May. 2015. Vol. 6. No. 5 ISSN2305-8269
International Journal of Engineering and Applied Sciences

www.eaas-journal.org

SYNTHESIS OF CIPROFLOXACIN LACTATE PROCAINAMIDE AS MUTUAL PRODRUG

FIRYAL MOHAMMED ALI *, FIRAS AZIZ RAHI, MOWAFAQ MOHAMMEDGHAREEB***AHMED YASEEN**** AND *****TAGHREED.H.AL-NOOR**

*Almustansiriya University, College of Science, Department Of Chemistry,
**Almustansiriya University, College of Pharmacy, Department Of Pharmaceutics,
***Baghdad University,College of Pharmacy, Department Of Pharmaceutics
****Baghdad College of Pharmacy
Corres. author : * E-mail: Abood121996@yahoo.com ,
** mt_iraq@yahoo.com ,*** mopharmacy@yahoo.com
**** ahmedpharma2009@gmail.com, ***** drtaghreed2@gmail.com

ABSTRACT

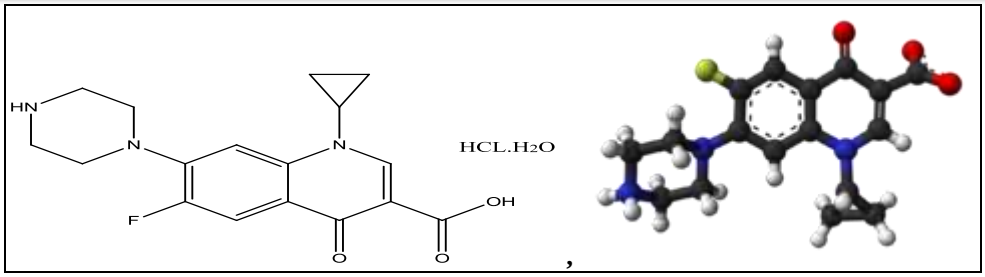
In this research, two drugs were bonded through amide and ester attachment, using lactic acid as aspacer binder, produced di pro drug such as Procain and Ciprofloxacin. Since Procain has ailocail anesthetic action and Ciprofloxacin as antibacterial drug was reacted with lactic acid produced ester compound (1), then the carboxylic acid of lactic acid could reacted with free Procain oil produced amide attachment, the controlled drug release in different pH values at 37C˚was studied to improve their characteristic and to minimize the side effect of the drug could be used in broad spectrum activity as atherapeutic material.This mutual prodrug was used with another biological active drug instead of single action. The prepared prodrug was characterized by FTIR, 1HNMR ,and UV. spectroscopies ,physical properties were determined and physical properties weremeasured.The biological assay were conducted for prepared prodrug using the microorganism such as E.coli, staphylococcus aureus, pseudomonas acuroginosoma, the prepared prodrug appear high biological activity,compared with standardGentamycin.

Keywords: Lactate, Ciprofloxacin, Procainamide.

INTRODUCTION

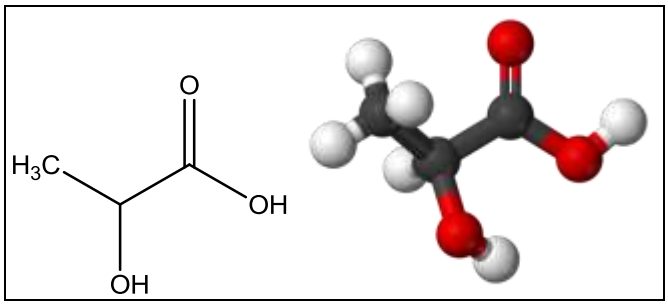
Mutual pro drug, where the carrier used is another biologically active drug instead of inert molecule. A mutual pro drug consists of two pharmacologically active agents coupled so that each acts as a promoiety for the other agent and vice versa [1, 2]. The carrier selected may have the same biological action as that of the parent drug and thus might give synergistic action .A Pro drug can be defined as "pharmacologically inert chemical derivative that can be converted *in vivo*, enzymatically and/or a chemical transformation, to the active drug that exerts the intended therapeutic effect(s) [3].The pharmacokinetics and behavioral effects of Procaine in the horse are of forensic importance because the presence of Procaine in the blood and urine of racing animals is forbidden by most racing authorities. The routes and forms in which Procaine may be administered to racehorses further complicate the pharmacokinetics of Procaine [4,5.6]. Thus, Procaine may be administered subcutaneously or intramuscularly for its local anesthetic action associated with minor surgery. Other possible uses of Procaine are regarded with disfavor by racing authorities [7].

