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Evaluation of Physical Characteristic in Ciprofloxacin Tablet Dosage Form and Quantitative Analysis by UV-Spectrophotometry and HPLC

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Abstract

Ciprofloxacin is used as an active ingredient in different dosage forms, including tablet, which is used for treatment of various infectious diseases. Quantitative determination was performed by UV/Visible spectrophotometer and high performance liquid chromatography. Evaluation of the physicochemical property of Ciprofloxacin was performed by various methods: weight uniformity test, friability, hardness, thickness, diameter, disintegration, dissolution test loss on drying and Karl Fischer titration. The results have shown that the percentages of the assay witch determined by UV/Visible spectrophotometry and HPLC are complying with United State Pharmacopeia. The results for weight variations, diameter, thickness, hardness, friability, water content, disintegration and dissolution are within the normal range. In addition, there is no significant difference between core and coated tablets. Ciproneer tablets (Ciprofloxacin, Pioneer, Iraq) of this batch can be marketed and prescribed for the patient because it is safe and effective.

Introduction:

Qualitative and quantitative analysis have a vast importance field of pharmaceuticals [1]. Physical characteristics applied to all dosage forms that are directly or indirectly affect the effectiveness and safety of the products [1]. Oral tablets have different size, weight, hardness, thickness, disintegration and dissolution, according to physicochemical characteristics and process of formulations [2-3]. The advantages of tablet dosage forms are: administration is easy and convenient for patients, inexpensive, easy formulation, more stable than liquid dosage forms and accurate in dosing [4, 5].

Ciprofloxacin (CIP) is a fluoroquinolones derivative has an important effect in the treatment of serious infections, multidrug-resistant tuberculosis, pseudomonas aurogenosa and *E.coli* [6, 7]. The structure of CIP consist of; 1-Cyclopropyl-6-fluoro-4-oxo-7-(piperazin-1-yl)-1, 4-dihydroquinoline-3-carboxylic acid [8]. The molecule of Quinolones (CIP) contains three types of rings: a naphthyridine, Quinolone nucleus with one N atom in position 1 and the piperazinyl group at C7 position [9]. In the present of (F) and (N) atoms which has π- electronic transition so delocalization occur at aromatic position. This phenomenon leads to hyper polarization and interact with a polar solvent as “like dissolve like”. This compound has carboxyl and amine groups with pKa (5.7-6.3 and 7.6-8.3) [8, 9]. The two aryl alkyl amines which are weak base do not undergo acid- base reaction in physiological fluid [8-10]. This characteristic gives an idea for the medicinal chemist for deciding the best solvent during preparation and identification or quantitative determinations [11]. Physicochemical property also has an important role in determining of dissolution and absorption *in*

vivo. In addition, CIP has some lipophilic characteristics that help to pass through biological membranes and easily reach the side of action [12, 13].

There are some assessment of quality, quantity and bio-availability according to compendial and non-compendial methods [14, 15]. The general assay of CIP tablet includes quantitative determination for ensuring that the drugs contain the labeled amount [16, 17]. CIP can be easily analyzed by a UV/visible detector, which is used in UV-spectrophotometry and HPLC [8, 18, 19]. Chromophore group present in the structure of CIP, which is unsaturated chemical bond formed by sharing electrons *(Figure: 1)* [8, 20]. These systems responsible for electronic absorption because it contain double bonds present as a highly conjugated system, which absorb light at λ 276-278 nm [7, 8].

