RECENT ADVANCES IN SYNTHETIC POLYAMPHOLYTES

Kudaibergenov S. E[1]. , Bekturov E. A[2].

[1]*Institute of Polymer Materails and Technology, Microregion "Atyrau 1" Bld. 3/1, Almaty, 050019, Kazakhstan*
[2]*Abay Kazakh National Pedagogical University, Dostyk ave., 13, Almaty, 050010, Kazakhstan*

Abstract. In this review the properties of annealed, quenched and betainic (or zwitterionic) polyampholytes in solution and gel state are analyzed. The stimuli-responsive character of water-soluble and water-swelling polyampholytes exhibiting temperature-, salt-, pH-, solvent-, DC- and bio-sensitivity is considered. Self-healing and shape memory behavior of polyampholyte gels is also in the eyeshot of this review. Catalytic properties of metal nanoparticles and enzymes immobilized within macroporous polyampholyte cryogels to design stable, active and selective flow through catalysts in reactions of hydrolysis, hydrogenation and oxidation of various substrates are highlighted. Future perspectives in development of polyampholytes are outlined.

Keywords: polyampholytes, stimuli-sensitivity, hydrogels, cryogels, catalysis

1. Historical background on development of polyampholytes

The intensive development of research on polyampholytes since 1950s has included the pioneering studies of Alfrey, Morawetz, Fuoss [1-3], Katchalsky [4-6], Ehrlich, Doty [7]. The interest to polyampholytes was due to several reasons. The first is the similarity of the hierarchical structure of amphoteric macromolecules to the structural organization of proteins [8,9]. The second is the modeling of the protein folding mechanism by synthetic polyampholytes [10]. The third is utilization of amino acids in obtaining amphoteric polypeptides [11-16], and forth is possibility of preparing semi-natural polyampholytes by modification of natural building blocks [17, 18].

One of the fundamental findings discovered in the last century with respect to polyampholytes is so-called *antipolyelectrolyte effect* [7, 19-26] that is related to the unfolding of annealed polyampholytes at the isoelectric point (IEP) or charge-balanced quenched polyampholytes in quasi-neutral state in the presence of salts. Another phenomenon is so-called *isoelectric effect* [27-32] that is connected with cooperative release (or detachment) of preliminary bondedlow- or high-molecular-weight substances at the IEP of annealed polyampholytes or at quasi-neutral state of charge-balanced quenched polyampholytes as a result of competition between inter- and intra-macromolecular (inter-MMC and intra-MMC) complexation.

The third finding is the structural and behavioral similarity of blockpolyampholytes and interpolyelectrolyte complexes (IPEC) –the products of interaction between oppositely charged linear polyelectrolytes [33-40].

Definitions and classifications of polyampholytes are given in Refs. [41-44]. Annealed polyampholytes consisting of acid-base monomers are ionized as a function of pH, whereas quenched polyampholytes retain their respective charges independently of pH. Combination of the weak acid/cationic or weak base/anionic monomers along macromolecular chains is defined as semi-annealed or semi-quenched polyampholytes. Polymeric betaines or synonymously zwitterionic polymers represent a unique subclass of polyampholytes containing acid/base or anionic/cationic groups on the same pendant monomeric units. In polybetaines the cationic group

is represented by quaternary ammonium groups, while the anionic groups can be carboxylate [poly(carbobetaines)], sulfonate [poly(sulfo betaines)] and phosphate (or phosphinate, phosphonate) groups [poly(phosphobetaines)].If the cationic-anionic monomer pairs compensate each other without counterions they can be represented as zwitterionic polymers [45-50] or polyampholytic ionic liquids [51, 52]. In the current literature, the terms "zwitterionic polyampholytes", "polybetaines", "zwitterionic polyelectrolytes", "polyzwitterions" are also widely used. Examples of annealed quenched, betainic, semi-annealed or semi-quenched polyampholytes are shown in Fig. 1.
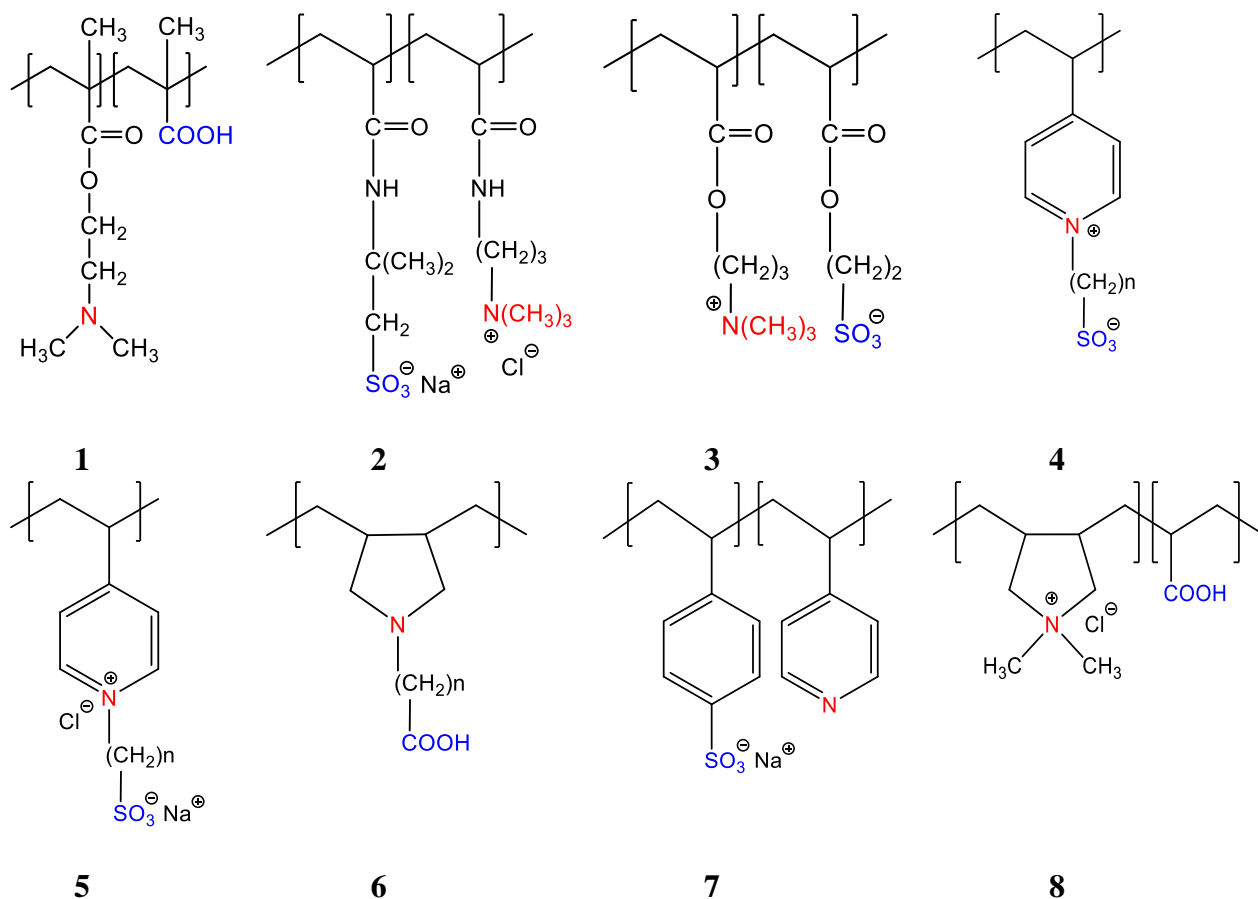

Figure 1. Repeating monomeric units of annealed (1), quenched (2), zwitterionic (3, 4), quenched betainic (5), annealed betainic (6) and self-annealed (or self-quenched) (7, 8) polyampholytes

The behavior of polyelectrolytes and polyampholytes in aqueous-salt solutions is quite different [53]. For instance, in salt solution the electrostatic repulsion between uniformly charged macroions is screenedand the macromolecular chain is coiled and its behavior tends to be close to neutralpolymer (*polyelectrolyte effect*). In contrast, annealed polyampholytes at the IEP and quenched polyampholytes as well as polymeric betaines and zwitterionic polymers in a quasi-neutral state unfold in salt solution due to screening of the electrostatic attraction between oppositely charged moieties (*antipolyelectrolyte effect*).

A renewed interest in polyampholytes is expected to result from evaluation of the microstructure and properties of linear and crosslinked amphoteric macromolecules,

accompanied by systematic evaluation of the literature [54-59].

