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Metal Complexes of Amphoteric Cryogels Based on Allylamine and Methacrylic Acid

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Summary: A series of macroporous amphoteric cryogels based on allylamine, methacrylic acid and acrylamide were synthesized by radical copolymerization of monomers in cryoconditions. The average molar composition of amphoteric cryogels was found from the potentiometric and conductimetric titration curves. The morphology of cryogels was evaluated by SEM. Cryogels are highly elastic and have continuous macroporous structure with 50–200 μm pores. The values of the isoelectric pH determined from the swelling experiments arranged between 3.5 and 4.3. Complexation of amphoteric with transition metal ions was studied. Cryogels with adsorbed copper, nickel, or cobalt ions have an intense colour due to formation of coordination and ionic bonds between metal ions and amine and/or carboxylic groups of cryogels. Metal ions entrapped within the pores of cryogels were reduced by treating aqueous solution of $NaBH_4$. Formation of micron sized metals on the inner surface of cryogels was observed by SEM.

Keywords: amphoteric cryogels; catalysis; macroporous polymers; metal-polymer complexes; swelling

Cryogels are gel matrices that are formed in moderately frozen solutions of monomeric and polymeric precursors.[1–3] The morphology of macroporous cryogels is determined by solvent crystallization when the temperature is kept below the freezing point of solvent. According to cryopolymerization concept the freezing of the initially homogeneous system results in crystallization of pure solvent (water) and accumulation of monomers and initiators in unfrozen micro zones (so-called "cryoconcentration"). The polymerization reaction proceeds in this non-frozen part of the reaction mixture. Water crystals grow in the course of freezing and interconnections with other crystals take place until a continuous system of porous is formed. Thawing of the system leads to formation of a monolithic gel matrix with continuous macroporous channels filled with liquid solvent. The gel has a sponge-like morphology and pore size of 10–100 μm. A system of large interconnected pores is a main characteristic feature of cryogels. Thus cryogels have a combination of unique properties allowing to use them as monolith adsorbents directly operational with particulate containing fluxes. The pore system in such sponge-like gels ensures unhindered convectional transport of solutes within the cryogels, contrary to diffusion of solutes in traditional homophase gels. Amphoteric gels due to response to temperature, pH, ionic strength, water-organic solvent composition, electric field, etc. belong to "smart" materials.[4,5] Last years the most attention was paid to amphoteric nanogels[6–13] and microgels[14–18] however to our best knowledge amphoteric cryogels

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have been introduced only recently by our group[19].

Experimental Part

Materials and Methods

Monomers and initiators – acrylamide (AAm, 99% purity), allylamine (AA, 99% purity), methacrylic acid (MAA, 99% purity), N,N,N′,N′-tetramethylethylenediamine (TMED), ammonium persulfate (APS), and crosslinking agent N,N′-methylenbisacrylamide (MBAA) were purchased from Aldrich and used without further purification. Reagent-grade metal salts – nitrates of copper, nickel and cobalt (99% purity) were used. Distilled, bidistilled and deionized water was used for preparation of cryogels, washing of samples and preparation of aqueous solutions of metal salts and 0.1N HCl and NaOH. Potentiometric and conductimetric titrations were carried out on the pH/conductivity meter "Mettler Toledo MPC 227" (Switzerland) at room temperature.

Synthesis of Amphoteric Cryogels

Amphoteric cryogels were synthesized as following. Mixture of AA (33 mg or 0.057 mmol), MAA (50 mg or 0.057 mmol), AAm (330 mg or 5.65 mmol), MBAA (41 mg) corresponding to molar ratio of monomers [AA]:[MAA]:[AAm] = 10:10:80 mol.%/mol.%/mol.% was dissolved in 5 mL of deionized water, bubbled by nitrogen during 10 min and degassed under vacuum for about 5 min to eliminate the dissolved oxygen. After addition of 10 μL of TMED the solution was cooled in an ice bath for 4–5 min. Then 0.1 mL of aqueous solution of APS (10 wt.%) preliminary cooled in an ice bath for 4–5 min was added and the reaction mixture was stirred for 1 min. The total volume of reaction mixture was divided into 5 parts and each part containing 1 mL of reaction mixture was placed into the 5 plastic syringes with diameter 5 mm with closed outlet at the bottom. The solution in syringe was frozen within 10 min at −12 °C and was kept frozen during 48 h. After completion of the reaction the sample was thawed at room temperature. The prepared cryogel sample was washed out by distilled, bidistilled and deionized water during several days, after washed out by 25, 50, 75 and 96% ethanol to dehydrate then dried in air and finally in vacuum oven to constant mass at room temperature. Thus a series of amphoteric cryogels (ACG) with initial molar ratio of [AA]:[MAA]:[AAm] = 10:10:80; 20:20:60; 30:30:40; 40:40:20 and 50:50:0 mol.%/mol.%/mol.% were synthesized. They were abbreviated as ACG-118, ACG-226, ACG-334, ACG-442 and ACG-550 (Figure 1).