Ciprofloxacin hydrochloride (USP) is the monohydrochloride monohydrate salt of Ciprofloxacin. [(IUPAC) name: 1-Cyclopropyl-6-fluoro-1,4-dihydro-4-oxo-7-(1-piperazinyl)-3-quinolinecarboxylic acid]= $C\ H_{18}FN_3O_3$. It is a faintly yellowish to light yellow crystalline substance with a molecular weight of 385.8 g/mol. Its empirical formula is $C_{17}H_{18}FN_3O_3HCl{\bullet}H_2O$. [8, 9]Quinolones are bactericidal and inhibit DNA Topoisomerases II, specifically DNA Gyrase. These enzymes are responsible for negative supercoiling of DNA, which is important in the packing of DNA as well as replication and transcription. [10]



Ciprofloxacin Hydrochloride

Quinolones are bactericidal and inhibit DNA Topoisomerases II, specifically DNA Gyrase. These enzymes are responsible for negative supercoiling of DNA, which is important in the packing of DNA as well as replication and transcription. [11, 12].



L-Lactic acid

L-Lactic acid, IUPAC name 2-Hydroxypropanoic acid = HO-CH (CH_3)-COOH, is an alpha-hydroxyl acid, which comprises two functional groups that undergo a condensation reaction by releasing water. This feature is particularly relevant, as it ensures stoichiometric concentration of both carboxyl, –COOH, and hydroxyl, –OH; ends groups [13].Lactic acid is a compound that plays a key role in several biochemical processes. For instance, lactate is constantly produced and eliminated during normal metabolism and physical exercise. Lactic acid has been produced on an industrial scale since the end of the nineteenth century and is mainly used in the food industry to act, for example, as an acidity regulator, but also in cosmetics, pharmaceuticals and animal feed [14, 15].

The objective of this work aimed to synthesis of mutual pro drug contain two drugs such as Ciprofloxacin and Procain which they bonded through lactate as spacer which has difunctional ester-amide, in vitro study was carry out in different pH values at 37˚C

EXPERIMENTAL WORK

Materials and Instruments

Lactic acid was purchased from BDH, Ciprofloxacinand Procaine were pruchased from Alderich, and DMF was obtained from Merck.All chemical materials were used with out further purification.

FTIR spectra were recorded by a4300 Shimatzu Spectrophotometer.UV-Shimatzu recorded VIS.Spectra.1H NMR spectra were recorded on Shimadzu Spectrophotometer in DMSO. Melting point were determined on CallenKamp MF.B-600 melting point apparatus.

May. 2015. Vol. 6. No. 5 ISSN2305-8269
International Journal of Engineering and Applied Sciences

www.eaas-journal.org

Ascending TLC was run on silica gel F254, pre-coated aluminum sheets. The final products and their intermediates were detected by irradiation with UV light by using the UV light detector (254). The Chromatograms were eluted by the mixed solvent

Controlled release study [16]

A 100mg of prepared prodrug(2) was kept in acylinder containing of 100ml of buffer solution at 37 ˚C without stirring .the sample was periodically withdrawn and analyzed by UV.Spectrophotometer at suitable (λmax) for every prepared sample to determine the amount of the released of Ciprofloxacin and Procainas prodrug, directly from the software built for many times using different pH values at 37˚C

Synthesis of Ciprofloxacinlactate (1)[1]

Ciprofloxacin acyl chloride was prepare firstly by using Thionyl chloride [18]

