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Copolymerization of N-vinylpyrrolidone with N,N'-methylen-*bis*-acrylamide: Properties and Structure

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Abstract The reaction of copolymerization of N-vinylpyrrolidone with N,N'-methylen-*bis*-acrylamide by mass ratio 1, 2 and 3% with the presence of initiator has been conducted. It is determined that along with cross- linked polymers there are also non-cross linked homo-polymer of poly-N-vinylpyrrolidone and copolymer of N-vinylpyrrolidone with N,N'-methylen-*bis*-acrylamide are obtained. The structures of obtained products by FTIR and ^{1}H, ^{13}C NMR spectroscopy have been investigated as well.

Keywords N-vinylpyrrolidone, Copolymerization, Cross-linking hydrogels, FTIR, NMR, Immobilization, Trypsin

1. Introduction

Recently it is very interesting to synthesize gel-forming polymers swelling in water which consider various functional groups [1-3]. Sensitive to pH - condition, temperature, ionic strength, electric field, ultra-violet radiation, such polymeric materials are also called ***smart*** or ***intelligent*** materials [4-6]. Such systems sharply changing the volume of interactions of the specified environments and result these polymers are widely used in medicine and biotechnology for purify biologically active compounds and also an immobilization of biocatalysts [7-9].

Synthesis of hydrogels on basis of synthetic polymers and immobilization on the biologically active compounds for the purpose of receiving complex compounds are in attention of many researchers today [10]. Ability of hydrogels to absorb a great amount of water is to give immobilization to antibiotics, enzymes, alkaloids and other biologically active compounds [11].

To regulate separation speed of biologically active compounds it is possible to change parameters of the - pH, temperature, radiation or change the molar ratio of monomer and cross-linked agent in the processes of gel synthesis [12, 13].

In this case, biologically active compounds don't have chemical bond with hydrogel and its separation depends on hydrogel and its structure [14]. The main demand made to hydrogels, used for the purpose of transportation of biologically active agents is a regulation of extent of their swelling degree from the irrespective of environment pH [15-17].

With the purpose of this scientific research we study to synthesize the three-dimensional polymeric hydrogels received by copolymerization of N-vinylpyrrolidone (VPr) with various molar ratio of N, N'-methylene-*bis*-acrylamide.

We studied the influence of parameters the yield of the obtained products, to investigate hydrogels properties and structure and to check transportation possibility of trypsin as well.

2. Experimental Part

2.1. Materials

VPr and N, N'-methylene-*bis*-acrylamide (MBAA) were purchased from Sigma Aldrich. Azobisisobutyronitrile (AIBN) (E. Merck) was purified by recrystallization twice from methanol and then dried in the dark (m.p. 104℃). For the preparation of mixture solution, deionized water was used and we used acetone for precipitation of poly-N-vinylpyrrolidone (PVPr) from Aldrich, too. The other reagents were used as obtained and the solvents were purified according to conventional methods.

2.2. Copolymerization Procedure

Copolymerization VPr with MBAA for to synthesizing cross-linked polymer has happened by the following method: At first prepared the solution of monomer with MBAA in 1, 2 and 3% mass quantity and mixture till formation of homogonous solution. Add in system as initiator AIBN. Initiator's concentration forming 4.5×10^{-3} mol/L. After full dissolution of the initiator, to avoid destruction process of polymer by air oxygen, the reactor was vacuum 5 mm. Hg. The copolymerization reaction was taken in 2 hours, at the 323 K. The obtained fractions were separated with acetone

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and washing in deionized water. The yield of obtained fractional are defined by a gravimetric method. For studying yield of products depend on the time of copolymerization reaction was taken (VPr:MBAA=44:1, $C_{Init.}$= 4.5×10^{-3} mol/L, T=343 K) in during 2,5; 5; 10; 15; 20; 60 and 120 minutes. For the purpose of studying the influence of temperature on yield of hydrogel copolymerization reaction taken at a 323; 343; 363 and 383 K temperature. In this process the molar ratio monomer and cross-linking agent are VPr:MBAA=44:1, the initiator concentration 4.5×10^{-3} mol/L and reaction was taken during 60 minute.

