JETIR.ORG ISSN: 2349-5162 | ESTD Year : 2014 | Monthly Issue
JOURNAL OF EMERGING TECHNOLOGIES AND INNOVATIVE RESEARCH (JETIR)
An International Scholarly Open Access, Peer-reviewed, Refereed Journal

ANALYTICAL ESTIMATION AND VALIDATION OF CIPROFLOXACIN HCL BY USING NEW STRATEGIES

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ABSTRACT
The development of a drug product is a complex endeavour, encompassing phases such as drug discovery, laboratory testing, animal studies, clinical trials, and regulatory approval. Process controls, including inspection of raw materials, in-process monitoring, and setting targets for the final product. Process validation, as mandated by the cGMP regulations, is essential at both general and specific levels as outlined in parts 210 and 211. Analytical estimation and process validation of Ciprofloxacin HCl tablet dosage form would be carried out. Three consecutive batches of Ciprofloxacin HCl are F1, F2& F3 will be taken for process validation studies. All the critical parameters would be evaluated for fixing the optimum process parameter. Analytical evaluation of Pre-formulations parameters, Post formulations parameters, preparing validation protocol, Execution of validation protocol.

KEYWORDS: Validation, Antibiotics, Ciprofloxacin, Tablets, Analysis.

INTRODUCTION

VALIDATION

A program called pharmaceutical validation verifies that apparatus, systems, procedures, or methods work as anticipated. It covers all the important parameters during the manufacturing process of any pharmaceutical dosage form. In the end, validation is a program that shows how procedures, tools, systems, and equipment are supposed to be used during the entire manufacturing(Gheen, 2022).

In essence, validation entails assessing the validity or proving the effectiveness of processes. It is a collaborative effort involving professionals from diverse disciplines within a plant. This approach rests on the premise that quality, safety, and efficacy are integral to the product's design or construction. Quality assurance

extends beyond mere inspection or testing of in-process and finished products; it involves controlling every step of the manufacturing process to ensure that the final product meets all quality specifications.

The development of a drug product is a complex endeavour, encompassing phases such as drug discovery, laboratory testing, animal studies, clinical trials, and regulatory approval. Process controls, including inspection of raw materials, in-process monitoring, and setting targets for the final product, are vital elements. The aim is to continuously monitor and validate the manufacturing process, even after validation is achieved. Moreover, adherence to current Good Manufacturing Practice necessitates the establishment of well-documented procedures for process controls to ensure ongoing performance monitoring.

Fig 1: Basic concept of Process Validation

STAGES OF PROCESS VALIDATION

Stage-1 Process Design

In the initial phase, the focus is on delineating the manufacturing process in a manner that ensures consistent production of a medicinal product meeting predetermined quality standards. Achieving this goal requires the validation team to thoroughly understand the intricacies of the process. Various sources and techniques are employed to gather pertinent process information:

- Product development endeavours provide valuable insights.
- Understanding the functionalities and limitations of production equipment is crucial.
- Predictions regarding factors contributing to variability are considered.
- Design of experiment (DOE) studies are conducted to explore different variables.

Stage-2 Process Qualification

In the second stage, known as Process Qualification, the focus shifts to evaluating the effectiveness of the process design for ensuring quality production. This involves a comprehensive assessment conducted through several steps:

a. Facility Design
b. Utilities and Equipment Qualification
c. Process Performance Qualification

Stage-3 Continued Process Verification

Following process design and qualification, the third stage of process validation focuses on establishing mechanisms to consistently uphold the validated process during routine production. Continued process verification involves several key practices:

a) Utilizing statistical process control (SPC) to monitor and sample process parameters and quality attributes continuously.
b) Conducting scheduled maintenance of the facility, utilities, equipment, and associated assets.
c) Implementing good documentation practices throughout the validation process.

SCOPE OF VALIDATION

d) Defining the scope of pharmaceutical validation presents a considerable challenge due to its extensive coverage across various aspects of pharmaceutical manufacturing. Nonetheless, upon close inspection of pharmaceutical operations, several key areas emerge for validation consideration (Pant, Hussain and Ashok, 2023).

IMPORTANCE OF VALIDATION

Validation holds significant importance across various facets of pharmaceutical manufacturing:

a) Ensuring quality assurance
b) Meeting time constraints effectively
c) Optimizing processes for efficiency
d) Ensuring the quality of raw materials
e) Validating the reliability of packaging materials
f) Verifying the performance of equipment
g) Validating the suitability of facilities

TYPES OF PROCESS VALIDATION

a. **Prospective Validation-** Prospective validation, conducted during the developmental phase (also known as premarket validation), aids in identifying potential risk factors within the production process. This method involves breaking down the process into distinct steps to scrutinize critical aspects like mixing time, relative humidity (RH), and temperature.
b. Concurrent Validation- Concurrent validation occurs during routine production stages and builds upon the thorough examination of the process conducted during prospective validation. Close

monitoring of the initial three production-scale batches is essential. Concurrent validation is conducted under the following circumstances.

c. **Retrospective Validation-** Retrospective Validation relies on historical and testing data from batches manufactured in the past. It involves analysing trends and verifying whether the process adheres to the permissible range of parameters. This analysis can be conducted through computer-based systems or manual methods.

d. **Process Re-Validation-** Revalidation is necessary to ensure that any alterations made to the process environment, whether deliberate or inadvertent, do not negatively impact process attributes and product quality.

MASTER FORMULA RECORD

The Master Formula Record (MFR) serves as a comprehensive document for pharmaceutical products, encompassing details of their manufacturing processes. It is crafted by the research and development team within the company. Subsequently, manufacturing units utilize the MFR as a basis to create other documents such as Batch Manufacturing Records (BMR) and Batch Packaging Records (BPR).

PROCESS VALIDATION PROTOCOL

A documented strategy outlining the approach for conducting process validation; it will detail the individuals responsible for different tasks and establish parameters for testing, including sampling plans, testing methodologies, and specifications. This plan will delineate product attributes and equipment requirements. It will also define the minimum number of batches required for validation studies and establish acceptance criteria.

Table 1: Process Validation Protocol

S.No.	Process Validation Protocol
i.	Objective
ii.	Label claim
iii.	Validation team and responsibility
iv.	Scope of protocol
v.	Manufacturing formula
vi.	Process flow chart
vii.	Critical steps, variables to be studied & acceptance criteria
viii.	Pre requisites for process validation
ix.	General tests for raw material
x.	List and qualification of equipment's/instruments
xi.	Manufacturing and sampling procedures

xii.	Process validation report
xiii.	Discussion
xiv.	Future scope

ANTIBIOTICS

Antibiotics are tailored to specific bacterial infections and cannot be indiscriminately swapped between different types of infections. When used correctly, antibiotics are generally safe with minimal side effects, as healthcare providers can customize treatment based on individual patient needs, including the type of antibiotic, dosage, and duration.

Mechanism of Action

Inhibition of cell wall synthesis: The structure of the bacterial cell wall includes numerous tiers of peptidoglycan, which are made up of peptide chains and sugars known as glycans. These glycans, consisting of N-acetyl muramic acid (NAM) and N-acetyl glucosamine (NAG), are linked together with the assistance of trans glycosidases. Additionally, the connection between the D-alanyl-D-alanine segment of the peptide chain and glycine residues occurs in the presence of penicillin-binding proteins (PBPs), contributing to the strengthening of the cell wall(Khanal, 2022).

Inhibition of membrane function: The cytoplasmic membrane regulates the passage of molecules and ions in and out of the cell through integral transporter proteins, maintaining selective permeability. Antibiotics targeting fatty acid synthesis and membrane phospholipids disrupt the function of this membrane. These antibiotics exert a detergent-like effect, creating random pores that result in the leakage of cellular molecules, inhibition of respiration, increased water uptake, and eventual cell death. Gram-positive bacteria, with their thicker cell walls providing protection, demonstrate a natural resistance to such antibiotics.

TABLETS

Tablets are solid medications comprising active pharmaceutical ingredients and additional substances known as excipients. They typically possess a solid, circular shape with a flat and convex surface. These dosage forms provide a specific quantity of medication in a single unit and are categorized as solid unit dosage forms.

