Open Access
Baghdad Science Journal **Vol.12(3)2015**

Methionine as a Spacer between Poly Acrylic acid and Ampicillin

Firyal M. Ali* ***Saif M. Ali***

Department of Chemistry, College of Science, University of AL-Mustansiriyah Baghdad-Iraq
***E-mail:** Abood121996@yahoo.com

Received 22, July, 2014
Accepted 26, October, 2014

Abstract:

In this work a novel drug delivery system through modification of poly acrylic acid with Methionine as a spacer between the poly acrylic acid which was converted to its acyl chloride and reacted with Methionine as spacer unit which has been reacted with Ampicillin drug. In vitro drug release study had been conducted successfully in basic medium in pH 7.4 and acidic medium in pH 1.1 at 37℃. Due to many problems associated with drug release and, this modification could decrease the side effect of drug. The prepared prodrug polymer was characterized by spectra method [FTIR and ^{1}H−NMR]. Physical properties and intrinsic viscosity of drug polymer were determined. The good results were obtained in the presence of spacer unit with comparing without spacer unit.

Key words: Poly acrylic acid, Methionine, Ampicillin.

Introduction:

Methionine is an essential amino acid that is required in the diet of humans and livestock. Plant proteins are frequently deficient in Methionine and consequently an exclusively vegetable diet may fail to meet nutritional requirements [1]. Polyacrylic acid based polymers are mainly used for oral and mucosal contact applications such as controlled release tablets, oral suspensions and bio-adhesives. It is also used as a thickening, suspending and emulsion stabilizing agent in low viscosity systems for topical applications [2]. Acrylic acid (AA) is deemed to form a super absorbent polymer which can absorb very large amount of water and retain it even under high pressure. As a result of this unique characteristic, it has been used in various controlled drug delivery systems [3].Many synthetic polymers are biologically inert. However, some exhibit toxicity, while others exhibit a wide range of therapeutic activities [4]. Some investigators have focused their attention on the preparation of bioactive polymeric materials, by bounding the drug to a polymer through covalent linking, e.g. chloramphenicol was previously attached to a meth acrylic by an acetal function and then copolymerized with 2-hydroxyl methacrylate [5].Polymers are becoming increasingly important in pharmaceutical applications especially in the field of drug delivery. Polymers range from their use as binders in tablets to viscosity and flow controlling agents in liquids,

suspensions and emulsions; can also be used as film Coatings, to disguise the unpleasant taste of a drug, to enhance drug stability and, to modify the release characteristics [6, 7]. Prodrugs can be used to afford drugs that would be too toxic to be given directly, a feature of the slow release. The prodrug that is converted to the active drug at the target site itself greatly reduced side effects of highly toxic drugs[8]. The fundamental macromolecular transport theory of biopolymers through tissues has been successfully applied to the design, fabrication, and prediction of in vivo performance of polymer drug-delivery systems. Consequently, based on the molecular weight, Conjugate polymer drug carriers have been developed to perform more specifically than the drug alone. Many other variables, such as composition of the polymer chain, structure, polyelectrolyte character, and solubility, can also affect polymer behavior [9].

Methods:

Materials and Instruments

Ampicillin was purchased from Samarra Company; Thionyl chloride was obtained from Fluka. Methionine and Acrylic acid were obtained from Aldrich. Dimethylformamide (DMF) was purchased from Merck. Triethylamine was purchased from Fluka.^{1}H-NMR spectra were recorded on a Shimatzu spectrophotometer in Dimethylsulphoxide (DMSO). The FTIR spectra were recorded by (4000-600cm^{-1}) on a Shimatzu spectrophotometer. Melting points were determined on callenkamp MF B-600 Melting point apparatus. Electronic spectra measurement using CINTRA5-UV.Visble spectrophotometer.

***Polymerization of Acrylic Acid*. [10]**

In a screw capped polymerization bottle (3g.), of acrylic acid was dissolved in (10 ml) of DMF, 0.05% of the monomer weight of di benzoyl peroxide was added as an initiator. The bottle was flashed with nitrogen for few minutes inside a glove and firmly stopped. The solution was maintained at 90℃, using water bath for 1 hr. The solvent was evaporated under vacuum; the product was obtained, washed three times with ether. Dried in a vacuum oven at 50℃, produced 95% of polymer with μ_{in} = 0.46 dL /g.

Preparation of polyacryloyl chloride. [11]

A thionyl chloride (5ml., 0.04mole) was added gradually to a mixture(2.48g, 0.03mole) of poly acrylic acid which was dissolved in 15ml of dioxane placed in a round-bottom flask provided with condenser, the contents were stirred with a magnetic bar at 50℃. The excess of thionyl chloride was distilled off and the poly acryloyl chloride was obtained and dried by vacuum oven. Producing white polymer, it was collected on a glass filter, washed repeatedly with ether giving 90%.