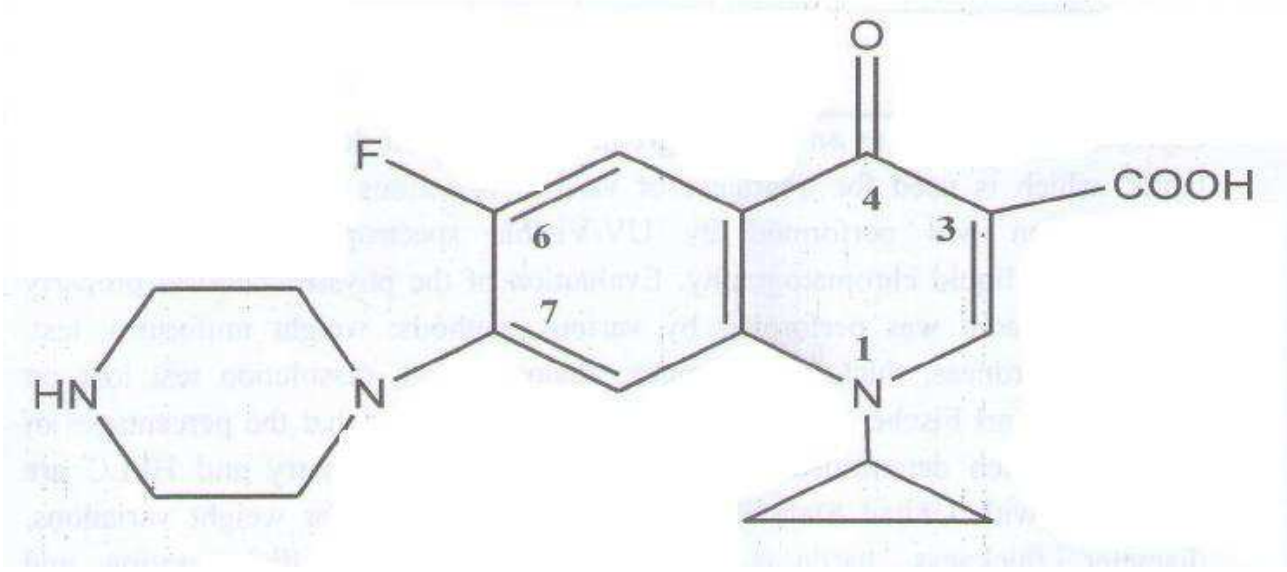

Figure-1: Structure of Ciprofloxacin

HPLC and UV/Spectrophotometry are widely used technique for determination of the amount of CIP. HPLC method is most official in United State Pharmacopeia (USP), British Pharmacopeia (BP) and Indian Pharmacopeia (IP) for analysis of CIP [8, 21]. The column that mostly used for analysis of CIP is reverse-phase C18 packed with silica [7]. The disadvantage of silica alone is that it interacts with quinolone group and cause peak widening and poor resolution [22].

Physical characteristics of tablet can assess the quality of drugs by: uniformity test, friability and variation in dimension [19]. Moisture content and residual solvent can be determined by loss on drying (LOD) and Karl Fischer (KF) tests that has an effect on half-life of drugs [23, 24]. The evaluation of bioavailability tests of tablet are dissolution and disintegration tests [25].

Materials and Methods

Quantitative Analysis of CIP:

- *HPLC method*

Solution A: consist of 0.025 M of orthophosphoric acid 85 % (Scarlab Company, Spanish) and was adjusted to pH 2 ± 0.1 by tri-ethylamine (Merck, Germany). Solution B: contains 13:87 (v/v) acetonitrile (ACN, Merck, Germany): solution A. Solution C: consist of 0.025M of phosphoric acid adjusted with tri-ethylamine to pH 3 ±0.1 and followed by filtration. Standard solution was prepared by dissolving 23.75 mg of CIP hydrochloride (working standard, 93.9%, USP, in USA) in 100 ml of solution B. Sample solution was prepared by dissolving 5 tablets of CIP (Ciproneer, Pioneer Company, Sulaimanyah, Iraq) in 500 ml of solution B, and sonicated for 20 min. The final prepared solution mixed and passed through filter paper with pore size 0.45µm, to remove any excipient contain in the sample. Four milliliter of this solution was diluted with solution B to give a final concentration of 0.2 mg/ml. The chromatographic conditions: Reverse phase chromatography, C18 column (4.6 mm×25 cm), 5 µm of pore size. Mobile phase: ACN: solution C (13:87, v/v), 1.5 ml/min flow rate, 10 µL injection volume. Detection was carried out by reading absorbance at 278 nm using HPLC with UV detector (Agilent technology 1200 series, Agilent Company, Germany). The

sample solution was injected 3 times and standard solution was injected 5 times independently by HPLC [26,].

- *UV/Visible-Spectrophotometry method*

Sample solution was prepared using 5 CIP (Ciproneer) tablets and dissolved in 1 L of 0.1 M HCl (Merck, Darmstadt, Germany). Working solution was at concentration of 0.005 mg/ml. The standard solution was prepared from CIP hydrochloride (working standard, 93.9%, USP, in USA) and dissolved in o.1 M HCl to have a final concentration of 0.00476 mg/ml. The absorbance was measured by UV-Spectrophotometry (Spercord 50, Analytik Gena, Germany) at a single wavelength of 276 nm. Each sample and standard was repeated reading three times independently [13, 26].

Evaluation of the quality of tablets:

- *Weight variation test*

Twenty tablets from a batch of Ciproneer tablets were weighed individually by sensitive analytical weighing balance (Sartorius balance, Germany). The average weight of the core and coated tablets was then calculated [27].

- *Hardness test*

Ten tablets were randomly selected from the core and coated tablet of one batch. The hardness was measured manually by hardener machine (Break force, Dr. Schleuniger Pharmatrone company, USA). This hardness value is obtained by unit of kPa (kilopascal***)*** [28].

- *Friability test*

Ten tablets for each batch were randomly selected and weighed. The tablets were placed into Friability tester (Dr. Schleuniger Pharmatrone Friabilator´s Company, USA). The friabilator was operated at 25 rpm for 4 min, the tablets were weighed again and the percentage of friability was then calculated to determine the percentage of weight loss [29].

- *Thickness and dimension tests*

Thickness and diameter tester (Dr. Schleuniger Pharmatrone machine, USP) were used to measure the thickness and diameter of 10 core and coated tablets. The percentage of deviation for thickness and diameter was then calculated [30].

- *Loss on drying (LOD)*

Prior this test, the melting point of CIP tablet was determined to insure that the temperature used for loss on drying test is below the melting point (decomposing point) of the tablets. LOD was then determined by Sartorius (MA150 moisture, Sartorius Company, Germany). Start heating the device below the melting point of CIP at 100 ±2°C. Then continuously the weight was measured at various times (10, 20, 40, 60, 80, 100, 120, 140 and 160) min [31].

- *Karl Fischer test*

This test was determined by KF instrument (870 KF Titrino plus, Metroham Autolab Company, Switzerland). The amount of water consumed by Karl Fischer reagent was determined.