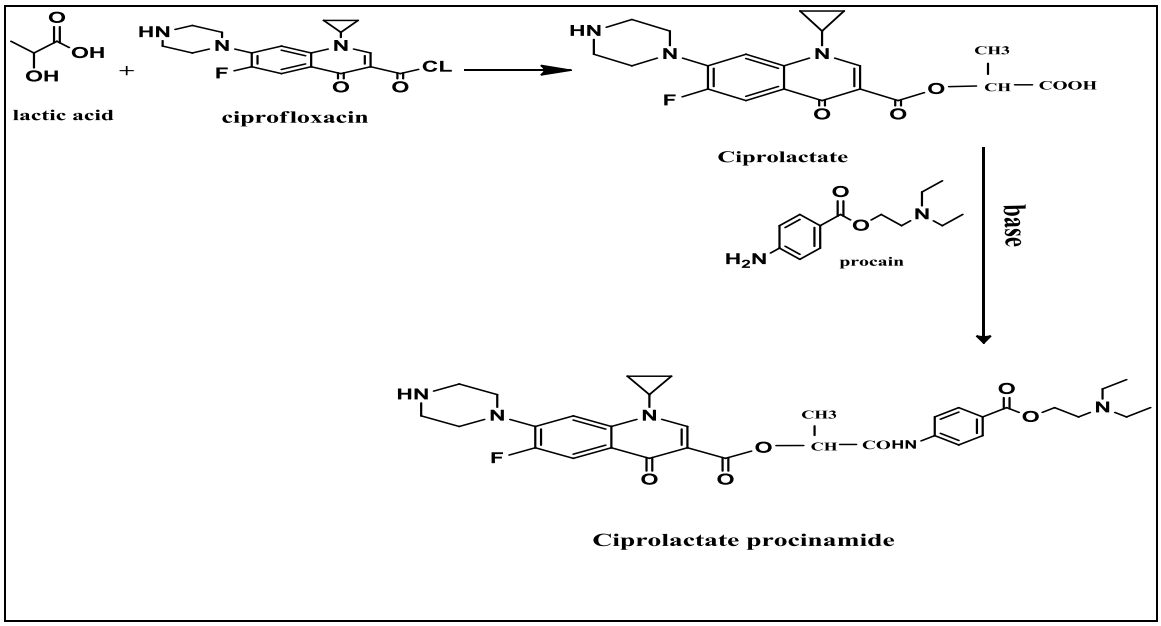
(2g, 0.036 mole) of Ciprofloxacin acyl chloride was dissolve in 5ml Dioxane, iiiiand (3.34g, 0.036 mole) of lactic acid was added gradually with continuous stirring about 1 hr. In the presence of tri ethylamine, the solvent was evaporated, the product was washed with ether and dried at room temperature,and the yield was 75% as a viscous material, RF =0.66in Di chloromethane

Synthesis ofCiprofloxacinlactate Procaine amide(2)

3 gm of Procain HCL was converted to free amine by adding NaHCO3 5% solution gradually until all CO2 gas was liberated, the oil phase was extracted by 10 ml of chloroform. (2.3 g, 0.036 mole) of Procain, oil was added to dissolve (2g,0.036 mole) of Ciprofloxacin lactate (1) in 10 ml of Dioxan, the mixture was stirred about 2 hr. The solvent was evaporated, the yellowish product was obtained, washed and recrystallized from ethanol. 80% yield was collected, Rf=0.59in chloroform.

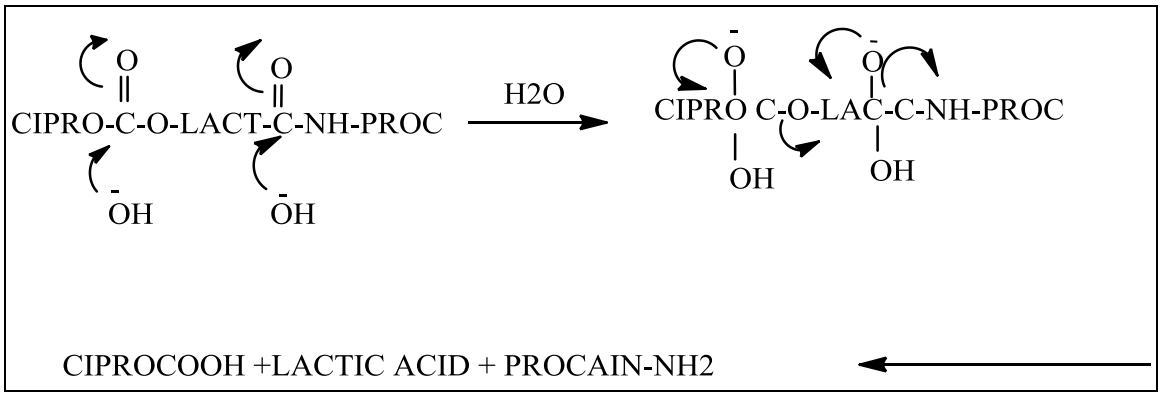
Result and discussion

Procain lactamide(1) was prepared by reaction of free amine Procaine oil with lactic acid produced amide derivative (1) , then the free hydroxy group of lactic acid was esterified with Ciprofloxacin acyl chloride produced derivative (2) , as shown below :-



Scheme 1

The prepared pro drug (2) contains two pharmacologically activity agents such as Procain has local anesthetic and Ciprofloxacin was used as antibacterial drug , the lactic acid was comprises two functional groups that undergo a condensation reaction by releasing water . Lactic acid is compound that play a key role in several biochemical process. For instance, lactate is constantly produced and eliminated during normal metabolism and physical exercise, also the two active agents coupled together by lactic acid could hydrolysis and release as the same biological action as parent drug with sustained release in specific site to prevent any side effect, with longterm drug delivery and highly desirable situation, because it could analysis through amide and ester groups in different pH value at 37 ˚c. The hydrolysis was explained as shown in the following Scheme (2):-



