2 REVIEW OF LITERATURE

There are many methods available on the analysis of these Cephalosporins. They include both classical and analytical and sophisticated instrument based procedures. The procedures are used to develop and validate the new methods that are very useful to society and industry for production. Consequently, special procedures have been used to develop the process of preparation and analysis of drug from biological fluids.

- **Revathi E et al (2014)** reported Spectrophotometric method for the determination of ceftriaxone sodium in different pharmaceutical dosage forms that also indicate stability of the drug. The proposed procedure is validated with all parameters for the assay of ceftriazone sodium drug contents. The developed method is simple selective and indicate stability to many dosage forms. In the given method the absorbance is measured at wavelength maximum of at 241 nm and linearity is in between from 5-50 microgram/mili-liter with correlation coefficient to 0.9983. Study shows that drug is degraded in acidic medium and underwent oxidation but R.S.D coefficient is less than two. It shows compliance with ICH guidelines.

- **Most. Umme Bushra et al (2014)** did experimental work on analytical method estimation & validation of Cefotaxime sodium by Ultra Violet-(200-400 nm to 800 nm) visible Double Beam Spectrophotometer for the estimation of bulk drug and its different dosages form available in market. Water and methanol were used as a solvent in method of determination of active constituent. The assay method is simple, reproducible, accurate, precise, ecofriendly and cost effective for routine analysis of drug. The wavelength maxima for absorption is 260 nanometer. This is determined by scanning the drug in solvent system and then analysis of spectra for maximum peak. Beer’s law was given concentration range with the following equation of trend line $y = 0.025x + 0.0028$ and the value of coefficient of correlation is 0.9995. the recovery studies was found to be in the range of 99.95% to 100.21%. The R. S. D. (Relatitive Standard Dev.) readings to interday and intra day reproducibility are 0.099 to 0.140 and 0.098 to 0.132. The value of LOD & LOQ are 0.079 µg/ml & 0.154 µg/ml.
percent of relative standard deviation for robustness & ruggedness were 0.142 – 0.221%.

- **Rajeev V Jadhav et al (2013)** have reported a method to assay of cephixime & its different types of dosage by spectrophotometer. technique is new, easy, specific, selective & cost effective. The selected wavelength was 290.60 for the estimation of drug content. The linearity range was found to be in the limit of 2-40 µ/ml with the correlation coefficient of \( R^2 = 0.9997 \) for regression equation.

- **Durga M R T et al (2011)** described high pressure Liq. Chromatographical or H.P.L.C. technique to determine cefotaxime as a pure drug & injection. High performance Liqu. Chromatographical (H.P.L.C.) procedure developed to determine cefotaxime sodium with impurity of its degraded chemicals is very easy, simple, stability showing, specific and precise. The Octadecylsilane (ODS) C-18 (150 × 4.6 mm) 5 µm column was used for analysis of cefotaxime sodium bulk drug & Injection. The mobile phase was used in the ratio of a mixture of buffers : acetonitrile : methanol (80 : 15 : 05). Mobile phase movement or velocity was 1.3 milliter / minute & injection volume was 20 µL with ultraviolet (UV) detector. The wavelength maxima for measurement of absorption was 254 nm and run time of mobile phase was 10 minutes. The method was proven for stability indicating capability by subjecting the active constituent (cefotaxime sodium) to stress conditions which includes acid hydrolysis, alkali hydrolysis, oxidation, photolysis, thermal degradation & resolution of compounds formed their in processes. Value of concentration limit to procedure is simply from 51.348 to 359.438 µ gram / milliter. The developed technique has been perfectly used in determination of cefotaxime sodium from injection dosage forms & reported the average assay data of 98.86% with a RSD (Relative standard deviation) of 0.48% for six samples. The cefotaxime sodium solution was stable upto 14 hrs at ambient temperature. The method validation protocol revealed excellent result for precision, linearity & Specificity. The present method can be utilized for routine analysis of drug with stability studies & Quality control.
• **Jhansi L M et al (2011)** reported Spectrophotometric procedure for effective quantity wise determination to cefotaxime sod. in pure medicament & different all other formulations. An effective, simple easy Ultraviolet (U.V.) Spectrophotometer technique was developed to determine pure drug cefotaxime sodium and pharma formulations and preparations (Injectable preparations. Beer’s was obeyed in the proposed method with a concentration limit from 5 to 30 microgram per milliliter. Value for coefficient of correlation $R^2$ 0.998 & maximum absorbance was measured at the maximum wavelength of 238 nm and having apparent molar absorptivity of $1.68092 \times 10^3$ Lit/Mol x cm. There is no interference of commonly used excipients in the injectable formulations. The result of analysis were validated by different statistical tools and recovery studies. The method is precise, accurate, linear, specific, cheap, cost effective, rapid and can be utilized in daily routine analysis of the drug and injectable preparations.

• **Devkhile A B & Shaikh K A (2011)** developed UV-Visible Spectrophotometric method for quantitative estimation of Cefperazone third generation cephalosporin antibiotics. The technique is easy, correct & cost effective to drug assay and may used in routine analysis of different dosages forms. In the developed method, water is used as solvent and the absorbance is measured at wavelength maxima of 275 nm. The developed method was validated with recovery studies, stability studies, LOD and LOQ, RSD and other all parameters. The linearity of the drug was found between 2-16 $\mu$g/ml. Weight basis measurement of concentration was carried out in the method. The correlation coefficient has a value of 0.9999 in the developed method. The result of recovery studies are 99.8 -110.3%. The result of LOD & LOQ are 0.12 microgram/milliliter and 0.46 microgram/milliliter.