2. Behaviors of annealed, quenched and zwitterionicpolyampholytes in aqueous-salt solutions

The composition, microstructure, charge distribution and density, molecular weight are the key parameters for a comprehensive analysis of the structure-properties-function relationships of polyampholytes [60-68]. Conformational change of annealed polyampholyte as a function of pH and ionic strength of the solution (μ) is shown in Fig. 2a [23]. The quenched polyampholyte gel exhibits a minimal swelling degree when the total charge is close to zero (Fig. 2b) [69]. Deviation from the IEPfor linear annealed polyampholyteand/or excess of anionic and cationic monomers in quenched polyampholyte gel leads to unfolding and swelling of macromolecular chains and the macromolecules behave like ordinary polyelectrolytes, e.g. the viscosity (or swelling degree) decreases with increasing of the μ (Fig.2, blue arrow). An opposite, so-called *antipolyelectrolyte effect* is observed at the IEP oflinear annealed polyampholyte and crosslinked charge-balanced quenched polyampholyte. The intrinsic viscosity [η] ofannealed polyampholyte increases with increasing of the μ(Fig.2, red arrow). Hydrogels of quenched polyampholyte prepared from 2-(methacryloyloxy)ethyltrimethylammonium chloride (MADQUAT) and sodium 2-acrylamido-2-methylpropanesulfonate (AMPS) (MADQUAT-*co*-AMPS) are in swollen and collapsed states in dependence of the net charges expressed as $\Delta f = f^+ - f^-$, where f^+ and f^- are the molar ratios of cationic and anionic monomers to the total monomer concentration, respectively [69]. In pure water when the molar ratio of cationic to anionic groups approaches to unity the opposite charges act as physical crosslinks stabilized by inter-MMC and/or intra-MMC and the liberated free counterions are effectively "dialyzed" from the hydrogel interior. As the concentration of the anionic and cationic parts deviates from the equimolar ones $\Delta f \neq 0$ the swelling ratio Q_e increases rapidly because the number of osmotically active ions in the hydrogel phase increases according to Donnan equilibrium.
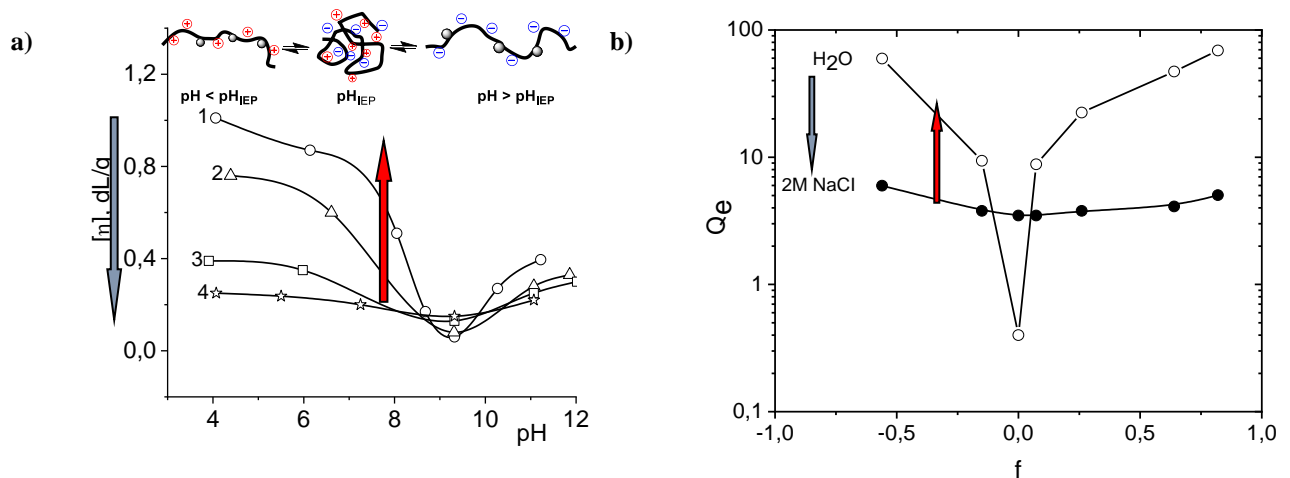

Figure 2.a) pH-dependent changing of the [η]of annealed polyampholyte in the presence of 0.005 (1), 0.01 (2), 0.1 (3), and 0.3M KBr (4). Reprinted from Ref. 23;**b**) Behavior of quenched polyampholyte MADQUAT-*co*-AMPS in pure water and 2 mol·L^{-1} NaCl solution. Redrawn from ref. 69

The behaviors of random quenched polyampholytes composed of anionic monomer – sodium salt of 2-acrylamido-2-methyl-1-propanesulfonic acid (AMPS) and cationic monomer – (3-acrylamidopropyl)trimethylammonium chloride (APTAC) (abbreviated as AMPS$_m$-*co*-APTAC$_n$ where m = 25, 50, and 75 mol.%, n = 75, 50, and 25 mol.%) were evaluated in aqueous-

salt solutions [70]. It was found that in aqueous KCl solutions, the $[\eta]$ of charge-unbalanced polyampholytes − AMPS$_{25}$-*co*-APTAC$_{75}$ and AMPS$_{75}$-*co*-APTAC$_{25}$ decrease while the $[\eta]$ of charge-balanced polyampholyte − AMPS$_{50}$-*co*-APTAC$_{50}$ increases demonstrating *antipolyelectrolyteeffect*. Increasing of the $[\eta]$ of charge-balanced AMPS$_{50}$-*co*-APTAC$_{50}$ in salt solution is due to destruction of intra- and interionic contacts between oppositely charged monomers. In high saline water, the electrostatic attraction between positively and negatively charged monomers is shielded by anions and cations of salts and the macromolecular chain unfolds (Fig.3).
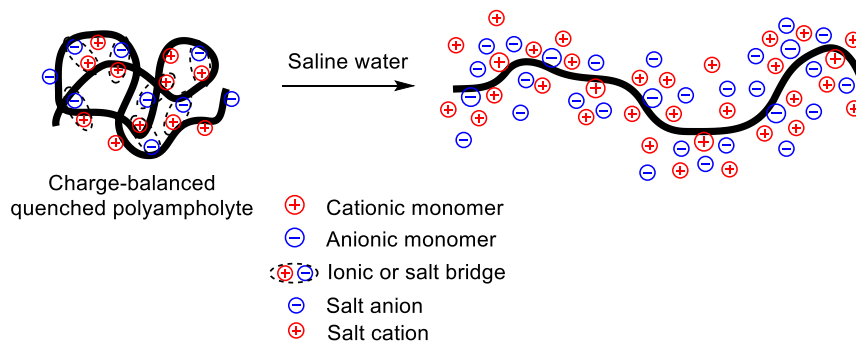

Figure 3. Unfolding of charge-balanced quenched polyampholyte AMPS$_{50}$-*co*-APTAC$_{50}$ in saline water

The unique properties of polymeric betaines are the tendency of the zwitterionic fragments to form intragroup, intrachainand interchain contacts as demonstrated in Fig. 4. In some cases, interchain crosslinking may cause gelation of polyzwitterions [45].
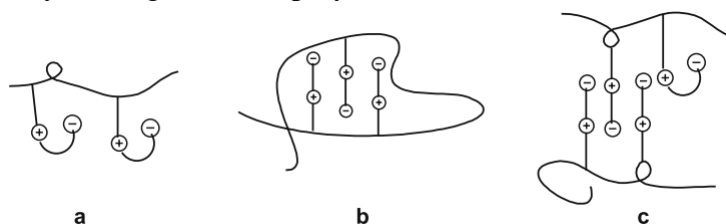

Fig.4. Formation of intragroup (a), intrachain (b), and interchain (c) salt bonds in polybetaines

Addition of salt leads to disintegration of inter-MMC and/or intra-MMC and to exhibiting of *antipolyelectrolyte effect*.Swelling of polyzwitterions is due to screening of the electrostatic attractions between opposite charges that diminishes both charge and crosslink density.Thus, the *antipolyelectrolyte effect* is a common phenomenon for annealed, quenched and betaine type polyampholytes in aqueous-salt solution [41-44].