2.3. Measurements and Characterization

After a copolymerization the structures of the hydrogels and polymers were identified by FTIR and ^{1}H, ^{13}C of a nuclear magnetic resonance (NMR) spectroscopy. The FTIR spectra of the prepared monomer and its polymer were recorded by IR spectroscopy (Varian). ^{1}H NMR spectra were recorded in dimethyl sulfoxide (DMSO) for monomer and polymer with tetramethylsilane as internal standard on Bruker NMR spectrometer. ^{1}H NMR spectra were run at 300 MHz, whereas ^{13}C NMR spectra were recorded in DMSO for monomer and were run at 125 MHz.

3. Results and Discussion

3.1. Effect of the Monomer Concentration on Yield of Copolymerization Products

Copolymerization reaction of VPr with MBAA is carried out 1, 2 and 3% a mass ratio with participation of the initiator. It is revealed that at allocation of reaction products and research of their structure definite that there are homo-polymer of VPr, copolymer VPr with MBAA and cross-linked polymer among the products. Yields of products in copolymerization reaction are given in table 1.

Apparently from table 1, with growth of quantity of MBAA a yield of the cross-linked polymer and copolymer increase. In shows that process activity of MBAA is identical during in cross-linking and copolymerization reaction.

When studying structure of the obtained cross-linking polymer it was revealed that its structure not completely coincides with a cross-linking homo-polymer PVPr with MBAA. During growth process of quantity of MBAA to get decreased yield of homo-polymer it was connected with as a result a rupture of double bond in structure of copolymer and transformation of cross-linking polymer. Comparison by the IR spectroscopy method structure of the cross-linking polymer with initial and other products of reaction shows that the structure of the hydrogel is similar with a copolymer structure.

Defined swelling degree of the samples which synthesize copolymerization VPr with MBAA in the deionized water depend on 1, 2 and 3% initiator weight. It is determined that swelling degree of all three samples was stabilized in 24 hours. The main changes happen within the first 180-200 minutes.

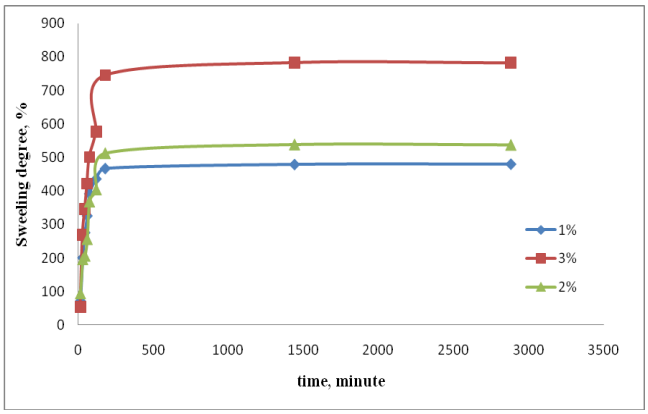

Figure 1. The swelling kinetics of basis VPr-MBAA gels in 0.9% NaCl solution. T=293 K

The equilibrium form of swelling degree formation speedy and was connected with hydrophilic of a polymeric macromolecule that increases the speed of adsorption or diffusion water molecules. Purpose of this hydrogels basis of synthesized VPr for transportation of biologically active substances is to study swelling degree of these gels in 0.9% NaCl solution [18]. These results differ from the results obtained in the pure water environment (Fig.1). From the fig.1 it is possible to show that emergence of the ionic force in the environment influences swelling degree of hydrogel. As a result, the Na^{+}, Cl^{-} hydrate molecules speedy diffuse into hydrogels inside comparison with the free molecules of water. Because the negative charges >C=O functional groups in pyrrolidones circle have more sensitivity of positive charged ions.