• **R K Nanda et al (2010)** developed simultaneous determination of cefotaxime sodium and sulbactum sodium by using double beam UV-Visible Spectrophotometer. Three accurate, very simple, easy and reproducible methods developed for simultaneous determination of cefotaxime sodium & sulbactum sodium in different pharmaceutical dosage forms by ultraviolet spectrosopy.the first procedure include estimation
utilizing the simultaneous equation method, the wavelength maxima selected for samples are 233.5 nm & 264 nm in the concentration limit of 5-35 µg/ml & 2.5 – 17.5 µg/ml for cefotaxime sodium & sulbactum sodium accordingly. The second procedure is AUC (area under curve) method. The wavelength selected for measuring absorbance are 238.5-228.5 nm & 269-259 nm. With linearity limit from 5 to 35 micro g per milliter & 2.5 – 17.5 micro gram / ml for both cephotoxime sod. & sulbactum continuously. The third procedure involves estimation utilizing the multicompoment method. In this method the sampling wavelength were selected at 233.5 nm & 264 nm over the concentrat³ from 5 to thirty five microg per ml and 2.5 to 17.5 mc gram per ml for respective drugs cefotaxime sodium & sulbactum. Statistical validation & recovery studies were perfomed and checked for verification of validation parameters according to ICH Guidelines.

• Palnikumar B et al (2010) reported an Reverse Phase Liquid Chromatographic H.P.L.C. technique to estimate simultaneously ceftriazone sod. and sulbactum sodi. from different injectable dosage forms. In Isocratic Liquid Chromatographic (H.P.L.C.) procedure developed to determine simultaneously in ceftiaxone sod. and sulbactum sod (Na) in pharmaceutical formulations & preparations (Cetriax-1.5 mg injection). Hypersil Octadecyl silan C-18 chromatograph (two hundred fifty m.m. x 4.6 m.m as internal diameter to 0.5 µm) was used for chromatographic separation of these two drugs ceftriaxone sodium & sulbactum sodium in different pharmaceutical dosage forms. Mobile phase of chromatographic method is composed of 10 mM potassium dihydrogen orthophosphate : acetonitrile (90 : 10 v/v) adjusted to pH 5 by potassium hydroxide. The optimum separation was obtained within 15 minutes. The determined high performance Liquid Chromatographic technique provide regular area peak, reasonable retention time and optimum resolution to each medicines ceftriaxone sodium & sulbactum sodium. The procedure was validated according to ICH (International conference of Hormonization) by using statistical tools like linearity, precision, L.O. D (Detection limit), L. O. Q (Quantification limit), Relative standard Deviation (RSD), Robustness, ruggudness, specificity and stability studies. The
developed method follows the Beer’s law in the concentration range of 140-250 mcg/ml for ceftriaxone sodium & 75-160 mcg/ml for sulbactum sodium.

- **Brett C Macwhinney et al (2010)** reported analysis of 12 β-lactam antibiotics to the serum of individual by H. P. L. C. utilizing Ultraviolet (U.V) identification. A very easy, accurate, ecofriendly and cost effective HPLC (high pressure Liq. Chromatography) procedure had been determined and authenticated to analysis of cephalosporin, twelve penicillins & carbopenams antibiotics in human serum to 200 µL. Following drugs (Ceftazime, meropenam, ceftriaxone, cefazoline, cefalothin, dioxacilline, ticarcilline, ertapenam, penicillin-G, flucloxacillin, Dicloxacillin) were analyzed by proposed. The sample is prepared by common precipitation method involving protein precipitation with CH₃CN and lipid soluble components were separated by washing with CHCl₃. Separations were done on waters X-bridge C-18 column according to analytes, one of three CH₃CN buffer mobile phases. The detection of active component were done on the wavelength maxima of ultraviolet UV detection at 210nm, 260 nm and 304 nm. Validation protocol had determined the method to be linear, accurate and well précised. Therapeutic drug monitoring has been done by this method in a pathophysiology & pathology laboratory of β-lactam antibiotics in critically ill patients.

- **Hafiz Muhammad Arsad et al (2009)** developed a very easy, accurate, fast, sensitive, precise H. P. Liquid Charomatography detection to estimate cefixime and its tablet int the market. The HPLC consists of LC-10 AT VT pump, SPD-10 AVP UV detector. The Bondapak C18 column was used to separate the drug at room temperat. utilizing M. phage to MeOH: Phosphate system of buffer (Sodium Dihydrogen Phosphate) 35 : 65 at pH = 2.75 adjusted with phosphoric acid. The flow rate of mobile phase was 1 ml/min with the retention time of six minutes. This assay is selective for cefixime and able to separate drug peak from the expients. The linearity showed between the range from 0.039 – 20 µg/ml with $r^2 = 0.9998$. the system suitability parameters were 5.819 ± 0.51 (Mean ± %CV). Interday & intraday variations were between RSD of 0.53 – 1.64%. The limit of detection & quantification were
0.0195 & 0.039 µ/ml respectively with the coefficient of correlation 0.9996. the accuracy result of seventy percent drug was 99.82%, for 100% was 99.89% and for 130% was 100.12% so the method is more convenient and efficient option for the analysis of cefixime bulk drug, tablet and capsule dosage form.

- **Sharon Shen Nee (SSN) Ling et al (2003)** described simple and easy HPLC (Liquid Chromatographic) method for the estimation of cefotaxime sodium in rate & human plasma. For measurement for cefotaxim in person & mouse serum, a new HPLC (Liqu. Chromatographical) technique along to ultra violet U.V detector had been estimated and validated. This method the drug (Cefotaxime) is directly injected from supernatant plasma after deprotonation. Cefotaxime sodium degradation in acid pH had been prevented by mixing buffer of phosphate type centrifusing the drug. CH₃COONH₄ in water (0.05M)-CH₃CN-tetra hydro furan (volume by volume 87:11:2) ratio was used into mobile phase with 5.5 pH adjustment. The flow rate of mobile phase had been 1 milliter per minute. the \( \lambda_{\text{max}} \) wave length maxim for determination had been 254 nano meter used. Quantification limit (LOQ) had been 0.02 micro g per ml. Intraday & inter day coefficient of variations & accurately results had been found to be < 8% & ±3%. The recovery studies were done by adding 40 mg, 80 mg and 120 mg drugs. The results were > eighty seven percent with the concentrat\(^n\) ranges from 0.2 to 0.50 microgram per ml. Sensivity, specific, reproducibility & rapidity to the procedure make it appropriate for the daily routine analysis of drug in human plasma. The sample is required relatively in very small amount. This makes the method suitable for neonate plasma analysis.