In Vitro bioavailability tests:

- *Disintegration test*

Randomly six tablets of CIP were selected from one batch and each tablet placed in different tube and covered with 900 ml of distilled water at 37±0.5°C. The basket was moved upward and downward at 29-32 cycles / min. The time necessary for the complete disintegration of each tablet was recorded using disintegration instrument (Pharmatest's Company, Germany) [32].

- *Dissolution test*

The dissolution profile was performed for the ciprofloxacin coated tablet at various times (15, 30, 45 and 60) min using dissolution tester (paddle method, Pharmatest Company, Germany). The dissolution rate for six coated and core CIP tablets were performed at 30 min. The medium used for this test was 900 ml of 0.01M HCL maintained at 37±0.5°C and 50 rpm. Five milliliter of the sample was withdrawn from the

apparatus by sampling tube at desired times; and the volumes of the medium were maintained constant. The absorbance read by UV-Spectrophotometry at 276 nm. The working standard solution used in this test was at the concentration of 0.0047 mg/ml. Each sample and working standard solution was repeated three times independently [33].

Results and Discussion

Quantitative analysis of CIP:

- *HPLC method*

HPLC chromatograms were showed excellent identification of sample (CIP tablet) and standard (CIP hydrochloride) solutions without interfering peaks *(Figure: 2 and 3).* Peak was identified by direct comparison of retention times with a single standard. The peaks were very sharp and the symmetry value was less than 1. The retention times under the selected conditions of CIP hydrochloride reference standard and CIP tablet were 15.72 and 15.77 min respectively.

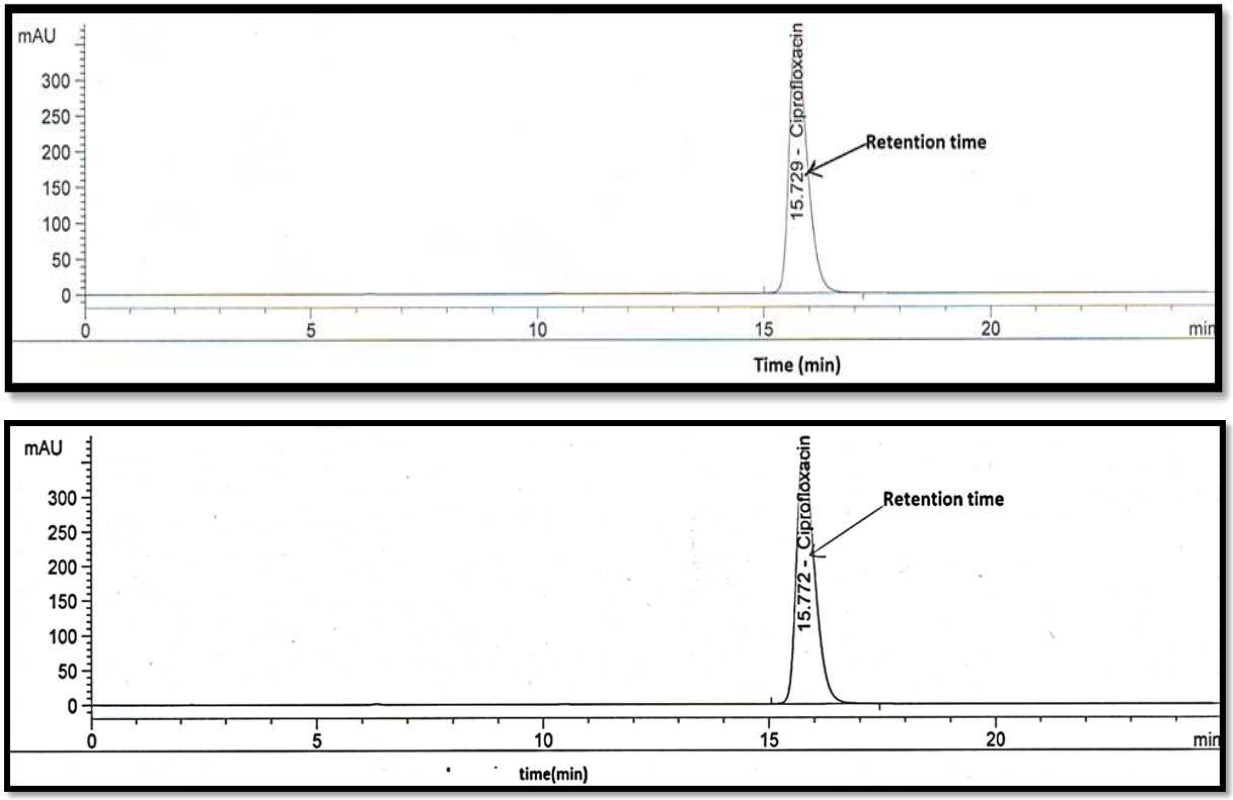

Figure-2 Illustrates the identification of 0.2375 mg/ml CIP hydrochloride (reference standard):: Retention time of 15.72 min. C18 column (4.6mm×25 cm), 5 µm of pore size. Mobile phase: ACN: solution C (13:87, v/v), flow rate 1.5 ml/min, injection volume 10 µL. UV/ Visible detector at 278nm, using HPLC.

Figure-3: Illustrates the identification and quantification of 0.2mg/ml CIP tablet. Retention time at 15.77 min. C18 column (4.6 mm ×25 cm), 5 µm of pore size. Mobile phase: ACN: solution C (13:87, v/v), flow rate 1.5 ml/min, injection volume 10 µL. UV/ Visible detector at 278nm, using HPLC.