Table 1. Yield of copolymerization products of VPr with different dose (%) of MBAA. $C_{Init.}$= 4.5×10^{-3} mol/L, T=343 K, t=120 min

MBAA %	ν_M:ν_{mbaa}	Degree transformation of monomer, %	Yield of cross-linked polymer, %	Yield of homopolymer, %	Yield of copolymer, %	Degree of cross-linked, %
1	133:1	97.74	30.0	51.78	16.42	36.67
2	66:1	98.32	40.2	32.42	26.54	56.38
3	44:1	99.83	49.7	16.94	33.72	74.60

3.2. Effect of the Initiator Concentration on the Rate of Copolymerization

The influence of initiator's concentration yield of gel fraction in system with the highest transformation of VPr participation with 3% (mass) of MBAA. At the result, it is observed increase of the gel fraction and decrease yield of copolymer fraction. Polymerization reaction direct towards to homo-polymer and forming gel process when increased the concentration of initiator and result yield of their fractions respectively increased. Although increased concentration of the initiator the yield of gel fraction increased but decreased the stability and swelling degree of cross-linking polymer, therefore the concentration of the initiator of 4.5 mmol/L is accepted the optimum. As with increases the concentration of the initiator the molecular mass of the polymer decreases and the cross-linking polymer consist a low molecular weight macromolecules. It was the cause of the little swelling degree which connected with the size of polymeric net.

3.3. Effect of Copolymerization Time

For the research dependence of yield of cross-linking polymer on time, reaction of copolymerization VPr with MBAA the molar ratio was 44:1, concentration the initiator 4.5×10^{-3} in mol/L and copolymerization become during 2.5; 5; 10; 15; 20; 60 and 120 minutes. It was shown that, the yield of gel fraction within 60 minutes makes 48.64%. During the reaction it was observed that the yield of copolymer and polymer fractions decreases and a more amount of monomer consumption to the gel-forming process. It was determined that in 60-70 minutes the yield of cross-linking polymers is stabilized (Fig.2) and the gradient of time doesn't impact the gels yield.

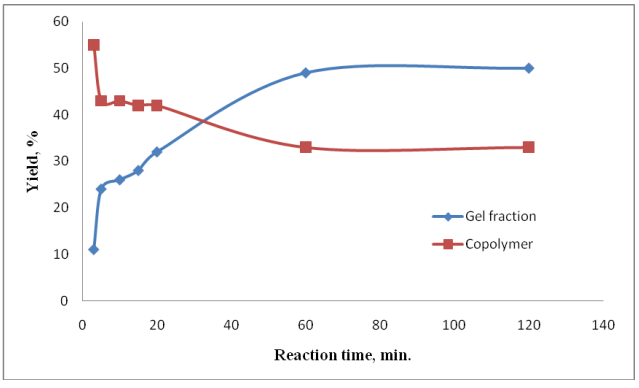

Figure 2. Depend of yield of cross-linking polymer and copolymer fraction on the reaction time

The basis results of process the optimum yield of cross-linking polymer accepted in 3% MBAA and during 60 minutes.

3.4. Effect of Temperature

The influence of the temperature on a gel yield was studied. Experiments were for this purpose made under the above described optimum conditions of a cross-linking (VPr:MBAA=44:1 molar ratio, $C_{Init.}=4.5\times10^{-3}$ mol/L, t=60 minutes). After completion of process all fractions were separated and the yield of products was determined by a gravimetric method (Fig.3).