METHODOLOGY

Determination of melting point: Small quantity of drug was placed into a sealed capillary tube. The tube was placed in the melting point apparatus. The temperature in the apparatus was gradually increased and the temperature at which entire drug gets melted was noted.

Solubility studies: The Solubility of ciprofloxacin hydrochloride was determined in distilled Water, methanol, dimethyl formamide and dilute alkali and in dilute acids.

Preparation of standard curve in distilled water: Accurately weighed 100 mg of ciprofloxacin and was dissolved in 100 ml of distilled water, from this stock solution 10 ml was withdrawn and transferred into 100 ml volumetric flask. Volume was made with distilled water in order to get standard stock solution containing 100 ug/ml. Form this standard stock solution, a series of dilution (10, 20, 30, 40, 50 ug/ml) were prepared using distilled water. The absorbance of these solutions was measured spectrophotometrically against blank of ciprofloxacin at 278 nm for ciprofloxacin HCL. Absorbance of drug at different concentrations were calculated and graph was plotted (Adama Diaman, K. 2023).

Infrared spectroscopic analysis: The FTIR spectrums of moisture free samples of Drug, all excipients and physical mixture recorded on IR spectrophotometer. The scanning range varies from 4000 - 400 cm-1 and resolution was 1 cm-1.

Preparation of tablets: Different formulations of Ciprofloxacin (F1-F3) were prepared by Wet Granulation method by using different excipients like Microcrystalline cellulose, Starch, Magnesium Stearate, Sodium Benzoate, Talc and Water.

Table 2: Formulation Containing

Ingredients	F1	F2	F3
Ciprofloxacin	1.460Kg	1.460Kg	1.460Kg
MCC	75gm	75gm	75gm
Starch	210gm	210gm	210gm
Sodium starch glycolate	34.5gm	34.5gm	34.5gm
Titanium dioxide	4.5gm	4.5gm	4.5gm
Benzoate	2.25gm	2.25gm	2.25gm

Angle of repose: Angle of repose is an indication of fractional forces existing between granules particles. The maximum angle possible between the surface of the pile of granules and the horizontal plane gives the angle of repose.

Bulk density: Bulk density of powder is the ratio of the mass of an untapped powder sample and its volume indicating the contribution of the intra-particulate void volume. The bulk density is expressed in g/ml. Bulk density is determined by weighing powder into a dry graduated 250 ml cylinder

Bulk density= Mass of the Blend powder/ Volume occupied by the powder blend

Tapped density: Tapped density is obtained by mechanically tapping a graduated measuring cylinder or vessel containing a powder sample. After observing the initial powder volume to weight, the measuring cylinder or

vessel is mechanically tapped and volume readings are taken until little less than l% further volume change is observed. The mechanical tapping is achieved by raising the cylinder or vessel and allowing it to drop under its own weight at specified distance. Secure the cylinder in the holder of the apparatus with weighed powder sample. Measure 100-200 taps and observe the corresponding volumes to the nearest graduated unit.

Tapped density = Mass of the powder Blend taken/ Tapped Volume of the powder blend

Carr's index: The Carr's Index and Hausner's ratio are measures of the porosity of a powder to be compressed. They measure the relative importance of interparticle interactions. For poor flow

materials there are frequently greater interparticle interactions and a greater difference

between the bulk and tapped densities. These differences are reflected in the compressibility

Index and Hausner's Ratio. Car's Index was calculated using the following formula.

Carr's Index-100* (TD-BD)/ TD

Hausner's Ratio: The Hausner's Ratio is a number that is correlated to the flow ability of a powder or granular material Hausner's ratio is calculate using following formula:

Hausner's ratio= TD/BD

EVALUATION OF TABLETS

The formulation tablets were evaluated for the following physical parameters.

Thickness: Thickness depends on the die filling, physical properties of material to be compared. There is possible of small variation in the thickness of individual tablet in a batch. But it should not be appear to the unaided eye. The thickness and diameter can be measure by vernier caliper.

Weight variation test: Twenty tablets were selected randomly and weighed individually. Calculate average weight compare the individual tablet weight to the average. Not more than two of individual weight derivate from the average weight by more than percentage shown in tablet and none derivate by more than twice the percentage.

Hardness: Tablets must possess sufficient strength or hardness and can be measured by Monsanto hardness tester. Ten tablets were randomly picked from each formulation and were evaluated for hardness and can be expressed in kg/cm2.

Friability: Friability can be performed in Roche friabilator, Pre weighed ten tablets were introduced in the friabilator. Then the machine was operated for 100 revolutions. Tablets were dropped from a distance of six

inches each revolution. Tablets were then dusted and reweighed. LOSs of less than 1% in weight is considered to be within the specification and acceptable.

Disintegration Time: This test determines whether tablet disintegrates with in prescribed time when placed in liquid medium.

Drug content: Assay: (BY HPLC)

Mobile Phase preparation: A mixture of 87 volume of 0.025 M phosphoric acid, previously adjusted with triethylamine to a pH of 3.0 + 0.1.

Chromatic condition

Apparatus: HPLC
Column: C18
Wave length: 278nm
Injection volume: 10 ul Flow
Rate: 1,5ml/min
Column Temperature: 30°C
Type of detector: UV

Standard Preparation: Weigh accurately 50 (mg of Ciprofloxacin into 100 ml volumetric flask and makeup volume with 0.01 M HCI.

Sample Preparation: Weight and powder 20 tablets. Weight accurately a quantity of the powder containing about 50 mg of ciprofloxacin and make up the volume with 0.01 M HCI (Abdu Hussen 2022).

In vitro Dissolution studies:

Procedure of Dissolution of tablets: Six tablets of Ciprofloxacin (Film Coated tablets) were introduced in the dissolution apparatus. The medium used was 900 ml of water and the dissolution medium were maintained at the temperature of 37.5, the RPM was set at 50. The Dissolution was carried out for 30 minutes and sample withdrawn at predetermined intervals. The estimation was carried out by UV method.

Standard Preparation: Weigh accurately 50 mg of Ciprofloxacin and transfer into a 100 ml volumetric flask. Dissolve and dilute up to volume with dissolution media. Take 1ml from above solution and transfer into 100ml volumetric flask, dissolve and dilute up to volume with dissolution media.

Sample Preparation: The dissolution parameters were set and one tablet is placed in each basket and care was taken to exclude air bubbles from the surface of the tablets and immediately the apparatus was started, 10ml of the sample was withdrawn and filter through whatmann filter paper, 10ml of solution was replaced in to dissolution medium, the same procedure was repeated at other time intervals. Take 1ml from above solution and transfer into 100 ml volumetric flask and volume make up (Gao, Z. 2022).

Drug release kinetics: The release kinetics was studied by various kinetic models such as zero- order plot, first-order plot, Higuchi plot, and Korsmeyer-Peppas plot. To study the release kinetics of the nanoparticle gel data obtained from in vitro drug release studies was plotted in various kinetic models: Zero-order as cumulative amount of drug releases versus time, first order as long cumulative % of drug remaining versus time, Higuchi model as cumulative % of drug released versus square root of time, and Korsmeyer-Peppas model as log cumulative % drug release versus long time. The best fit model was confirmed by the value of correlation coefficient near to one (Shripathy 2023).

RESULT

Pre-formulation Parameters:

Organoleptic properties: The following studies were performed for ciprofloxacin HCl for its various physical parameters.

Table - 3: Observation of organoleptic properties

TEST	SPECIFICATION	OBSERVATION
Colour	Pale yellow	Pale yellow
Odour	Odour less	Odour less

Solubility analysis: Ciprofloxacin hydrochloride samples are examined and it was Practically found to be soluble in distilled water and slightly soluble methanol, soluble in dimethyl formamide. It also dissolves in dilute alkali and in dilute acids.

