Electric conductivity of deoxyribonucleic acid water solutions

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Electric Conductivity of Deoxyribonucleic Acid Water Solutions

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List of Abbreviations

Chemical substances:

DNA ..... Deoxyribonucleic Acid

NaCl ..... Sodium chloride

$MgCl_2$ ..... Magnesium chloride

RNA ..... Ribonucleic Acid

Physical quantities

A ..... Cross section area

D ..... Diffusion coefficient

F ..... Faraday's constant

I ..... Electrical current

L ..... Length

N ..... Degree of polymerisation

R ..... Resistance

Q ..... Charge

V ..... Voltage

c ..... Molar concentration

e ..... Unit charge

l ..... Polymer length

f ..... Fraction of free counterions

k_b ..... Boltzmann constant

r ..... Distance

z ..... Valency

Λ ..... Molar conductivity

γ ..... Mass concentration

ε ..... Electrical permittivity

μ ..... Mobility

ρ ..... Resistivity

σ ..... Conductivity

φ ..... Electric potential

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Abstract

Deoxyribonucleic acids (DNA) are one of most important naturally occurring macromolecules. Today we know that this molecule is present in every living being carrying its genetic code that defines its species, gender, physical attributes and more. Modern research has come as far as using artificially constructed DNA even in nanotechnology making understanding of its properties evermore important. From its structure one can observe DNA as a polymeric structure and thus attributing it with general properties from this group. In the past models that simulate polymer behaviour were described one of which is Manning Oosawa condensation model, widely accepted for polyelectrolytes dissolved in water. From this model equations for solution conductivity can be derived. This dissertation compares this calculations with experimental measurements.

Key words: DNA, conductivity, DNA solutions, Manning Oosawa model

Sažetak

Deoksiribonukleinska kiselina (DNA) je jedna od najvažnijih makromolekula u prirodi. Prema današnjim saznanjima DNA molekula je prisutna u stanicama svakog živog bića, a nosi genetski kod čime definira vrstu, spol, fizička obilježja i mnoge druge karakteristike. Najnovija istraživanja pokazala su već da je moguća *proizvodnja* DNA, kakva se se koristi u nanotehnologiji. Promatrajući strukturu DNA kao polimernu strukturu mogu joj se dodjeliti opće karakteristike navedene grupe. Povijesno su bili opisani modeli koji simuliraju ponašanje polimera među kojima se nalazi i Manning – Oosawa kondenzacijski model, koji opisuje vodene otopine polielektrolita. Iz tog modela proizlaze jednadžbe koje koreliraju koncentracju istovrsnih otopina s njihovom vodljivošću. Ovaj diplomski rad uspoređuje te izračune sa eksperimentalnim mjerenjima.

Ključne riječi: DNA, vodljivost, DNA otopine, Manning Oosawa model

Theoretical Introduction

Polymers

The word polymer comes from Greek words “polu” meaning many and “meros” meaning parts and refers to molecules consisting of many elementary units called monomers. These units are structural repeating units of polymer that are connected through covalent bonds to each other.[1] Reaction through which polymers are created is called polymerisation. This is the process where building blocks (monomers) from which this macromolecule is created are interconnected into final structure. Physical as well as chemical characteristics of certain polymer depends not only on the monomer used but also on the number unit repeats. This number is called degree of polymerisation.

Structure of polymers

Polymeric microstructure is dependant intrinsic as well as extrinsic factors. When considering internal factors structure building units (monomers) arrangement is a major factor. Since polymeric structure cannot be changed without breaking covalent bonds, once monomers are polymerised it is impossible to change polymeric characteristics without chemical reaction. Additionally, because monomers can interconnect in different way there are several isomeric structure can be produced in the polymerisation process. Depending on the position of the radical sequence isomerism can be expected (head to head or tail to tail), if more than one double covalent bond (C=C) in monomer is present structural isomerism (tran- and cis- forms) is also a possibility. Finally, there are also cases of stereoisomerism depending on the angulation of the radical group.

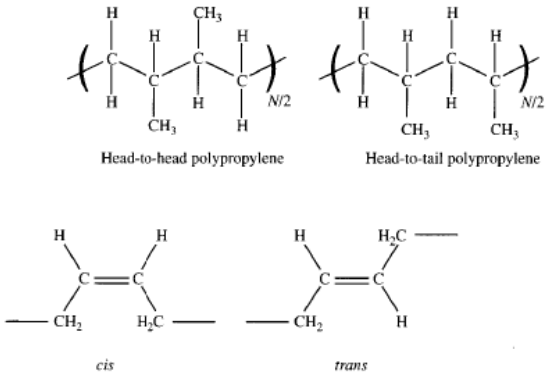

Figure 1: Schematic display of sequence isomerism (top structures) and structural isomerism (bottom structures)[1]

Besides microstructural differences that influence polymer properties its macrostructural shapes can also differ additionally defining its characteristics. These variations can be as simple as differences in number of repeats and as complex as formation of diverse architectures. Best

example of length influencing physical characteristics of polymer can be provided by observing melting temperatures of alkane hydrocarbons (methane repeats) that increase with the number of C atoms in the chain. On the other hand changes in architecture also provide completely different molecules as one can simply see by observing their structure shown in Figure 2.