The percentage of assay and standard deviation of CIP was104.061% ± 0.558 and it is in an acceptable limited range of USP (not less than 90% and not more than 110%).

- *UV/Visible-Spectrophotometry method*

The percentage of assay that found by UV/spectrophotometry was 102.16% ±0.25, which is in an acceptable range of USP [26].The structure of CIP shows that it contains a highly conjugated system that can be detected at visible region and does not need any reagent for analysis.

In this study both sample and standard solutions were prepared without complexion with ions and other reagents and determined at 276 nm, this in agreements with works reported by A. Kishore and P. Amareshwar [2]. E. Cazedey and coworkers reported that (Fe III) can be used for complexion with CIP and the result has been obtained at 386.4 nm (λ max) [34]. The result show complexion of CIP with ferric ion

lead to shifting of the λ max to higher value which is more accurate and eliminate all interference from excipient of the drug [34]. After the comparison no significant difference was observed between method without complexation and method with complexation. The results of both studies were so close to each other this mean CIP not need the complexion and there is no any interference with other reagents.

In addition, the spectrophotometric result was compared with HPLC result. Although HPLC is more accurate and more specific than UV/spectrophotometry, but the results of both methods were so close to each other.

Evaluation of the quality of tablets:

In his study the evaluation of weight variation, hardness, friability, thickness and diameter in CIP tablets of Ciproneer (500 mg) from pioneer company in Iraq was compared with Cipro-J (500 mg) from Nigeria:

- *Weight variation test*

The average weight variation of Ciproneer for the core and coated tablet was 747±1.15 mg and 731±1 mg respectively, therefore coating of the tablet does not have any effect on weight variation. All tablets of core and coated tablet were within the limited acceptable value according to USP <905> [26, 35]. The standard percentage of deviation for each tablet (weighted more than 375 mg) should not be more than ± 5%.

The weight variation for uncoated tablet Cipro-J was 597±2.79 mg. Therefor; none of the tablets departs from the percentage of deviations but the deviation for Cipro-J was more than Ciproneer [14].

- *Hardness test*

The results for core and coated CIP tablet were obtained and comply with normal range of Brand Company (Cipro). The normal range for core and coated tablet should not be less than 15 kPa. The average strength and STD of coated and core Ciproneer tablets are more than the acceptable standard value which are 22.87±1.97 and 20.51±1.86 kPa respectively. As observed the coated tablet is harder than core tablet, this reveals that coating of the tablet increase the hardness. The hardness of Cipro-J was less than Ciproneer which is 11.3 kPa.

- *Friability test*

This value expressed in percentage and the standard value should be less than 1% w/w. The result showed that the percentage of friability of Ciproneer was 0% and for Cipro-J was 0.7. Therefore, Ciproneer and Cipro-J were complied with USP <1216> [14].

- *Thickness and dimension tests*

These two tests are non compendial method so the standard deviation range should compare to BP. The percentage of deviation of thickness for the tablets should not show higher than 5% [30]. Thickness and their relative deviation of core and coated tablets were 6.228 ±0.57 and 6.279 ±0.47 mm respectively. Coated and core tablets were complied with BP. Thickness was increased by coating, but there is no any significant change in percentage of deviations.

Change in diameter is less often happens because it is not changed by the force of compression. According to BP the percentage of deviations should not deviate by ±3% for a tablet with a diameter greater than 15 mm [30]. Therefore, the results were complied with normal range as shown in table 1.

Table-1: Summary of quality evaluation of Ciproneer core and coated tablets

	Qualitative evaluation of Ciproneer tablets			
Type of Ciproneer tablets	Average Weight (mg)±STD	Average Hardness (kPa)	Average Thickness (mm)	Average Diameter (mm)±STD
Uncoated tablets	731±1.2	20.51±1.86	6.27±0.58	18.04±0.15
Coated tablets	741±1.15	22.87±1.97	6.228±0.47	18.13±0.2

- *Loss on drying (LOD)*

This test is to evaluate the water content as a source of humidity or excipient used during tablet preparation. In this test the temperature used should be lower than the melting point of CIP (253°C),

therefore the heating starts from 100°C. As the temperature increases the water removal increase, because the moisture undergoes evaporation by heating *(Table: 1)*. Weight reduction, percentage of moisture content and loss on drying are shown in figures 4, 5 and 6.

Table-2: Demonstrate the changes in amount of water, moisture content and amount of weight loss of powdered CIP heated at 100°C.

Duration of heating (min)	Weight of sample	Moisture content %	Loss on drying %
0	2.117	0	0
10	2.068	2.36	2.31
20	2.06	2.76	2.76
40	2.054	3.067	3.067
60	2.049	3.31	3.21
80	2.044	3.57	3.57
100	2.039	3.82	3.68
120	2.037	3.92	3.77
140	2.035	4.03	3.895
160	2.034	4.08	3.92
180	2.034	4.08	3.92

The weight of the sample was decreased with an increase in duration due to removal of water in the sample *(Figure: 4)*. The results showed that the weight of sample was decreased from 2.117 g to 2.034 g, which is the dry weight of sample.