Melting point of drug: The melting point of the drug was determined using Differential Scanning Calorimeter and was found to be 256 ^{O}C

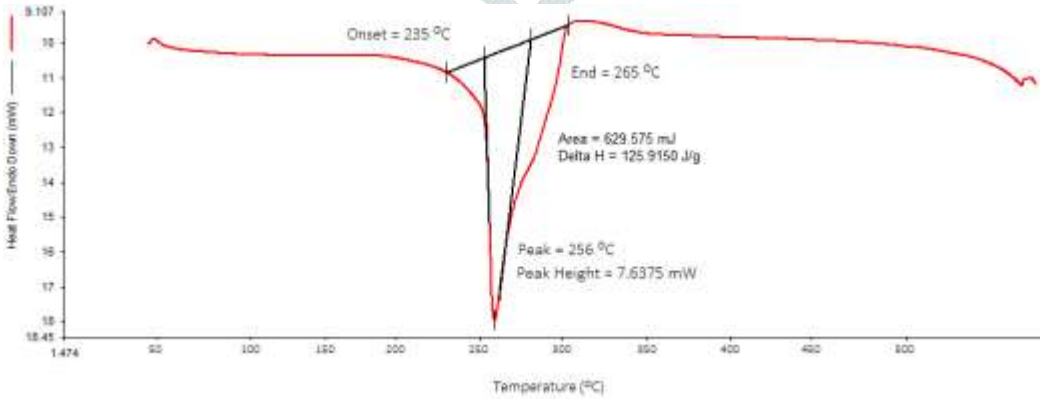


Fig. 2: DSC of Ciprofloxacin HCl

Determination of pH: The pH was measured three times and mean was noted. Hence pH of Ciprofloxacin HCl was found to be 4.2.

FT-IR analysis: FT-IR spectroscopic analysis was carried out to characterize drug. The FT-IR spectra obtained was compared with that given in pharmacopoeia for Ciprofloxacin HCl. Diagnostic peaks and finger print regions were found identical. These characteristics peaks are useful in identification of drug. FT-IR of Ciprofloxacin HCl, all excipients and mixture containing Ciprofloxacin HCl and all excipients was done for drug compatibility studies. The results obtained showed that there occur no interactions between the components when taken together.

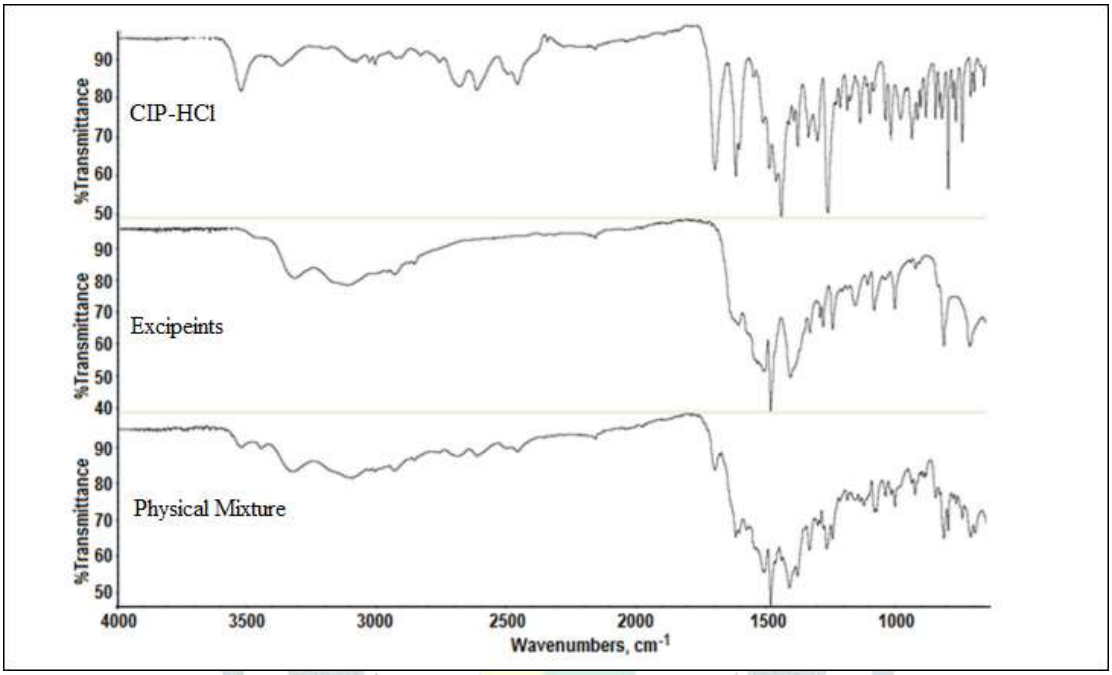


Fig. 3: FTIR graph of Ciprofloxacin, Excipients and Physical Mixture

Analysis by UV-Visible spectrophotometry

Preparation of standard graph- Stock solution of Ciprofloxacin HCl

Stock solution of 100 µg/ml was prepared by dissolving 10 mg of Ciprofloxacin HCl in 100 ml of distilled water. Dilution in the range of 10 of 100 µg/ml were scanned for determining λ max from 200-400 through UV spectrophotometer and λ max was found to be at 278 nm for Ciprofloxacin HCl.

Calibration curve of ciprofloxacin hydrochloride was determined by plotting absorbance (nm) versus concentration (µg/ml) at 276 nm. The results obtained are as follows.

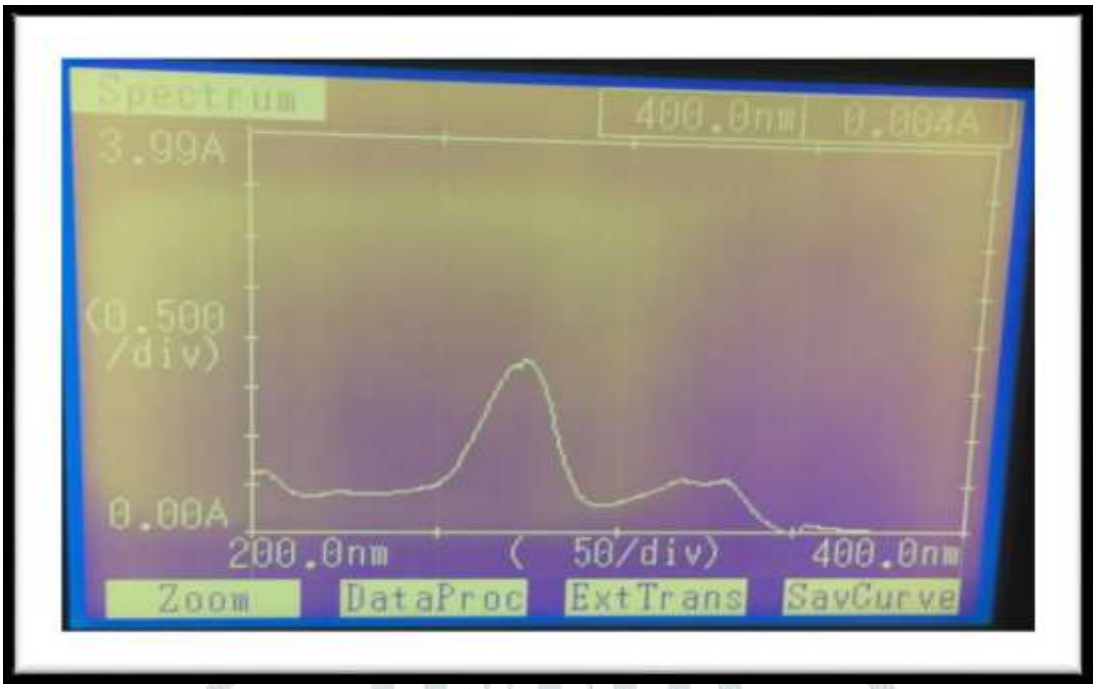


Fig. 3: Standard Curve of Ciprofloxacin

Preparation of calibration curve in distilled water

From these solutions of conc. 2µg/ml, 4µg/ml, 6µg/ml, 8µg/ml, 10µg/ml were prepared.