Modification of polyacryloly chloride with Methionine (P_2). [12]

In a round bottom flask provided with condenser (2g., 0.02mole) of poly acryloyl chloride was dissolved in (10ml) of DMF, Then (4.13g., 0.02mole) of Methionine the mixture was refluxed with stirring for 2hrs, the viscous product was obtained, the solvent was evaporated, washed with ether and dried at room temperature. The polymer (P_2) was obtained with 82% as a bright yellow viscous polymer.

Substitution of Poly [2-(4methylthio butanoic acid) acrylamide] with Ampicillin(P_3). [13]

(1.5g., 0.007mole) of prepared polymer (P_2) was dissolved in of dioxane: DMF mixture (10:1vol.), and (1ml)

thionyl chloride was added, the mixture was heated at 50℃ the prepared acyl chloride and (1ml) of triethylamine was added to dissolved (2.58g., 0.007mole) Ampicillin, the mixture was refluxed with stirring for 2hrs. The solvent was evaporated under vacuum; the product was washed with water three times, dried under vacuum oven. The reddish brown polymer (P_3) was obtained with 78%. The softening point of the drug polymer (P_3) was (95-100) ℃.

Determination of degree of Methionine substitution.[14]

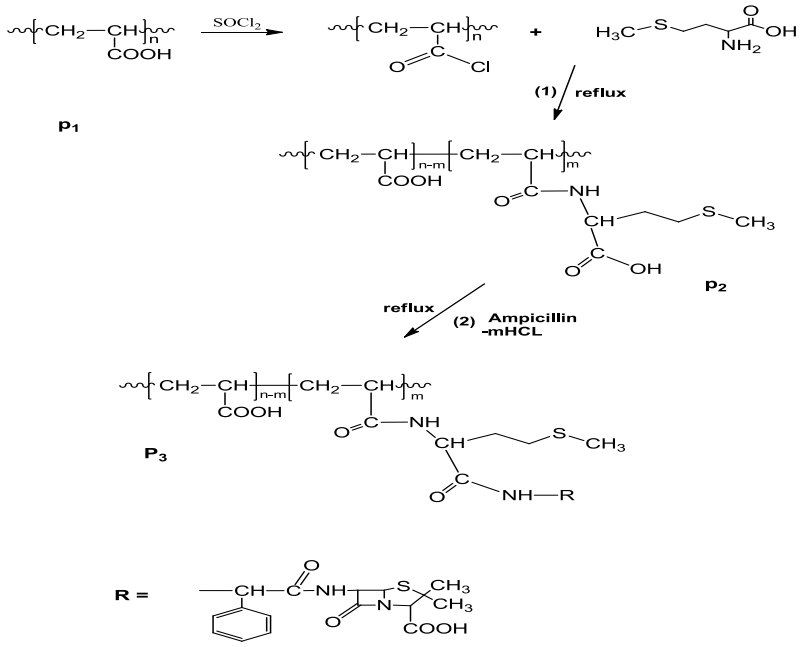
5mg of prepared prodrug polymer (P_3) was dissolved in 2ml of 0.1 N NaOH, the solution was heated to 70℃, for 15min in a water bath, cooled and the resulting solution was titration with 0.1N HCL to determine the excess of NaOH solution.

Controlled Drug Release. [15-19]

(0.1g.) of dried prepared prodrug polymer (P_3) was poured in 100ml of aqueous buffer solution such as (phosphate buffer pH 7.4) or acidic (solution pH 1.1). The buffer solution maintained at 37℃. with continuously stirred and 3ml of sample was analyzed by UV spectrophotometer and compared with calibration curve which was obtained computerized under similar medium. Fig. (5). Showed controlled Ampicillin release in different pH values at 37℃.