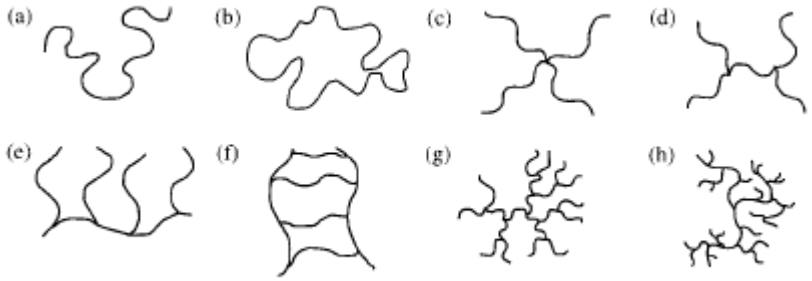

Figure 2: E*xamples of polymeric architecture (a) linear, (b) ring, (c) star, (d) H, (e) comb, (f) ladder, (g) dendrimer, (h) randomly branched* [1]

There is another important distinction in structure of polymers depending on number and arrangement of different monomeric subunits. If one type of monomer is present then this macromolecules are called homopolymers however if more than one type of monomer is present then this macromolecules are heteropolymers. (Figure 3)

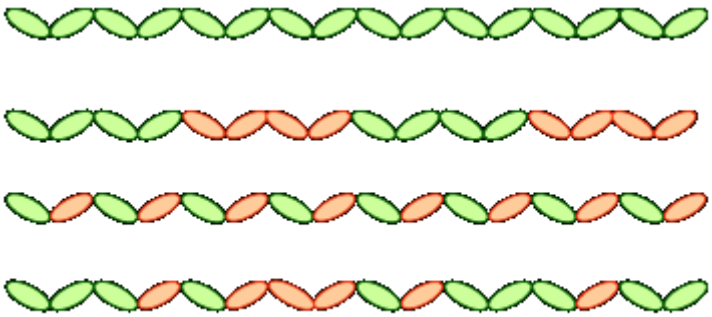

Figure 3: Comparison of homopolymer structure (top chain) to block heteropolymer (second chain from the top) alternating heteropolymer (third chain from the top) and random heteropolymer (bottom chain)[2]

The properties of heteropolymers depend both on composition (fraction of each type of monomer present) and on the sequence in which these different monomers are combined.[1] Number of different type of monomers present in macromolecule. If two types are present polymer is called copolymer and if three types are present than its called terpolymer. Also according to arrangement

polymers can be alternating, random, bloc or graft. Many biopolymers are heteropolymers, such as proteins, nucleic acids, sugars, etc.

Polymer conformations

As already mentioned above monomeric units play a crucial role in defining chemical and physical properties of polymers. Once these units polymerise macromolecules form conformation based on interactions between monomers. In order to understand complex real chains knowing intrinsic properties universal to polymers is essential and these are best explained by hypothesising an ideal chain. In this case at certain temperature it is considered that repulsive and attractive forces between monomers nullify each other. Thus the multitude of conformations an ideal polymer depends on flexibility of the between monomers and on stiffness of the macromolecule as a whole.

To better understand flexibility of the chain consider a polyethylene molecule where under ideal conditions bond lengths are considered to be constant and minor fluctuations do not affect changes in conformation. Left only with rotational component of neighbouring monomers. The angle between neighbouring bonds is 68° in tetrahedral configuration such as polyethylene. When spatial orientation of three neighbouring bonds is observed there are several rotational configurations that can take place. State, where two non-neighbouring bonds are on the opposite sides of rotational axis that is represented by the bond between the two, is called trans state. Conformation with trans positioning of molecules is one with the lowest energy. When one of the bonds is rotated around the afore defined axis while another is fixed in the plane that it has formed with the neighbouring bond rotational angle for this three bond sequence is defined by torsion angle. For any torsion angle compared to trans state energy of the conformation is increased. However, two secondary minimums called gauche-plus and gauche-minus can be seen. This two states present secondary formation of the bonds that can occur. The likelihood of this state taking place in the polymer depends on energy difference between the energy barrier between trans state and gauche states.

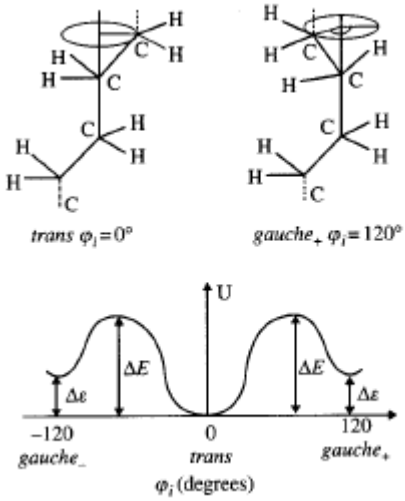

Figure 5: Top two schemes show trans and gauche positioning of adjacent monomeric units while the bottom graph is showing changes in energy state depending on rotation of one monomeric unit around the axis of another. Δε designates energy difference between trans and gauche states and ΔE defines energy barrier that needs to be overcome in order for polymer to change conformation.[1]

Qualitatively different mechanism of flexibility for many polymers such as DNA is uniform flexibility that stretches throughout whole chain. These chains are best described by worm-like chain models. This model also called Kratky-Porod model is best for describing stiffness of polymeric structures. This model focuses on fluctuations of the contour of the chain rather than onto trans-gauche bond rotation. Persistence length is the parameter defining this effect. It represents length of persistence segment that is best defined as segment of the polymer over which longitudinal tangent becomes nearly uncorrelated to the shape of the polymer.

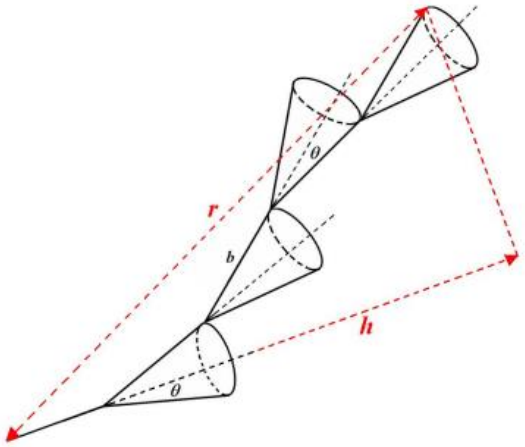

Figure 6: Schematic presentation of Kratky-Porod model (worm-like model)[3]

Deoxyribonucleic acid as a polymer

DNA has various importance in modern science. Firstly, it is a polymer coding for genetic information that is present in almost every cell of every organism on earth. Secondly, DNA has also recently emerged as an important material for nanotechnology. Its exceptional self-assembly capabilities have enabled the development of nanostructured systems with unparalleled precision at this scale.[4] Finally, deeper understanding of its chemical as well as physical properties provides bases for advancements in medical practice as it plays key roles in many devastating diseases such as effects of teratogens, cancer cell formation, radiation sickness etc.