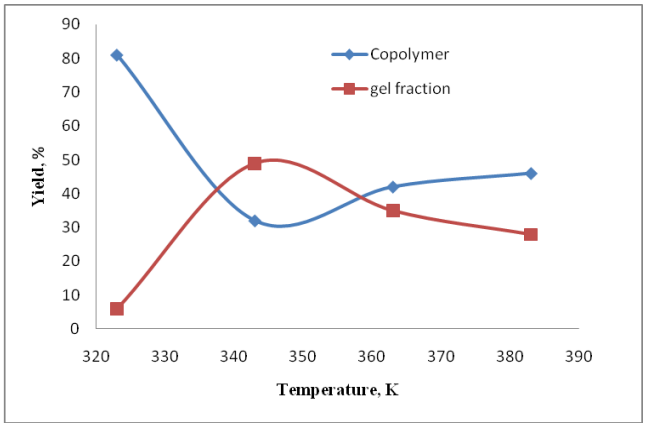

Figure 3. Yield of gel fraction and copolymer in different temperature

It is visible that the optimum yield of cross-linking polymer (48.64%) is observed at the 343 K. The low yield of gel fraction at a lowered temperature is connected with insufficiency of the radicals which doesn't continue process of direction to form the gel. At temperatures over 343-363 K decreases the yield of cross-linking polymers connected with destruction of polymer a macromolecules and obtaining low-molecular soluble fractions (an olygomer or a dimer). Thus the optimum temperature for the synthesis of cross-linking polymers at block is 343 K.

3.5. Characterization

The FTIR spectrum of cross-linking gel absorption bands at 1420–1410, 1650 and 1580-1475 cm^{-1} represent the CH_2=CH-, >C=O and -NH- groups (Fig.4). This shows that the double CH_2=CH- bands at MBAA don't complete during the copolymerization reaction.

As a result of spectral research ^{1}H and ^{13}C nuclear magnetic resonance of the dissolved fractions in acetone, it was established that 2.8; 2.9 and 3.0 ppm are the maximum chemical shifts. These shifts are characteristic, generally for polymers and the protons relating to groups -CH_2-, >CH- of a lateral chain. Chemical shifts 3.9; 4.1 ppm and 4.8; 5.0 ppm in ranges belong to protons of a macromolecule of CH_2=CH- group.

The maximum chemical shifts ^{13}C a nuclear magnetic resonance a range in these fractions at 18,0-19,0 ppm, 30-45 ppm, characterize the atoms ^{13}C entering into a chain of the basic fragments (Fig.5).

In addition, in a range relating to strong area (95 ppm, 130 ppm) chemical shifts are observed, characteristic for carbonic atoms of vinyl group. The carbon atom >C=O group of a lateral chain has shift to 175 ppm. Thus, based on FTIR received spectral analyses and a nuclear magnetic resonance spectroscopy it is possible to tell that the unrealized vinyl group of the cross-linking agent remains free in copolymer.