- **Joshi Shalini (2002)** reported HPLC (Liquid Chromatographic) separation of antibiotics present in formulated and unformulated drug samples. A high specific and fastly acting method for antibiotic determination produce stability type problems. HPLC (Liquid Chromatographic) could be used to generate highly pure method for characterizing the antibacterial activity in the present review articles. Mobile phase & column conditions for the different classes of antibiotics viz. cephalosporins, macrolides, rifampicin, penicillins, chloramphenicol, tetracyclines, aminoglycosides,
polyene, quinolines, urinary antiseptics etc. have been represented from April 1998 to November 2000. The brief spectrum of activity, structure and mechanism of action of each classes have been also discussed.

- **Marie-Clemence-vedier et al (2002)** reported estimation of 12 antibiotics containing beta lactam ring (cephalosporins & Penicillins) to serum of human with HPLC (Liq. Chromatographic) method using Ultra Violet detectors. The following antibiotics (Cefepime, cefotaxime, ceftriaxone, Cefperazone, amoxicillin, cloxacillin, impenam, meropenam, oxacillin, pencillin-G, piperacillin and ticarcillin). The extraction of drug was done by precipitation technique of proteins using MeCN. Atlantics T3 chromatograph (analytical) with straight gradients of MeCN & two pH H₃PO₄ soln was used for separation. Photodiode array detection type UV detector is used at wavelength of two hundred ten nano meter, two hundred thirty nanometer / 290 nanometer. This procedure is specific, correct & precise (C. V. /variation coefficient < 8%) permitting quantificatn of β-latam serum concentration 5 - 250 micro gram per ml no change in readings with various compounds. These techniques are very simple in daily regular therapeutics medicament handling to penicillin and cefalosporins.

- **Jolanta J. Bafeltowska et al (2002)** determined cefotaxime and desacetylcefotaxime in cerebrospinal fluid by solid phase extraction and HPLC (Liquid Chromatographic) method. A HPLC (Liquid Chromatographic) method determined for the finding concentration of desacetyl cephotaxime and cephotaxime in cerebrospinal fluid or CSF. Desacetyl cephotsaxime & cephtaxime drugs have been separated & collected from C.S.F. sample utilizing S. P. E. (Extraction of solid phase). Chromatograph of Lichrocot Reverse phase-18 (22 miligram and 3 ml) & mix up of MeOH : Phosphate buffer with 7 pH (1:1) had been used to move cephtaxime & metabolite of desacetylcefotaxime. The separatn had been done on Lichrochrow 100 RP C - 18 (05 µm x250 x 4 m.m. internal diameter) chromatograph. Phase of solvent movement was o.01 Mol/lit CH₃COO⁻ buffers 4.8 acidic pH : MeOH in the ration of 85:15. Solvent flow was 1.56-100 µg per ml. the LOQ (Limit of Quantification) are 0.78 for desacetylcefotaxime and 70.39 micro gram per ml for cefotaxime. Recovery of drug
from cebrospinal fluid mised cephotaxime & desacetylcetofaxme 6.5 and 6.8%. the
detemined S.P.E.- H.P.L.C (Liquid Chromatographic) procedure had been used to find out cefotaxim & des acetyl cefotaxime determinations.

- **Okarmoto Y et al (1990)** reported degradation study of cefnidir drug by water or hydrolysis. The analysis was carried out at different pH solutions for example solution of pH=1 acidic, solution of pH =6-7 neutral solution and basic pH= 10 sol. The drug was degraded to two product one product is due to cleavage of beta lactam ring and another product is due to epimerization or isomerization of C-6 and C-7 position of the ring by different anvironment or lactonization of oxime product. The degradated product were identified and isolated by preparative and high pressure liquid chromatography with parallel reaction of product monitoring. The analytical technique is very useful and easily identify the degradation product of cefdinir in the pharmaceutical formulation and preparation of various dosage forms.

- **Okamoto Y et al (1996)** reported development and validation of cefnidir and related substance present in the preparation. The high performance liquid chromatographic method was used to develop and validate the drug and impurities found within the formulation. The method was checked for accuracy, precision, correctness & reproducibility, limit of detection and quantification, robustness & ruggedness, specificity & linearity. The method developed was specific and stability indicating for the drug cefdinir. The value of standard deviation is less then one percentage and range of reproducibility is within the limit and drug can be easily quantify by this method.

- **Kees F et al (1996)** reported In a cross-over study on twelve healthy volunteers cefpodoxime prosetil (CAS 87239-81-4) and acetylcysteine (CAS 616-91-1) were evaluated for possible pharmacokinetic interactions. After a standardized breakfast, the subjects received p.o. either 200 mg cefpodoxime administered as cefpodoxime prosetil (reference) or 200 mg cefpodoxime and 200 mg acetylcysteine (test). To determine the pharmacokinetic profile of cefpodoxime the plasma concentrations were
determined by HPLC. The plasma concentration-time curve of cefpodoxime was very similar after both regimens, and with respect to cefpodoxime bioequivalence has been proven. The narrow range of 90% confidence intervals for the quotient test/reference for Cmax and AUC indicate reliable bioavailability of cefpodoxime proxetil independent of co-administered acetylcysteine.

- **F Camus et al (1994)** reported the high performance liq. Chromatographic method for the analysis of cefpodoxime in human serum and sinus mucosa. The method was developed using ternary mobile phase of acetate (CH₃COO⁻) buffer and methanol (MeOH)- acetonitile (ACN) in ratio of 87 to 103. The method was validated as per international conference of harmonization for ruggedness & robustness, reproducibility & correctness, LOD & LOQ, system suitability and stability study, linearity & range. The sample preparation was done with C-8 cartridge of solid phase extraction. The method was developed with the reference of cefclor as internal standard. The stationary phase was C-18 and mobile phase is determined with the wavelength maxima of 235 nanometer. The both between and with the day reproducibility is in the limit & the linearity is also checked for plasma & serum with mucosa of sinus. The C.V. is less than 13.6% where n=10 and method is optimum for routine analysis of cefpodoxime in the pharmaceutical preparations and formulations.