Table 4: Absorbance in different dilutions of drug at 278 nm

S.No.	Concentration (µg/ml)	Absorbance
1	0	0
2	2	0.200
3	4	0.372
4	6	0.537
5	8	0.723
6	10	0.876

The linear regression analysis was done on absorbance data points. A straight line generated to facilitate the calculation of amount of drug, the equation is as follows:

$$\mathbf{Y = mx + c}$$

Where Y= absorbance, m = slope, x = concentration

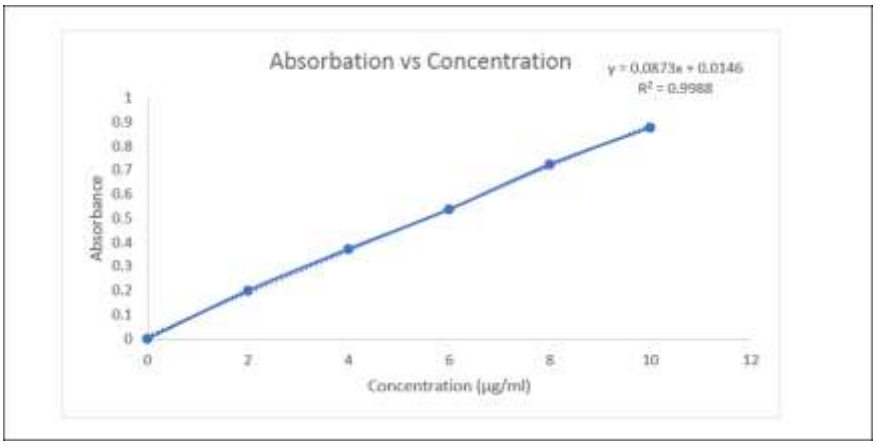


Fig. 4: Standard Calibration Curve of Ciprofloxacin

Linearity of Ciprofloxacin HCl by HPLC method

Preparation of standard graph- Stock solution of Ciprofloxacin HCl

Accurately weighed 100 mg of Ciprofloxacin HCl was dissolved in 100 ml of mobile phase. 1ml pipette out from above solution and taken in 10 ml volumetric flask and volume make up with mobile phase. Standard stock solution containing 100 µg/ml.

Standard graph of Ciprofloxacin HCl

Form this standard stock solution, a series of dilution (10, 20, 30, 40,50µg/ml) were prepared using mobile phase.

Table 5: Area of different dilutions of Ciprofloxacin HCl

S.No.	Concentration (µg/ml)	Peak Area
1	0	0
2	10	3377138
3	20	6754231
4	30	10131562
5	40	13508521
6	50	16885678

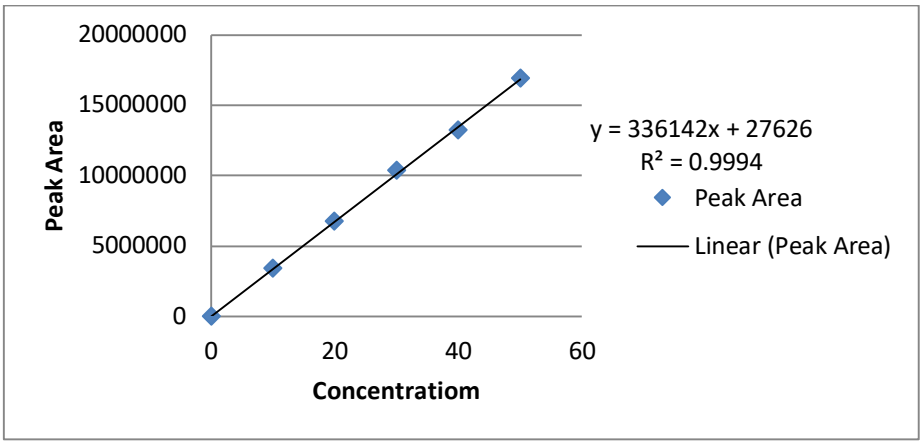


Fig. 5: Standard calibration curve of Ciprofloxacin HCl

Evaluation of Ciprofloxacin HCl:

The blended granules of different formulation were evaluated for angle of repose, bulk density, tapped density, carr's index and Hausner's ratio. The results of these evaluations were as.

Table 6: Pre-Compression Parameters

Form. Code	Angle of Repose (θ) (± SD)	Bulk Density (g/cc) (± SD)	Tapped Density (g/cc) (± SD)	Carr's Index (%) (± SD)	Hausner's Ratio(± SD)
F1	25.5±0.31	0.735±0.12	0.836±0.08	14.52±0.06	1.13±0.09
F2	25.1±0.45	0.781±0.09	0.899±0.09	15.10±0.05	1.15±0.07
F3	26.3±0.98	0.782±0.08	0.902±0.08	15.34±0.08	1.15±0.09

(± S.D) (S.D= Standard deviation), n=3

Post Compression parameters:

Table 7: **Post compression parameter of three batches**

S. No.	Form. Code	Thickness (mm)*	Hardness (Kg/cm^2)	Friablity (%)*	Weight Variation(mg)**
1	**F1**	5.63	9	0.25	821.7
2	**F2**	6.69	10	0.27	822.3
3	**F3**	5.67	9	0.24	821.2

Disintegration Test of Three batches at post compression stages

Table 8: Disintegration Test of three batches

S. No.	Form. Code	Disintegration Time
1	F1	2
2	F2	3
3	F3	3

Disintegration Test of Three batches after coating

Table 9: Disintegration Test of three batches after coating

S. No.	Form. Code	Disintegration Time
1	F1	6
2	F2	5
3	F3	7

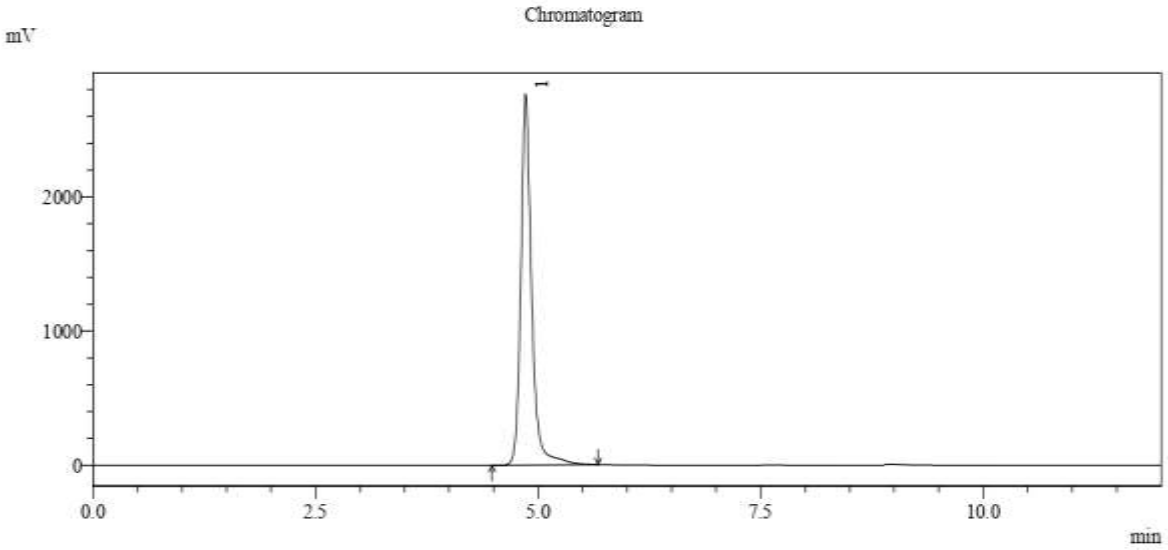
Assay: Potency of all three batches was found to be in below table