As already discussed DNA is a heteropolymer that is best observed via Kratky-Porod model. DNA is best known naturally occurring heteropolymeric structure consisting of four different types of monomers (nucleotides).[1] Each of these nucleotides consist of one phosphate group ($PO4^-$) a pentose monosaccharide and a nitrogenous base that present monomers and are schematically shown in Figure 7. DNA chain best follows Kratky-Porod model of configuration because of high energy barriers limiting rotational changes of its conformation. Nonetheless, it has been observed that DNA chain can be found in at least two types of conformation depending on the surrounding environment.

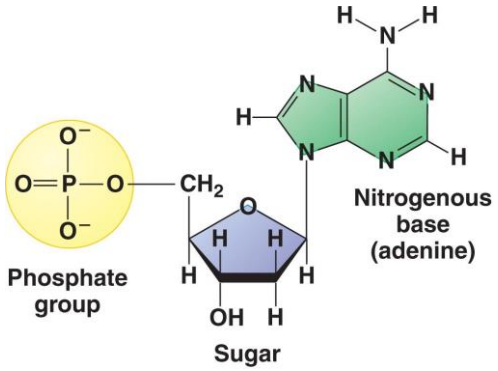

Figure 7: Display of structure of DNA monomeric unit/nucleotide[5]

Starting with its structure DNA is composed of monomeric molecules nucleotides interconnect into large polymers trough covalent bonds between phosphate group of one molecule to the pentose sugar of the adjacent molecule. Through this type of connection a single strand DNA is formed. It binds to the other complementary single strand in precise manner through hydrogen bonds always pairing in the same manner adenine to thymine and cytosine to guanine. This standard conformation of DNA nicely translates to small fragments as well. These are called oligonucleotide and are often used in experiments for that reason. Oligonucleotides can also adopt different conformations in solution depending on the sequence and environment. The best characterized form of DNA is the right-handed, B-form, helix. This form is normally adopted by double stranded DNA in

physiological conditions.[4] A thicker right-handed duplex with a shorter distance between the base pairs has been described for RNA-DNA duplexes and RNA-RNA duplexes and is called A-form nucleic acid.[6] Such conformational change also changes chemical and physical properties of the same molecule warranting for further experimentation.

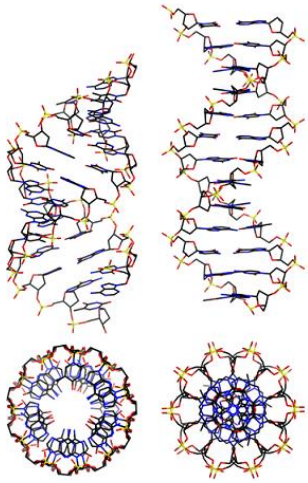

Figure 7: Models are showing B form of DNA (models left) and A form of DNA (model right)[6]

Polyelectrolyte

Many biological macromolecules such as proteins, polysaccharides, ribonucleic acids and DNAs act as polyelectrolyte when their solutions are prepared. The main characteristic separating polyelectrolytes from other polymers is the fact that they contain radical groups that easily ionize in water solutions. DNA is unique type of biological polyelectrolyte due to its ionized phosphate radicals. Observation of structure, dynamics and ionic charge of these macromolecules when dissolved in salty solutions is of importance because this simulation of the natural environment provides insight into their biological function. Polyions usually form rod like structure due to strong repulsive ionic forces from neighbouring charges. Once salt ions are added to the solution, counterions neutralize these repulsive forces influencing conformational flexibility of polyelectrolyte. Current models explaining polyelectrolyte solutions such as Imai and Onishi[7], Oosawa[8] and later Manning[9], are based on infinite linear chain and are accurate only in cases of infinitely small concentrations. This model dictates that when concentration of charged particles along the polyionic chain is high enough part of dissociated ions condense back to the proximity of polyelectrolyte. Due to this condensation charge of polyelectrolyte decreases consequently decreasing the repulsive forces and increasing flexibility of the polyion. The complete explanation of this phenomenon is much more complex and multifactorial mostly due to intrinsic asymmetric nature of polyelectrolyte solution. Thus it is impossible to define magnitude of electrostatic effect

for any particular polyion neither is it possible to define in a same manner the effect of any particular dissociated counterion.

Another aspect that needs to be taken into account when discussing polyelectrolyte solutions is concentration of polyelectrolyte itself. These solutions can be diluted or semidilute which greatly influences the structure and consequently electrostatic effects.

Manning Oosawa condensation

In this model linear distribution of charge in polyelectrolyte is crucial. This presumption dictates strong electrostatic attraction between dissociated ions and polyion chain. This attraction results with condensation of ions at the polyelectrolyte chains. This effect decreases charge density across the length of polyelectrolyte causing increased flexibility of polymer and decreasing the attraction of polyelectrolyte itself to dissociated ions thus creating a limit to which linear charge density can decrease. Electric potential of polyelectrolyte in the Manning-Oosawa condensation is presented by the following equation:

$$\varphi(r) = \frac{2e}{4\pi\epsilon_0\epsilon_r b} ln(r)$$

Eq. 1.: Electric potential (φ) as a function of distance of counterion (r) from polyion where b presents average distance between two neighbouring elemental charges on the polyelectrolyte and ε_r stands for relative substance permittivity. Additionally, constants ε_0 for absolute permittivity and e for unit charge.

This logarithmic correlation shows mathematical limit described earlier. Manning Oosawa condensation is characteristic for cylindrical and symmetrical shaped systems, due to their logarithmic dependence of electrostatic potential[10]. This model is good enough for explaining electrical properties for double stranded DNA water solutions.