- **N. H. Vadia et al (2009)** reported two method of analysis as colorimetry and derivative spectroscopy for the analysis of cefetamet pivoxil. The colorimetric method is very easy, simple, correct, rapid and sensitive for the drug. The drug was reacted with folin reagent and sodium hydroxide to develop a color. The wavelength maxima was noted to measure the concentration of colored product. The concentration of formed complex was measured within one hour of color development and analyzed. The other method was derivative spectroscopy. The first derivative spectra was obtained as different sensitivity for two standard drugs. Both the methods were checked for recovery studies at the concentration of 50, 100 and 150 mg for spiked drugs. Other standard parameters of ICH guideline were tested and found within the
limit. So both colorimetery and first derivative spectroscopy was used for the analysis of drug in drug testing laboratories and industries.

- **T. Madhusudana Reddy et al (2003)** reported analysis of cefixime and cefpodoxime proxetil by electrochemical reduction behavior of the both drugs in the cell. The different voltameteric techniques have been used to analyze these both cefpodoxime proxetil and cefixime drugs using Britton Robinson buffer system. The both compounds produce two cathodic waves from the cell within complete range of pH. The electrons were transferred from one electrode to another electrode and reduction process was measured and reaction process has been proposed. The process of both the compounds cefixime and cefpodoxime were reversible and diffusion controlled as observed in the results. The linearity of current was found within range. The drugs were determined by differential pulse voltametric method and can be used for the daily routine analysis of the drug. The method is correct, simple, precise and accurate, stability indicating, suitable for marketed preparations of both the drugs cefixime and cefpodoxime proxetil.

- **Silber Michael B., et al. (1987)** reported an easy, accurate & reproducible isocratic R. P. H.P.L.C. of a new cephalosporin in human serum and urine. Mobile phase was prepared fresh on the day of analysis. Mobile phase A was prepared by combining 170ml of acetonitrile, 1.36g of monobasic sodium phosphate, 2ml of 85% phosphoric acid and 828ml of purified H2O. M phase B (used for detection of urine) was prepared by combining 200ml of acetonitrile, 1.36g of monobasic sodium phosphate, 2ml of 85% phosphoric acid and 798ml of distilled water at pH 2.7 with flow rate of 2.0ml/min.

- **Castillo M, et al. (1988)** reported the degradation rate constants for ampicillin and for dicloxacillin in the suspension filtrate, and their solubility coefficients (at 25\(^0\)) by spectrophotometry employing a multicomponent computer program.

- **R. Jain, V. K. Gupta et al (1998)** had been determined procedure to the medicament
cephixime in biological and pharmaceutical formulation & fluid by voltammetric analysis. The study of cepixime was done through electric reduction and adsorption in buffer solution of phosphate by C.V. or voltammetry of cycle type, D.P.Cad.S.V. or strip voltammetry of differentive pulse cathodic adsorptive type and strip voltammetry of squar wave cathodic adsorptive type S.W. Cad. S. V. at mercury drop electrode of hanging type. Traces or minute estimation of the pure medicamente in pharmaceutical and general preparation and formulations were done usinig stripping voltammetry of cathodic type.

- **S. F. Choragud et al (2007)** had been estimated concentration of cepixime by colorimetry analysis. For development and validation of method a colored complex was prepared by suing ferric chloride as reagent. The wavelength maxima was found to be 370 nanometer. The blue colored complex was prepared by using potassium ferrocyanide and FeCl3 solution. The drug was estimated from the complex.

- **D. G. Shankar et al (2006)** had been estimated the concentration of cepixime in pure drug and pharmaceutical formulations by two ultraviolet and visible spectroscopy. The double beam spectrophotometer of Shimadzu was used to develop and validate the both method. The drug was scanned in whole range. The wavelength of maxima ($\lambda_{\text{max}}$) was found to be 290 nanometer for ultraviolet spectroscopy and the colored complex was prepared with the help of FC (Follin Ciucaltea) reagent using alkali media to develop. The drug was analyzed and validate according to new ICH guidelines. The wavelength maxima of the colored compex was 720 nanometer.

- **B. S. Virupaxappa et al (2005)** had been determined the concentration of cefixime drug in pure form and pharmaceutical preparations and formulations. The spectrophotometric method was developed using variamine. The drug cefixime was treated with alkali solution to breakdown of betalactam ring of cephalosporin. The degraded product was treated with iodate to librate free iodine in acidic media. The
colored complex was formed at 572 nanometer using variamine that convert from blue to violet. The standard curve was prepared and drug was analyzed. The recovery studies was performed by the standard guidelines.

• **R. K. Maheshwari et al (2004)** had been determined the drug content of cephixime in pure drug and different dosage form and formulations like tablet, capsules, syrup, injection, powder etc by using hydrotopic solubilization techniques. The hydrotopes are sodium benzoate, sodium citrate, urea, sodium acetate (higher concentration) are used to solubilize the drug cefixime and then analyzed by double beam spectrophotometer. The reagent sodium tartarate is used to solubilize and analyzed by preparing standard curve. The hydrotopes solubilize by salting in phenomenon and the major advantage of the technique is the hydrotopes are non toxic, non inflammable, non evaporable, low in cost, harmless to environment & ecofriendly. The developed method of cefixim is rapid, sensitive, not time consuming and can be used estimation of pure drug and formulations by different dosages forms. The method was validated by recovery studies, robustness, ruggedness, limit of detection, limit of quantification, system suitability parameters, interday and intraday study of the drug stability, precision and repeatability, reproducibility etc. the wavelength maxima \( \lambda_{\text{max}} \) for cefixime drug is 288 nanometer and the linearity for the drug is between 5 to 30 microgram per milliliter.