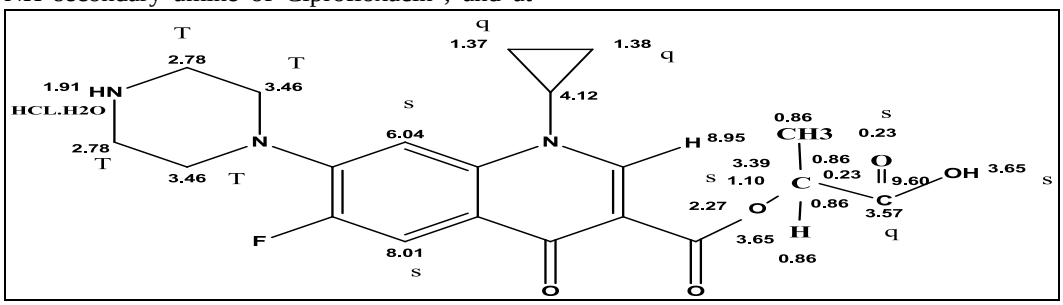
Scheme (2) hydrolysis of amide and ester groups of prodrug(2)

FTIR spectrum Fig (1) showed the beak at 3410 cm-1 assigned to the OH alcoholic of lactic acid, the other beak appeared at 2949 cm-1 of C-H aliphatic and 3160 cm-1 represented to CH aromatic, 1635 cm-1 represented to C=O of cyclic group in Ciprofloxacin, and the other was observed at 1714 cm-1 due to carbonyl of lactate ester, and at 3500-3000 cm-1 due to ammonium HCL of Ciprof loxacin.

FTIR spectrum ,Fig (2) of Ciprofloxacin lactate Procain amide (2) show the band at 3350cm-1 of NH secondary amine of Ciprofloxacin , and at 3207cm-1 is attributed to NH of Procain amide , the stretching vibration band at 3100 and 2980 cm-1 were assigned to CH aromatic and aliphatic respectively.

At 1710cm-1 and 1637cm-1 were assigned to carbonyl ester and amide respectively. The band appeared at 1394cm-1 is indicated the C-F of Ciprofloxacin as shown in FIG (1) and (2).

1H-NMR spectrum Fig (3) showed the signals of compound (1) indicated the presence of the following signal at structure below:

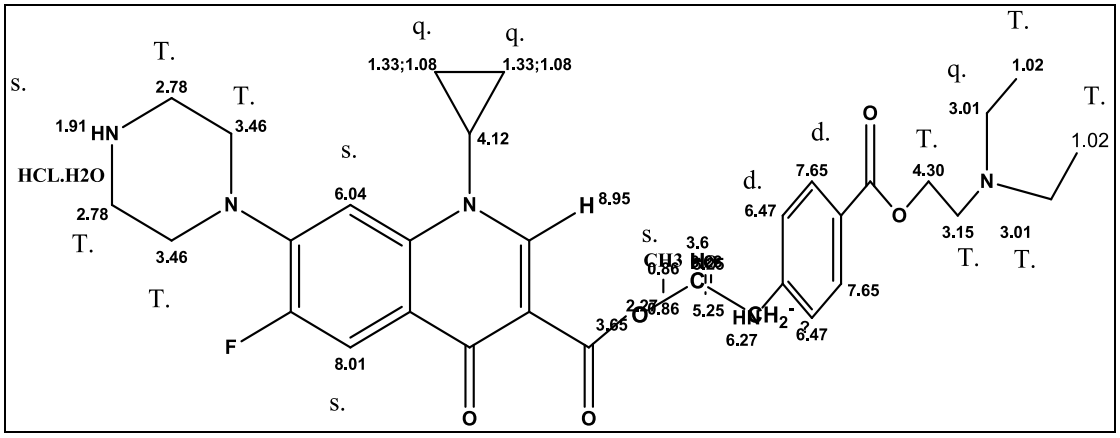


1HNMR signals of Cipro lactate (1)

May. 2015. Vol. 6. No. 5 ISSN2305-8269
International Journal of Engineering and Applied Sciences

www.eaas-journal.org

1H-NMR spectrum pro drug (2) as shown in Fig (4) indicated the signals as explained in structures below:



1HNMR signals of Ciprofloxacin lactate Procainamide (2)

Antibacterial assay

The inhibitory effect was done for prepared Ciprofloxacin lactate Procainamide pro

drug in the growth medium at concentration mg/mL of nutrient agar in petri dishes. The agar was inoculated the bacteria plugged out front old culture of *E .coli, staphylococcus and pseudomonas aecuroginosa*, on nutrient agar the plate incubated at(37C˚) and the colony was estimated by measuring perpendicular diameter of colony ,were compared with Gentamycin, the analysis of variance to show the statistical significance of the data as shown in certificate analysis ,it appeared high biological activity, it inhibit the growth of gram negative bacteria with high effective of used as antibacterial medicine,[22] also it is suggested that may play as an important role in antimicrobial activity, with high successfully control with more development as a new derivative.(2)

The difference in anti-bacterial activities of the investigated **Ciprofloxacin lactate Procainamide** and their control drug (Gentamycin) were studied and the results are presented in **table-1**. and the certificate of analysis was listed as shown below:

Table-1 :Minimum inhibitory concentrations of investigated Ciprofloxacin lactate Procainamide and the Gentamycin **drug (in** mg/mL**)**

Bacteria	Inhibition Zone Diameter	
	Extract mg/mL	Positive control (mm)
E. coli	38	14
staphylococcus	42	18
pseudomonas aecuroginosa	41	17

The cursory view of the data indicates the following trend in antibacterial activity of the substances under investigation: *staphylococcus pseudomonas aecuroginosa* ***E. coli***

CONCLUSION

In this work the lactic acid was used as a spacer bioactive material with other two agents with different active drugs Procain could use for local anesthetic action and Ciprofloxacin as antibiotic effect , also the lactic acid as it is known play a key role during metabolism and physical exercise . This new derivative was used and characterized; it was found a high biological activity with respect to Gentamycin. The hydrolysis of ester and amide groups were carried out in basic medium pH 7.4 are higher than acidic medium pH 1.1, this attributed to more nucleophile attack on OH^- alkaline carbonyl groups than water molecule and H^+proton..

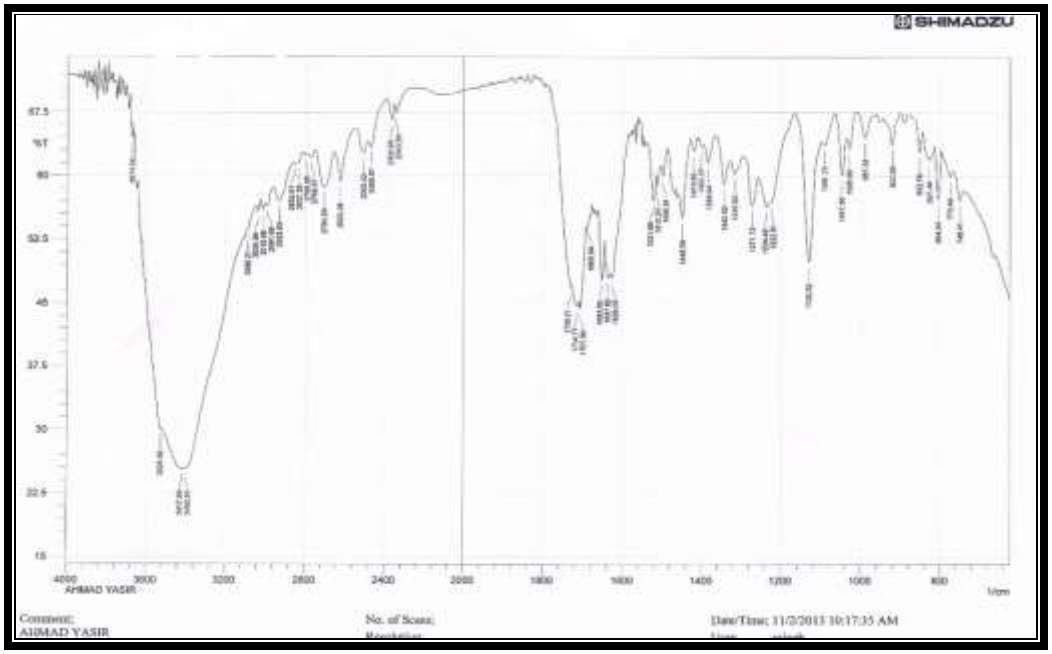


Fig (1) FTIR Spectrum of Cipro-lactate(1)