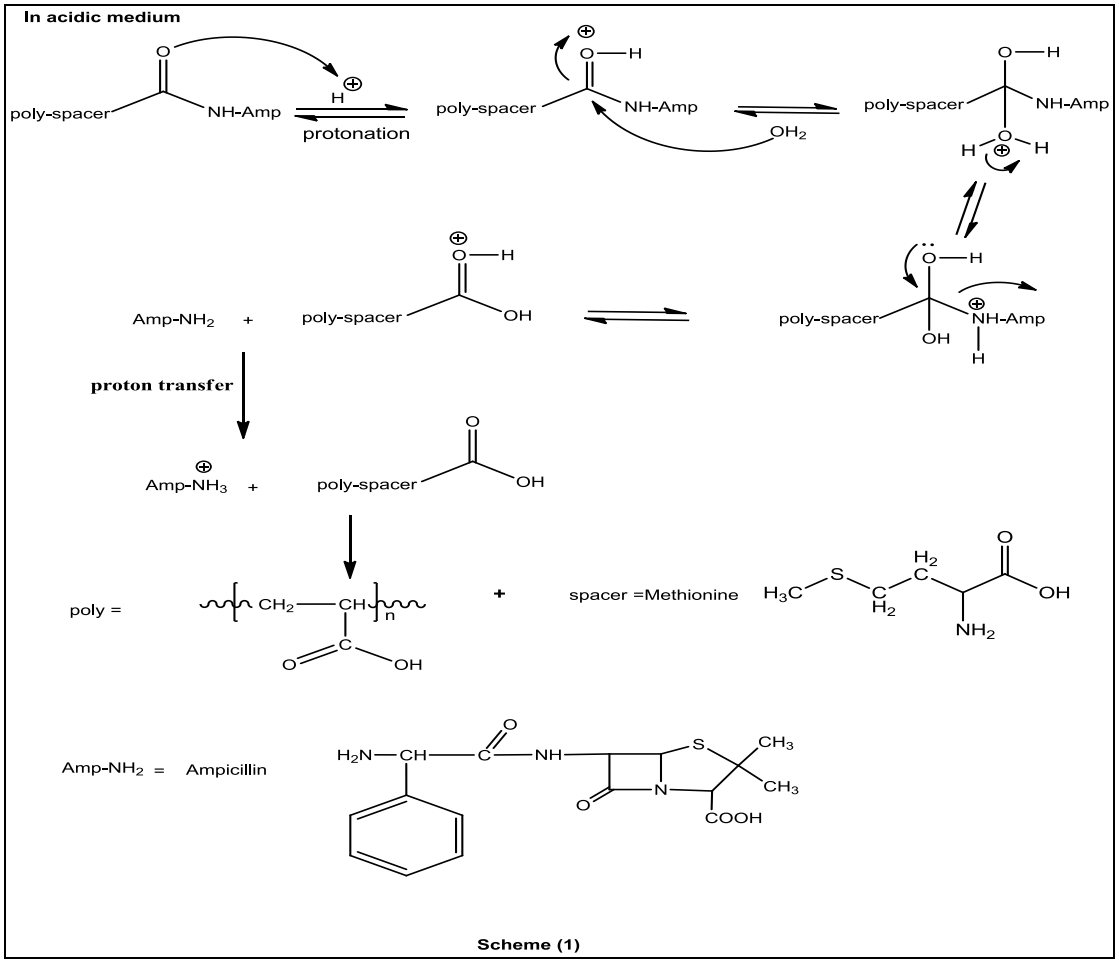
Results and Discussion:

In this research the prodrug was prepared using di functional spacer groups such as Methionine which was inserted between the Methionine and poly acryloyl chloride. The carboxylic acid groups were reacted with ($-NH_2$) groups of Methionine, produced amide attachment group, and the other carboxyl groups were reacted with Ampicillin which could produce amide arm groups. This work aimed to extend the drug pended units to be easy hydrolysis through polymer chains. The high yield was obtained by reaction of poly acrylic acid and Methionine as spacer side arm units as shown below:

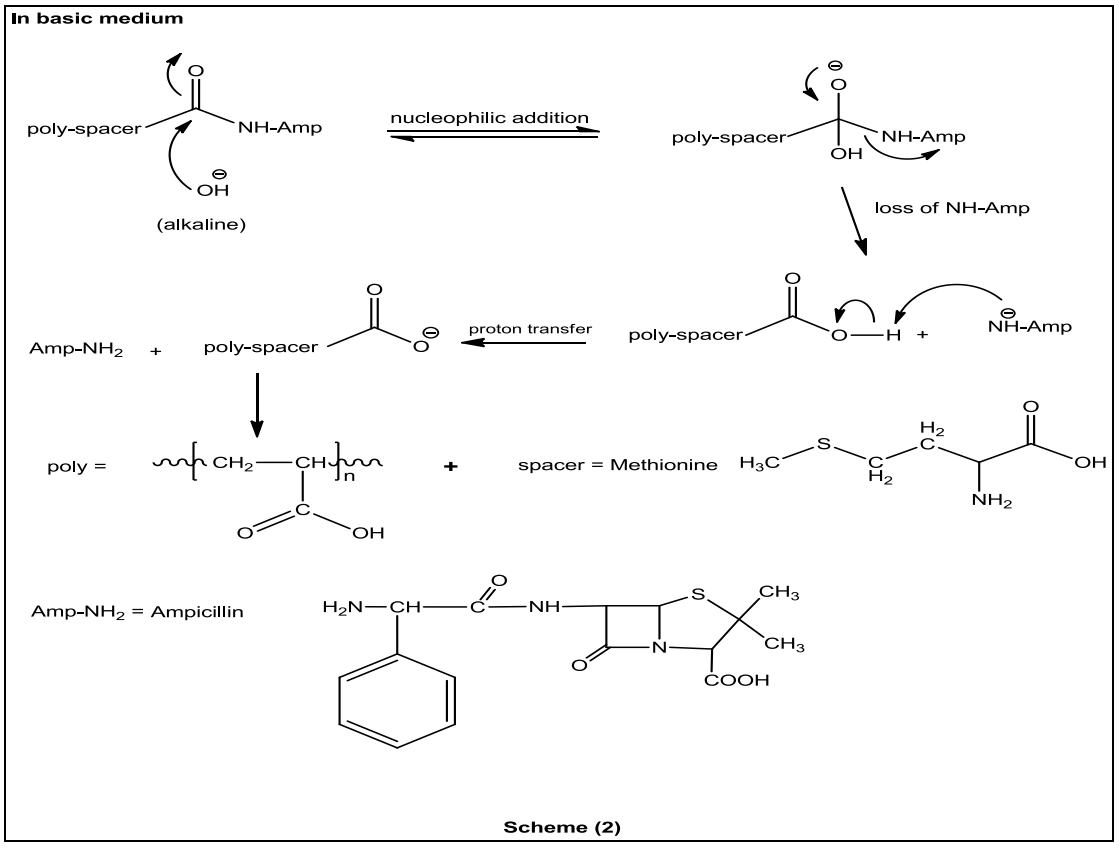


The modified polymer P_2 and P_3 were characterized, by FTIR spectrum, Fig (1) shows the broad beak around 3462-2571 cm^{-1} broad assigned of OH stretching carboxylic group 3200-3500 cm^{-1}, peaks at 2814-2931 cm^{-1} were

asymmetrical and symmetrical stretching of C-H aliphatic, peak around 1728 cm-1 represents stretching vibration of C=O from carboxylic groups. Fig (2) shows the broad beak around 3462-2700 cm^{-1} broad assigned of remained OH stretching carboxylic group 3200-3500 cm^{-1}, overlap ,and N-H stretching from an amide group, peaks at 2918-2987 cm^{-1} were asymmetrical and symmetrical stretching of C-H aliphatic, 1647 cm^{-1} represented to (amide carbonyl), peak around 1728 cm-1 represents stretching vibration of C=O from carboxylic groups of Methionine and 1728 cm^{-1} due to carbonyl of carboxylic group of unreacted poly acrylic acid. Fig (3) ^{1}H−NMR spectrum of P_2 shows the signals at δ: 1.5 ppm (2CH2−CH, 4H, d.) polymer, δ: 2.4 ppm (CH−COOH, 1H, T.), δ: 3.4 ppm (CH−CO,1H, T.), δ: 3.2 ppm (CH2−S, 2H, T.), δ: 1.2 ppm (CH2−CH, 2H, q.), δ: 2.8 ppm (CH3−S, 3H, S.), δ: 4.1 ppm (CH−CO, 1H, T.) of Methionine, δ: 8.2 ppm (NH, 1H, S.), δ: 11.4 ppm (COOH, 1H, S.).The remained carboxylic acid and Methionine. FTIR spectrum, Fig (4) of Ampicillin Methionine acryl amide polymer P_3 shows the beak at around 3398cm^{-1} assigned of remained OH stretching carboxylic group 3500-2600 cm^{-1}, and 3200cm^{-1} as shoulder beak due to NH amide , peaks at 2785-2985 cm^{-1} were asymmetrical and symmetrical stretching of C-H aliphatic, 3050 cm^{-1} of C−H aromatic, 1691cm^{-1} is attributed to (carbonyl-amide) and the other absorption appeared at 1722 cm^{-1} is for carbonyl acid and the new absorption were appeared at the beak appeared at 1641 cm^{-1} is due to carbonyl−amide. Fig (5) ^{1}H−NMR spectrum of polymer P_3 shows the signals δ: 2.1 ppm of (2CH2−CH, 4H, d.), δ: 3.2 ppm (CH−COOH, 1H, T.), δ: 3.6 ppm (CH−CO, 1H, T.), δ: 2.8 ppm (CH2−S, 2H, T.), δ: 3.4 ppm (CH3-S, 3H, S.), δ: 2.3 ppm (CH2−CH, 2H, q.), δ: 4.3 ppm (CH−CO,1H, T.) of Methionine, δ: 2.0 ppm (2CH3 terminal, 6H, S.), δ: 3.8 ppm (CH−CO, 1H, T.), δ: 4.8 ppm due to(2CH cyclic, 2H, d.), δ:5.4 ppm (CH−Ar, 1H, S.), δ: 7.2 ppm (2H)d. of ortho aromatic ring, δ: 7.9 ppm of (3H) T., of meta and para, δ: 8.1 (NH, 1H, S.), 11.2 ppm (COOH,1H, S) The remained carboxylic acid was 45% was tested by titration of polymeric sample with 0.1N of NaOH in the presence of phenonaphthalene as an indicator. The concept of polymeric drug has been subjected with medicine chemists as long consideration synthetic polymers. The polymer which is substituted by drug groups enhanced the using as prodrug polymers. The UV. Spectra of P_3 gave absorptions at 250 and 230 nm due to. (n-π*) and (π-π*) due to electron transition for dreg conjugation structures. The controlled release rates were studied as drug polymers which could be hydrolyzed in basic and acidic medium due to ester bonds as shown in the following mechanism:-