3. Stimuli-responsive properties of polyampholytes

3.1 Temperature and salt sensitive polyampholytes

The polyampholytes possess thermoresponsive character and exhibit both lower (LCST) and upper critical solution temperature (UCST) at definite salt concentration, polymer concentration and molecular weight of polymers. The thermoresponsive behavior of quenched diblock polyampholytes composed of anionic block (AMPS)$_{82}$ (where the lower subscript 82

indicates the polymerization degree) and cationic blocks (APTAC)$_n$ (where n = 37, 83 and 181) (abbreviated as AMPS$_{82}$-*b*-APTAC$_n$) was studied in aqueous and aqueous-salt solutions [71]. All three quenched polyampholytes, namely PAMPS$_{82}$-*b*-PAPTAC$_{37}$, PAMPS$_{82}$-*b*-PAPTAC$_{181}$, and PAMPS$_{82}$-*b*-PAPTAC$_{83}$ exhibit thermo-responsive character with LCST at definite polymer (C$_p$) and NaCl concentrations (Fig.5a). Phase transitions for PAMPS$_{82}$-*b*-PAPTAC$_{37}$, PAMPS$_{82}$-*b*-PAPTAC$_{83}$ and PAMPS$_{82}$-*b*-PAPTAC$_{181}$ are observed for [NaCl] = 0.5-0.75, 0.8-1.1, and 0.7-0.9 M, respectively. The phase transition temperature of PAMPS$_{82}$-PAPTAC$_{83}$ in 0.9 M NaCl shifts toward higher values with decreasing C$_p$ from 10 to 1 g L^{-1}.
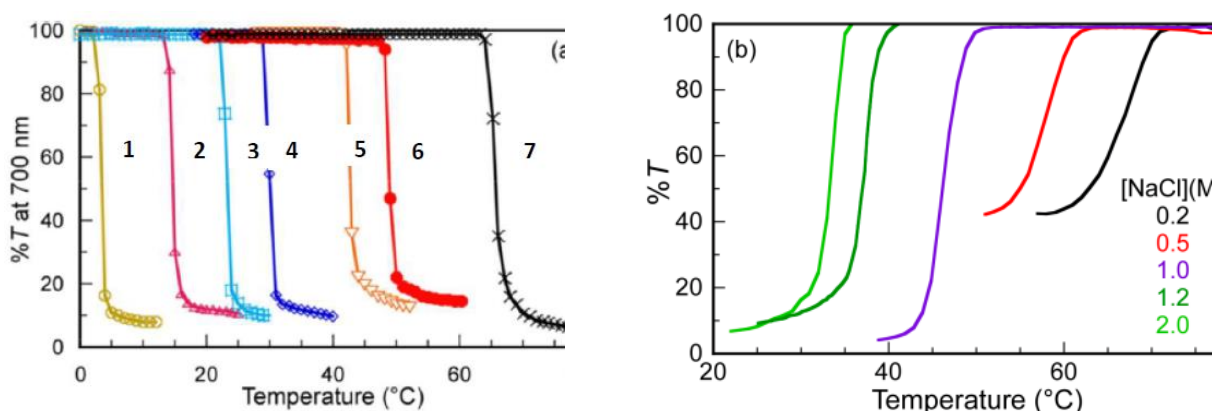

Figure 5. **a**) Temperature dependent transmittance (%T) of aqueous solution of PAMPS$_{82}$-*b*-PAPTAC$_{83}$ at C$_p$ = 10 g·L^{-1} and [NaCl] = 0.80 (1), 0.85 (2) 0.90 (3), 0.95 (4), 1.00 (5), 1.05 (6), 1.10 (7). Reproduced from Ref. 71;**b**) Temperature dependent transmittance (%T) at 700 nm for aqueous solutions of P(VBTAC/NaSS)$_{97}$ at C$_p$ = 2.0 g·L^{-1} as a function of temperature at different NaCl concentrations with cooling. Reproduced from Ref. 72

Quenched polyampholytes composed of vinylbenzyl trimethylammonium chloride (VBTAC) and sodium p-styrenesulfonate (NaSS) [P(VBTAC/NaSS)$_n$] exhibits UCST behavior in aqueous NaCl solutions at C$_p$= 0.5-5.0 g·L^{-1} [72]. The shifting of the UCST to higher temperature takes place upon increasing the polymer concentration and molecular weight. However, the UCST decreases by increasing the salt concentration (Fig.5b).

3.2 pH and solventsensitive polyampholyte gels

The pH [73, 74] and solvent-responsive [75, 76] properties of polyampholytes are demonstrated for [3-(methacryloylamino)propyl]trimethylammonium chloride-*co*-acrylic acid (MAPTAC-*co*-AA), MAPTAC-*co*-sodium acrylate (MAPTAC-*co*-SA) and polybetaine gel (Fig. 6).
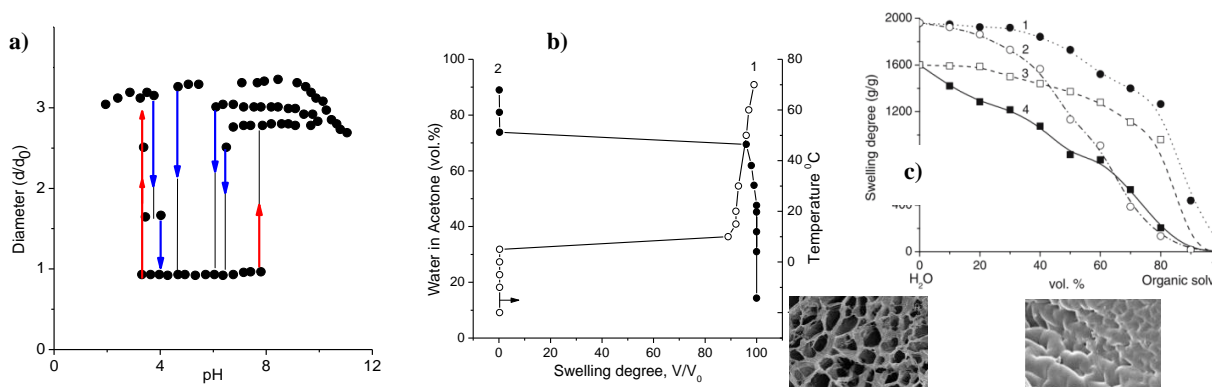
_____

_____

Figure 6. **a**) Equilibrium swelling degree d/d_0 of MAPTAC-*co*-AA gel in water as a function of pH (Reproduced with permission from Ref. 73). **b**) Temperature- (1) and solvent-dependent (2) volume phase transition of MAPTAC-*co*-SA gel. (Reproduced with permission from Ref. 74). **c**) Swelling-shrinking of polybetaine gel in water-acetone (1,3) and water-ethanol (2,4) mixtures. SEM pictures of polybetaine gel in water and acetone. (Reproduced with permission from Ref. 75).

At pH 6.4 the MAPTAC-*co*-AA has a diameter $d = d_0$ that is referred to phase **1** ($d/d_0 = 1$). At pH = 8.5 the gel swells discontinuously to phase **2.7** (Fig.6a). If the pH decreases from 8.5 to 7.0 the gel returns to phase **1** discontinuously. If instead the pH increases further from 8.5 to 9.8 and goes back from this point, the gel collapses into phase **1** at pH 6.4. Multiphasic pH-dependent behavior of MAPTAC-*co*-AA [73] and 4VP-AA [77] in a wide range of pH including the pH_{IEP} along the plateau regime is due to the fact that pH inside the hydrogel remains at the pH_{IEP} although the external pH in bath changes over a wide range. This swelling-collapsing behavior of polyampholyte gels due to pH difference between the inside and outside of amphoteric network is adequately described by Flory-Rehner swelling theory coupled to the Donnan equilibrium. Thermoshrinking of amphoteric MAPTAC-*co*-SA gel [74] is originated from a relative ratio of hydrophobic group to hydrophilic group while discontinuous volume-phase transition of MAPTAC-*co*-SA in water-acetone mixture is characterized by the specific binding between the oppositely charges (Fig. 6b).In pure organic solvents (ethanol or acetone) the polybetaine gel collapses due to significant suppression of ionization and poor thermodynamic quality of solvents with respect to ionizable groups of polyampholytes. SEM pictures taken in water and acetone are also in good agreement with swelling-shrinking behavior of such hydrogels (Fig. 6c).

3.3 DC responsive polyampholyte gels

The direct current (DC) responsivity of polyampholyte gel specimens was considered in Refs. [78-80]. It has been found that the polyampholyte gel shrinks, swells or oscillates at the pH_{IEP}. These results were interpreted in terms of redistribution of fixed and mobile ions, the Donnan equilibrium and water dissociation. The appearance of pH gradient inside of polyampholyte gel samples induced by DC electric field was tested for anionic, cationic and amphoteric gels [81, 82]. The pH profiles of anionic polyelectrolyte – poly(sodium acrylate) and cationic polyelectrolyte – poly(vinyl ether of ethanolamine) have no any considerable pH deviation in an applied external DC electric field. Whereas for equimolar amphoteric gel composed of sodium acrylate and vinyl ether of ethanolamine (SA-*co*-VEMEA) the applied DC electric field leads to appearance of linear stable pH gradient. The central part of gel specimen adopts neutral pH while near of cathode and anode sides the pH value is more acidic and basic respectively (Fig. 7a). Based on this principle it is suggested a novel approach for separation of protein mixture within monolithic polyampholyte gel [81, 82]. In a new approach the proteins to be separated are concentrated in appropriate pH zones of amphoteric gel according to their isoionic points (pH_{IIP}) (Fig. 7b). The advantage of this method is that the gel matrix and amphoteric buffer is combined in one and the same sample[83].
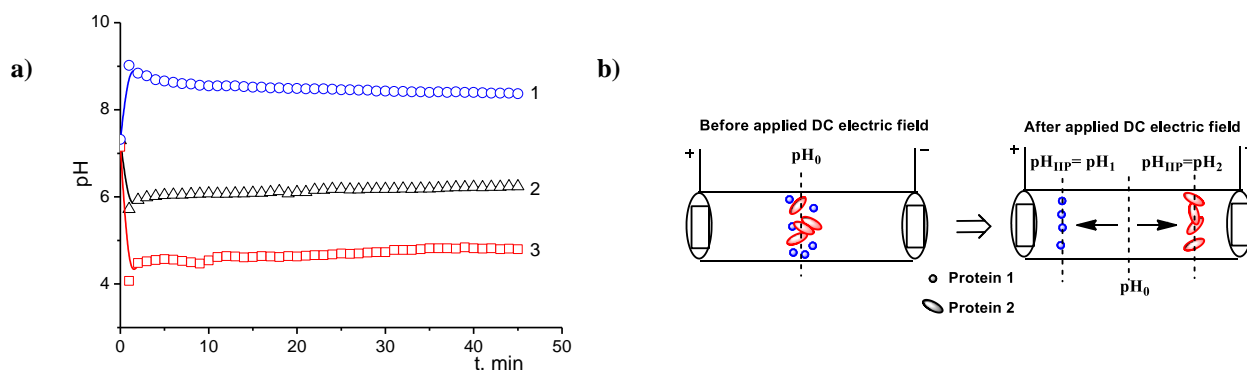
Figure 7. **a**) Appearance of pH gradient near of anode (1), central (2), and cathode (3) sides of SA-*co*-VEMEA amphoteric gels specimen at E = 10 V. Ref. 111;**b**) Scheme of separation of proteins within monolithic amphoteric gel under externally imposed DC. (Reproduced from Ref. 82)