Table 10: Assay of all three batches

S.No.	Form. Code	Assay
1	**F1**	99.2
2	**F2**	99.7
3	**F3**	99.4

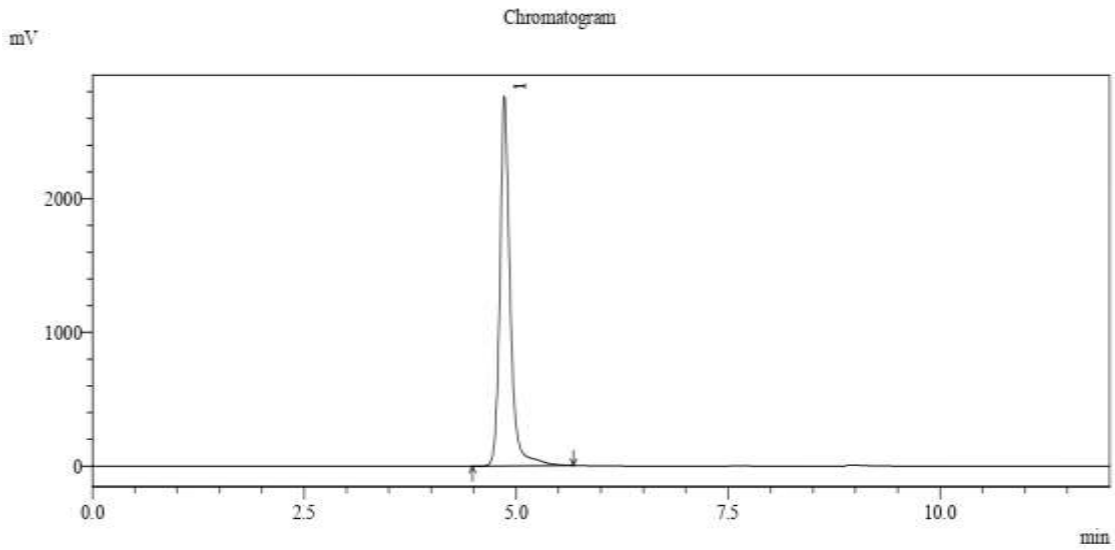
Peak#	Name	Area	Area%	Ret. Time
1	Std Ciprofloxacin	1688571	100.000	4.861
Total		1688571	100.000	

Fig. 6: Chromatogram of Std. Ciprofloxacin



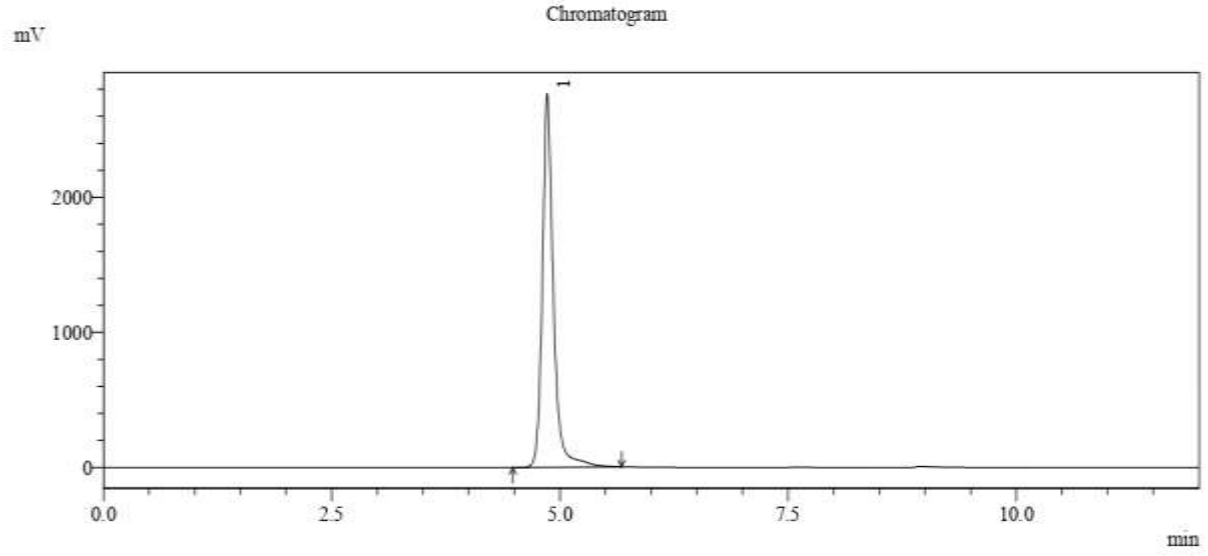
Peak#	Name	Area	Area%	Ret. Time
1	F1 Formulation	1692362	100.000	4.866
Total		1692362	100.000	

Fig. 7: Chromatogram of F1 Formulation



Peak#	Name	Area	Area%	Ret. Time
1	F2 Formulation	1703722	100.000	4.864
Total		1703722	100.000	

Fig . 8: Chromatogram of F2 Formulation



Peak#	Name	Area	Area%	Ret. Time
1	F3 Formulation	1694622	100.000	4.879
Total		1694622	100.000	

Fig 9: Chromatogram of F3 Formulation

***In Vitro* Dissolution studies:**

Table 11: *In-Vitro* drug release of formulations at different time intervals

S.No.	Time intervals	F1	F2	F3
1	0	0	0	0
2	5	10.2	11.7	10.5
3	10	25.7	27.1	26.8
4	15	47.8	46.2	48.0
5	20	69.4	71.3	70.4
6	25	85.1	86.6	87.3
7	30	97.2	96.9	97.4

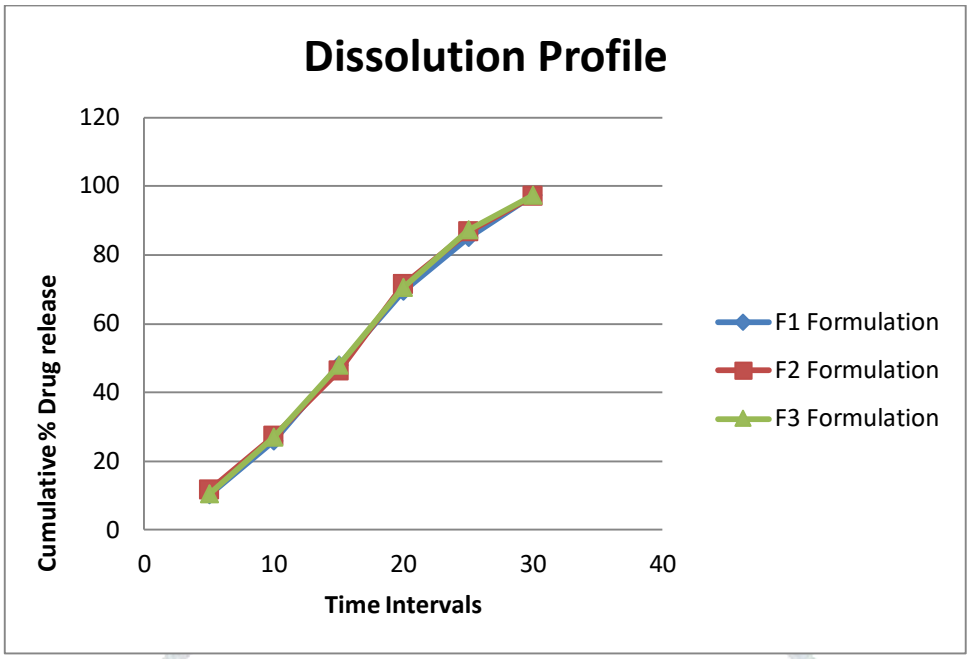


Fig. 10: Drug release pattern of F1 to F3

Kinetic Study of F2 Formulation: Evaluation of mechanism of drug release was done for the F2 Formulation. In vitro drug release date was fitted into various kinetic models.