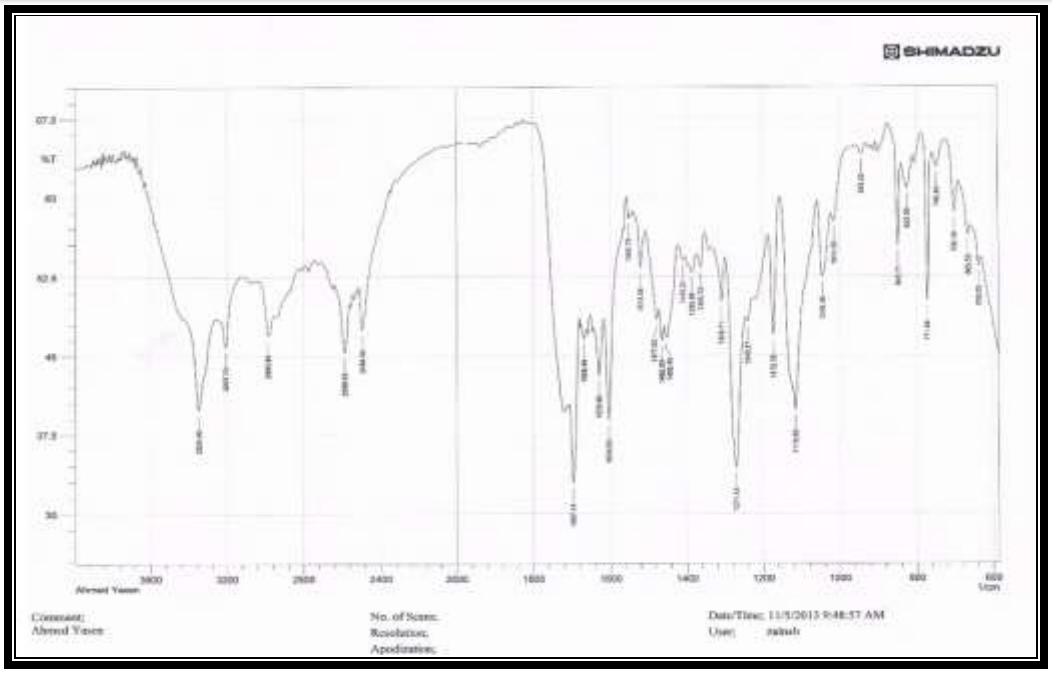


Fig (2) FTIR of Ciprofloxacin lactate Procainamide (2)

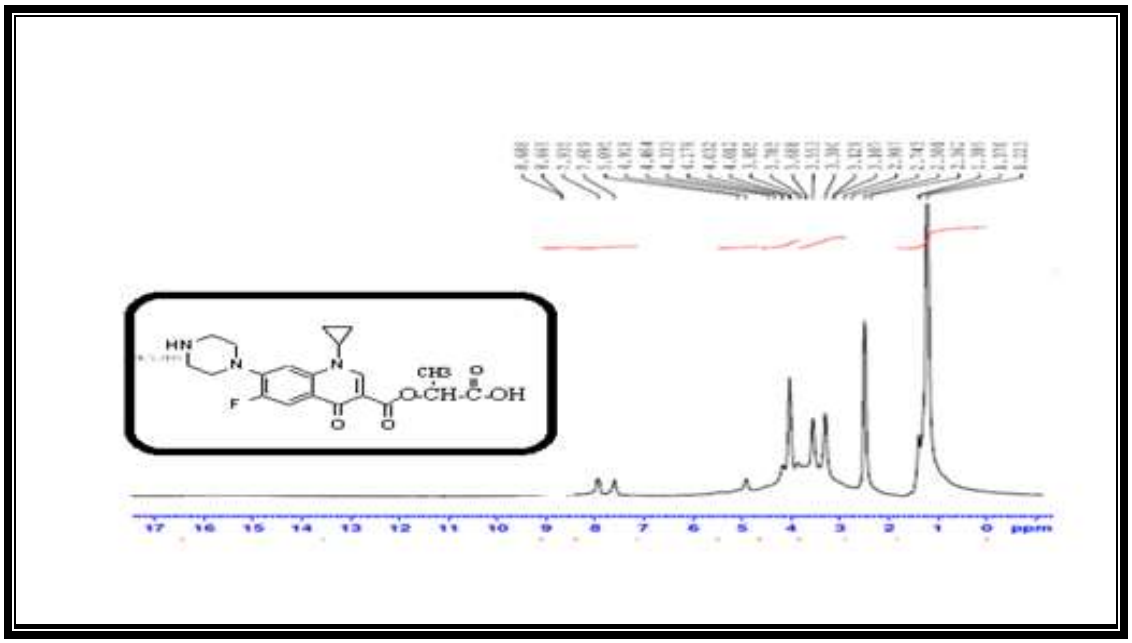


Fig (3) 1H-NMR Spectrum of Ciprolactate (1)