Potentiometric and Conductimetric Titrations

The fine powdered cryogel samples (10 mg) were put to 10 mL of distilled water, stirred 1 h and titrated by 0.1M HCl or KOH. Potentiometric and conductimetric titrations were carried out on the pH/conductivity meter "Mettler Toledo MFC 227" (Switzerland) at room temperature. For calculation of pK_b of allylamine groups the Henderson-Hasselbalch equation $pOH = pK_b + n \cdot \log(\beta/(1 - \beta)$ (where β is the degree of ionization of primary amine groups of cryogels, n is the specific parameter for polyelectrolytes) was used.

Swelling-Deswelling Measurements

The swelling capacity of cryogel samples as a function of pH was evaluated from the height measurements. For this dry gel sample with diameter 5 mm and height 10 mm was placed into a glass tube with diameter 10 mm the bottom of which was closed by Shott filter ended by valve. After passing of 100 mL of aqueous solutions through a gel sample with desired pH, that was adjusted by addition of 0,1N HCl or KOH to distilled water to avoid the influence of buffer on swelling behavior, its height was measured accurately three times and averaged. The ratio of L_s/L_0 (where L_s is the length of swollen gel, L_0 is the length of dry gel) on pH was plotted. The microstructure of cryogel samples was investigated with Scanning Electron

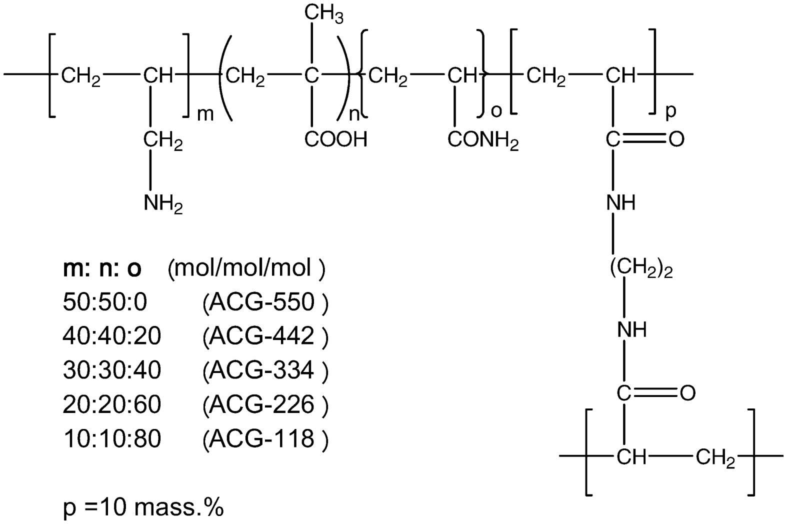

Figure 1.
Structural units of amphoteric cryogels derived from AA, MAA and AAm crosslinked by MBAA.

Microscopy (JEOL, JSM5800). The flow-rate of water passing through the cryogel samples was determined by the procedure described in[20].

Adsorption and Desorption of Metal Ions

Sorption of Cu^{2+}, Ni^{2+}, and Co^{2+} ions by ACG-334 sample was performed as follows: 10 mg of dry sample ($1.3 \cdot 10^{-4}$ mol) swollen in water up to 5 mm in diameter and 10 mm in height was placed inside of the glass tube with diameter 5 mm and height 50 mm, then 1 L aqueous solution of metal salts with concentrations either 10^{-5} or 10^{-3} mol $\cdot$ L^{-1} was passed through the sample during 1 or 2 days. The residual concentration of metal ions was determined with the help of ion-plasma coupled emission spectrometer "Optima 5100 DV" (Perkin Elmer, USA). Desorption of metal ions from the gel matrix was carried out by 0.1N HCl.

Results and Discussion

Potentiometric and Conductimetric Titration

Figure 2 shows the potentiometric and conductimetric titration curves of amphoteric cryogels.