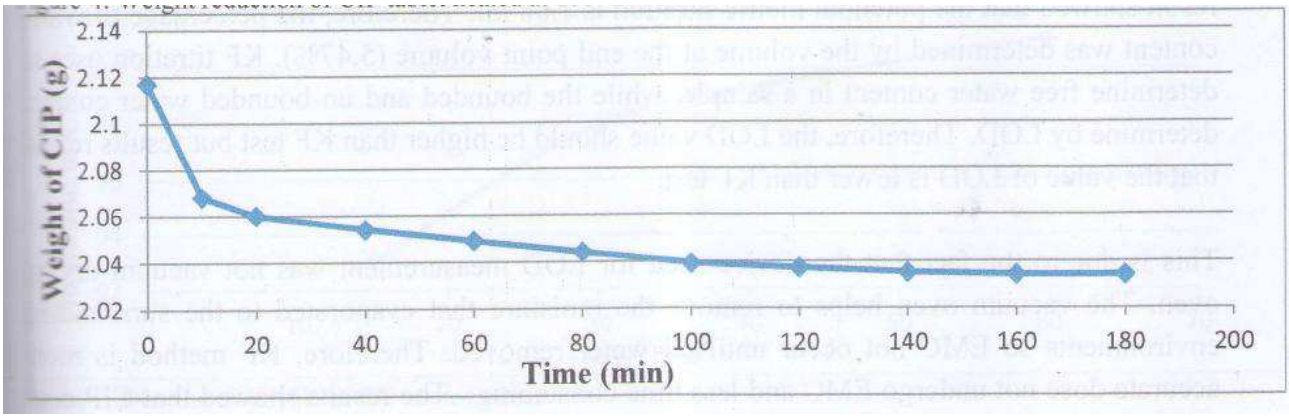

Figure-4: Weight reduction of CIP tablet heated at 100°C / min

Percentage of moisture content and loss on drying are shown in figures 5 and 6.

As heating times increase the percentage of moisture content and loss on drying were increased. The result of moisture content % and LOD % at the equilibrium point become 4.08% and 3.92% respectively. The more changing in results does not happen due to reaching the equilibrium point with surrounding medium called equilibrium moisture content (EMC) [36].

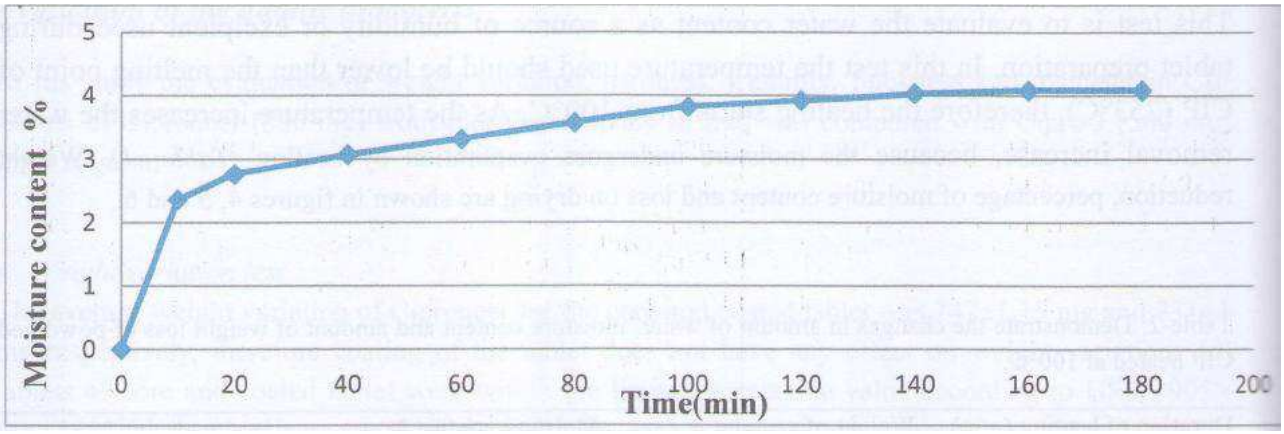

Figure-5: Illustrate percentage of moisture content in CIP tablet heated at 100°C at different time.

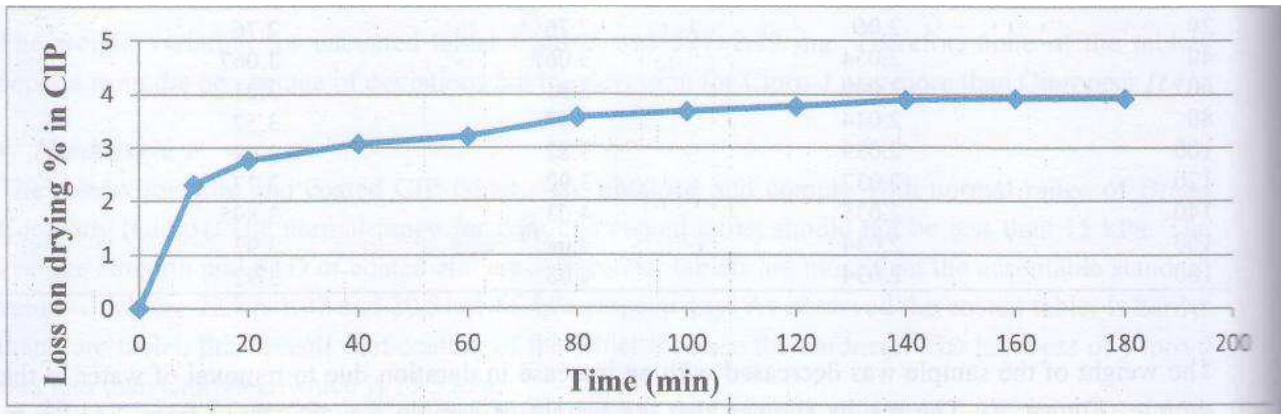

Figure-6: Illustrate the percentage of loss on drying in CIP tablet heated at 100°C at different time.