Scheme (1)



Scheme (2)

Conclusion:

In this work, it was concluded that the extended side arm of polyacrylic acid with suitable spacer di-functional such as Methionine as amino acid unit with Ampicillin as antibiotic drug through amide attachment, it appeared in basic medium, the rate of hydrolysis is higher than acidic medium this is due to the presence of OH^- in alkaline, which acts as a stronger nucleophilic attack to carbonyl group with respect to water, and the H_2O takes place faster hydrolysis than acidic medium, H^+ which is bonded to oxygen atom of amide as shown in Scheme (2). The spacer effect appeared more enhancements in hydrolysis of amide groups.

Fig (6) shows the release profile of drug release (mole fraction) versus time A swelling percentage of the prepared polymer was studied which equals to 16%.

The swelling% was calculated according to the following equation.

$$\Delta m = m_1 - m_\circ / m_\circ \times 100$$

When:-

$m_\circ$ is the weight of dry drug polymer.

m_1 is the swallowed polymer in water.

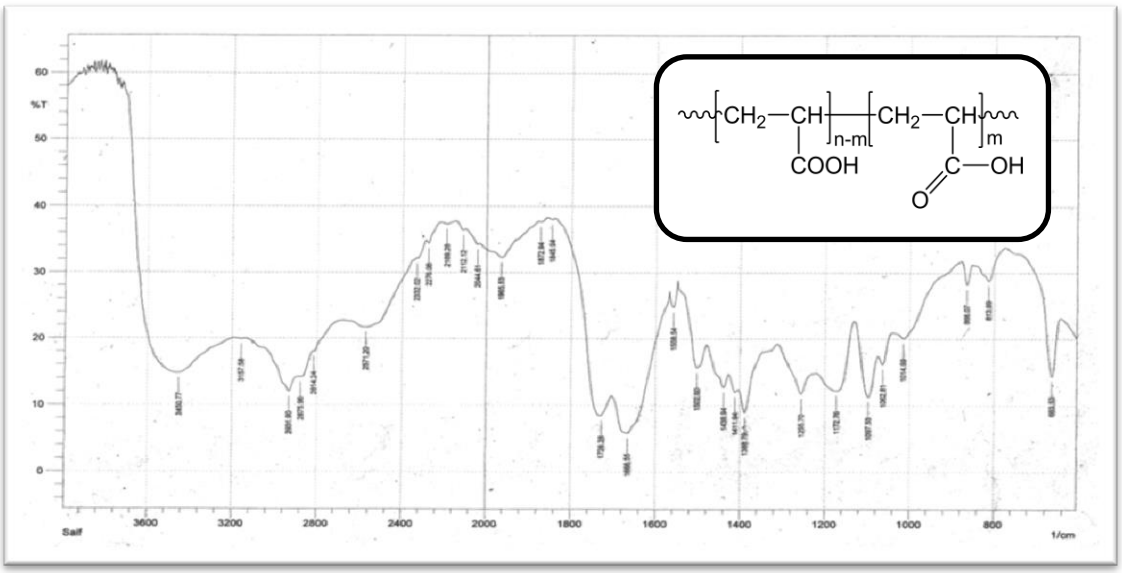


Fig (1) FTIR spectrum of polymer (P_1)