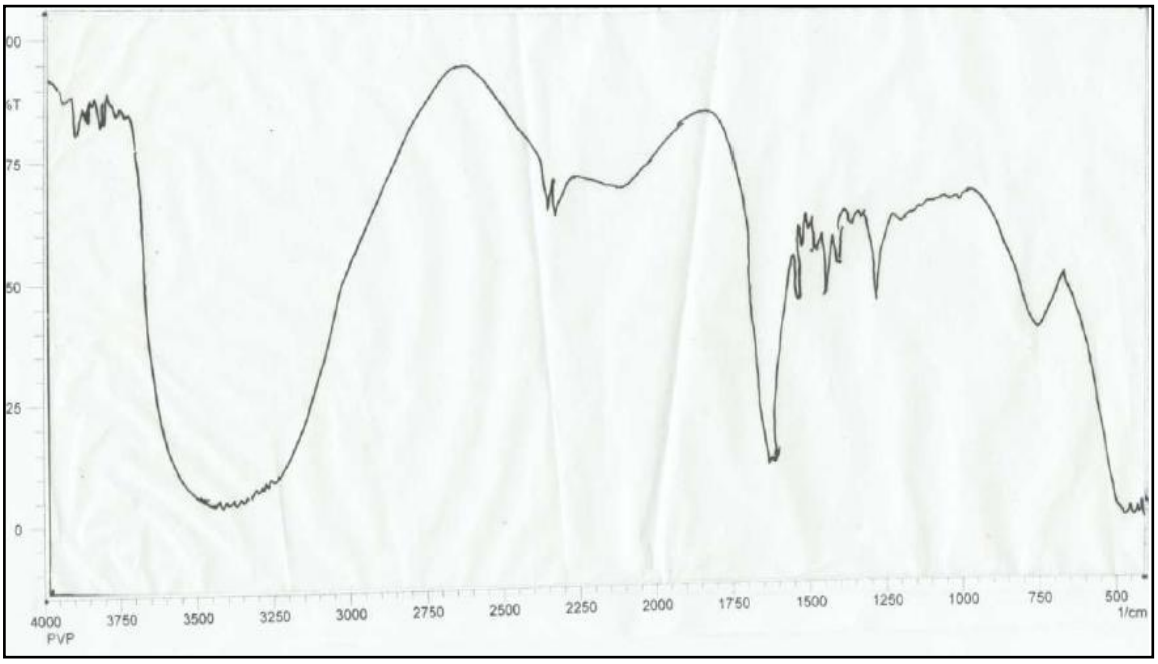

Figure 4. The FTIR spectrum of PVPr (separated from the copolymers products)

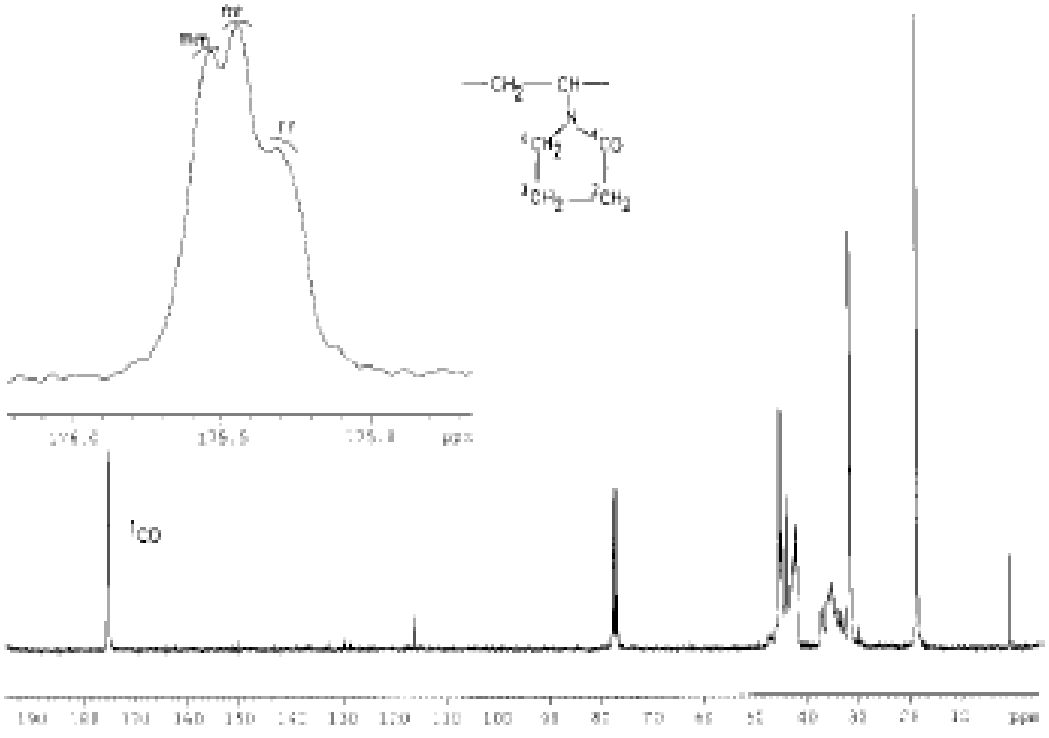

Figure 5. The NMR spectrum of pure PVPr (separated from the copolymers products)

During the cross-linking VPr with MBAA in block formed the fraction which wasn't dissolution in acetone. The received in such reaction products contain CH_2=CH-, >C=O, -NH- functional groups in a lateral chain of copolymer. As a result synthesized cross-linking gel containing double carbon band in lateral chain of copolymer which isn't soluble in water and acetone. Its structure was defined in the following form (Fig. 6).