- *Karl Fischer test*

Free water content was determined by volumetric titration using Karl Fischer reagent. The result showed that the potential metric titration is 1.68 ml. Therefore, the percentage of water content was determined by the volume at the end point volume (5.47%). KF titration uses to determine free water content in a sample, while the bounded and un-bounded water content determine by LOD. Therefore, the LOD value should be higher than KF test but results reveal that the value of LOD is lower than KF test.

This is due to the fact that the device used for LOD measurement was not vacuum drying oven. The vacuum oven helps to remove the moisture that evaporated to the surrounding environments so EMC not occur until all water removed. Therefore, KF method is more accurate dose not undergo EMC and less time consuming. The results showed that CIP does not comply with LOD and KF methods according to USP [37] while, water content comply with BP standard value; it should be less than 6.7% [38].

In Vitro bioavailability tests

- *Disintegration test*

The average rate of disintegration was 2.27 ± 0.9 min which is less than disintegration time for Cipro-J [14]. The results demonstrate that all tablets are within limited value of USP <701>, (less than 30 min) [39].

- *Dissolution test*

The time required for maximum dissolution was determined for CIP coated tablet in 0.01M HCl *(Table: 3)*.

Table-3: Dissolution profile of ciprofloxacin coated tablet at different time duration

Dissolution time (min)	Dissolution %
15	93.035
30	97.99
45	99.77
60	101.07

The result demonstrates that the percentage of dissolution increase as the dissolving times increase *(Figure: 7)*.

The percentage of dissolution of CIP at 15 min was 93%, while at 60 min was 101%. This indicates that CIP is highly soluble in acidic medium of the stomach. The results indicate that there is no significant difference in the average dissolution rate of CIP coated (98.137 %) and core tablets (98.7808 %).

In immediate release tablet 80% of it should dissolve in 30 min according to USP <711> [26], but rate of dissolution of Cipro-J was 80% in 30 min which is less than dissolution rate of uncoated Ciproneer.

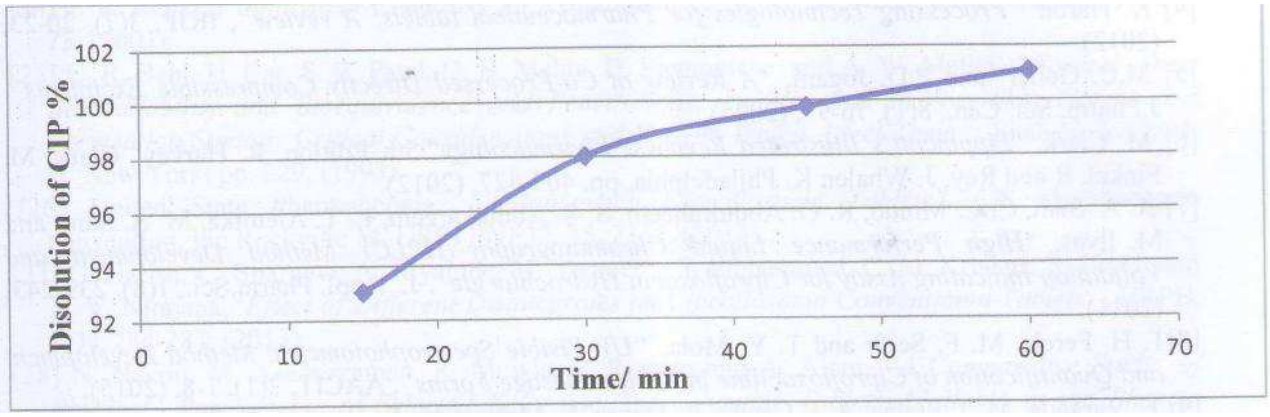

Figure-7: Dissolution profile of CIP coated tablet

In-Vitro dissolution of CIP is very important for predicting *in-vivo* absorption of CIP tablets [39]. Therefore, post marketing evaluation of *in-vivo* bioavailability is not necessary and it is very expensive [39].

Conclusion

This is an evaluation study for qualitative and quantitative determination (by HPLC and UV/ spectrophotometry) of CIP contain in a locally product (Pioneer Company) and comparing the result with that obtained from other product Cipro-J (Nigeria). The percentage of assay for both techniques was complied with USP. The results obtained showed that weight variations, diameter, thickness, hardness and friability for core and coated CIP tablets were complied with USP official specification. The water content of CIP was obeying BP. In addition, CIP tablets have a better bioavailability, which was determined by dissolution and disintegration tests, both tests was complied with USP national formulary- monograph. Therefore, Ciproneer tablets of this batch can be marketed and prescribed for the patient because it is more convenient and therapeutically effective than Cipro-J.

Acknowledgment

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Referencing

[1] A. Kasture , S. Wadodkar, K. Mahadic and H. More,*"Pharmaceutical Chemistry -I"*: 25th Edition. Abhyudaya Pragati, N. India, pp. 1-2, (2008).