3.4 Biosensitive system based on polyampholyte gels

Kokufuta et al. [84] developed polyampholyte hydrogels consisting of N-isopropylacrylamide (NIPAM), acrylic acid (AA) and N-vinylimidazole (VI) monomeric units with co-immobilized glucose oxidase and urease. Such system is able to convert biochemical energy to mechanical work and serve as a biocatalyst through the positive feedback mechanism. A swollen gel shrinks at the IEP (pH ~ 6-7) and swells upon deviation from the IEP. Swelling and shrinking of amphoteric gel containing both glucose oxidase and urease is demonstrated in Fig.8.
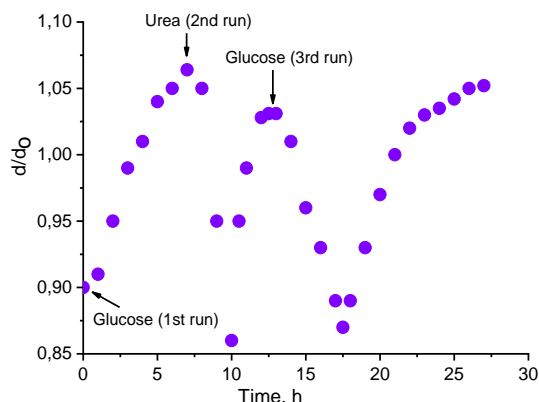

Figure 8. Swelling and shrinkingof NIPAM-*co*-AA-*co*-VI polyampholyte gels with co-immobilized enzymes upon alternative addition of urea and glucose. (Redrawn from Ref. 84)

Ureasa decomposes the urea to ammonia and carbon dioxide and increases the pH while glucose oxidase produces gluconic acid and decreases the pH. Consequentlyin the presence of glucose the hydrogel swells due to ionization of imidazole groups, in the presence of urea the hydrogel swells due to ionization of carboxylic groups. Thus, permanent addition of urease and/or glucose oxidase to NIPAM-*co*-AA-*co*-VI containing co-immobilized enzymesdemonstrates time-dependent oscillation of amphoteric gel.

The impact of pH-coupled with oxygen stimuli on osmotic pressure and electrical potential responses of hemoglobin-loaded polyampholyte hydogel was studied by authors [85] for developing biocompatible pH- and oxygen-reactive gas carrier, bio-sensing and actuating materials.

_____

_____

3.5 Self-healing and shape memory behavior of polyampholyte gels

Authors [86-89] developed a new class of tough, viscoelastic and self-healing polyampholytehydrogels by random copolymerization of oppositely charged ionic monomers at high monomers concentration and equimolar monomers ratio. The unique property of these gels is that the strong ionic bonds play the role of permanent crosslink whilethe weak ionic bonds are responsible for elongation, deformation, adhesion, self-healing, and shape memory. The sense of self-healing is that a virgin sample after cutting the weak ionic bonds can be re-formed again and healed in the vicinity of surfaces (Fig.9).
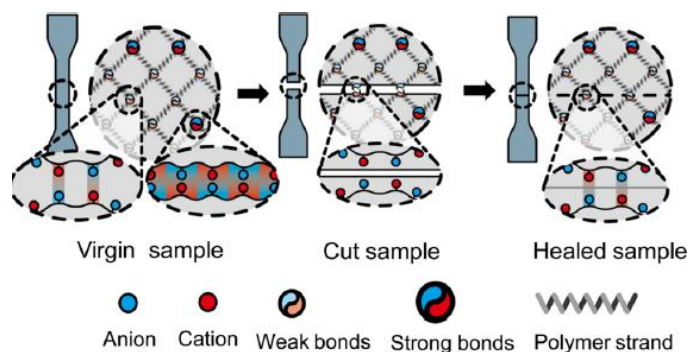

Figure 9. Schematic representation of self-healing of quenched polyampholyte hydrogels. Reproduced from ref. 88

Introduction of hydrophilic or hydrophobic groups into the macromolecular chain of polyampholyte hydrogels drastically changes their physico-chemical and mechanical properties [77, 90, 91]. "Dilution" of continuous charges of polyampholyte sequences by hydrophilic (or hydrophobic) monomers may control the formation of intra-MMC or inter-MMC leading to either more relaxation of chains or produce inter-MMC between the polymer chains acting as physical cross-link. Hydrophobically modified physical gels were prepared from annealed (equimolar copolymer of acrylic acid and 4-vinylpyridine (AA-*co*-4VP)) [77] and charge-balanced quenched (AMPS-*co*-APTAC) polyampholytes [91]. As hydrophobic monomer, copolymer of AA-*co*-4VP contained stearyl methacrylate (SMA), AMPS-*co*-APTAC–octadecylacrylate (ODA). Both hydrogels exhibited a high tensile strength and stretchability. Cut-and heal testes conducted at 50 C° revealed a complete healing efficiency with respect to Young's modulus for quenched polyampholyte gels within 4 h (Fig. 10).
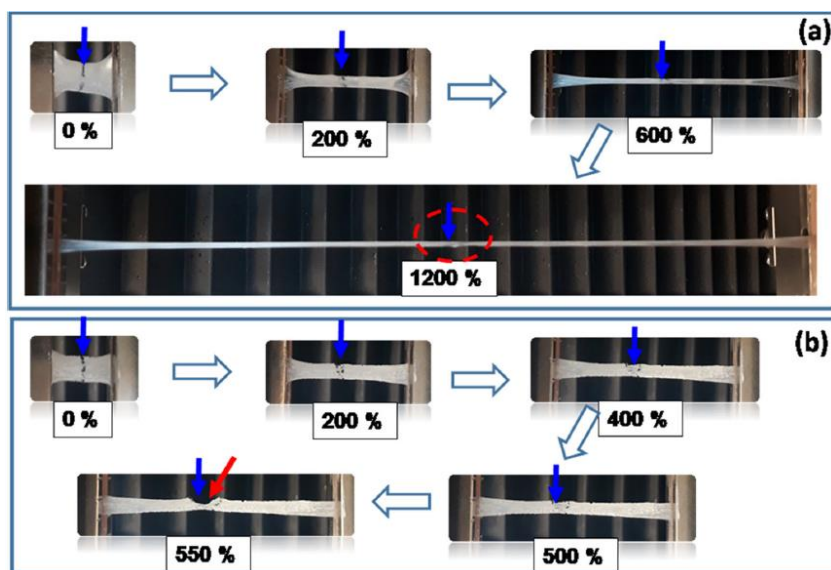
Figure 10. Images of healed AMPS-APTAC-ODA hydrogel specimens with 5 (a) and 20 mol.% (b) ODA during stretching. Reproduced from Ref. 91

Shape memory behavior of quenched polyampholyte hydrogels derived from MAPTAC-*co*-NaSS [86] and MAPTAC-*co*-NaSS-*co*-MAA [92] is compared in Figure 11.
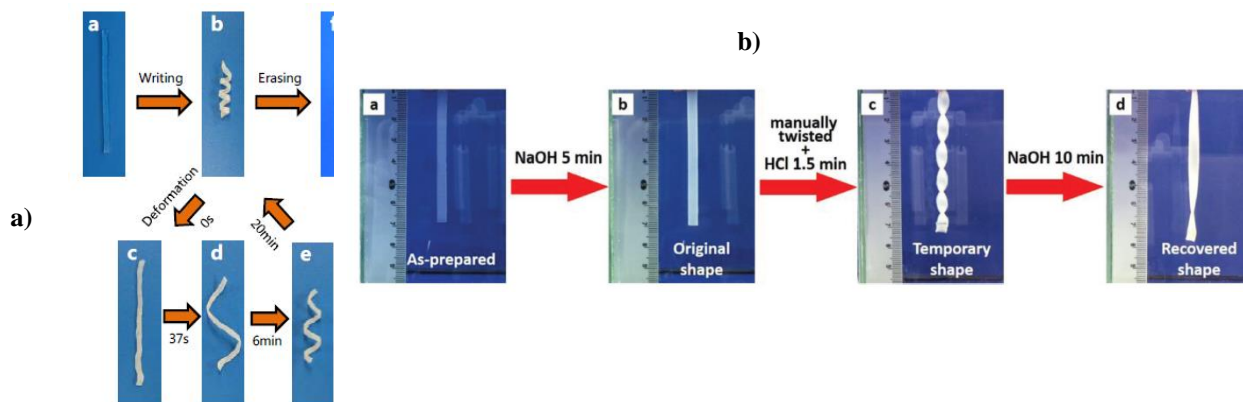