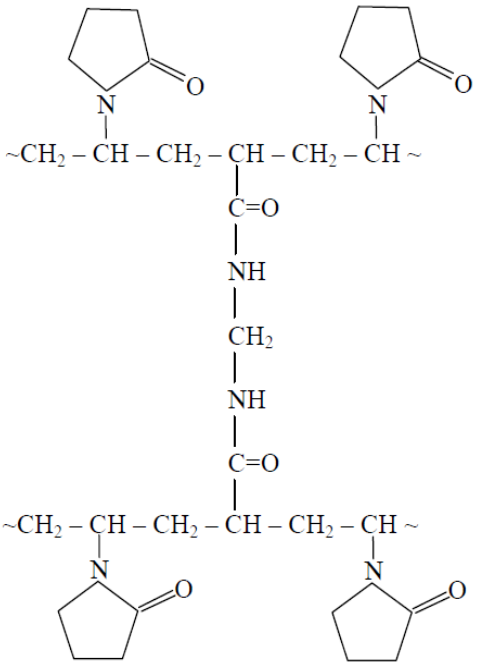

Figure 6. Schematic illustration of gel fraction based of VPr-MBAA

The investigated immobilization trypsin into gel, which this gel synthesized copolymerization VPr with 3% MBAA. It also was studied dependence of quantity and activity of trypsin on time and environment of pH. It was determined that amount and the specific activity of the immobilized trypsin into gel makes 0.38 mg/g and 42 ED/mg respectively. Research of the immobilized trypsin into gel in the static conditions, allocated in solution in the range of pH=3÷11, shows that at pH=8 desorption was optimum and makes 84.22%. At above the pH=9, it causes formation the collapse of the swelling degree of hydrogels decreases, trypsin separated from the gel at the end of macromolecule of enzyme began disintegration [19, 20]. The time factor of separation trypsin into solution which immobilized in the received polymeric gel was studied at pH=8. It was shown that the separation degree of immobilized trypsin the first 6 hours makes about 32.46% and 18 hours later the amount of the enzyme which separated into solution was stabilized [21, 22], and changed about ~89.36% (±0,8-1,2). Thus, it was shown that the change of synthesis conditions doesn't lead to special changes in structure and swelling degree of the obtained gels basis of VPr and MBAA.

4. Conclusions

It was shown that the materials of three-dimensional structure obtained by copolymerization of VPr with MBAA, possess ability of gel forming in water and they meet the requirements, shown to transportation of biologically active compounds.

REFERENCES

[1] Schacht E.H. Biodegradable polymers for biomedical applications // European Cells and Materials, 2003; 5, (1), 58.

[2] Tuncer C., Simin K., Gokhan D. Thermosensitive poly-N-isopropylacrylamide-co-acrylamide hydrogels: Synthesis, swelling and interaction with ionic surfactants // European Polymer J., 2006; 42, (2), 348-355.

[3] Brannon P.L. Polymers in Controlled Drug Delivery // Medical Plastics and Biomaterials, 1997; 4, 34-44.

[4] Tapdiqov, S. Z.; Mammedova, S. M.; Humbatova, S. R.; Zeynalov, N. A. The Obtaining and Stabilization of Silver Nanocomposites in Gum Arabic Environment. Journal the News of Baku State University 2013, 2, 22-27.

[5] Caykara T., Melike D. The effect of solvent composition on swelling and shrinking properties of polyacrylamide-co-itaconic acid hydrogels // European Polymer J., 2004; 40(11), 2605-2609.