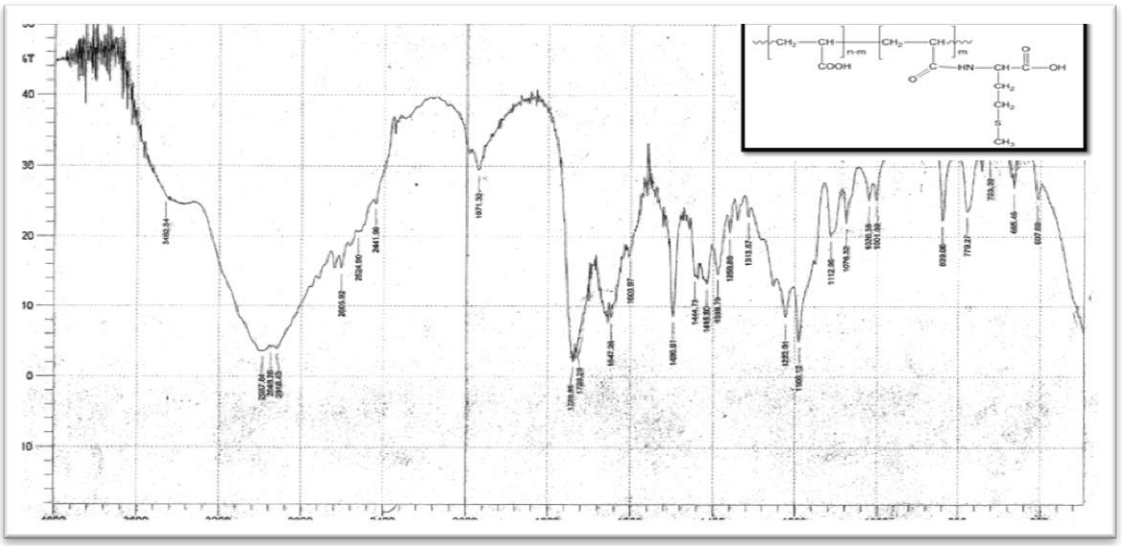


Fig (2) FTIR spectrum of polymer (P_2)

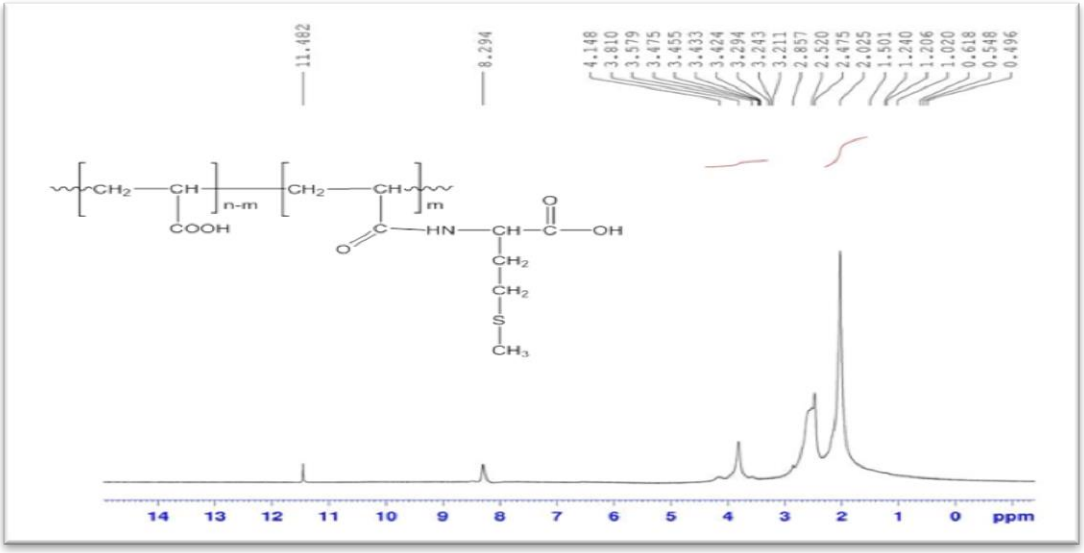


Fig (3) ^{1}H-NMR spectrum of polymer (P_2)