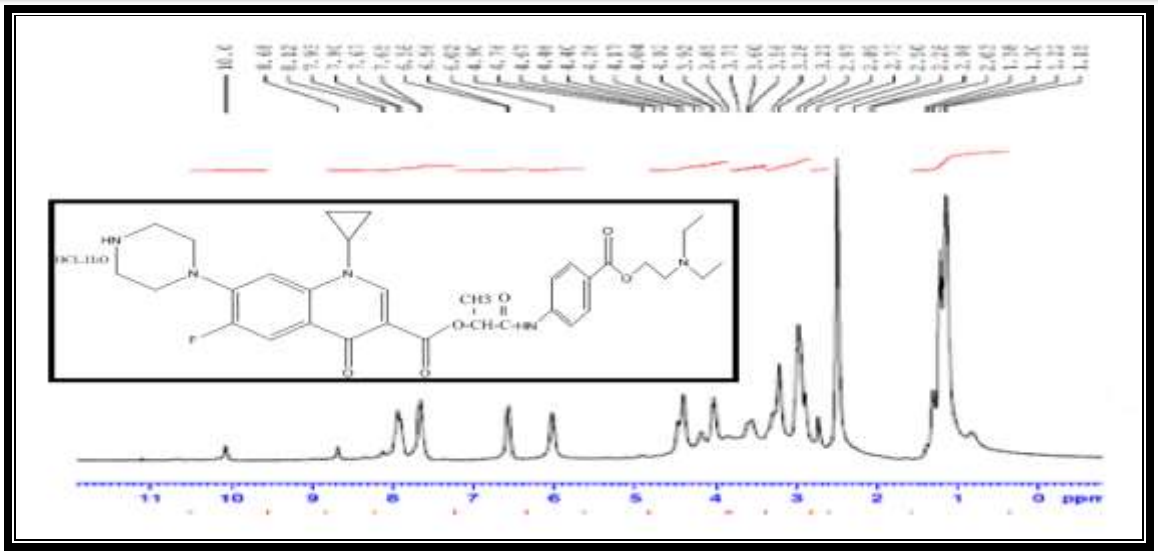


Fig (4) 1H-NMR Spectrum of Ciprofloxacin lactate Procainamide (2)

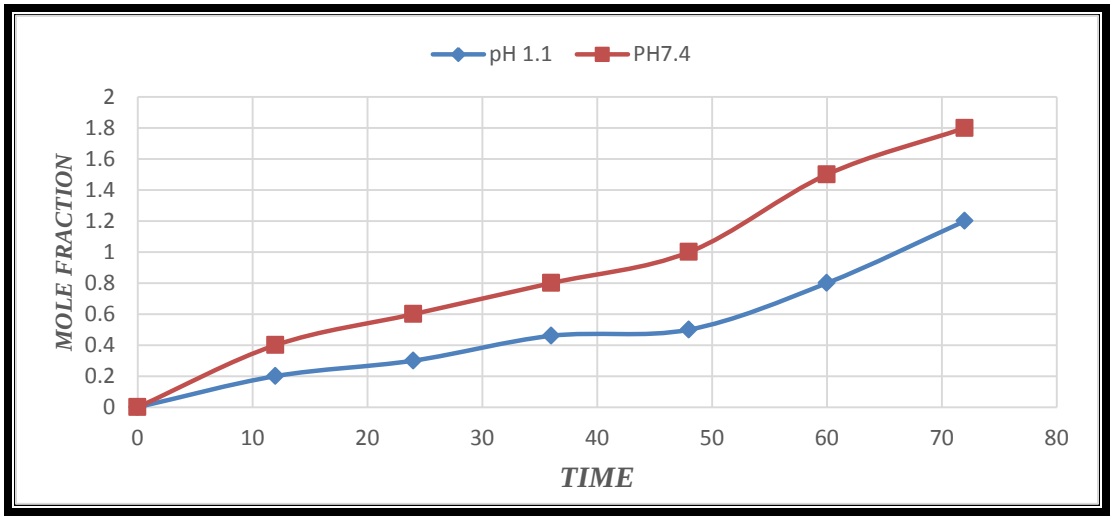


Fig (5) Controlled drug release of a mutual prodrug

ACKNOWLEDGMENT

With deep sense for respect and gratitude, we express out thanks to college of pharmacy,Almustansiriya University