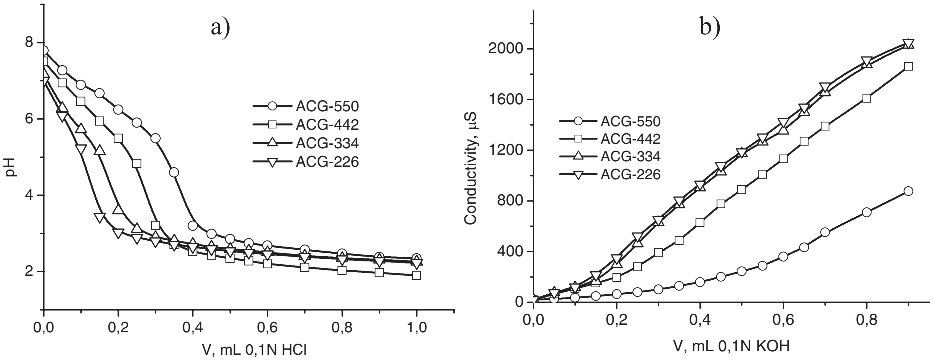

Figure 2.
Potentiometric (a) and conductimetric (b) titration curves of amphoteric cryogels by 0.1M HCl (a) and KOH (b).

Gradually shifting of inflection points to left side confirms overall decreasing of the content of carboxylic and amine groups in cryogels in the following order: ACG-550 > ACG-442 > ACG-334 > ACG-226 > ACG-118 and it is in good agreement with decreasing of the content of acid and base monomers in the feed. Compositions of amphoteric cryogels found from potentiometric and conductimetric titration curves together with ionization constant of allylamine groups (pK_b) and isoelectric pH (pH_{IEP}) are shown in Table 1. The average molar composition of amphoteric cryogels found from the potentiometric titration curves is equal to 55.5 ± 2.0 mol.% of COOH group and 44.5 ± 2.0 mol.% of NH_2 group while acid-base content found from the conductimetric titration curves corresponds to 63 ± 4 mol.% of COOH group and 37 ± 4 mol.% NH_2 group. In spite of inconsistency of potentiometric and conductimetric titration data they indicate that the composition of amphoteric cryogels is enriched by acidic monomer. This is probably due to relatively low activity of AA monomer in copolymerization reaction. According to literature data[21] copolymerization rate (r_1) of allyl-, diallylamine and other ammonium derivatives is in the range of 0,1 and 0,2 while r_2 for unsaturated carboxylic acids (acrylic or methacrylic acid) is 3–4 times higher.

It should also be taken into account that AA and MAA in aqueous solution forms so called inner salts that is moderately active than that of precursors. Due to low activity of AA the composition of amphoteric cryogels is probably enriched by methacrylic acid.

It is commonly accepted[4] that the position of the IEP depends on the acid/base ratio. For example, enrichment of the composition of polyampholyte by acidic monomer shifts the position of the IEP to acidic region and *vice versa*. For all ACG samples the average value of the pH_{IEP} is replaced in acidic region and equal to 3.9 ± 0.4 (pH unit) due to excess of carboxylic groups. The values of pK_b for ACG-550, ACG-442 ACG-334 and ACG-226 determined from Henderson-Hasselbalch equation are equal to 5.44; 5.25; 5.78 and 5.62 respectively. Such values of pK_b suggest effective complexation of allylamine groups with transition metal ions and formation of coordination bonds.

Morphology of Cryogels

Cross- and longitudinal sections of dry ACG-118 show sponge-like porous structure with pore size ranging from 50 to 200 μm (Figure 3a) and the interconnected channels (Figure 3b).

As seen from Figure 3c,d, the ACG-442 containing 40 mol.% of both acidic and basic monomers has more closed pores than that of ACG-118 which composed of 10 mol.% of acidic and 10 mol.% of basic monomers. In the case of ACG-442 the electrostatic attraction between acidic and basic groups makes the pores of cryogels more closed.

Dynamics of Swelling and Water Flux

Dynamics of swelling of ACG samples are shown in Figure 4. In comparison with amphoteric microgels, swelling kinetics of macroporous amphoteric gels is consider-

Table 1.
Composition, ionization constant of allylamine groups (pK_b) and the isoelectric pH (pH_{IEP}) of cryogels.