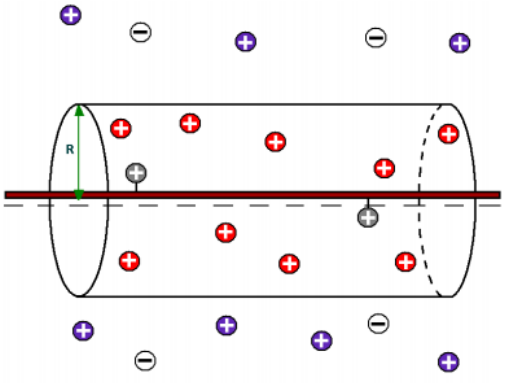

Figure 8: Display of Manning-Oosawa model depicting positions of counterions in polyelectrolyte solutions in relation to polymer radius and cylinder of radius (R) within which

polyion exerts electrostatic forces restricting movements of counterions. In polyelectrolyte solution we differ: free floating ions (blue ions), condensed counterions (red ions) and non-dislocated counterions (gray ions)[3]

Conductivity

Electrical conductivity is a fundamental physical quantity of any substance that quantifies how strongly it conducts the flow of electric current. In this dissertation it is signified by the Greek letter σ and units used according to SI are siemens per meter [S/m]. Both conductivity and conductance describe material's ability to enable electric current to flow through itself, however only conductivity is intrinsic property of the substance.

Conductivity is reciprocal of electrical resistivity, a physical property describing substance's resistance to flow of electric current. This relation is mathematically described in equation 2.

$$\sigma = \frac{1}{\rho}$$

Eq. 2: Conductivity (σ) is inversely proportional to resistivity (ρ)

According to Pouillet's law resistivity can be calculated with following equation:

$$\rho = R\frac{A}{l}$$

Eq. 3: Relation of resistivity (ρ) to substance's resistance (R) and length (l) of the specimen and its cross-sectional area (A)

Once parameters from Pouillet's law are inserted into equation 2 and Ohm's law is applied one can calculate conductivity of the material from experimental data.

$$\sigma = \frac{l}{AR} = \frac{lI}{AV}$$

Eq. 4: Relation of substance conductivity to electric current (I) obtained from experimental data collected at certain voltage (V).

Conductivity of polyelectrolyte solutions

In diluted polyelectrolyte solutions where distance between polyions is assumed to be large enough not to interact with each other. This is due to repulsive forces between same negative or positive charges on monomeric units. This repulsion straightening of polyion structure and forms rod structures where total length (L_p) of polyelectrolyte equals to product of monomeric length (L_m) and number of units/degree of polymerisation (N).

$$L_p = NL_m$$

Eq. 5: Length of polyelectrolyte in diluted solution

If polyelectrolyte solution is placed in external uniform electric field polyions and counter ions will move in opposite direction this provides evidence of conductivity. This solution's conductivity can be mathematically expressed as a sum of conductivity for every type of ion. Each ionic contribution can be calculated as a product of particle valency (z), its molar concentration (c[mol/L]) and its molar conductivity (Λ[Sm^2/mol]).

$$\sigma = \sum_i \sigma_i = \sum_i |z_i|\, c_i \Lambda_i$$

Eq. 6: Expression for calculating solution conductivity in general according to all type of charges (i) present.

This approach can also be used in polyelectrolyte solutions where charge is carried via polyion (designation p) and counterion (designation c).

$$\sigma = z_c c_c \Lambda_c + Z_p c_p \Lambda_p$$

Eq. 7: Formula for calculating conductivity of electrolyte solution in which Z_p signifies valency of polyion.

With introducing fraction of free counter ions (f) that are in theory sole carriers of electric current[11] and some further theoretical investigation that follow Manning-Oosawa condensation model one can express polyelectrolyte conductivity as:

$$\sigma = z_m c_m f\left(\Lambda_c + \Lambda_p\right)$$

Eq 8: Relation of electrolyte solution's conductivity to charge of one monomer (z_m) whilst including theoretical fraction of free ions (f). Note that concentration included in this formula represents concentration of monomeric units (c_m) and not polyelectrolyte concentration (c_p).

From final equation (Eq. 8) it is easily seen that for calculating theoretical conductivity of any solution is enough to get information on molar conductivity for simple ions that can be taken from the standardized tables. Molar conductivity of infinitely diluted NaCl water solutions at 298 K is 126.88 mSm^2/mol.[12] Molar conductivity of polyion (Λ_p) is a more complex problem because it depends on polyion diffusion coefficient (D_p) in addition to its charge (z_p).

Conductivity of DNA solutions

When measuring conductivity of DNA solutions same general approach as described above is used. First step is to determine molar concentration of monomeric units (c_m) following relation is used:

$$c_m[mmol/L] = \gamma_p[g/L]1.5[mmol/g]$$

Eq. 9: Conversion of mass concentration of double stranded DNA solution into molar concentration of monomeric units.

Second step is to determine molar conductivity of DNA solution. This can be calculated using Faraday constant (F) and mobility of polyion (μ_p).

$$\Lambda_p = F\mu_p$$

Eq 10: Molar conductivity of polyion (Λ_p) as a function of polyion moblity (μ_p).

After further mathematical investigation and introduction of diffusion coefficient (D_p) final equation for calculating polyion conductivity is given:

$$\Lambda_p = FQ_p\frac{D}{k_bT}$$

Eq. 11: Final formula for calculating molar conductivity of polyion (Λ_p) using charge of the polyion (Q_p), diffusion coefficient (D), temperature (T), Faraday constant (F) and Boltzman constant (K_b).

Taking into account that double stranded DNA polyion charge (Q_{DNA}) equals to double times monomer unit charge (Q_m) multiplied by fraction of free counterions (f) and considering the fact that each monomer has one PO_4^- unit with charge of one electron/unit charge (e) molar conductivity for DNA polymer solution can be expressed as:

$$Q_{DNA} = f2Q_m = f2e$$

Eq. 12: Polyion charge of double stranded DNA expressed by fraction of free counterions(f) and monomer unit charge (Q_m).

$$\Lambda_{DNA} = F\frac{D}{k_bT}f2e$$

Eq. 13: Expressing molar conductivity of DNA polyion (Λ_{DNA}) by introducing equation 12.