[2] K. A. kishore and P. Amareshwar, *"Quality Evaluation and Comparative Study on Tablet Formulations of Different pharmaceutical Companies"*, J. Curr. Chem. Pharm. Sci., Vol.2, No.1, pp 24-31, (2012).

[3] A. Loyd, P. Nicholas and A. Howard, *"Ansel's Pharmaceutical Dosage Forms and Drug Delivery SystemsSolid Dosage Forms and Solid Modified-Release Drug Delivery Systems":* 9th Edition. Lippincott Williams and Wilkins, Philadelphia, pp. 232, (2011).

[4] K. Harbir *"Processing Technologies for Pharmaceutical tablets: A review"*, IRJP, Vol,3, No,7, pp 20-23, (2012).

[5] M.C. Gohel and P.D. Jogani, *"A Review of Co-Processed Directly Compressible Excipients"*, J.Pharm. Sci. Can., Vol,8, No,1, pp 76-93, (2005).

[6] M. Clark, *"Lippincott's Illustrated Reviews: Pharmacology":*5th Edition, R. Harvey Clark, M. Finkel, R and Rey, J. Whalen K. Philadelphia, pp 409-427, (2012).

[7] A. A. Sani, C. C. Mmuo, R. O. Abdulraheem, S. S. Abdulkareem, E. T. Alemika, M. A. Sani and M. Ilyas, *"High Performance Liquid Chromatography (HPLC) Method Development and Validation Indicating Assay for Ciprofloxacin Hydrochloride"*, J. Appl. Pharm.Sci.Vol.1, No.8, pp 239-243, (2011).

[8]T. H. Fereja, M. F. Seifu and T. Y. Mola, *"UV-Visible Spectrophotometric Method Development and Quantification of Ciprofloxaciline in Tablets Dosage Forms"*, AACIT, Vol. 2, No.1, pp 1-8, (2015).

[9] F. Varanda, M. J. Pratas, A. I. Cacüo, R. Dohrn, F. Makrydaki, E. Voutsas, D. Tassios and I. M. Marrucho, *"Solubility of Antibiotics in Different Solvents, 1.Hydrochloride Forms of Tetracycline, Moxifloxacin and Ciprofloxacin"*, Ind. Eng. Chem. Res., Vol.45, No.18, pp 6368-6374 ,(2006).

[10] F. William, L. Thomas and W. Daived, *"Foye's Principles of Medicinal Chemistry"*: 7th Edition. Williams and Wilkins, Philadelphia, pp. 32, (2012).

[11] A. I. CaÇo, F.Varanda, M. J. Pratas de melo, A. M. A. Dias and I. M. Marrucho, *"Solubility of Antibiotics in Different Solvents. Part II:Non-Hydrochloride Forms of Tetracycline and Ciprofloxacin"*, Ind. Eng.Chem. Res.,Vol. 47, No.21, pp 8083-8089, (2008).

[12] S. Harder, U. Fuhr, D. Beermann and A. Staib, *"Ciprofloxacin Absorption in Different Regions of the Human Gastrointestinal Tract Investigations with the hf-Capsule"*, Br. J. Clin. Pharmacol., Vol.30, No.1, pp 35-39, (1990).

[13] H. Hopkala and D. Kawalczuk, *"Application of Derivative of Uv-Spectrophotometry for the Determination of Ciprofloxacin, Norfloxacin, and Ofloxacin in Tablets"*: Acta Pol. Pharm., Vol.57, No.1, pp 3-31, (2000).

[14] N. C. Ngwuluka , K. Lawal, P. O. Olorunfemi and N. A. Ochekpe, *"Post-market in Vitro Bioequivalence Study of Six Brands of Ciprofloxacin Tablets/Caplets in Jos, Nigeria"*, Sci. Res. Essays., Vol.4, No.4, pp 298-305, (2009).

[15] S. Vinay, R. S. Bhat, V. Balamuralidhara and T. M. P. Kumar, "*Comparative Study of In-process and Finished Products Quality Control Tests of IP, BP & USP for Tablets*", IJPTP. Vol.2, No.4, pp 176-183, (2011).

[16] S. Nandipati, K. R. Vanka and S. Uba, *"The Separation and Quantitative Determination of Ciprofloxacin in a Pharmaceutical Formulation by Ultra Performance Liquid Chromatography"*, Int. J. Pharm. Pharm.Sci., Vol.5, No.3, pp 312-317, (2013).

[17] S. O. Sarr, S. M. Ndiaye, D. Fall, A. Diedhiou, A. Diop, B. Ndiaye and Y. M. Diop**,** "*Development and Validation of a Simple and Economical Spectrofluorimetric Method for Estimation of Ciprofloxacin in Pharmaceutical Dosage Forms"*, Int. J. Anal. Bioanal. Chem. Vol.3, No.3, pp 72-77, (2013).

[18] L. Levi, G. C. Walker and L. I. Pugsley, *"Quality Control of Pharmaceuticals"*, Can. Med. Assoc. J**.,** Vol.91, No.15, pp 781-785, (1964).

[19] G. Kahsay and A.G. Egziabher, *"Quality Assessment of the Commonly Prescribed Antimicrobial Drug, Ciprofloxacin Tablets, Marketed in Tigray, Ethiopia"*, MEJS, Vol.2, No.1, pp 93-107, (2010).