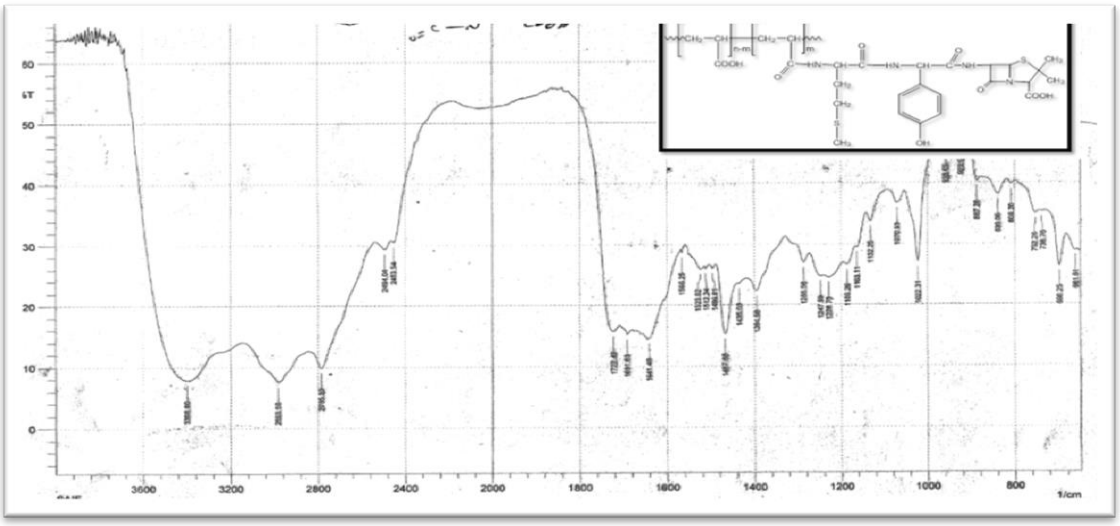


Fig (4) FTIR spectrum of polymer (P_3)

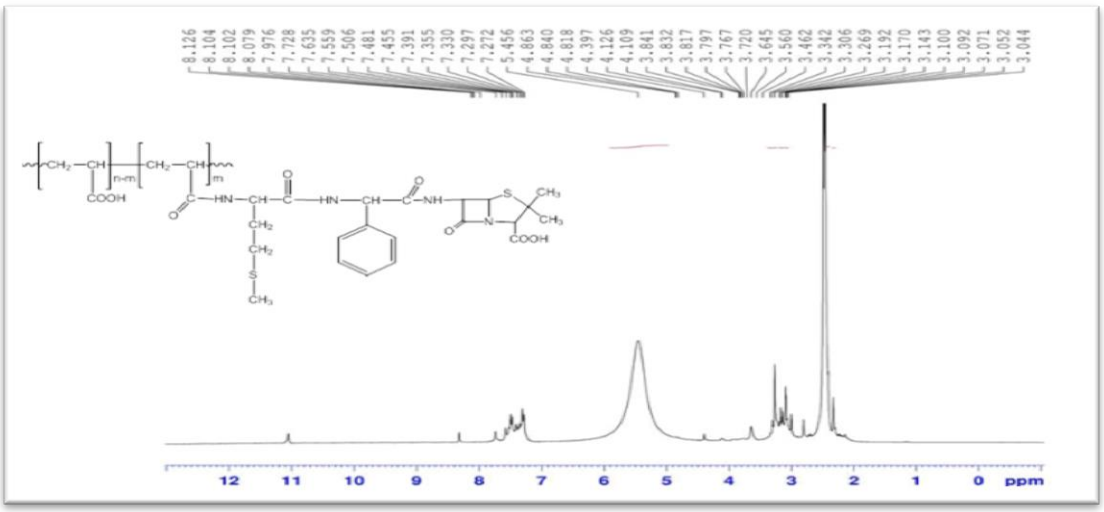


Fig (5) ^{1}H-NMR spectrum of polymer (P_3)

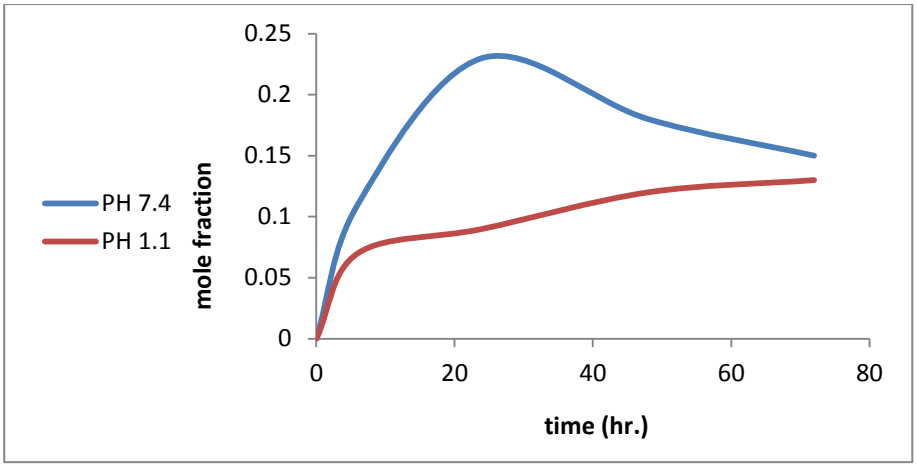


Fig (6) Drug release of P_3 in pH 1.1 and 7.4 at 37°C.

References:

[1] Dharmendra, K. and James, G. 2004. Methionine production by fermentation, Biotechnol Advan. 23: 41-61.