Combining equations 7, 8, 12 and 13 we can get mathematical expression for calculating conductivity of DNA solutions (σ_{DNA}).

$$\sigma_{DNA} = 2fz_{Na}c_{Na}\Lambda_{Na} + 2fZ_{DNA}c_{Dna}\frac{FeD}{k_bT}$$

Eq. 14: Inserting expression of for DNA molar conductivity (σ_{DNA}) from Eq. 13 into formula for polyelectrolyte conductivity (Eq. 7). Taking into account that amount of counterions equals to the ionic strength of DNA.

$$\sigma_{DNA} = 2fz_m c_{DNA}\left(\Lambda_{Na} + \frac{FeD}{k_bT}\right)$$

Eq. 15: Formula for calculation of conductivity of DNA solutions as a function of its concentration.

It is important to denote that molar conductivity of NaCl (Λ_{NaCl}) changes with molar concentration of NaCl (c_{NaCl}). For purposes of this dissertation values of NaCl molar conductivity were calculated using Debye–Hückel–Onsager equation taken from CRC Handbook of Chemistry and Physics, 93rd Edition.[12] Fraction of free counterions (f) for DNA solution is a instrinsic property calculated from ratio of maximal theoretical conductivity where fraction of free counterions is considered f = 1 and measured electrical conductivity of pure DNA solution water where no NaCl was added. Diffusion coefficient is also intrinsic property of polyelectrolyte which for Na-DNA solutions can be approximated with satisfactory precision to diffusion coefficient of counterions and amounts to $D = 1.33 \times 10^{-9}$ m^2/s.[13] For degree of polymerisation value of average number of base pairs in DNA in question $N = 10^4$ can be taken.[3] And for molar conductivity of Na^+ counterions value $\Lambda_{Na} = 50$ S cm^2/mol.[13]

Lastly, due to additive nature of conductivity (see Eq. 6) to calculate conductivity of NaCl DNA solution following equation is used:

$$\sigma_{solution} = z_{NaCl}c_{NaCl}\Lambda_{NaCl} + 2fz_m c_{DNA}\left(\Lambda_{Na} + \frac{FeD}{k_bT}\right)$$

Eq. 16: Formula for calculating solution conductivity.

Hypothesis

Measurements of electrical conductivity of DNA solutions with variable salt (NaCl) concentrations give results that are in accordance with calculated theoretical electrical conductivity defined by equation for DNA conductivity (Eq. 16),

Materials and methods

Materials used in this study where following solvents: sodium chloride (NaCl), magnesium chloride ($MgCl_2$) and genomic DNA, all of which were dissolved in Milli-Q water used as a solute. Milli-Q water was obtained by successive steps of filtration and deionization to achieve maximal purity in terms of low conductivity. Electrical conductivity of Milli-Q water was measured prior to preparation of all testing solutions. For NaCl samples conductivity of Milli-Q water measured was 4.63 μS/cm, for $MgCl_2$ samples conductivity 2.14 μS/cm was measured. Difference between two measured values was attributed to manipulation in laboratory setting. For more accurate results a different setting that provides a constant flow of sample solution trough the sensor is needed.

DNA samples

Primary DNA solution was prepared by dissolving dehydrated threads of Na-DNA threads. These were obtained from salmon testes (producer: Sigma-Aldrich, serial number D1626). DNA water solutions were then prepared according to following protocol. DNA fragments were dissolved in Milli-Q water over period of 48 hours at temperature 277 K. During second phase dissolved DNA was additionally diluted to the concentration of 0.2 g/L that was then used as primary solution for further steps in experiment.

Measurement setup

Solution concentrations for each of the afore mentioned electrolyte solution 50 mL primary solution was made that was later in the procedure diluted as needed. Primary solution concentrations are seen in table Tb.: 2. In order to achieve accurate concentrations of primary solutions solvent quantity was measured via analytical balance scale (producer: Mettler Toledo, serial number: 1125163141) with a precision of 0.01 mg.

Solvent	Concentration
NaCl	c(NaCl) = 20 mmol/L
$MgCl_2$	$c(MgCl_2)$ = 20 mmol/L
Na-DNA	γ(Na-DNA) = 0.2 g/L; c_m(Na-DNA) = 0.3 mmol/L

Tb. 2: Concentrations of primary solutions for solvents used in experiment.

From $MgCl_2$ primary solution testing samples were gathered by diluting it into a standardized 4 mL ampullae to attain desired range of molar concentrations: 0.025 – 5 mmol/L. Analogously, from NaCl primary solution testing samples were gathered by diluting it into a standardized 4 mL

ampullae to attain desired range of molar concentrations: 0.01 – 5 mmol/L. These were then used to preform conductivity measurement.

Na-DNA solution were prepared with constant concentration of 0.1 g/L DNA and variable concentrations of NaCl in range of molar concentrations: 0.01 – 2.5 mmol/L.

Measurements of conductivity were performed by conductivity sensor InLab® 741, produced by METTLER TOLEDO for all testing samples. This sensor measures electrical conductivity via conductivity measuring cell that consists of an electrode pair, so called electrodes, to which voltage is applied. The meter then measures the flowing current and calculates the conductivity according to the relationship presented in the equations Eq. 17 and Eq. 18.[14]

$$G = \frac{I}{V}$$

Eq. 17: Conductance (G) equals to ratio of electric current (I) and voltage (V)

$$\sigma = GK = \frac{Gl}{A}$$

Eq. 18: The linear relationship between conductivity (σ) and conductance (G) by a factor of a cell constant (K) which is dependent on the cross-sectional area of electrolyte between the electrodes (A) and the distance between the electrodes (*l*).