• **R. K. Nanda et al (2003)** had been founded out two methods for drug content of cephixime and erdosteine in pure drug and various types of pharmaceutical dosage forms.. the first method is based on area under curve calculation and it is calculated between the range of 294.5 to 284.4 nm range. The other method of estimation is first derivative spectroscopy. The simultaneous determination of cefixime and erdosteine wad done with the help of equation at the wavelength maxima of 309 nanometer for cefixime and 227.5 nanometer for erdosteine. The developed method for cefixime and erdosteine is very sensitive, accurate, reproducible, repeatable and usable in daily analysis of drugs in analytical laboratory and industries.

• **M. M. Deshpande et al (2002)** had been estimated concentration of drug cefixime
trihydrate and ambroxol hydrochloride simultaneously in various types of dosage forms such as capsule, syrup, tablet, powder, injection etc. the two methods were developed and validated are first derivative spectroscopy by spectrophotometer and second method is Vierordt’ method of analysis. The first method determined the wavelength maxima using zero crossing at 238 nanometer for cefixime trihydrate and 275.5 nanometer for ambroxol hydrochloride ($\lambda_{\text{max}}$). the Vierordt’s method found the wavelength maxima ($\lambda_{\text{max}}$) for cefixime trihydrate (285 nanometer) and for abroxol hydrochloride ($\lambda_{\text{max}}$) is 244.4 nanometer. The developed methods were validated as per international conference of harmonization guidelines for stability in different mediums, inter and intraday study, repeatability, LOD and LOQ study, accuracy & precision, system suitability, linearity and range, ruggedness and robustness. The methods are very simply, rapid, sensitivity, low in cost and can be used to determine the concentration of these pure drugs and its pharmaceutical formulations.

- **S. Caei, W. Fang** and **F. Lei (2001)** had been applied the finding of cefixime by high performance liquid chromatraphy in serum of human and plasma also. The mobile phase for the drug cefixime was determined on the basis of hit and trial basis. The chromatograph was thermo hypersil keystone octadecylsilane in the dimension of 25 mm x 2.5 mm and 5 micron pore size. The mobile phase was taken as acetonitrile and phosphate buffer system of pH 2.6 in the ratio of eighty five to fifteen for estimation. The wavelength maxima was taken as 291.0 nanometer. The internal standard was taken as tinidazole for developing the method to analyse the drug. The developed H.P.L.C. method was validated according to United States Pharmacopoeia as linearity, range, ruggedness, robustness, limit of detection & quantification, recovery studies, stability studies by various types of stress and degradation. The method is correct, reproducible and repeatable, very sensitive, quick, simple, precise for routine analysis of drug.

- **V. V. Pisarev et al (2000)** had been showed the high performance liquid chromatographic method for cefixime in two different capsule dosage form (100 mg capsules) cemidoxor and (500 mg capsules) supermax. The method was developed using mobile phase and ultraviolet type of detector. The blood plasma and serum have been used as sample for analysis. The reproducibility of the drugs were found to be in
the range of 0.06 to 1 microgram per ml. the accuracy and correctness was in the limit of ICH guidelines. The bioequivalence study was done with the developed method. The method was useful to find out the pharmacokinetic study of the drugs different dosage forms. The developed method for two dosage form 100 mg & 500 mg is very sensitive, correct, reproducible

- **B. Huo et al (1999)** had been developed a method of analysis for cephixime and its pharmacokinetic studies in blood serum and plasma by high performance liquid chromatographic techniques. The mobile phase was identified on the basis of hit and trial basis as acetonitrile (ACN) and buffer of phosphate ate pH=4.2 and the ratio is 87% acetonitrile and 13% buffer for pharmacokinetic study in blood plasma and pure drug in pharmaceutical formulations. The chromatograph for analysis is eclipsed X.D.B C-8 type. The procedure was validated for precision & accuracy, limit of detection and quantification, reproducibility & repeatability, stability at different pH and different temperature at many places. The method for the study of pharmacokinetic study of cefixime is good, easy, precise, fast, cheap and are used in daily analysis of drug cefixime and its various studies in the pharmaceutical formulations.

- **P. B. Shah et al (1999)** had been studies high pressure liquid chromatographic, spectrophotometric & different spectroscopic techniques for the estimation of cefixime bulk drugs and its pharmaceutical preparation and formulations. The three methods of analysis of cefixme developed are simple easy, correct, precise, very rapid and ecofriendly to the society (HPLC, spectroscopic & spectrophotometric). These three methods are given below as in the first method the drug cefixime is interacted wth 3-methyl-2-benzothiazolin-2-one hydrazone hydrochloride in the appearance of FeCl$_3$ (ferric chloride) that forms a complex with oxidative coupling to these reagents. The colored complex have wavelength maxima of ($\lambda_{max}$) 630 nanometer. The absorbance of the drug-reagents complex has been measure at six hundred thirty nanometer. The second method is base on the scanning of the drug in ultraviolet region and wavelength maxima is observered at 268 nanometer and 237 nanometer. The standard curve was contructed on the basis of dilution by Beer Lambert law and the linearity
The unknown concentration of drug was determined by these equations and validated according to ICH guidelines for cefixime drug and its formulation.

The third method is HPLC in which the mobile phase is taken on the hit and trial basis is phosphate buffer (dihydrogen phosphate buffer and methanol in the ratio of 78 to 22 vol by vol. the absorbance was measured on the $\lambda_{max}$ of 286 nm by the UV spectrophotometric detector and the chromatograph was lichrospher RPC-18. The method is validated by standards of United States of America USP pharmacopoeia and ICH and used in daily analysis in different laboratories and pharmaceutical industries.

