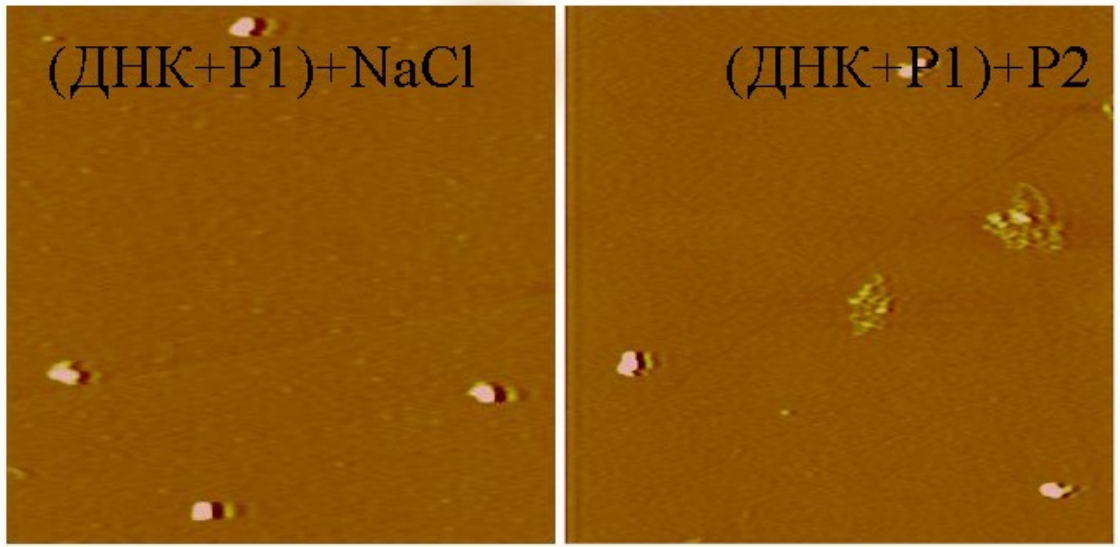

DNA
DNA+P2
DNA+P1
DNA+C1
DNA
1
4
3
2
1

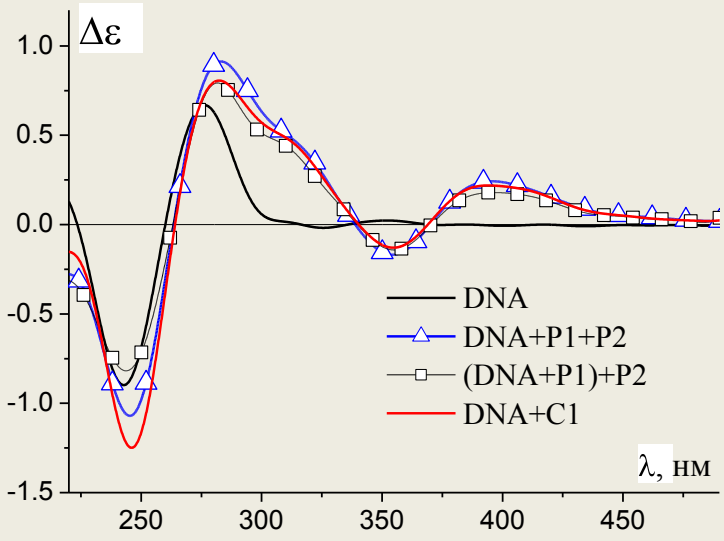
DNA-C1 complex can dissociate after dilution:



DNA DNA+C1 (1) DNA+C1 (2) DNA+C1 from(2)

DNA-C1 complex can be formed by the addition of P1 and P2 (one by one) into DNA solution





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Thank you for the attention