A) Zero order: Graph of cumulative percentage of drug released and time

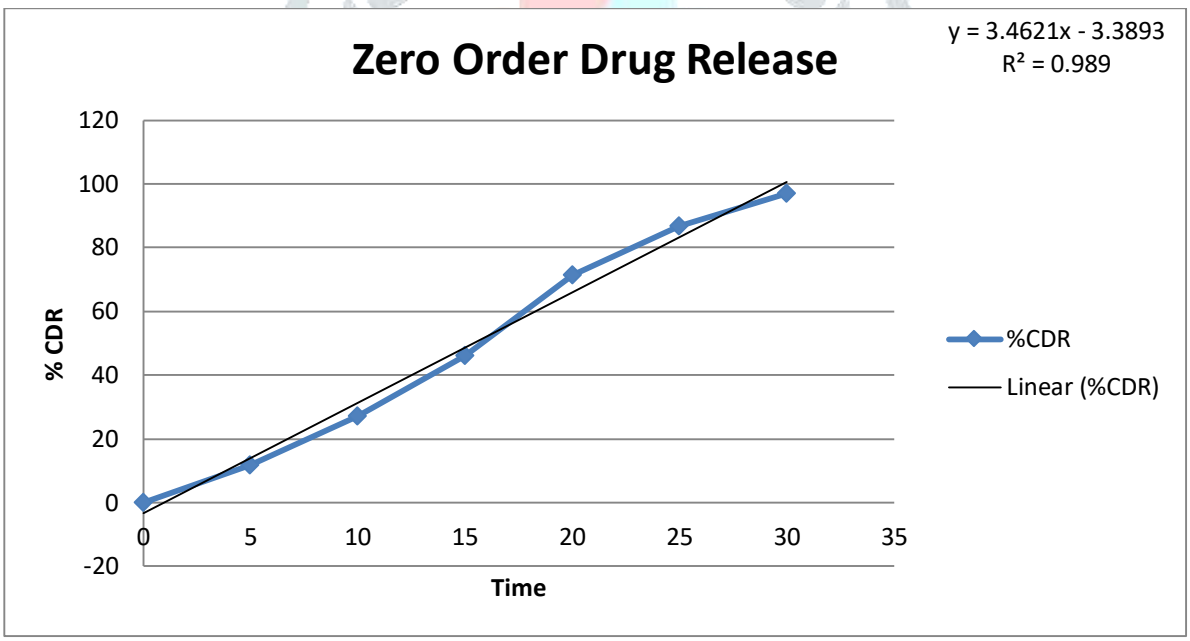


Fig. 11: Zero Order plot of drug release kinetics

B) First order: Graph of log cumulative percentage of drug remaining and time.

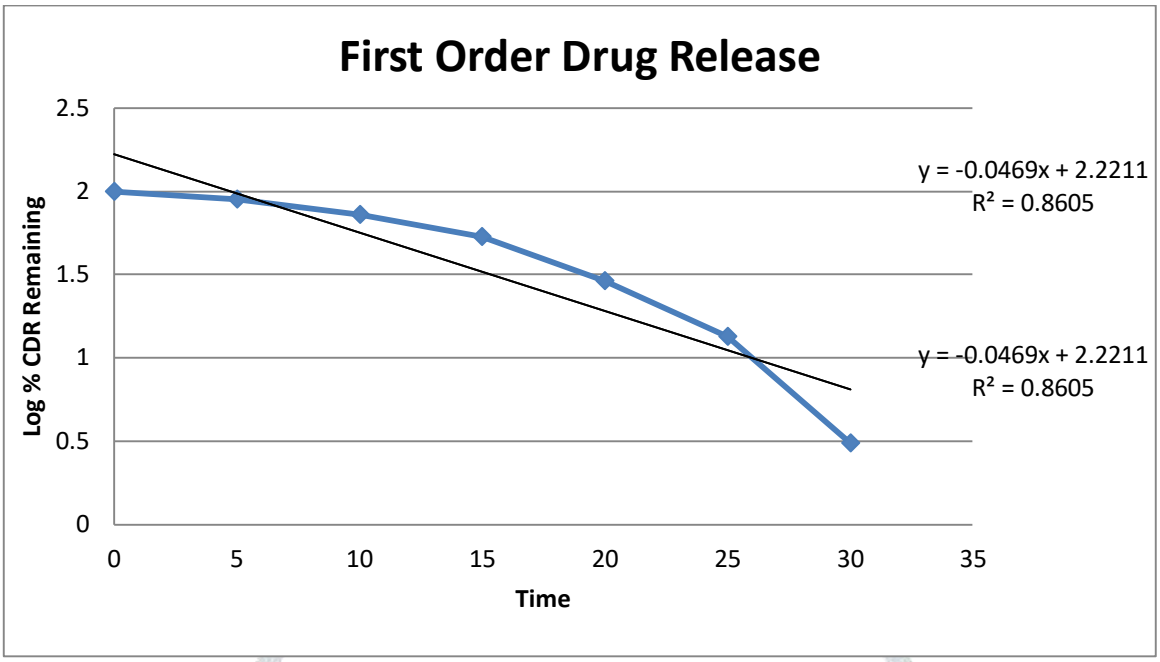


Fig. 12: First Order plot of drug release kinetics

C) Higuchi model: Graph of cumulative percentage drug release and square root of time

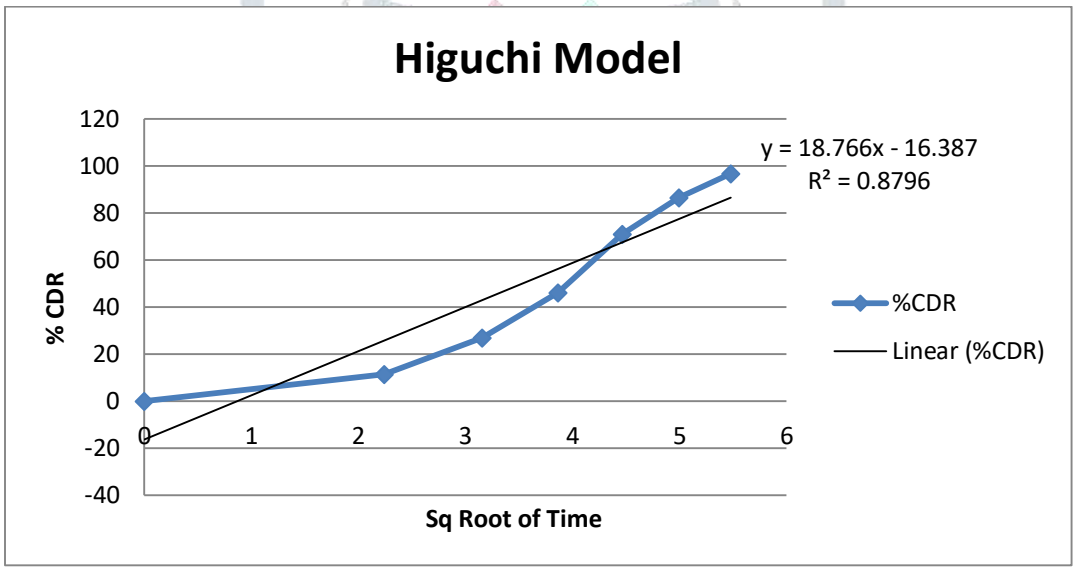


Fig. 13: Higuchi Model of drug release kinetics

D) Korsmeyer – peppas model: Graph of log cumulative percentage drug release and log tim

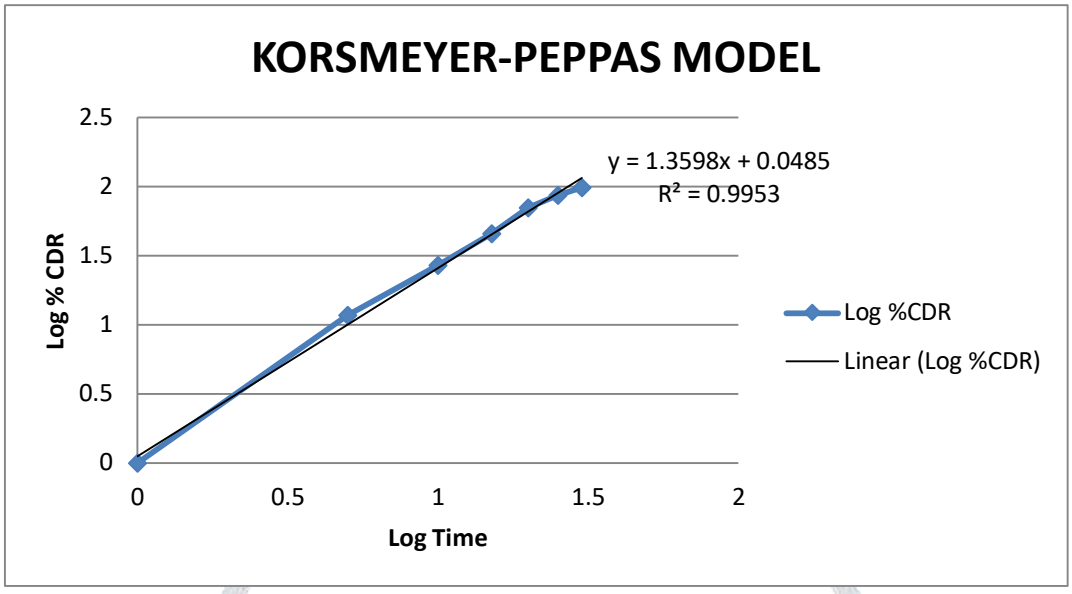


Fig. 14: Peppas Model of drug release kinetics

Table 12: Kinetics of drug release of F2 formulation

S. No.	Plot	R^2
1	Zero Order	0.989
2	First Order	0.860
3	Higuchi Model	0.879
4	Peppas Model	0.995

The data obtained for in vitro release were fitted into equations for zero order, first order, Higuchi and Korsmeyer Peppas release models. The interpretation of data was based on the value of the resulting regression coefficients.