Cryogels	$-NH_2$ groups		$-COOH$ groups		pK_b	pH_{IEP}
	mol.%		mol.%			
	Potentiometric titration	Conductimetric titration	Potentiometric titration	Conductimetric titration		
ACG-550	46.7	41.0	53.3	59.0	5.44	4.0
ACG-442	42.4	37.0	57.6	63.0	5.25	4.1
ACG-334	43.3	33.3	56.7	66.6	5.78	4.2
ACG-226	–	41.4	–	58.6	5.62	4.3
ACG-118	–	38.5	–	61.5	–	3.5

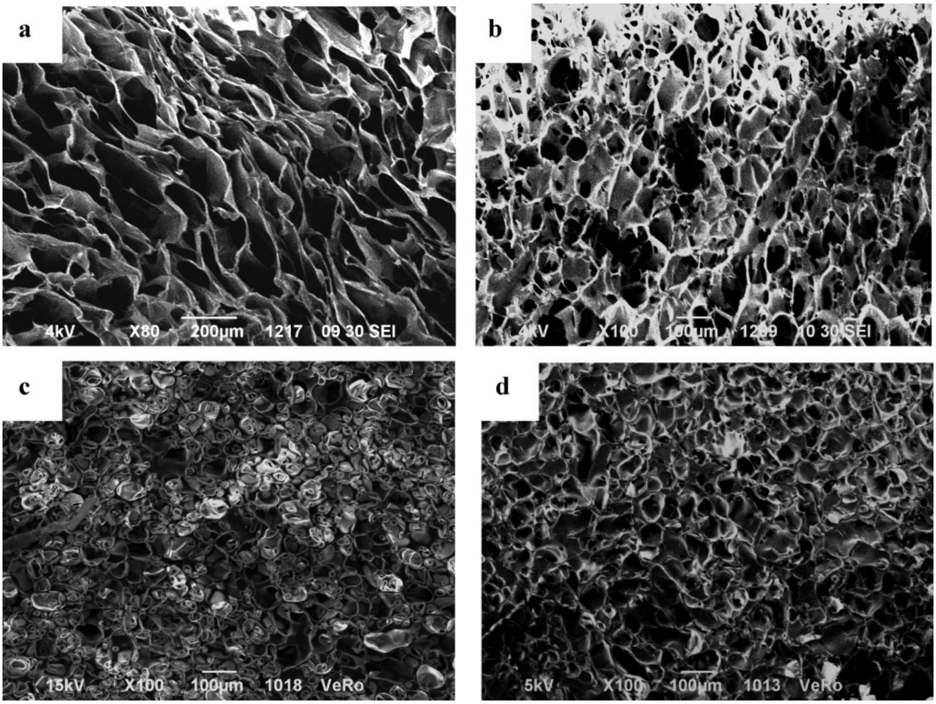

Figure 3.
SEM images of cross (a, c, d) and longitudinal (b) sections of ACG-118 (a, b) and ACG-442 (c, d).

ably fast due to porous structure. Increasing of the swelling rate in the order: ACG-118 > ACG-226 > ACG-334 > ACG-550 is in good agreement with dynamics of water flux through the cryogel samples (diameter 5 mm and height 10 mm) that are summarized in Table 2.

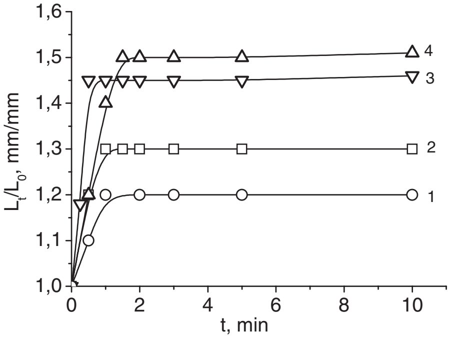

Figure 4.
Swelling rate of amphoteric cryogels ACG-550 (curve 1), ACG-334 (curve 2), ACG-226 (curve 3) and ACG-118 (curve 4) in water.

It is seen that amphoteric cryogels swell fast during 0.5–2 min. Increasing of the content of acidic and basic monomers in a series of ACGs leads to progressive decreasing of swelling degree and water flux probably due to formation of ionic contacts between oppositely charged groups of macromolecules leading to decrease of pore size of cryogels. For ACG-118 samples crosslinked at different concentration of [MBAA] = 10, 6 and 4 wt. % the exponential increase of water flowing is observed that reveals increasing of pore size of

Table 2.
Dynamics of water flow-rate through amphoteric cryogels.

Amphoteric cryogels	Water flux mL/min
ACG-118	1.20 ± 0.1
ACG-226	0.35 ± 0.05
ACG-334	0.18 ± 0.05
ACG-442	0.05 ± 0.01
ACG-550	0.02 ± 0.01

cryogels with decreasing of crosslinker concentrations.