- **K. Pasha et al (2004)** had been studies reverse phase high performance liquid chromatographic procedure for estimation of cefixime and its pharmaceutical dosage forms of different types. The chromatograph was SS aokosil C-18 type with the dimension of 4.5 mm x 250 mm with 5 micron particle size packing. The ratio of acetoCN, MeOH and ammonium acetate pH=5 in 44:16:40 was used in mobile phase. The flow rate of mobile phase was taken as 0.8 ml per minute.

- **H. Zhang et al (2003)** was found to identify cefixime and its preparations by reverse phase high performance chromatographic method. The mobile phase used for the procedure is the combination of 0.1 mol per ml ammonium acetate at pH=7 (this is adjusted by ammonia solution) & AcCN (acetonitrile) in the ratio of 94 to 6 of both solvents. The analysis was performed by the chromatograph hypersil B.D.S. C-18 with the dimension of 250 m.m x 4.6 m.m with 10 micron particle size. The wavelength maxima was 254 nanometer & temperature of chromatograph was 35ºC.

- **Sanjiv Arora et al., (2011)** demonstrated thermal and dissolution studies of Cefpodoxime proxetil drug and tablets. The study was authenticated and used in dosage form studies.
• **Umesh A. Nimbalkar et al., (2011)** prepared the solid dispersion of cefpodoxime proxetil with PEG 6000 to improve its solubility and dissolution rate. Cefpodoxime proxetil is class-IV drug according to BCS classification and it is having poor solubility and dissolution rate.

• **SN Borkar et al., (2010)** formulated floating bi layered tab. Of cefpodoxime proxetil. The direct compression method was utilized to prepare & formulated floated bi layered tablets of the drug cefpodoxime proxetil. Keeping site of tablet in the mind that the drug is put in the gastrointestinal tract by controlled of sustained way or manner after that into blood or systemic circulation. The formulated & prepared floated bi layered tablet of cefpodoxime proxetil was checked & evaluated for buoyancy, tablet floating time (T.F.T.), compatibility study, stability study and in vitro dissolution studies.

• **Madhusudan Sharma et al., (2011)** prepared and evaluated the hydrodynamically balanced system (floating tablets) of cefpodoxime proxetil by using HPMC of different grades. The polymer-medicament ratio, hydroxyl propyl methyl cellulose viscosity grades, various gas producing agents & diluents were being used to effect floating characteristics and release of medicaments to the formulated H.B.S.

• **Patel Sanket A et al., (2011)** developed simultaneous spectrophotometric technique for estimation of cefpodoxime proxetil & ofloxacin in formulations. The method was based on finding absorbance maxima for both drug at zero crossing level and further procedure is common to other methods as developed for various drugs.

• **U. A. Nimbalkar et al., (2011)** formulated solid lipid nanoparticles of an antibiotic i.e., Cefpodoxime Proxetil, with a view of its lymphatic as well as systemic absorption and controlled delivery of the drug. After observations of lipid nanoparticles, this can be stated that cost effective and bicompatible lipid i.e., Precirol could be utilized to prepare a powerful Solid Lipid Nanoparticulate formulation with smallest particle size to better percentage entrapment capacity & yield of practical. Interpretation of size of particles showed
that the particles had been of the range to size of 110-130 nm and had lymphatic as well as systemic absorption due lipid coating. *In-vitro* medicament flow studies revealed that flow from the SLN get successfully retarded for over 24 hrs.

- **Deepa Karthikeyan et al., (2010)** developed cefpodoxime proxetil or CP floating microspheres because of to receive increased retention in the gastrointestinal tract upper part. To make the safety of prodrug from attack of catalytic enzyme that can increase the absorption and increase the bioavailability. The spheres of micro size had been formulated by solvent evaporation of non aqueous type procedure utilizing various ratios of cefpodoxime proxetil and H. P. M. C. or hydroxyl propyl methyl cellulose K4M and ethyl cellulose (1:1:1, 1:1:2, 1:1:3, 1:1:4, 1:1:5 & 1:1:6) in the combination of DCM (methylenedichloride) and EtOH (C₂H₅OH/ ethanol) at ratio of 1:1 with tween eighty as surface active agent. This combination makes the floating microspheres of cefpodoxime proxetil pure drug in formulations.

- **Fahim Khan et al., (2010)** reported that cefpodoxime proxetil is poorly water soluble drug or we can say 400 microgram per ml is very low aqueous solubility of the drug that can produce various types of problem with the drug to develop and formulate different dosage forms of drugs. To solve this type of challenge the drug can be reduce to microparticle by utilizing high velocity homogenization of cefpodoxime proxetil. Application of salting out agents such as calcium chloride and sodium citrate with various types of polymers such as H.P.M.C (hydroxyl propyl methyl cellulose), chitosan, sodium alginate and methyl cellulose can precipitate these polymers on the surface of cefpodoxime proxetil. The pure drug and these formulated microparticles with different concentrations of polymers can be identified on the basis of solubility phenomenon morphology of surface (SEM or scanning electron microscope), drug release, drug content, thermal behavior (DSC or differential scanning calorimeter, stability studies and particle size). The in vivo study of these microparticles were used to study pharmacokinetic study of drugs. The formulations are best in the form of microparticles.
• **Vasu Kumar Kakumanu et al., (2006)** investigated the factors responsible for low oral bioavailability of cefpodoxime proxetil and demonstrated that hydrolysis of the drug in intestinal lumen and pH dependent solubility of cefpodoxime are the main factors responsible for its low oral bioavailability.

• **Sylvie Crauste-Manciet et al., (1997)** demonstrated hydrolysis of food material effects with the drug cefpodoxime proxetil into the lumen of intestine prior of absorption by comparing in vitro human food material & rabbit.

• **Nina saathoff et al., (1992)** investigated the pharmacokinetics of Cefpodoxime Proxetil and interactions with an antacid and an H2 Receptor Antagonist.