From these values, it was observed that the Peppas model was found to be best suited with R2 value of 0.995.

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ACKNOWLEDGEMENT

The author acknowledges the teacher's staffs and friends for their kind support.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

1 Anderson, L. (2023). Antibiotics Guide. Drugs.com. Available at: https://www.drugs.com/article/antibiotics.html.

2 Bhakar, N. (2019). 4 types Process Validation, Pharmaceutical FDA 2019, Pharmaguddu. [online] Pharmaguddu. Available at: https://pharmaguddu.com/process-validation-pharmaceutical/.

3 Brainaugh, A.D. (2021). Tablet dosage forms- Properties and types. [online] PharmaCampus. Available at: https://pharmacampus.in/pharmaceutics/tablet-dosage-forms/.

4 Brooks, A. (2019). Process Validation: A Guide to Ensuring Quality and Compliance.

5 Bow, E. J. (July 2013). "Infection in neutropenic patients with cancer". Critical Care Clinics, 29(3), 411-441. doi: 10.1016/j.ccc.2013.03.002. PMID: 23830647.

6 Cayuela, R. (2020). How to create a Validation Master Plan in 5 steps. Templates & more. [online] CIQA Validation Engineering - Dataloggers Rental and more. Available at: https://ciqa.net/validation-master-plan/.

7 Choudhary, A. (2005). Preparation of MFR for Pharmaceuticals. [online] Pharmaguideline.com. Available at: https://www.pharmaguideline.com/2016/07/preparation-of-mfr-for-pharmaceuticals.html.

8 Cochrane Library (2023). About the Cochrane Database of Systematic Reviews | Cochrane Library. [online] Cochranelibrary.com. Available at: https://www.cochranelibrary.com/cdsr/about-cdsr.

9 Colcol, S. (2024). Process Validation in the Pharmaceutical Industry. [online] SafetyCulture. Available at: https://safetyculture.com/topics/process-validation/DrugBank (2005). Ciprofloxacin.

10 Flowers, C. R., Seidenfeld, J., Bow, E. J., Karten, C., Gleason, C., Hawley, D. K., et al. (February 2013). "Antimicrobial prophylaxis and outpatient management of fever and neutropenia in adults treated for malignancy: American Society of Clinical Oncology clinical practice guideline". Journal of Clinical Oncology, 31(6), 794-810. doi: 10.1200/JCO.2012.45.8661. PMID: 23319691.

11 Gould, K. (March 2016). "Antibiotics: from prehistory to the present day". The Journal of Antimicrobial Chemotherapy, 71(3), 572-575. doi: 10.1093/jac/dkv484. PMID: 26851273.

12 Gualerzi, C. O., Brandi, L., Fabbretti, A., & Pon, C. L. (4 December 2013). Antibiotics: Targets, Mechanisms and Resistance. John Wiley & Sons. p. 1. ISBN 978-3-527-33305-9.

13 Institut, J. (n.d.). Process Validation: Definition & Examples ~ What to Look Out For. [online] johner-institute.com..

14 Jones & Bartlett Publishers. (2011). Antibiotics Simplified (pp. 15-17). ISBN 978-1-4496-1459-1.

15 Keyur, B., Ahir, K., Singh, Yadav, S., Patel, H. and Poyahari, C. (2014). Overview of Validation and Basic Concepts of Process Validation. Scholars Academic Journal of Pharmacy (SAJP), [online] 3(2), pp.178–190. Available at: https://saspublishers.com/media/articles/SAJP32-178-190.pdf.

16 Khanal, S. (2022). Mechanisms of Action of Antibiotics: An Overview.

17 Laxminarayan, R., Duse, A., Wattal, C., Zaidi, A. K., Wertheim, H. F., Sumpradit, N., et al. (December 2013). "Antibiotic resistance-the need for global solutions". The Lancet. Infectious Diseases, 13(12), 1057-1098. doi: 10.1016/S1473-3099(13)70318-9. PMID: 24252483

18 Leekha, S., Terrell, C. L., & Edson, R. S. (February 2011). "General principles of antimicrobial therapy". Mayo Clinic Proceedings, 86(2), 156-167. doi: 10.4065/mcp.2010.0639. PMID: 21282489. PMCID: PMC3031442.

19 Mahmoud, T.Y., Hamza, I.S. and Jarallah, A.L. (2024). Spectrophotometric Method for the Determination of Ciprofloxacin in Pure and Pharmaceutical Preparations: Development and Validation. Engineering Proceedings, [online] 59(1), p.164. doi:https://doi.org/10.3390/engproc2023059164.

20 Pant, D., Hussain, K. and Ashok, P. (2023). Overview of Validation and Basic Concepts of Process Validation. [online] 10(09). Available at: https://www.jetir.org/papers/JETIR2309206.pdf [Accessed 3 Jun. 2024].

21 Pangilinan, R., Tice, A., & Tillotson, G. (October 2009). "Topical antibiotic treatment for uncomplicated skin and skin structure infections: review of the literature". Expert Review of Anti-Infective Therapy, 7(8), 957-965. doi: 10.1586/eri.09.74. PMID: 19803705.

22 Rahangdale, K., Patle, M., Kumbhalwar, N., Thakre, T., Vaishnav, N., Pardhi, N. and Patle, S. (2021). OVERVIEW OF VALIDATION AND BASICS CONCEPTS OF PROCESS VALIDATION. www.wjpps.com │, [online] 10(10), p.1771. doi:https://doi.org/10.20959/wjpps202110-20314.

23 Rollins, K. E., Varadhan, K. K., Neal, K. R., & Lobo, D. N. (October 2016). "Antibiotics Versus Appendicectomy for the Treatment of Uncomplicated Acute Appendicitis: An Updated Meta-Analysis of Randomised Controlled Trials". World Journal of Surgery, 40(10), 2305-2318. doi: 10.1007/s00268-016-3561-7. PMID: 27199000. SCID: 4802473.

24 Sharma, A. and Dadhich, A. (2011). PHARMACEUTICAL PROCESS VALIDATION WHY TO DO, WHEN TO DO AND HOW TO DO IT. [online] PharmaTutor. Available at: https://www.pharmatutor.org/articles/pharmaceutical-process-validation [Accessed 3 Jun. 2024].

25 Sharma, P.C., Jain, A., Jain, S., Pahwa, R. and Yar, M.S. (2010). Ciprofloxacin: review on developments in synthetic, analytical, and medicinal aspects. Journal of Enzyme Inhibition and Medicinal Chemistry, 25(4), pp.577–589. doi:https://doi.org/10.3109/14756360903373350.

26 Singh, R.P. and Bhuva, C.K. (2011). PHARMACEUTICAL PROCESS VALIDATION WHY TO DO, WHEN TO DO AND HOW TO DO IT. [online] PharmaTutor.

27 Sethi, N. J., Saff, S., Korang, S. K., Hröbjartsson, A., Skoog, M., Gluud, C., & Jakobsen, J. C. (Cochrane Heart Group). (February 2021). "Antibiotics for secondary prevention of coronary heart disease". The Cochrane Database of Systematic Reviews, 2(5), CD003610. doi: 10.1002/14651858.CD003610.pub4. PMID: 33704780.

28 Tarlengco, J. (2024). Process Validation in the Pharmaceutical Industry. [online] SafetyCulture. Available at: https://safetyculture.com/topics/process-validation/.

29 Yeotikar, S. (2020). Process Validation - Master Your Concept in 30 mins. - Pharma GxP. [online] pharmagxp.com. Available at: https://pharmagxp.com/quality-management/process-validation.

30 "Metronidazole". The American Society of Health-System Pharmacists. Retrieved 31 July 2015.