Isoelectric Points (IEPs) of Amphoteric Cryogels

One of the specific features of linear and crosslinked polyampholytes is the existence of so-called isoelectric points (IEPs) where intra- and intermolecular attractions of opposite fixed charges lead to pseudo-neutral behavior and compact structure of amphoteric macromolecules[4]. Figure 5 shows the swelling and deswelling of amphoteric cryogels as a function of pH. It is seen that the deswelling of amphoteric cryogels is minimal at the IEPs. The isoelectric pH (pH_{IEP}) of amphoteric cryogels found from the swelling-deswelling measurements arranges between 3.5 and 4.3 (see Table 1).

The influence of ionic strength on the swelling behavior of amphoteric cryogels was examined (Figure 6 and 7). In acidic and alkaline media, amphoteric hydrogels behave like ordinary polyelectrolyte gels, e.g. the swelling degree falls with growth of the ionic strength. An opposite, so-called "antipolyelectrolyte", effect is observed at the IEP, the amphoteric cryogels swell with increasing of the ionic strength. This is due to the fact that the low molecular weight electrolyte screens the opposite charges on macromolecules unfolding the compact conformation. This behavior together with our previous results[22] entirely confirm the prediction of Ehrlich and Doty[23] and theoretical conclusions of authors[24] on the unfolding of amphoteric macromolecules at the IEP in the presence of neutral salts. Thus behavior of amphoteric cryogels with respect to ionic strength not deviates from common peculiarities of amphoteric macromolecules.

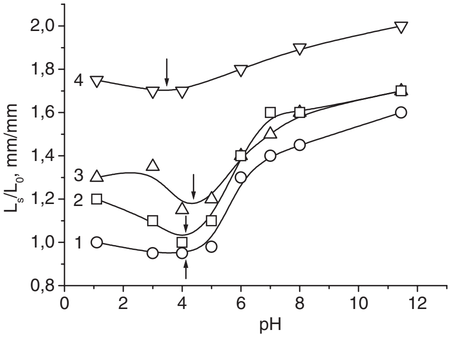

Figure 5.
Swelling-deswelling curves of cryogels ACG-550 (curve 1), ACG-334 (curve 2), ACG-226 (curve 3), and ACG-118 (curve 4) on pH. Arrows show on the position of the IEPs.

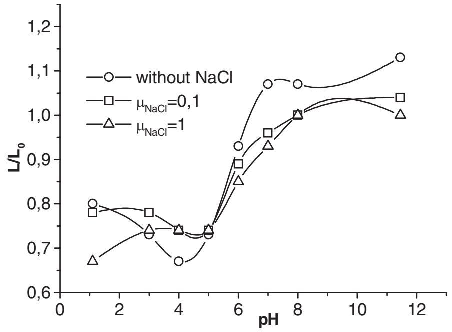

Figure 6.
The influence of the ionic strength on the swelling degree of ACG-334.

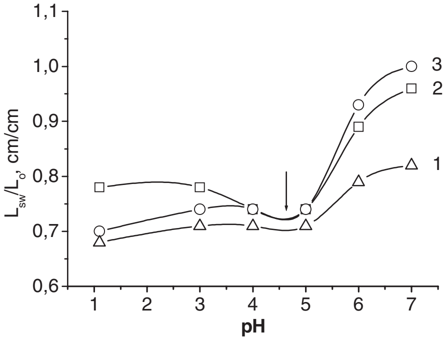

Figure 7.
Influence of $MgCl_2$ (curve 1), KCl (curve 2) and NaCl (curve 3) on the position of the IEP of ACG-334 at $\mu = 0.1$. Arrow shows on the position of the IEPs.

It is known[4] that the specific binding of cations and/or anions shifts the IEPs. In the presence of 0.1N NaCl the isoelectric pH (pH_{IEP}) of ACG-334 slightly shifts to basic region from 4.2 to 4.6 in comparison with salt-free solution (Figure 6). Shifting of the position of the IEP to more basic region in comparison with pure water may be connected by specific binding of low-molecular-weight cations to negatively charges of macromolecules. However in the presence of $MgCl_2$ and KCl the position pH_{IEP} as in the case of NaCl remains at $pH_{IEP} = 4.6$ meaning the indifferent nature of specific binding of cations with respect to carboxylate groups of cryogel (Figure 7). In the presence of KCl, KBr and KSCN the value of pH_{IEP} shifts to acidic region in the following sequence: $SCN^- > Br^- > Cl^-$ (not shown). Probably due to strong specific binding of SCN^- to charged amine groups, apparent disappearance of the IEP is observed. Thus behavior of amphoteric cryogels with respect to ionic strength and the nature of anions and cations not deviates from common peculiarities of amphoteric macromolecules.