[20] L. D. S. Yadav, *"Organic Spectroscopy"*: Kluwer B. New Delhi, India, pp. 29, (2005).

[21] E. C. L. Cazedey and H. R. N. Salgado, "*Spectrophotometric Determination of Ciprofloxacin Hydrochloride in Ophthalmic Solution"*, Adv. Anal. Chem., Vol.2,No.6, pp 74-79, (2012).

[22] B. H. Kim, N. Hee Choi and J. Hoa Ok, *"Comparison of Reversed-Phase Liquid Chromatographic Methods for the Separation of New Quinolones"*, J. Chromatogra. Sci., Vol.40, No.7, pp 305-380. (2002).

[23] R. Nogueira, S. M. Queiroz, G. E. B. Silva,W. F.C. Rocha, G. F. Sarmanho, R. R. R Almeidaa and G. F. Moreira, *"Determination of Volatiles in Pharmaceutical Certified Reference Materials"*, J. Braz. Chem. Soc., Vol.23, No.9, pp 1636- 1646, (2012).

[24] J. V. Kenkel, *"Analytical Chemistry for Technician":* 3rd Edition. United State America, pp. 34-75, (2003).

[25] C. R. Behl, H. Car, S. B. Patel, D. B. Mehta, D. Piemontese and A. W. Malick, "*Topical Drug Bioavailability and Bioequivalence and Penetrations: In-Vitro and In-Vivo Skin Uptake and Permeation Studies: Critical Considerations and Factors Which Affect Them":* Shah V, Maibach H. New York, pp. 229, (1993).

[26] United State Pharmacopeia. *" Monograph Ciprofloxacin Tablets"*, US Pharmacopria convention. Inc Rockville M. USP37/NF32. Vol.35, pp 2355, (2015).

[27] G. Harish, C. Bhargavi, A. Riyajune, M. Yasmeen, S. Mohammed, C. M. Krishna, S. Jilani and A. V. Nirajana,*"Effect of Different Disintegrates on Ciprofloxacin Conventional Tablets"*, IJRPB, Vol.1, No.3, pp 284-287, (2013).

[28] N. Nasrin, M. Asaduzzaman, R. Mowla, F. Rizwan and A. Alam, *"A Comparative Study of Physical Parameters of Selected Ketorolac Tromethamine Tablets Available in the Pharma Market of Bangladesh"*, JAPS, Vol.1, No.8, pp 101-103, (2011).

[29] A. D. Salman, M. Ghadiri, and M. J. Hounslow, *"Particle Breakage: Handbook of Powder Technology-12":* Elsevier. London, pp.960, (2007).

[30] B. N. V. Hari, V. Abbirami, P. Sainithya, A.Shobana and D. R. Devi, *"In-Vitro Bioequivalence Study and Effect of Food Beverages on Various Brands of Aceclofenac Available in Indian Market"*, JCPS, Vol.7, No.1, pp 1-7, (2014).

[31] L. L. Augsburger, and S. W. Hoag *"Pharmaceutical Dosage Forms – Tablets: Unit Operation and Mechanical Property-1":* 3rd Edition. CRC, Taylore and Francis, New York, pp. 220, (2008).

[32] S. Usman, A. Alam, R.Suleiman, K. Awad and I. Abudeek, *"Evaluation of Dissolution Testing for Ciprofloxacin (500mg) Tablets: Post Market Surveillance of Different Brands Availability in Ras Alkhaimah (UAE)"*, I. J. B., Vol.5, No.1, pp 65-72, (2014).

[33] S. Agrwal and R. Khana, *"Physical Pharmacy: Diffusion and Dissolution"*, 2nd Edition. Sharma B. India, pp. 239, (2006).

[34] E. C. L. Cazedey, R. Bonfilio, M. B. Araújo and H. R. N. Salgado, *"A First-Derivative Spectrophotometric Method for the Determination of Ciprofloxacin Hydrochloride in Ophthalmic Solution"*, JPC, Vol.2, No.6, pp 116-122, (2012).

[35] J. Mu'az, K. L. Gazali, U. G. Sadiq and M. G. Tom, "*Comparative in Vitro Evaluation of the Pharmaceutical and Chemical Equivalence of Multi-Source Generic Ciprofloxacin Hydrochloride Tablets Around Maiduguri Metropolitan Area"*, Nig. J. Pharm. Sci., Vol.8, No.2, pp 102- 106, (2009).

[36] J. C. Callahan, G. W. Cleary, M. Elefant, G. Kaplan, T. Kensler and R. A. Nash, "*Equilibrium Moisture Content of Pharmaceutical Excipients"*, Drug Dev. Ind. Pharm., Vol.8, No.3, pp 355-369, (1982).

[37] P. Thomas, *"Physicians' Desk Reference 2004: Supplemen"*. 58th Edition. Thmson, England, pp.78, (2004).

[38] British Pharmacopeia. *"Monographs Medicinal and Pharmaceutical Substances: Ciprofloxacin Hydrochloride"*, British Pharmacopeia. Volume (I and II), (2009).

[39] T. H. Fereja and S. B. Tufa, *"In Vitro Quality Assessment and Bioequivalence Studies on Four Brands of Ciprofloxacin Tablets, Marketed in Ambo, Ethiopia"*, Int. J. Pharm. Sci. Vol.5, No.2, pp 1007-1012,(2015).