Figure 11. **a**) Shape transformation of MAPTAC-*co*-NaSS hydrogels from straight to spiral shape. Reproduced from ref. 116;**b**) Evolution of the shape of as-prepared P(MAPTAC-*co*-NaSS-*co*-MAA) in 0.1M NaOH and 0.1M HCl. Photos of MAPTAC-*co*-NaSS-*co*-MAA hydrogels: a) as-prepared; b) original shape after treatment by 0.1M NaOH; c) fixed temporary shape in 0.1M HCl; d) recovered shape in 0.1M NaOH. Reproduced from ref. 92

The main difference between two amphoteric hydrogels is that MAPTAC-*co*-NaSS-*co*-MAA contains additional methacrylic acid (MAA) that is sensitive to pH. In the first case an as-prepared hydrogel MAPTAC-*co*-NaSS was deformed from initial straight shape to a helix shape and immersed in water memorizing the spiral shape. After deformation of spiral shape to straight shape the gel sample automatically returns to spiral shape. The memorized spiral shape is erased and gel recovers to its initial straight shape in 0.5M NaCl solution. This process can be repeated many times. In author's opinion using the ion complexation process in water it is possible to "write" any desired shape with the help of polyampholyte hydrogels and to "erase" the shape by destruction of polyionic complexes in NaCl. In case of MAPTAC-*co*-NaSS-*co*-MAA [92] the helical shape of gel was manually formed in 0.1M NaOH and fixed in 0.1M HCl. Immersing of

_____

gel sample again in 0.1M NaOH solution recovered its original straight shape. The "straight-helix" transformation was repeated at least for 10 times by alternate immersing in NaOH and HCl solutions. Taking into account that MAPTAC-*co*-NaSS itself exhibit shape memory ability one can suppose that the presence of pH sensitive MAA units in MAPTAC-*co*-NaSS-*co*-MAA synergistically enhance the shape memorizing process.

4. Catalytic properties of metal nanoparticles and enzymes immobilized within macroporous polyampholyte cryogels

Macroporous amphoteric cryogels are formed in the moderately frozen solutions of monomeric and polymeric precursors below the freezing point of solvent [93-97]. The cryogel has a sponge-like morphology and pore size of several tens and hundreds of microns that allows free flow of fluid through microporous channels. Modified polyacrylamide-based amphoteric cryogels were tested as catalytically active substances in transesterification of glyceryl oleate [98]. The highest conversion of glyceride was equal to 88.4% and the yield of methyl oleate is about 64.%. Macroporous amphoteric cryogels containing metal nanoparticles in pores can be used for catalytic reactions as batch-type and continuous-flow-type reactors. The idea of the use of macroporous and supermacroporous cryogels as flow-through catalytic reactors was for the first time formulated in [55] and experimentally realized in [99-102] for hydrogenation of nitroaromatic compounds (Fig. 12).
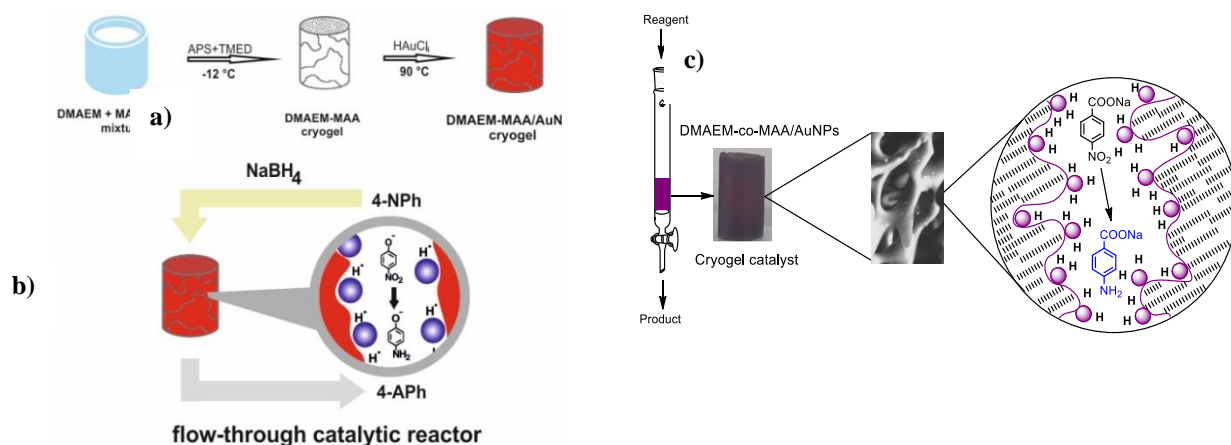

Figure 12.**a**) Immobilization of gold nanoparticles (AuNPs) within macroporous amphoteric cryogel derived from N,N-dimethylaminoethylmetacrylate and methacrylic acid (DMAEM-*co*-MAA);**b**) Hydrogenation of 4-NP over DMAEM-*co*-MAA/AuNPs catalyst;**c**) Hydrogenation of *p*-NBA over DMAEM-*co*-MAA/AuNPs catalyst (Compiled from Ref. 100)

Hydrogenation of 4-nitrophenol (4-NP) to 4-aminophenol (4-AP) [103] and reduction of *p*-nitrobenzoic acid (*p*-NBA) to *p*-aminobenzoic acid (*p*-ABA) [104] was performed with the help of DMAEM-*co*-MAA cryogel with immobilized gold (AuNPs) and palladium (PdNPs) nanoparticles.The kinetic parameters, turnover number (TON), turnover frequency (TOF) and activation energy for hydrogenation of 4-NP and*p*-NBA were determined.

Recently [105] we have demonstrated the oxidation of ethanol,n-propanoland iso-propanol by catalase encapsulated within amphoteric cryogel matrix (Fig. 13). Chromatographic and chromatomass spectroscopic analysis show that at optimal conditions (temperature range 5-20 °C and volume ratio of substrate to reducing agent is 1:1) ethanol is converted to acetaldehyde with

high yields ≈ 99 %, n-propanol and iso-propanol – to propionaldehyde and acetone, respectively, with 95-97% yields at room temperature and atmospheric pressure.
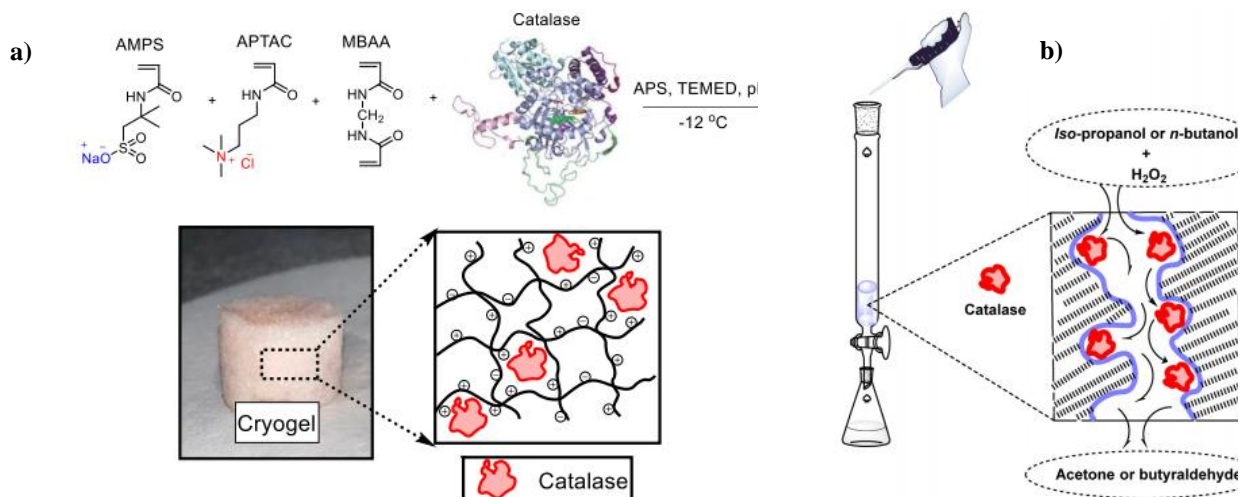