Metal Complexes of Amphoteric Cryogels

The advantages of amphoteric macroporous cryogels with respect to metal ions are: 1) the ability to provide the adsorption process in dynamic regime; 2) simplification of adsorption process, passing metal containing aqueous solution through the sample; 3) high adsorption capacity of metal ions due to presence of complex-forming ligands and high developed inner and outer surface (see SEM pictures of Figure 3a); 4) concentration of trace amount of metal ions; 5) immobilization of targeted metal ions within macropores in order to reduce them and use as flowing catalytic microreactor. Complexation of amphoteric cryogels with transition metal ions is accompanied by colourization of samples (Figure 8). This is due to formation of coordination and ionic bonds between metal ions and amine and/or carboxylic groups of cryogels when aqueous solutions of metal salts pass through the gel specimen.

Table 3.
Adsorbed and desorbed amounts of metal ions by ACG-334.

Type of metal ions	Concentration of metal ions passed through ACG-334 mol/L	Adsorbed %	Desorbed %
Cu^{2+}	10^{-3}	99.9	51.4
Ni^{2+}		99.9	67.2
Co^{2+}		99.9	62.0

As seen from Table 3 the ACG-334 adsorbs up to 99.9% of metal ions from aqueous solution containing $10^{-3}\,mol \cdot L^{-1}$ of copper, nickel, and cobalt ions.

Metal sorption capacity of 1 g dry ACG-334 sample with respect to $10^{-3}\,mol \cdot L^{-1}$ of copper, nickel, and cobalt ions was equal to 0.63; 0.58 and 0.59 g respectively. It was found that on average 4 repeating units of amphoteric cryogels are bound up with 3 metal ions. Preferentially adsorption of Cu^{2+} ions (79%) in comparison with

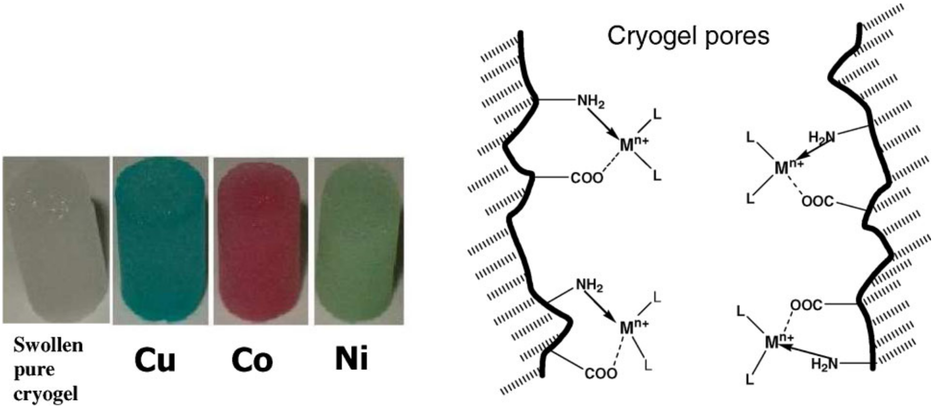

Figure 8.
Coloring of ACG-334 after adsorption of metal ions (left) and schematically representation of metal-polymer complexes in cryogel pores (right).

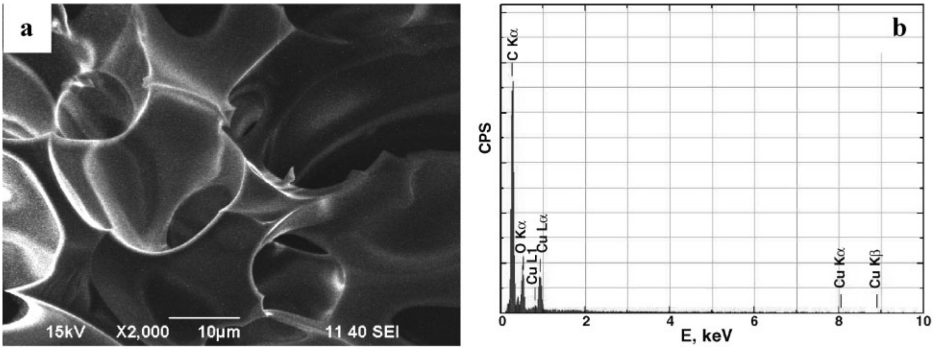

Figure 9.
SEM picture of ACG-334/Cu^{2+} complex (a) and its elemental composition (b).