[6] Chengiz O., Tuncher Ch., Omer K., Olgun G. Radiation synthesis of poly-N-vinyl-2-pyrrolidone-β-tartaric acid hydrogels and their swelling behaviors //Polymer Advance Technology, 2002; 13(2), 87-93.

[7] Jaya M., Vivek K.S. Cross-linking in Hydrogels - A Review. American J. Polymer Sciences, 2014; 4(2), 25-31.

[8] Kang D.Y., Tao P., Han B.F, Yu Y.H. Swelling kinetics and release characteristic of cross-linked chitosan: Polyether polymer network hydrogels //J. of Polymer Science, Part A: Polymer Chemistry, 1994; 32(7), 1213-1223.

[9] Ding L., Li Y., Jiang Y., Cao Z., Huang J. Properties and synthesis of new supports for immobilization of enzymes by copolymerization of vinylene carbonate and methacrylic acid. Chinese J. Polymer Science, 2000; 18(4), 343-349.

[10] Mammedova S.M., Tapdıqov S.Z., Humbatova S.F., Zeynalov N.A. Spectroscopic Investigated Interaction between Silver Nanocomposites Based of Poly-N-Vinylpyrrolidone and Doxorubicin for Drug Delivering. J. Chemistry and Chemical Engineering, 2014; 8, 800-804.

[11] Hsiang-Fa L., Min-Hao H., Rong-Ming H. etc. Novel Method Using a Temperature-Sensitive Polymer (Methylcellulose) to Thermally Gel Aqueous Alginate as a pH-Sensitive Hydrogel//Biomacromolecules, 2004; 5(5), 1917-1925.

[12] Fariba G., Ebrahim V.F. Hydrogels in Controlled Drug Delivery Systems//Iranian Polymer J., 2009; 18(1), 63-88.

[13] Xin C., Wenjun L., Wei Z., Yuhua L., Tongyin Y. pH sensitivity and ion sensitivity of hydrogels based on complex-forming chitosan/silk fibroin interpenetrating polymer network //J. of Applied Polymer Science, 1997; 65(11), 2257-2262.

[14] Kimberly A.S., Anne P.S., Diane L.H., Mark W.G. Synthesis of a novel polysachharide hydrogel // J. Macromolecular Science, Part A, 1999; 36(7), 981-989.

[15] Mani P., Jo ão F.M. Stimuli-Responsive Hydrogels Based on Polysaccharides Incorporated with Thermo-Responsive Polymers as Novel Biomaterials//Macromolecular Bioscience, 2006; 6(12), 991-1008.

[16] Nikolas A.P. Hydrogells // Ratner: Biomaterials Science, 2003; 12, 35-36.

[17] Pal K., Banthia A.K., Majumdar D.K. Polymers Hydrogels: Characterization and Biomedical Applications-A mini review. 2009; 12, 197-220.

[18] Violeta G.J., Harold E.S. Trypsin immobilization on derivatized cellulose beads by biospecific avidin-biotin interaction and characterization of the immobilized activity //J. Food Biochemistry, 2006; 26(2), 119-129.

[19] Zeynalov N.A., Akhmedov I.D., Tapdiqov Sh.Z. Carry basid of poly-N-vinylpyrrolidone and arabinogalactane for immobilization trypsine, J. Actually problem of Humanitarian and Natural Sciences, 2010; 8(1), 44-48.

[20] Purcena L.L., Caramori S.S., Mitidieri S.R., Fernandes K.F. The immobilization of trypsin onto polyaniline for protein digestion //Materials Science and Engineering, 2009; 29(4), 1077-1081.

[21] Martin M., Anders L. Immobilization of trypsin on porous glycidyl methacrylate beads: effects of polymer hydrophilization//Colloids and Surfaces B: Biointerfaces, 2000;18 (3-4), 277-281.

[22] Nouaimi M., Moschel K., Bissvanger H. Immobilization of trypsin on polyester fleece via different spacers //J.Enzyme and microbiological, 2001; 29(8), 567-574.