This simplified explanation neglects two very important aspects that must be considered. First is the accumulation of anions on anode and cations on cathode. This is called polarization which in turn this effect affects distribution of the solution and sensor detects falsely lower conductivity from the real. The greater the concentration of the solution means more ions and greater polarization effect thus this effect acts as an upper limit to the sensor. To prevent this undesirable effect alternating current can be applied this couses constant exchange of anode and cathode plate. Consequently, ions remain vibrating in place instead of migrating to either plate as shown in Figure 10. This method reduces the effect but does not eliminate polarization.

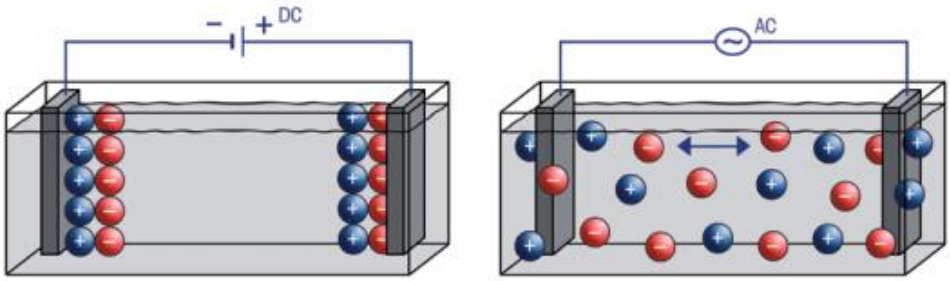

Figure 9: Migration of ions when direct current is applied (left) and oscillatory movement of ions when alternating current is applied (right)[14]

Second effect that presents the lower limit to every sensor is capacitance. This is the effect produced by two electrodes that are put in non-conductive medium. When direct current is applied to these electrodes no electric flow is produced causing the electrodes to behave like a capacitor.[1] The capacitive resistance becomes infinitely high for conductance is zero. However, when alternating current is applied to the electrodes the capacitive resistance drops and conductance rises correspondingly. This is highly dependent on measuring frequency which then has to be adapted according to expected conductivity of the sample.

Device

Device (InLab® 741) used for my research is a 2 steel pole steel shaft type of conductor. These with a measuring range of 0.001 – 500 µS/cm. Main advantage of the 2-pole conductivity cell is high accuracy of low conductivity measurement. This type of sensor is typically used for conductivity measurements of pure water and highly diluted aqueous solutions. Figure 11 shows schematics of 2-pole conductivity cell used.

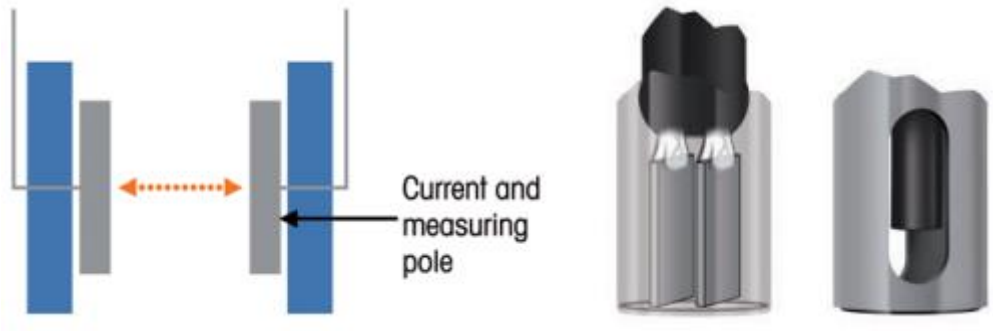

Figure 10: Schematics of a 2 pole conductivity cell[14]

To acquire most accurate results for each concentration of electrolyte solutions three measurements for each testing sample were performed while for DNA solution five measurements were performed per testing sample. Collected data was then statistically analysed and plotted.

Results

Figure 11 and figure 12 are showing conductivity measurements for NaCl and $MgCl_2$ water solutions. Three separate measurements were performed for each concentration and were statistically analysed using OriginLab® 2018 and plotted in order to define correlation between the two physical values.

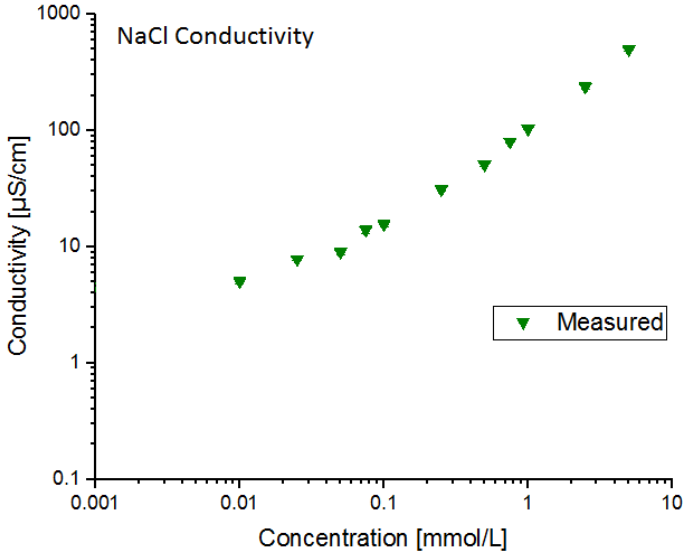

Figure 11: Graphical presentation of the average conductivity of NaCl solutions together with corresponding standard deviations plotted against its concentration on logarithmic scales.

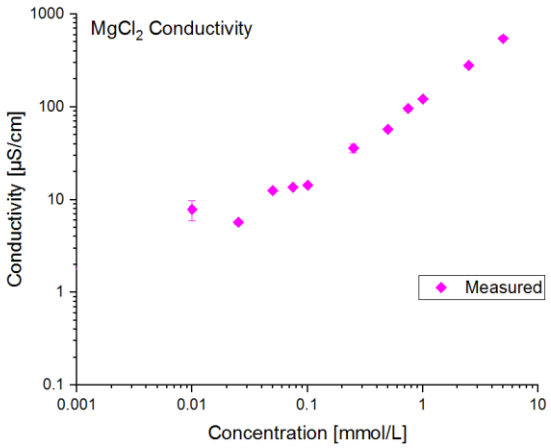

Figure 12: Graphical presentation of the average conductivity of $MgCl_2$ solutions together with corresponding standard deviations plotted against its concentration on logarithmic scales.