Figure 13. a) Encapsulation of catalase within the monolithic p(APTAC-co-AMPS) cryogel matrix; **b**) Oxidation of alcohols to corresponding key products. Reproduced from Ref. 105;

5. Research perspectives of polyampholytes

The statistical data surveyed during the half century demonstrate that the number of publications and citations on polyampholytes steadily increases starting from1970s (Figure 14).
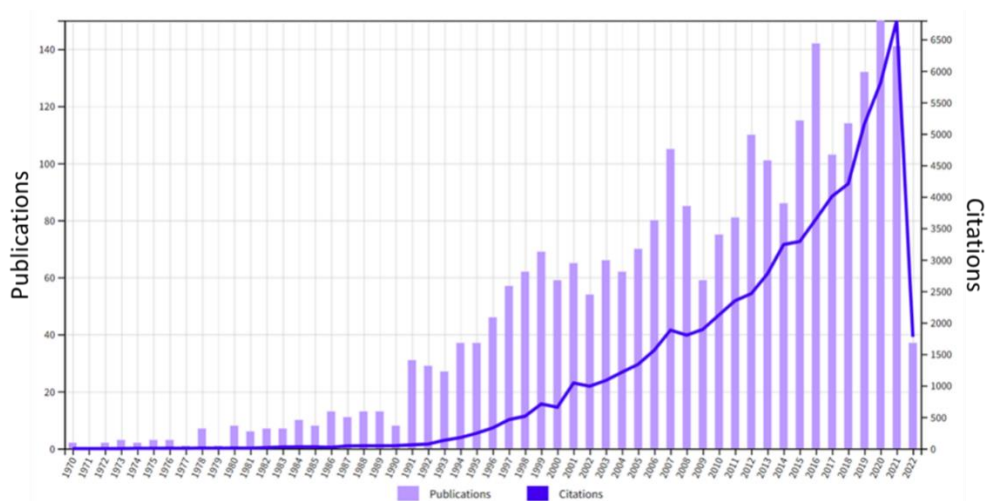

Figure 14. Progress of publications and citations on polyampholytes according to the data of Web of Knowledge, generated using the keywords "polyampholyte" or "amphoteric polymer"

A further potential development in polyampholytes includes intrinsically disordered proteins (IDPs), which belong to strong polyampholytes, polypeptide-based polyampholytes and polyampholytic ionic liquids. Future possibilities regarding polyampholytes may be related to semi-natural polyampholytes that can be prepared by functionalization of natural polysaccharides,such as chitosan, cellulose, starch, gellan, and alginic acid, among others,

through introduction of either carboxylic (sulfo, phospho) or amine (ammonium, phosphonium) groups, or both, into the macromolecular chains. Synthetic, natural, semi-natural polyampholytes, ampholytic polypeptides, and IDPs might be considered as a single focal point, indicating the multidisciplinary character of research into amphoteric macromolecules.Understanding the fundamental relationships between themicrostructures and properties of synthetic, natural and semi-natural polyampholytes is expected to attract the attention of researchersin advanced theoretical and experimental investigations.

References

[1] Alfrey T, Morawetz H, Fitzgerald EB, Fuoss RM. Synthetic electrical analog of proteins. *J Am Chem Soc* 1950; 72: 1864-1864. doi:10.1021/ja01160a532.

[2] Alfrey T, Morawetz H. Amphoteric Polyelectrolytes. I. 2-Vinylpyridine—Methacrylic Acid Copolymers. *J Am Chem Soc* 1952; 74:436-438. doi:10.1021/ja01122a046

[3] Alfrey T, Fuoss RM, Morawetz H, Pinner H. Amphoteric polyelectrolytes. II. Copolymers of methacrylic acid and diethylaminoethyl methacrylate. *J Am Chem Soc* 1952; 74:438-441. doi:10.1021/ja01122a047.

[4] Katchalsky A, Gillis J. Potentiometric behavior of polyampholytes. Copolymers of 2-vinylpyridine-acrylic acid. *Recl Trav Chim-Pays Bas* 1949; 68:871-877.

[5] Katchalsky A, Miller IR. Polyampholytes. *J Polym Sci.* 1954; 13:57-68. doi:10.1002/pol.1954.120136805.

[6] Mazur J, Silberberg A, Katchalsky A. Potentiometric behavior of polyampholytes. *J Polym Sci* 1959; 35:43-70.

[7] Ehrlich G, Doty P. Macro-ions. III. The solution behavior of a polymeric ampholyte. *J Am Chem Soc* 1954; 76:3764-3777.

[8] Ciferri A, Kudaibergenov SE. Natural and synthetic polyampholytes. 1. Theory and basic structure. *Makromol Rapid Commun* 2007; 28:1953-1968.

[9] Kudaibergenov SE, Ciferri A. Natural and synthetic polyampholytes. 2. Functions and applications. *Makromol Rapid Commun* 2007; 28:1969-1986.

[10] Edwards SF, King PR, Pincus P. Phase change in polyampholytes. *Ferroelectrics* (GB) 1980; 30:3-6.

[11] Roy SG, De P. pH responsive polymers with amino acids in the side chains and their potential applications. *J Appl Polym Sci* 2014; 131:41084(p.1-12).

[12] Mori H, Endo T. Amino-acid-based block copolymers by RAFT polymerization. *Macromol Rapid Commun* 2012; 33: 1090-1107. doi: 10.1002/marc.201100887.

[13] Kruse J-H, Biehl P, Schacher FH. Different routes to ampholytic polydehydroalanine: Orthogonal versus simultaneous deprotection. *Macromol Rapid Commun* 2019; 10:1800857(p.1-6). Doi: 10.1002/marc.201800857

[14] Evans AC, Skey J, Wright M, Qu W, Ondeck C, Longbottom DA, O'Reilly RK. Functional and tunable amino acid polymers prepared by RAFT polymerization. *J Polym Sci. Part A Polym Chem* 2009; 47:6814-6826

[15] Shirbin SJ, Lam SJ, Chan NJA, Ozmen MM, Fu Q, O'Brien-Simpson N, Reynolds EC, Qiao GG Polypeptide-based macroporous cryogels with inherent antimicrobial properties: The importance of a macroporous structure. *ACS Macro* 2016; 5:552-557. DOI: 10.1021/acsmacrole.6b00174.

[16] Tao Y, Wang S, Zhang X, Wang Z, Tao Y, Wang X. Synthesis and properties of alternating polypeptoids and polyampholytes as protein-resistant polymers. *Biomacromolecules*, 2018; 19:936-42. DOI:10.1021/acs.biomac.7b01719.

[17] Janciauskaite U, Visnevskij C, Radzevicius K, Makuska R. Polyampholytes from natural building blocks: synthesis and properties of chitosan-O-alginate copolymers. *Chemija* 2009; 20:128-135.

[18] Lin M, Liu H, Deng J, An R, Shen M, Li Y, Zhang X. Carboxymethyl chitosan as a polyampholyte mediating intrafibrillar mineralization of collage via collagen/ACP self-assembly. *J Mater Sci Technol* 2019; 33:1894-1905.

[19] Higgs PG, Joanny JF. Theory of polyampholyte solutions. *J Chem Phys* 1991; 94:1543-1554.

[20] Zheng G-Z, Meshitsuka G, Ishizu A. Inter- and intramolecular ionic interactions of polyampholyte: Carboxymethyl-2-diethylaminoethylcellulose.*PolymIntern* 1994;34:241-248.

[21] Patrickios CS, Hertler WR, Abbot NL, Hatton TA. Diblock, ABC triblock, and random methacrylic polyampholytes: Synthesis by group transfer polymerization and solution behavior. *Macromolecules* 1994; 27:930–937.

[22] Asayama S, Nogawa M, Takei Y, Akaike T, Maruyama A. Synthesis of novel polyampholyte comb-type copolymers consisting of a poly(L-lysine) backbone and hyaluronic acid side chains for a DNA carrier. *Bioconjug Chem* 1998; 9:476–481.

[23] Kudaibergenov SE, Shayakhmetov ShSh, Rafikov SR, Bekturov EA. On hydrodynamic properties of amphoteric copolymers. *Dokl Acad Nauk USSR* 1979; 246:147-149.

[24] Kudaibergenov SE, Shayakhmetov ShSh, Bekturov EA. Hydrodynamic properties of polyampholytes based on 1,2,5-trimethyl-4-vinylethynylpiperidinol-4 and acrylic acid. *Vysokomol Soedin* 1980; B22:91-95.

[24] Ibraeva ZhE, Hahn M, Jaeger W, Bimendina LA, Kudaibergenov SE. Solution properties and complexation of polyampholytes based on N,N-dimethyldiallylammonium chloride and maleic acid or alkyl (aryl) derivatives of maleamic acids. *Macromol Chem Phys* 2004; 205:2464–2472.