Ni^{2+} (38%) and Co^{2+} ions (32%) was observed for the mixture of metal ions containing $10^{-5}\,mol \cdot L^{-1}$ of Cu^{2+}, Ni^{2+} and Co^{2+} in accordance with Irving-Williams sequence. Figure 9a,b shows the morphology and elemental composition of ACG-334/Cu^{2+} complex taken from the SEM and the energy dispersive X-ray attached to SEM.

It is seen that the surface of cryogel-metal complex is smooth and contains 6.1%

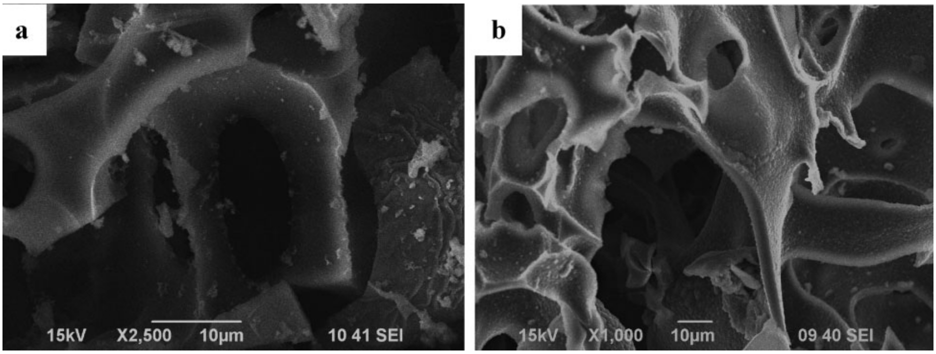

Figure 10.
SEM pictures of ACG-334/Ni^{2+} (a) and ACG-334/Co^{2+} (b) complexes after reduction by $NaBH_4$.

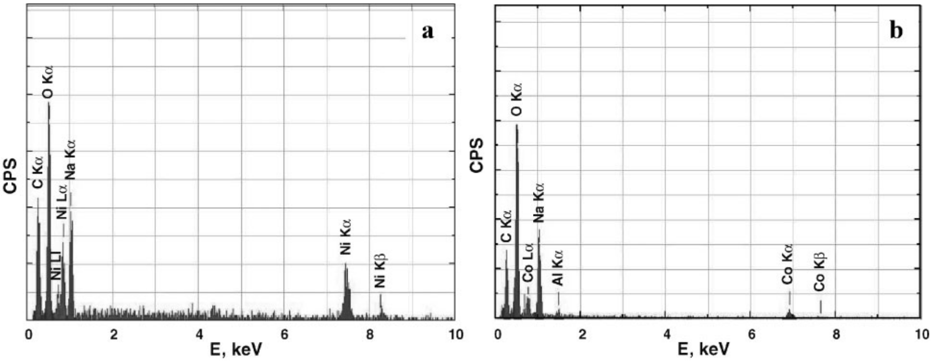

Figure 11.
Elemental composition of ACG-334/Ni^{2+} (a) and ACG-334/Co^{2+} (b) complexes after reduction by $NaBH_4$.

of copper. The reduction of cryogel-metal complexes by $NaBH_4$ leads to formation of nano- and micron-sized particles of metals and/or metal oxides immobilized on the inner and surface parts of amphoteric cryogels (Figure 10a,b). The chemical composition of the reduced by $NaBH_4$ complexes of ACG-334/Ni^{2+} and ACG-334/Co^{2+} revealed that up to 34.2% mass % of Ni and 9.7 mass % of Co particles are formed (Figure 11).

Thus amphoteric cryogels can serve as efficient heterogeneous supports for nano- and micron-sized zero-valent metal particles and as flowing microreactors in catalysis.