Figure 13 is showing conductivity measurements for 0.1 g/L DNA NaCl solutions where concentration of NaCl were altered. Five separate measurements were performed for each

concentration and were statistically analysed using OriginLab® 2018 and plotted in order to define correlation between the two physical properties.

Also conductivity of pure DNA solution without addition of NaCl salt was measured σ_{DNA} = 20,58 µS/cm.

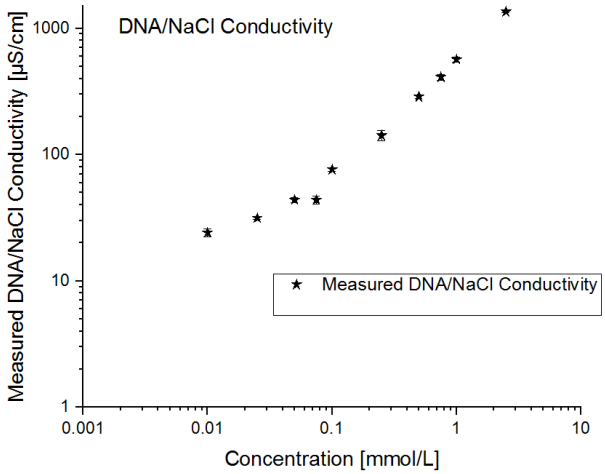

Figure 13: Graphical presentation of the average conductivity of the DNA/NaCl solutions together with corresponding standard deviations plotted against NaCl concentration.

Discussion

According to eq. 7 theoretical values for conductivity of NaCl and $MgCl_2$ solutions were calculated and plotted together with measured values. This is shown in figure 14 for NaCl solution and in figure 15 for $MgCl_2$ solution.

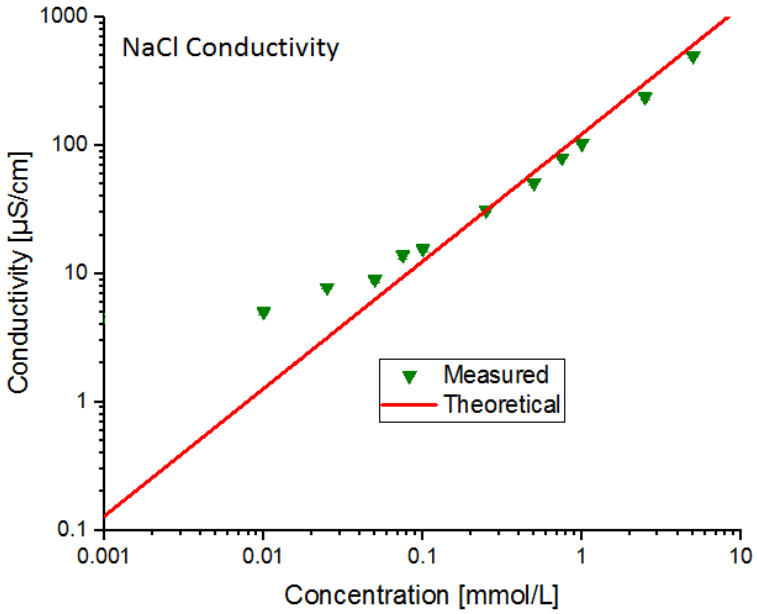

Figure 14: Triangular marks show measured conductivities for NaCl solutions while the red graph presents theoretical values for conductivities both plotted against concentration of the electrolyte solution question.

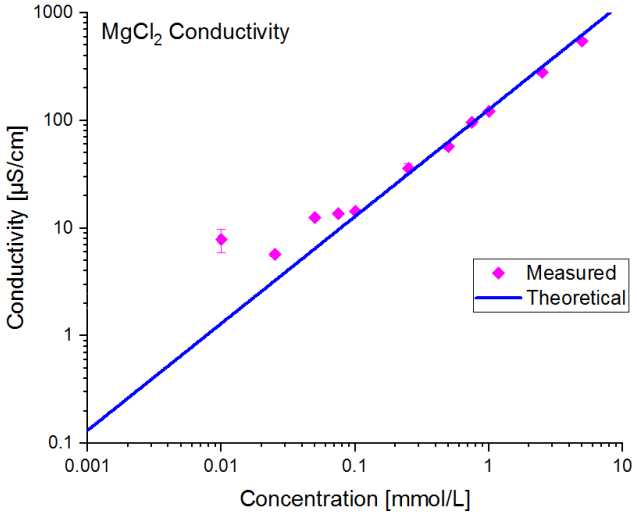

Figure 15: Diamond marks show measured conductivities for $MgCl_2$ solutions while the blue graph presents theoretical values for conductivities both plotted against concentration of the electrolyte solutions in question.

From observing both graphs it is easily recognized that at lower concentrations there is mismatch of measured versus theoretical conductivities. This can be attributed to dissolved carbon dioxide (CO_2) from the environment. It is well established that once CO_2 is dissolved in water it forms carbonic acid that dissociates into hydronium ions, bicarbonate ions that contribute to increased conductivity. Additionally, unlike other gases solubility of CO_2 increases with decreased temperature. Trruman S. Light et al claim that conductivity of Milli-Q water increases approximately 1 µS/cm when exposed to air with CO_2 content of 0.033%, which is typical value for pure air.[15] Considering all the above mentioned it is obvious that for values of conductivity measured bellow 20 µS/cm this causes a mistake of over 5% which is then seen as false higher conductivity and causes deviation from calculated values. This is also seen in both Figures 14 and 15 for NaCl and $MgCl_2$ solutions respectively. Note that once measured values increase above 20 µS/cm measured results follow theoretical predictions and are not significantly affected by described effect.