• **J. Bi et al (2004)** had been described procedure of estimation of cefixime in solid dosage form (Capsules) by reverse phase liquid chromatographic techniques. A ZORBAX C-18 chromatograph of dimension 250 m.m x 4.6 millimeter with 5 micron particle packing was utilized for analysis of cefixime in capsules. The flow rate of mobile phase was kept one milliliter per minute and the concentration of drug was detected by UV spectrophotometer detector at wavelength maxima or λmax of 288 nanometers. The mobile phase was selected with different combination of solvents like acetonitrile (AcCN) and 0.25 percent tetra butyl ammonia or TBA (0.4 mol per liter tetra butyl ammonia or TBA was diluted to thousand milliliter water; 25 ml) the pH value of mobile phase was adjusted to pH=7 with 1.5 mol per ml phosphoric acid) in the ratio of 1 volume to 2 volume of each solvents. The temperature of chromatograph was taken 25ºC.

• **S. A. Khan et al (2002)** had been described development and validation of the procedure for simultaneous estimation of cloxacillin and cefixime in tablet dosage form. The chromatograph was Kromasil C-18 Reverse Phase type was used with the dimensions of 250 m.m x 4.6 mm with particle packing of 4 microparticles. The mobile phase is combination of buffer of pH= 6.5 and MeCN or acetonitrile in the ratio of sixty five to thirty five of each solvents. The concentration of combination was measured at 225 nm
wavelength maxima. The R.T. of the both drugs cefixime and cloxacillin was 4.99 % 13.887 minutes.

- **G. Rathinavel et al (2005)** had been described the development and validation of procedure of simultaneous estimation of drug cefixime and drug cloxacillin by RP high pressure liq. Chromatographic method. The mobile was developed by using various trials and finally buffer of phosphahate at pH=5.0:acetonitrile: methanol in the ratio of eighty: seventeen: three. The chromatoraph was in 250 m.m in length, 4.6 in internal diameter and packing of particles were 5 microparticles. The flow of mobile solvents phase is 2 milliliter per minutes. 5.65 and 6.20 minutes were the retention time of cefixime and cloxacillin drugs in the combination. The developed method was accurate, precise, robustic, ruggedless, suitable for routine analysis.

- **M. V. Dhoka et al (2007)** had been reported the development and validation of cefixime and erdosystein in different dosage form of pharmaceutical preparationsby RP H.P.L.C. the medicament was separated on the HiQ sil C-8 chromatograph with 25 cm length, 4.6 mm internl diameter and 5 micron particle size packing. The selection of mobile phase was made on the combination of TBAH (0.1 N aq. Tertra butyl ammonium hydroxide) of pH = 6.5 with orthophosphoric acid: acetonitrile or MeCN in the ratio of two to one and the F. rate of m. phase was taken 1 ml per min. the separation was completed in eleven minutes and the detection wavelength was taken two fifty four nanometer.

- **K. Kathiresan et al (2005)** had been mentioned development and validation of cefixme drug and dicloxacillin drug simultaneously in combination with reverse phase high pressure liq. Chromatographic procedure in tablets and different dosage forms. The mobile phase was selected the mixture of potassium hydroxide buffer and acetonitrlre in the ratio of sixty to fourty of each type solvent. The rate of mobile liquid is one ml per minute in the range of 20 ml. the intersil C-18 chromatograph was used for the analysis of this combination. The detector wavelength was two hundred twenty nm for the measurement of concentration of drug.
• **Eric-jovanovic S, et al. (1998)** reported a H.P.T.L.C procedure to the estimation of ceftaxime, cefatriazone and cefixime high pressure thin layered chromatographic plates with pre coated silica gel. This contains combination of mobile phase ethyl acetate: acetone: MeOH and H2O in different ratios. The method was validated for the international conference of hormonazation for all quality control parameters to analyze this triple combination.

• **Gehad G. Mohamed, et al. (2006)** reported an easy rvery fast & correct spectroscopic procedure to the estimation of β-lactum drugs, fluoro cloxacillin and dicloxacillin in bulk drug & different preparations. The absorption of Fluclox and Diclox are recorded in different pH values ranged from 2 to 12 and the curves at pH 2-12 are characterized by two absorption bands at 225 - 270, and 225 - 274nm. for Fluclox and Diclox respectively. The developed method was validated for correctness, repeatability, stability to degradation, system suitability, reproducibility and limit of detection & quantification.

• **Nanda RK, et al. (2009)** reported correct, very rapid, reproducible, repeatable, economic, cheap and ecofriendly spectrophotometric method for estimation of cefixime and ornidazole in combination of pharmaceutical dosage forms. The technique is base on the simultaneous equation and wavelength selection of both drugs at zero crossing system and the wave length maxima was found to be for both drugs was two hundred ninty nanometer for cefixme and 312 nm for the drug ornidazole. The simultaneouse determination of cefixime and ornidazole was validated for various quality control parameters lime accuracy, precision, limit of detection, limit of quantification, robustness, ruggedness, stability and suitability according to United States of Pharmacopeia & ICH guidelines and after that method may be used for routine analysis of different dosage forms of these drugs in pharamacutical formulations.

• **Shahnaz Gauhar, et al. (2009)** reported a Revese Phase –High Pressure Liq. Chromatographic procedure to the detection of Cefixime in pure drug & in Capsule
Consisting of a LC-10 AT VP pump, SPD-10AVP UV/visible detector with Column as Bondapak C18 with m. phases composed of MeOH: Buffer solution (sod. dihydrogen phosphate) in the ratio of 35:65 at a flow rate of 1ml/min.

- **Dhoka Madhura V, et al. (2010)** reported a simple, precise, accurate and sensitive Reverse phase high liquid chromatographic method for simultaneous estimation of Cefixime trihydrate and Erdosteine in combined capsules dosage form. Drugs were resolved on a HiQ Sil C8 column (25x4.6mm) utilizing mobile phase of TetraButyl Ammonium Hydroxide (0.1N) pH adjusted to 6.5 with Orthophosphoric acid (10% aqueous) in a ratio of 2:1. Flow rate 1.0ml/min. at 254nm.