REFERENCES

1. Bhosle.D, Bharambe.S, Gairola.N, Dhaneshwar.S.**Mutual prodrug concept: Fundamentals and applications**. Indian Journal of Pharmaceutical Science.2006;68(3): P 286-294
2. Yasser F .**Synthesis and in vitro kinetic study of new mutual prodrugfor colon cancer associated with constipation**. Tikrit Journal of Pharmaceutical Sciences. 2012; 8(1)35.
3. Patil S., ShiroteP. **Prodrug Approach: An Effective solution to overcome side effects**. International Journal of Medical and Pharmaceutical Sciences, 2011; 1 (7).
4. Takahiro S. , Toru E. , Toyoshi Y. , Hirotaka D. , Masaru I. **Stimulatory**

Effect of Procaine on the Growth of Several Microalgae and Cyanobacteria. Journal of Pharmacy and Pharmacology.2000; 52(2): p 243–251.

5. Natasha L. Kuchembuck .M, Patrick T. Callahan, D, Keith D. , David A. P, Kirsten W., Cynthia A. **Plasma concentration and local anesthetic activity of procaine hydrochloride following subcutaneous administration to horses**. 2007; 86(5).
6. Sinnott C, Cogs w, Johnson A, et al. **On the mechanism by, which epinephrine potentiates lidocaine's peripheral nerve block? Anesthesiology** 2003; (98):p181–188
7. Seifen A, Ferrari A, Seifen E, Thompson D, Chapman J.**Pharmacokinetics of intravenous Procaine infusion in humans**.Anesth Analg. 1979 Sep-Oct; 58(5):p382-6.
8. **Ciprofloxacin. In clinical pharmacology Gold Standard/ Elsevier** (2008Nov22).
9. **Cipro Labeling Revision 04/06/2009 Supplement 073**". US Food and Drug Administration. 6 April 2009. Retrieved 8 September 2009
10. Montagnac R, Briat C, Schillinger F, Sartelet H, Birembaut P,Daudon M. 2005. **Fluor quinolone induced acute renal failure**. General review about a case report with crystalluria due to Ciprofloxacin. Nephrol Ther 2005;(10): 44–51.
11. **European Directorate for the Quality of Medicines & Health-Care**. 2008. European Pharmacopoeia, 6th edition. Strasbourg,France: Council of Europe
12. 1**Pharmacopoeia of India, Ministry of Health and Family Welfare**, 2007;(2)p 938.
13. Dina A. , Susana J, Cristina M.. G. Baptista, M.. Gil. **Poly (lactic acid) Synthesis in Solution Polymerization.** Macromol. Symp. 2010;(296): 63–71 .
14. L. Avérous, **Polylactic Acid, Synthesis, Properties and Applications**, CH.21.indd 435 (2011).
15. Whiteman, N., DeLassus, P., and Gunderson, J., **New flavor and aroma barrier thermoplastic, polylactide, Polyolefins**, International Conference on polyolefins, Houston Texas USA.2002; 24-27 .
16. Marcin S.**Synthesis and characterization of polyester conjugates of Ciprofloxacin.European Journal of Medicinal Chemistry**.2010; 45(9): P 3844–3849.
17. Jian B, Chun S, Fen L, Xin Y and Ming X, **Rapid determination of Ciprofloxacin lactate in drugs by the Rayleigh light scattering technique**. Measurement Science and Technology.2007;18(3).
18. Yusuf M., Inas S, Ashok K., Rula M. andRana A.**Synthesis and Antibacterial Properties of New8-Nitrofluoroquinolone Derivatives**.Molecules,2007.
19. Sandhya A. Maratha, Rupesh Kumar, Parthasarathi Ajitkumar, Valakunja Nagaraja and Dipshikha Chakravortty.Curcumin **reduces the antimicrobial activity of ciprofloxacin againstSalmonella Typhimurium and Salmonella Typhi**. Journal of Antimicrobial Chemotherapy Advance Access published October 15, 2012
20. Getu K, and Awot G.**Quality Assessment of the Commonly Prescribed Antimicrobial Drug, Ciprofloxacin Tablets, Marketed in Tigray, Ethiopia**.Ethiopian Drug Administration and Control Authority.2010; 2 (1): 93-107
21. Brown KA, Khanafer N, Daneman N, Fisman DN; Khanafer; Daneman; Fisman (May 2013). **"Meta-analysis of antibiotics and the risk of community-associated Clostridium difficile infection**". Antimicrob. Agents Chemother. 57 (5): 2326–32.