Conclusion

Macroporous amphoteric cryogels based on allylamine, methacrylic acid and acrylamide have been synthesized for the first time. They exhibited sponge-like porous structure with pore size ranging from 50 to 200 μm. The swelling rate and the dynamics of water flux through the cryogel samples were in good agreement and changed in the following sequence: ACG-118 > ACG-226 > ACG-334 > ACG-550. The values of the IEP found from the swelling experiments were in the range 3.5–4.3. The specific binding of cations shifts the IEPs to basic region while the specific binding of anions – to acidic region. Macroporous amphoteric cryogels adsorb efficiently copper, nickel, and cobalt ions from dilute (10^{-3} mol $\cdot$ L^{-1}) solutions. The sorption capacity of ACG-334 for copper, nickel, and cobalt ions is equal to 0.63; 0.58 and 0.59 g/g dry cryogel,respectively. Cu^{2+} ions are adsorbed preferentially (79%) in comparison with Ni^{2+} (38%) and Co^{2+} ions (32%), respectively, in accordance with Irving-Williams sequence. Reduction of cryogel-metal complexes by $NaBH_4$ leads to formation within the pores of amphoteric cryogels nano- and micron-sized particles of metals that might be used as efficient flowing microreactors in catalysis. Cryogels are produced in a cheap and environmentally friendly way from aqueous solutions. The high adsorption capacity of trace metal ions in combination with unique cryogel properties like interconnected macropores (and hence low flow resistance) and sponge-like nature make cryogel monoliths promising for processing particulate containing streams like waste water treatment, removal of inhibiting ions from fermentation feeds e.g. for bioethanol production, pretreatment of process water for different industries.

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[1] B. Mattiasson, A. Kumar, I. Yu. Galaev, in: *Macroporous polymers: Production, Properties and Biotechnological/Biomedical Applications*, Eds., CRC Press, Boca Raton **2010**, p 513.

[2] V. I. Lozinsky, *Uspekhi Khimii* **2002**, *71*, 559.

[3] V. I. Lozinsky, I. Yu. Galaev, F. M. Plieva, I. N. Savina, H. Jungvid, B. Mattiasson, *Trends Biotechnol.* **2003**, *21*, 445.

[4] S. E. Kudaibergenov, in: *Polyampholytes. Synthesis, Characterization and Application*, Kluwer Academic/ Plenum Publishers, New York **2002**.

[5] S. E. Kudaibergenov, in: *Encyc. Polym. Mater. Techn*, John Wiley & Sons, Inc., **2008**.

[6] K. Ogawa, A. Nakayama, E. Kokufuta, *Langmuir* **2003**, *19*, 3178.

[7] K. Ogawa, A. Nakayama, E. Kokufuta, *J. Phys. Chem. B* **2003**, *107*, 8223.

[8] L. Deng, Y. Zhai, S. Guo, F. Jin, Z. Xie, X. He, A. Dong, *J. Nanopart Res.* **2009**, *11*, 365.

[9] J. Hu, Sh. Yu, P. Yao, *Langmuir* **2007**, *23*, 6358.

[10] Sh. Yu, J. Hu, X. Pan, P. Yao, M. Jiang, *Langmuir* **2006**, *22*, 2754.

[11] M. Miyake, K. Ogawa, E. Kokufuta, *Langmuir* **2006**, *22*, 7335.

[12] R. Doi, E. Kokufuta, *Langmuir* **2010**, *26*, 13579.

[13] W. Wu, N. Mitra, E. C. Y. Yan, S. Zhou, *ACS Nano* **2010**, *4*, 4831.

[14] B. H. Tan, P. Ravi, K. C. Tam, *Macromol. Rapid Commun.* **2006**, *27*, 522.

[15] K. E. Christodoulakis, M. Vamvakaki, *Langmuir* **2010**, *26*, 639.

[16] S. Schachschal, A. Balaceanu, C. Melian, D. E. Demco, T. Eckert, W. Richtering, A. Pich, *Macromolecules* **2010**, *43*, 4331.

[17] H. Ni, H. Kawaguchi, T. Endo, *Macromolecules* **2007**, *40*, 6370.

[18] T. Hoare, R. Pelton, *Biomacromolecules* **2008**, *9*, 733.
[19] S. Kudaibergenov, Zh. Adilov, D. Berillo, Zh. Sadakbayeva, G. Tatykhanova, Kh. Abdullin, I. Galaev, *eXPRESS POLYMER LETTERS* **2012**, *6*, 346.
[20] P. Arvidsson, F. M. Plieva, I. N. Savina, S. Fexby, L. Bulow, V. I. Lozinsky, Yu. I. Galaev, B. Mattiasson, *Journal of Chromatography A* **2002**, *977*, 27.
[21] V. A. Kabanov, D. A. Topchiev, in: *Polymerization of Ionizing Monomers*, Nauka, Moscow **1975**.
[22] S. E. Kudaibergenov, Sh. Sh. Shayakhmetov, S. R. Rafikov, E. A. Bekturov, *Dokl.Acad.Nauk SSSR* **1979**, *246*, 147.
[23] G. Ehrlich, P. Doty, *J. Am. Chem.Soc.* **1954**, *76*, 3764.
[24] P. G. Higgs, J. F. Joanny, *J. Chem. Phys.* **1991**, *94*, 1543.