[26] Ibraeva ZhE, Hahn M, Jaeger W, Laschewsky A, Bimendina LA, Kudaibergenov SE. Swelling behavior and complex formation ability of ternary amphoteric gels based on allylamine derivatives and maleic acid. *Macromol Mater Eng* 2005; 290:769–777.

[27] Kudaibergenov SE, Bekturov EA. Influence of the coil-globule confromational transition in polyampholytes on sorption and desorption of polyelectrolytes and human serum albumin. *Vysokomol Soedin Ser A* 1989;31:2614-2617.

[28] Kudaibergenov SE, Tatykhanova GS, Klivenko AN. Complexation of macroporous amphoteric cryogels based on N,N-dimethylaminoethylmethacrylate and methacrylic acid with dyes, surfactant, and protein. *J Appl Polym Sci* 2016;133:43784-1–43784-9.

[29] Kudaibergenov SE, Nuraje N. Intra- and interpolyelectrolyte complexes of polyampholytes. *Polymers* 2018; 10:1146(p.1-34).

[30] Kudaibergenov SE. Physico-chemical, complexation and catalytic properties of polyampholyte cryogels. *Gels* 2019; 5:8(p.1-22).

[31] Koetz J, Hahn M, Philipp B, Bekturov EA, Kudaibergenov SE. Inter- and intramolecular interactions in polyelectrolyte complex formation with polyampholytes. *Makromol Chem* 1993;194:397-409.

[32] Rogacheva VB, Panova TV, Bykova EV, Zezin AB, Joosten J, Brekman J. Interaction of ampholyte dendrimers with network polycations and polyanions. *Polymer Science Series A* 2009;51:242-249.

[33] Varoqui R, Tran Q, Pefferkorn E. Polycation-polyanion complexes in the linear diblock copolymer of poly(styrene sulfonate)/poly(2-vinylpyridinium) salt. *Macromolecules* 1979;12:831-835.

[34] KudaibergenovSE, Frolova VA, Bekturov EA., Rafikov SR. Interpolymer reactions with participation of blockpolyampholyte poly(1-methyl-4-vinylpyridinium chloride)-poly(methacrylic acid). *DoklAkadNauk USSR* 1990;311:396-398.

[35] Bekturov EA, Kudaibergenov SE, Frolova VA, Schultz RS, Zoller J. Conformational and complex formation ability of poly(methacrylic acid)-*block*-poly(1-methyl-4-vinylpyridinium chloride). *Makromol Chem* 1990;191:1329-1333.

[36] Koetz J, Philipp B, Schultz W, Kudaibergenov SE, Bekturov EA. Amphoteric character of polyelectrolyte complex particles as revealed by isotachophoresis and viscometry. *ColloidPolymSci* 1988;266:906-912.

[37] Katchalsky A, Spangler R. Dynamics of membrane processes. *QuartRevBiophys* 1968;1:137-175.

[38] Wang H, Li W, Wang Z. Studies on chitosan and poly(acrylic acid) interpolymer complex. 1. Preparation, structure, pH-sensitivity, and salt sensitivity of complex forming-poly(acrylic acid): chitosan semi-interpenetrating polymer network. *J Appl Polym Sci* 1997;65:1445-1450.

[39] Madinya JJ, Chang LW, Perry SL, Sing CE. Sequence-dependent self-coacervation in high charge-density polyampholytes. *Mol Syst Des Eng* 2020; 5:632-44.

[40] Lin YH, Brady JP, Chan HS, Ghosh K. A unified analytical theory on heteropolymers for sequence-specific phase behaviors of polyelectrolytes and polyampholytes. *J Chem Phys* 2020; 152:045102.

[41] Kudaibergenov SE. *Polyampholytes: Synthesis, Characterization and Application*. New York: Kluwer Academic/Plenum Publishers; 2002.

[42] Lowe AB, McCormick CL. Synthesis and solution properties of zwitterionic polymers. *Chem Rev* 2002; 102:4177-4190. doi:10.1021/cr020371t.

[43] Kudaibergenov SE. Recent advances in studying of synthetic polyampholytes in solutions. *Adv Polym Sci* 1999; 144:115-197.

[44] Kudaibergenov SE. Polyampholytes: past, present, perspectives. Almaty: cop-almaty.kz; 2021.

[45] Kudaibergenov S, Jaeger W, Laschewsky A. Polymeric betaines: Synthesis, characterization and application. *Adv Polym Sci* 2006; 201:157-224.

[46] Laschewsky A. Structures and synthesis of zwitterionic polymers. *Polymers* 2014; 6:1544-1601. doi:10.3390/polym6051544.

[47] Singh Paresh K, Singh Vinay K, Singh M. Zwitterionic polyelectrolytes: A Review. *e-Polymers* 2007; 7:1-34. DOI:10.1515/epoly.2007.7.1.335

[48] He XY, Zhou WR, Xu XJ, Yang W. Preparation and application of zwitterionic polymers. *Progress in Chemistry* 2013; 25:1023-1030.

[49] Taylor ME, Lounder SJ, Asetkin A, Panzer MJ. Synthesis and self-assembly of fully zwitterionic triblock copolymers. *ACS Materials* 2020; 2:261-65.

[50] Liu Hongyan ZJ. Biological applications of zwitterionic polymers. *Progress in Chemistry* 2012; 24:2187-2197.

[51] Ohno H, Yoshizawa-Fujita M, Ogihara W. *Electrochemical Aspects of Ionic Liquids*, in: Ohno H, editor, John Hoboken, New Jersey: Wiley & Sons, Inc; 2011, p.433-439.

[52] Fouillet CCJ, Greaves TL, Quinn JF, Davis TP, Adamcik T, Sani MA, Separovic F, Drummond CJ, Mezzenga R. Copolyampholytes produced from RAFT polymerization of protic ionic liquids. *Macromolecules* 2017;50:8965-78.

[53] Ramos DP, Sarjinsky S, Alizadehgiashi M, Moebus J, Kumacheva E. Polyelectrolyte vs polyampholyte behavior of composite chitosan/gelatin films *ACS Omega* 2019; 4:8795-8803.

[54] Dobrynin AV, Colby RH, Rubinstein M. Polyampholytes. *J Polym Sci Part B Polym Phys* 2004; 42:3513-3538.

[55] Kudaibergenov S, Nueraje N, Khutoryanskiy V. Amphoteric nano-, micro-, and macrogels, membranes, and thin films. *Soft Matter* 2012; 8:9302-9321. Doi:10.1039/C2SM25766A.

[56] Kudaibergenov SE. Macromolecular complexes of polyampholytes. *Pure Appl Chem* 2020; 92:839-858.

[57] Kudaibergenov S, Okay O. Behaviors of quenched polyampholytes in solution and gel state. *Polym Adv Technol* 2021; 32:2639-2654.

[58] Kudaibergenov SE. Synthetic and natural polyampholytes: Structural and behavioral similarity. *Polym Adv Technol* 2021; 32:906-918.

[59] Kudaibergenov SE. Advances in synthetic polyampholytes for biotechnology and medicine. *Review Journal of Chemistry* 2020; 10:12-39.

[60] Ali SA, Rasheed A, Wazeer M I. Synthesis and solution properties of a quaternary ammonium polyampholyte. *Polymer* 1999; 40**:**2439-2446.

[61] Puglia G P. Solution properties of polymers. 1. A measure of the ionic strength dependence of polyampholyte solutions. *Annu Tech Conf Soc Plast Eng* 1992:2280-2281.

[62] Bekturov EA, Kudaibergenov SE. Solution properties of (co)polyvinylethynylpiperidols. *J Macromol Sci Phys* 1986; B25:133-151.

[63] Bekturov EA, Kudaibergenov SE. Physico-chemical properties of polyampholytes. *Makromol Chem Macromol Symp* 1989; 26:281-295.

[64] Bekturov EA, Kudaibergenov SE, Rafikov SR. Synthetic polymeric ampholytes in solution. *J Macromol Sci Rev* 1990; C30:233-303.

[65] Bekturov EA, Kudaibergenov SE, Rafikov SR. Solution properties and complexation of amphoteric polyelectrolytes. *Uspekhi Khimii* 1991; 60:835-851.

[66] Candau F, Joanny JF. Polyampholytes (Properties in aqueous solution), In: Salamone JC, editor. *Polymeric Materials Encyclopedia*, New York: CRC Press; 1996, p.5476-5488

[67] Kathmann EE, McCormick CL. Polyampholytes, in: Salamone JC editor. *Polymeric Materials Encyclopedia*, New York: CRC Press Boca Raton; 1996, p. 5462-5476.

[68] Zhang LM, Tan YB, Li ZM. New amphoteric graft copolymers of sodium carboxymethyl cellulose with acrylamide and dimethylaminoethyl methacrylate: Aqueous solution properties. *J Appl Polym Sci* 2000; 76:305-310.

[69] Nisato G, Munch JP, Candau SJ. Swelling, structure, and elasticity of polyampholyte hydrogels. *Langmuir* 1999; 15:4236-4244.