[2] Brecksville, R. 2010. Polymers for Pharmaceutical Applications, Pharmaceutical Bulletin1, Lubrizol, Wickliffe, USA.

[3] Talib, H.; Mahvash, A.; Nahzar, M. R.; Ikram, U. K. and Yasser, S. 2013. Chemically Cross-linked Poly "acrylic-co-vinylsulfonic" Acid Hydrogel for the Delivery of Isosorbide Mononitrate, The Sci. World J. 2013(2): 9.

[4] Vogl, L.G. and Eds, O. 1978. Polymeric Drugs. Donaruma, Academic Press, New York, NY.

[5] Julia, D. M. and Abramson, M. D. 2003. Cancer Center of the University of Pennsylvania, Posting Date.

[6] Davis, S. S. and Hium, L. 1998. Drug delivery systems for challenging molecules, Int. J. Pharm 176: 1-8.

[7] Henry, CM. 2002. Materials scientists look for new materials to fulfill unmet needs, C&E News 80(34): 39-47.

[8] Alka, V.; Bhupesh, V.; Sunil, K. P. and Kishu T. 2009. Prodrug as a chemical delivery system, A Review, Asian J. Research Chem. 2(2): 100-103.

[9] Larsen,C. 1989. Dextran prodrugs-Structure and stability in relation to therapeutic activity, Adv. Drug Delivery Rev. 3:103-154.

[10] Miller, M. L. 1964. Encyclopedia of Polymer Science and technology, Interscience publishers, a division of John Wiley & Sons, New York, 1st Ed., 197.

[11] Firyal, M. A. 2005. Modification of acryloyl chlorides polymers, Um-Salama Sci. J. 2 (4): 645-652.

[12] Peter, K. and Renata, T. 2002. Reactive and Functional polymers, John Wiley and Sons, New York, 52(1), 11.

[13] Callery, P. and Peter, G. 2002. Cancer and cancer chemotherapy. In: Foye's principles of medical chemistry (5^{th} Ed.). David, A.W. and Thomas, L.L. (EDs), Lippincott, Williams and Wilkins, Philadelphia, 924.

[14] Soudabeh, D- and Ali, A. 1990. Synthesis and hydrolysis of modified PVA, Iranian polym. J. 5(3): 183-191.

[15] Torchilin, V.P. 2001. Structure and design of polymeric surfactant-based drug delivery systems, J. Cont. Rel. 73: 137-72.

[16] Fang, J.; Nakamura, H. and Maeda H. 2011. The EPR effect,

unique features of tumor blood vessels for drug delivery, factors involved, and limitations and augmentation of the effect, Adv. Drug Deliv Rev 63(3):136-151.

[17] Mahammad, R. S.; Madhuri, K. and Dinakar, P. 2012. Polymers in Controlled Drug Delivery Systems, International J. of Pharm Sci. 2 (4):112-116.

[18] Ronald, A. and Michael, J. 2012. Overview of Controlled Release Mechanisms Controlled Release Society, J. Rathbone 33 (1): 19-42.

[19] Debjit, B.; Harish, G.; Pragati, B.; Kumar, S.; Duraivel, K. P. and Sampath, K. 2012. The pharm innovation Controlled Release, J. Drug Deliv. Systems 1:10.

الميثيونين كفاصل بين بولي حامض الاكريليك و الأمبيسلين

فريال محمد علي **سيف محسن علي**

الجامعة المستنصريه- كلية العلوم- قسم الكيمياء.

الخلاصة:

تضمن العمل تصميم نظام جديد لإيصال الدواء من خلال تحوير بولي حامض الأكريليك مع الميثيونين كفاصل بين بولي حامض الأكريليك الذي تم تحويلة إلى كلوريد الأسيل وتفاعلة مع الميثيونين كوحدة فاصلة من خلال تفاعله مع دواء الأمبيسلين. وقد أجريت في المختبر بنجاح دراسه التحرر الدوائي في الوسط القاعدي في درجة الحامضيه 7.4 والوسط الحامضي في درجة الحامضيه 1.1 عند 37م°.بسبب العديد من المشاكل المرتبطة لتحرر الأدواء، وهذا التعديل يمكن ان يقلل من الأثار الجانبية للأدويه. تم تشخيص دواء مساعد البوليمر المحضر بواسطة مطياف الأشعة تحت الحمراء وطيف الرنين النووي المغناطيسي. تم تحديد الخصائص الفيزيائية واللزوجة الجوهرية للبوليمر الدوائي. تم الحصول على نتائج جيدة في حالة وجود وحدة فاصلة مع المقارنة من دون وجود وحدة فاصلة.

الكلمات المفتاحية: بولي أكريليك أسد، الميثيونين، الأمبيسلين.