31 Chemical Analysis of Antibiotic Residues in Food. John Wiley & Sons, Inc. (2012). pp. 1-60. ISBN 978-1-4496-1459-1.

32 "Why antibiotics can't be used to treat your cold or flu". www.health.qld.gov.au. 6 May 2017. Retrieved 13 May 2020.

33 "General Background: Antibiotic Agents". Alliance for the Prudent Use of Antibiotics. Archived from the original on 14 December 2014. Retrieved 21 December 2014.

34 Here are the references converted into Harvard citation style:

35 PIC/S. (2007). Recommendations on Validation Master Plan, Installation and Operational Qualification, Non-Sterile Process Validation, Cleaning Validation. September.

36 Berman, J., & Planchard, J. A. (1995). Blend Uniformity and Unit Dose Sampling. *Drug Development and Industrial Pharmacy*, 21(11), 1257-1283.

37 Goutte, F., Guemguem, F., Dragan, C., Vergnault, G., &Wehrl, P. (2002). Power of Experimental Design Studies for the validation of pharmaceutical process: Case study of a Multilayer Tablet Manufacturing Process. *Drug Development and Industrial Pharmacy*, 28(7), 841-848.

38 Emori, H., Sakuraba, Y., Takahashi, K., Nishihata, T., & Mayumi, T. (1997). Prospective Validation of High Shear Wet Granulation Process by Wet Granule Sieving Method. II. Utility of Wet Granule Sieving Method. *Drug Development and Industrial Pharmacy*, 23(2), 203-215.

39 Newnes, L. B. (1996). Flexible Pharmaceutical Powder Production. *International Journal of Computer Integrated Manufacturing*, 9(3), 227-233.

40 Nash, R. A. (1996). Process Validation: A 17-year Retrospective of Solid-Dosage forms. *Drug Development and Industrial Pharmacy*, 22(1), 25-34.

41 Ferrante, M. (1999). A Simple way to Establish Acceptance Criteria for Validation Studies. *Catalytica Pharmaceutical*. *Journal of Validation Technology*, S(2), 1-3.

42 Klausner, E. A., Lavy, E., Friedman, M., & Hoffman, A. (2003). Expandable Gastroretentive dosage forms. *Journal of Controlled Release*, 90, 143-162.

43 Hwang, S. J., Park, M., & Park, K. (1998). Gastric retentive drug-delivery systems. *Critical Reviews in Therapeutic Drug Carrier Systems*, 15, 243-284.

44 Garg, S., & Sharma, S. (2003). Gastroretentive drug delivery systems. *Drug Delivery Technology*, 160-166.

45 Song, N. N. LL., Huang, Y. R., Zheng, B. Z., Feng, P., & Liv, C. X. (2006). Pharmacokinetics and relative bioavailability of metformin hydrochloride extended-release tablets in healthy volunteers. *Asian Journal of Pharmacodynamic and Pharmacokinetics*, 6(1), 55-62.

46 Gupta, P. K., & Robinson, J. R. (1998). Gastric emptying of liquids in the fasted dog. *International Journal of Pharmaceutics*, 43-45.

47 Washington, N., Washington, C., & Wilson, C. G. (2001). *Physiological Pharmaceutics II*. New York: Taylor and Francis.

48 Hasler, W. L. (1995). *Textbook of Gastroenterology II*. Lippincott JB editor. In: Yamada T

49 Nayak, A. K., Maji, R., & Das, B. (2010). Gastroretentive drug delivery systems: a review. *Asian Journal of Pharmacy and Clinical Research*, 3(1), 23-45.

50 Stepensky, D., Friedman, M., Sour, W., Raz, I., & Hoffman, A. (2001). Preclinical evaluation of pharmacokinetic-pharmacodynamic rationale for oral CR metformin formulation. *Journal of Controlled Release*, 71, 107-115.

51 Arora, S., Ali, J., Ahuja, A., Khar, R. K., &Baboota, S. (2005). Floating drug delivery systems: a review. *AAPS Pharm Sci Tech*, 47, 372-390.

52 Dressman, J. B., Berardi, R. R., Dermentzoglass, L. C., Russell, T. L., Schmeltz, S. P., Barnett, J. L., Jaruenpoa, K. M. (1990). Upper gastrointestinal pH in young, healthy men and women. *Pharmaceutical Research*, 7, 756-761.

53 Russell, T. L., BR, Barnett, J. L., Dermentzoglov, L. C., Jarvenpoa, K. M., Schmaltz, S. P., Dressman, J. B., et al. (1993). Upper gastrointestinal pH in seventy-nine healthy, elderly, North American men and women. *Pharmaceutical Research*, 187, 1092-1096.

54 NHS. (2014, June 5). Antibiotics. Retrieved from [NHS website](link).

55 European Centre for Disease Prevention and Control. (n.d.). Factsheet for experts. Retrieved from [European Centre for Disease Prevention and Control website]

56 Metronidazole. (n.d.). In *The American Society of Health-System Pharmacists*. Retrieved from link.

57 *Chemical Analysis of Antibiotic Residues in Food*. John Wiley & Sons, Inc. (2012). pp. 1-60. ISBN 978-1-4496-1459-1.

58 G. Sowjanya*, P. Ramaa Bharathi, Dr.A.M.S.Sudhakar Babu (2023). Film Coating Technology: An Over View | PharmaTutor. [online] www.pharmatutor.org. Available at: https://www.pharmatutor.org/articles/film-coating-technology-over-view.

59 Gupta Ankit, Ajay Bilandi, Mahesh Kumar Kataria and Khatri Neetu (2012). Tablet Coating techniques: Concepts and recent trends. [online] ResearchGate. Available at: https://www.researchgate.net/publication/285856849_Tablet_Coating_techniques_Concepts_and_recent_trends.

60 P Saiesh, Shabaraya AR and Shripathy (2023). kinetic modelling of drug release from gel,. pp.92–104.

61 Ousmane, T., Harouna, K., Mahamadou, K., Younoussa, C., Bernard, C., Ousmane Lansenou, B., Hawa, T. and Adama Diaman, K. (2023). Crossed Ectopy Of The Kidney Without Fusion Of The Extremities In

The Radiology And Medical Imaging Department Of The Chu Point G : About A Case. International Journal of Advanced Research, 11(06), pp.835–840. doi:https://doi.org/10.21474/ijar01/17133.

62 R Poduri, K. (2024). Medications and safety. International Physical Medicine & Rehabilitation Journal, 4(6). doi:https://doi.org/10.15406/ipmrj.2019.04.00219.

63 Beakawi Al-Hashemi, H.M. and Baghabra Al-Amoudi, O.S. (2018). A review on the angle of repose of granular materials. Powder Technology, [online] 330, pp.397–417. doi:https://doi.org/10.1016/j.powtec.2018.02.003.

64 D. Kaur (2024). Pre And Post Formulation Compatibility Study Of Diacerein Based On Atr-Ftir Study For The Design Of Transfersomal Carriers | International Journal Of Pharmaceutical Sciences And Research. [Online] International Journal Of Pharmaceutical Sciences And Research | IJPSR. Available at: https://ijpsr.com/bft-article/pre-and-post-formulation-compatibility-study-of-diacerein-based-on-atr-ftir-study-for-the-design-of-transfersomal-carriers/ [Accessed 21 Nov. 2024].

65 Abdu Hussen, A. (2022). High-Performance Liquid Chromatography (HPLC): A review. Annals of Advances in Chemistry, [online] 6(1), pp.010–020. doi:https://doi.org/10.29328/journal.aac.1001026.

66 Lex, T.R., Rodriguez, J.D., Zhang, L., Jiang, W. and Gao, Z. (2022). Development of In Vitro Dissolution Testing Methods to Simulate Fed Conditions for Immediate Release Solid Oral Dosage Forms. The AAPS journal, [online] 24(2), p.40. doi:https://doi.org/10.1208/s12248-022-00690-5.

67 Chen, P., Ansari, M.J., Bokov, D., Suksatan, W., Rahman, M.L. and Sarjadi, M.S. (2022). A review on key aspects of wet granulation process for continuous pharmaceutical manufacturing of solid dosage oral formulations. Arabian Journal of Chemistry, 15(2), p.103598. doi:https://doi.org/10.1016/j.arabjc.2021.103598.