To approximate contributions of NaCl electrolyte to the total conductivity of DNA/NaCl solutions testing samples, graph displaying measured conductivities for pure NaCl solutions and graph displaying measured conductivities for DNA/NaCl solutions were combined (see Figure 16).

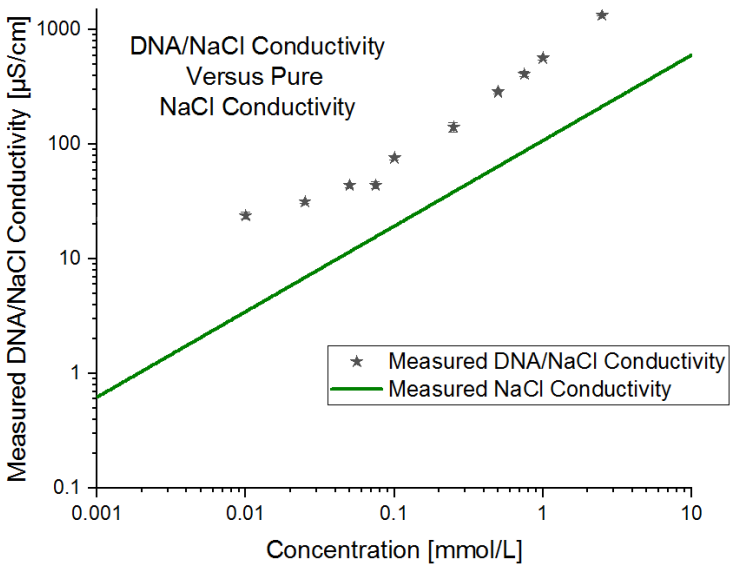

Figure 16: Star marks represent measured conductivity values of DNA/NaCl solutions while green graph is showing fitted curve for conductivity values of NaCl solutions.

With careful observation of figure 16 one can already see that for increase of DNA/NaCl solution's conductivity pure contribution of NaCl ions is not the only factor, even though it is the only variable parameter in this particular experiment setup. This conclusion can be drawn by the

fact that measured parameters are not parallel, which would be the case if the conductivities of both solvents were additive.

Figure 17 displays comparison between theoretical values of DNA/NaCl solutions calculated using equation 16 and measured values of this solutions. Fraction of free counterions (f) used was estimated by calculating the ratio between maximal theoretical conductivity for Na-DNA polyion (σ_{DNA}= 30 μS/cm when f = 1) and conductivity of Na-DNA solution measured when no NaCl was added (σ_{DNA}= 20,58 μS/cm) resulting with fraction of free counterion f = 0.686.

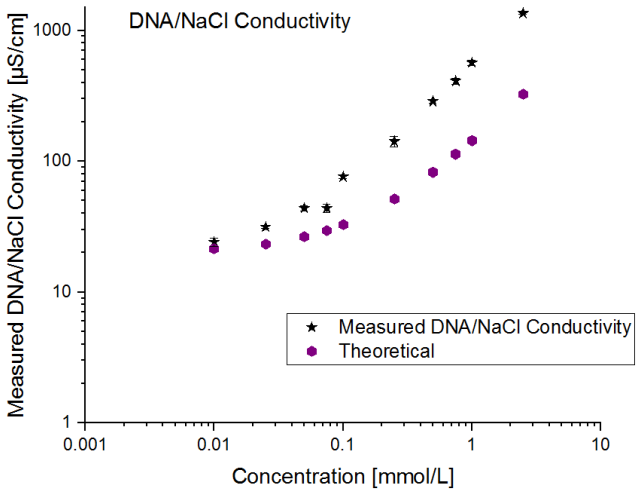

Figure 17: Star marks represent measured conductivity values of DNA/NaCl solutions while purple hexagons are showing calculated conductivity values of DNA/NaCl solutions.

It is clear from this comparison that theoretical calculation of conductivity are not accurate in predicting measured values. In figure 17 it is also clearly seen that with increasing concentration difference between projected values and measured values keeps increasing making it evermore inaccurate. Another thing that may be observed is that measured conductivity values change slope somewhere between NaCl concentration 0.75 mmol/L and 1 mmol/L. Within this range it can be speculated that DNA changes conformation. Also data at NaCl concentrations higher than 1 mmol/L display certain consistency that can be speculated to mean stabilization of DNA macromolecule. This data is insufficient to provide undisputable proof and thus further research of this suspected conformational change is needed. Also since even at higher NaCl concentrations difference between theoretical and measured conductivity is not constant it is probable that with changing environment fraction of free counterions changes thus changing molar conductivity of DNA polyelectrolyte. In that case molar conductivity of DNA is not only dependant on its intrinsic properties but also on the environment that surrounds this macromolecule. Additional investigations are needed in order to provide more accurate model that includes correlation between environmental factors and molar

conductivity of DNA. Note that results from this dissertation are only applicable for γ_{DNA} = 0.1 g/L and in order to create such model more similar experiments that include variable DNA concentrations are needed.

Conclusion

To conclude based on the results measured by the device (InLab® 741) the hypothesis is refuted. The equation 16 does not accurately predict conductivity values of DNA/NaCl solutions. This may be attributed to changes in conformation that are speculated to occur somewhere in range between NaCl concentration 0.75 and 1 mmol/L. Furthermore, due to inconsistent increase in difference between measured and theoretical values it is possible that changes in molar conductivity of DNA occur with changing environment due to changes in fraction of free counterions. Finally, gathered data shows need for further research in this area so that better model for conductivity of DNA can be established.

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In this dissertation I would foremost like to thank my mentor for the guidance provided for completing this research and teachings she provided when confronted by challenges during my investigation. I would also like to thank my parents for their belief in me and everlasting support throughout my studies. Finally, I would also like to thank my friends who picked me up when I was down and motivated me through numerous hardships that I encountered during when preparing for exams and when pressured by deadlines.

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