[70] Toleutay G, Shakhvorostov A, Kabdrakhmanova S, Kudaibergenov S. Solution behavior of quenched or strongly charged polyampholytes in aqueous-salt solutions. *Bulletin of Karaganda University. Ser. Chem* 2019; 2(94):35-44.

[71] Kawata Y, Kozuka S, Yusa S. Thermo-responsive behavior of amphoteric diblock copolymers bearing sulfonate and quaternary amino pendant groups. *Langmuir* 2019; 35:1458-1464. Doi: 10.1021/acs.langmuir.8b01684.

[72] Sharker KK, Ohara Y, Shigeta Y, Ozoe S, Yusa S-I. Upper critical solution temperature (UCST) behavior of polystyrene-based polyampholytes in aqueous solution. *Polymers* 2019; 11:265.

[73] Annaka M, Tanaka T. Multiple phases of polymer gels. *Nature* 1992; 355:430-432.

[74] Shibayama M, Tanaka T. Volume phase transition and related phenomena of polymer gels. *Adv Polym Sci* 1992; 109:1-62.

[75] Noh JG, Sung YJ, Geckeler KE, Kudaibergenov SE.Synthesis, characterization, and stimuli-sensitive properties of novel polycarbobetaines. *Polymer* 2005; 46:2183-2190.

[76] Kudaibergenov SE, Bimendina LA, Yashkarova MG. Preparation and characterization of novel polymeric betaines based on aminocrotonates. *J Macromol Sci Pure Appl Chem* 2007; 44: 899-913.

[77] Su E, Okay O. Polyampholyte hydrogels formed via electrostataic and hydrophobic interactions. *Eur Polym J* 2017; 88:191-204.

[78] Suleimenov IE, Sigitov VB, Kudaibergenov SE. Influence of the electric current on the swelling of polyampholyte hydrogels, *Polymer Sci Ser B* 1998; 40:67-70.

[79] Kudaibergenov SE, Sigitov VB, Didukh AG, Bekturov EA, Suleimenov IE. Behavior of polyelectrolyte gels under the influence of d.c. electric and magnetic fields. *Polym Adv Technol* 2000; 11:805-809.

[80] Sigitov VB, Kudaibergenov SE, Suleimenov IE. Oscillation-relaxation properties of polyampholyte hydrogels, *Dokl Akad Nauk SSSR Ser Khim* 1998; 361:654-656.

[81] Kudaibergenov SE, Sigitov VB, Didukh AG, Moldakarimov SB. Measurement of pH gradient of polyampholyte gels under the action of DC electric field. *Polym Prepr* 2000; 41:724-725.

[82] Kudaibergenov SE, Didukh AG, Moldakarimov SB. Structure of polyampholyte gels and their behavior with respect to applied external DC electric field. In: Bohidar HB, Dubin P, Osada Y. editors; *Polymer Gels. Fundamentals and ApplicationsACS Symp. Ser. 833*, Washington DC: ACS; 2002, p.149-162.

[83] Chiou S-H, Wu S-H. Evaluation of commonly used electrophoretic methods for analysis of proteins and peptides and the application in biotechnology. *Analytica Chimica Acta* 1999; 383:47-60.

[84] Ogawa Y, Ogawa K, Wang B, Kokufuta E. A biochemo-mechanical system consisting of polyampholyte gels with coimmobilized glucose oxidase and urease. *Langmuir* 2001; 17:2670-2674.

[85] Goh KB, Li H, Lam KY. Modeling the impact of pH- and oxygen-coupled stimuli on osmotic pressure and electrical potentila repsonses of hemoglobin-loaded polyampholyte hydrogel. *ACS Appl Bio Mater* 2018; 1:318-27.

[86] Sun TL, Kurokawa T, Kuroda S, Ihsan AB, Akasaki T, Sato K, Haque MdA, Nakajima T, GongJP. Physical hydrogels composed of polyampholytes demonstrate high toughness and viscoselasticity. *NatureMaterials*2013; 12:932-937.

[87] Sun TL, Luo F, Kurokawa T, Karobi SN, Nakajima T, Gong JP. Molecular structure of self-healing polyampholyte hydrogels analyzed from tensile behaviors. *Soft Matter* 2015; 11:9355-9366. Doi: 10.1002/pi.4907.

[88] Sun TL, Luo F, Hong W, Cui K, Huang Y, Zang HJ, King DR, Kurokawa T, Nakajima T, Gong JP. Bulk energy dissipation mechanism for the fracture of tough and self-healing hydrogels. *Macromolecules* 2017; 50: 2923-2931. doi: 10.1021/acs.macromol.7b00162.

[89] Ihsan AB, Sun TL, Kurokawa T, Karobi SN, Nakajima T, Nonoyama T, Roy CK, Luo F, Gong JP. Self-healing behaviors of tough polyampholyte hydrogels. *Macromolecules* 2016; 49:4245-52. Doi: 10.1021/acs.macromol.6b00437.

[90] Ekici S. Thermoresponsive crosslinked amphoteric terpolymers (CATs): Effect of salt concentration on porosity, thermal, mechanical, electrical conduction and imprintable properties. *J Mater Sci Nanotechnol* 2019; 7:1-11.

[91] Toleutay G, Su E, Kudaibergenov S, Okay O. Highly stretchable and thermally healable polyampholyte hydrogels via hydrophobic modification. *Colloid Polym Sci* 2020; 298:273-284.

[92] Zhang Y, Liao J, Sun W, Tong Z. Polyampholyte hydrogels with pH modulated shape memory and spontaneous actuation. *Adv Funct Mater* 2018; 28:1707245(p1-9). DOI: 10.1002/adfm.201707245.

[93] Lozinsky VI. Cryogels on the basis of natural and synthetic polymers: Preparation, properties and areas of implementation. *Russ Chem Rev* 2002; 71:489-511. http://dx.doi.org/10.1070/RC2002v071n06ABEH000720.

[94] Lozinsky VI. Cryostructuring of polymeric systems. 50. Cryogels and cryotropic gel-formation: Terms and definitions. *Gels* 2018; 4:77, doi:10.3390/gels4030077.

[95] Lozinsky VI. A brief history of polymeric cryogels. *Adv Polym Sci* 2014; 263:1-48. https://doi.org/10.1007/978-3-319-05846-7_1.

[96] Lozinsky VI, Okay O. Basic principles of cryotropic gelation. *Adv Polym Sci* 2014, 263:48-102. https://doi.org/10.1007/978-3-319-05846-7_2.

[97] Okay O, Lozinsky VI. Synthesis, structure-property relationships of cryogels. *Adv Polym Sci* 2014; 263: 103-157. https://doi.org/10.1007/978-3-319-05846-7_3.

[98] Yang C, Liu GF, Zhou XL, Liu YR, Wang J, Tian LL, Hu XY, Wang YY. Polyacrylamide based cryogels as catalysts for biodiesel. *Catalysis Letters* 2015; 145:1778-1783.

[99] Kudaibergenov SE. Physicochemical, complexation and catalytic properties of polyampholyte cryogels.

Gels 2019; 5:8. Doi: 10.3390/gels5010008.

[100] Kudaibergenov SE, Tatykhanova GS, Selenova BS. Polymer protected and gel immobilized gold and silver nanoparticles in catalysis. *J Inorg Organomet Polym Mater* 2016; 26:1198-1211. doi 10.1007/s10904-016-0373-z.

[101] Dzhardimalieva GI, Zharmagambetova AK, Kudaibergenov SE, Uflyand IE. Polymer-immobilized clusters and metal nanoparticles in catalysis. *Kinetics and Catalysis* 2020; 61:198-223.

[102] Dolya NA, Kudaibergenov SE. Catalysis by thermoresponsive polymers. In: Khutoryanskiy VV, Georgiou T. editors; *Temperature-Responsive Polymers: Chemistry, Properties and Applications*; World Scientific Co Inc: Singapore, 2018, p.357–377.

[103] Klivenko AN, Yergazieva E, Kudaibergenov SE. Gold nanoparticles stabilized by amphoteric cryogel-perspective flow-through catalytic reactor for oxidation and reduction processes. *International Conference on Nanomaterials: Application & Properties (NAP)*, 2016:02NSA03-1-02NSA03-5. Doi: 10.1109/NAP.2016.7757304

[104] Kudaibergenov S, Dauletbekova M, Toleutay G, Kabdrakhmanova S, Seilkhanov T, Abdullin Kh. Hydrogenation of *p*-nitrobenzoic acid by gold and palladium nanoparticles immobilized within macroporous amphoteric cryogels in aqueous solution. *J Inorg Organomet Polym* 2018; 28:2427-2438. https://doi.org/10.1007/s10904-018-0930-8.ee.

[105] Smagulova I, Tatykhanova G, Shakhvorostov A, Akbayeva D, Kudaibergenov S. Oxidation of *iso*-propanol and *n*-butanol by catalase encapsulated within macroporous polyampholytic cryogel matrix. *Polym Adv Technol* 2021; 32:3817-26