- **Kumudhavalli M.K, et al. (2010)** reported a R. P.-H.P.L.C. procedure for simultaneous determination of potassium clavanate and cefixmie in tablet formulations and pharmaceutical preparations by m. phase is composed to O.3 mol buffer of phosphate & MeOH in the combination of eighty four and sixteen with λmax of two hundred twenty nanometer and rate of flow is one ml per minutes. The method was validated for ICH guidelines.

- **Wankhede Ajit R, et al. (2010)** reported a method development and validation for estimation of cefixime and cloxacillin simultaneously in pharmacytucal preparations and formulations by reverse phase high pressure liquid chromatography procedure (RP-HPLC). The m. phase was the combination of tetra butyl ammonium hydroxide buffer and acetonitrile in the ratio of forty five to fifty five of both solvents. The adjustment of pH was done with orthphosphoric acid at pH=4 and the wavelength maxima was two hundred twenty five for the combination of the drugs. The flow rate was one ml per minutes and the method was validated for ICH schedules.

- **Yost RL et al (1985)** described the method development and validation for the estimation of cefotaxime and its metabolite desacetylcefotoxime simultaneously in serum and urine of human sample by utilizing reverse phase (RP) high pressure liquid chromatographic (HPLC) techniques in the laboratories. Acetonitrile or MeCN was used
to deproteinized the serum plasma. the supernatant was separated with the mixture of CH$_3$Cl (chloroform) and 1-butanol or BuOH after deproteinization. The cephalosporine and its metabolites (cefotaxime and its metabolite desacetylcefotaxime) was separated in one phase and the other phase is discarded from the phase solution. The aqueous phase is directly injected to the chromatograph by the injection to the running mobile phase of reverse phase high pressure liquid chromatography (RP-HPLC). A part of serum plasma water was dissolved in acetonitrile MeCN, 1-butanol and chloroform layer. The aqueous phase has higher concentration of cephalosporin (cefotaxime and its metabolites) as compared to original serum plasma samples. The similar procedure was used to measure the concentration of cefotaxime and its metabolites in the urine.

The standard calibration curve was made in the range of fifty to 250 microgram per ml range of concentration. This procedure was used to study pharmacokinetic study of cefotaxime and its metabolites in healthy person. This simple deproteinization procedure can be applied for the study of other cephalosporins like cephazoline, ceftriazone, cefposoxime, ceftazitidine, cephalaxine, cefprozil. Cefetamet etc.

- **Beeby et al (1978)** synthesized the following compound which are to be used opposite to a broad variety of G +ve & G – ve bacteria.

![Chemical structure](image-url)
• **Hoshi et al (1987)** provides a novel cephalosporin intermediate, 7-β-amino-3-[[Z-1-propenyl]cephem-4-carboxylic acid and ester thereof having the general formula.

![Chemical Structure](attachment:image)

...Wherein the general configuration of the 3-propenyl group is Z sometimes referred to as cis and R is hydrogen or a conventional carboxy protected group and acid addition salt thereof and the metal salt of the forgoing substance. wherein R is hydrogen. These compound are useful as intermediate for preparation of orally useful cephalosporin.


![Chemical Structure](attachment:image)

7-[2-amino(4-hydroxyphenyl)acetamido]-8-oxo-3-[(1Z)-prop-1-en-1-yl]-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid

B. M. Y. 28100
These derivatives are useful to the different pharmacy dosage form & are intermediate to separate thereof combination related to medicament that has isomer of E-1-propenyl group.

- **Ling SS et al (2003)** reported development and validation of cefotaxime for determination of its concentration in human and mouse serum plasma by reverse phase high performance liquid chromatographic technique using ultraviolet type of detector. The flow rate of mobile phase was taken to one milliliter per minute. The other degraded products of cefotaxime and its metabolites are deproteinized by using seventy percent of perchloric acid. The supernatant of serum plasma was directly injected into the chromatograph using microml syringe for injection of sample. Cefotaxime is degraded in acid medium and its degraded product are was remove by adding phosphate buffer before centrifuging the sample for detection. The mobile phase was identified on the basis of hit and trial basis and was taken as combination of 0.05 Mol per liter ammonium acetate, MeCN (acetonitrile), THF or tetra hydro furan in the ratio of eighty seven: eleven volume by volume and two of each above mentioned solvents. The detection wavelength maxima ($\lambda_{\text{max}} = 254 \text{ nm}$) was taken as two hundred fifty four hundred nanometer. The LOQ or limit of quantification for the procedure is $0.20 \mu g$ per ml. the recovery study was found to be more then eighty seven percentage for the drug concentration $0.20$ to $0.50 \mu g$ per ml. the interday and intraday variation or precision was to be found to eight percentage and accuracy was found to be three percentage. The speed, specificity, sensitivity and precision of this procedure make the technique for daily analysis of cefotaxime in mouse and human serum plasma suitably. The sample is required in $100 \mu l$ so it can be applied for analysis of plasma sample analysis of neonates.

- **Martinez LG et al (1998)** reported determination of cefotaxime with derivatization technique to naphthoquinoline sulphate (NQS) to form complexes and validate the method according to ICH guidelines. The derivatized 1,2 naphthaquinolone 4-sulphonate – cefotaxime complex was extracted with solid phase cartridge C-18 chromatograph and detected by ultraviolet visible system. Proper condition for newly
developed technique is using sodium bicarbonate buffer of 5.5 pH & reaction time is five minutes at 25ºC. the concentration of 1,2 naphthaquinone 4-suphonate is $7.1 \times 10^{-3}$ mol per liter. the procedure was used to determine concentration of cefotaxime in urine. This was checked for accuracy and precision. H-point standard method was used to determine concentration and compare results with reverse phase high performance method to analyze the urine sample of cefotaxime.

- **Nuevas L et al (1998)** reported extended wide spectrum third generation cephalosporin cefotaxime sod. Is devative of 7-amino cefalosporanic acid. The drug is prepared by reaction of S-2-benzthozoyl $\alpha$-methoxyimino-4-thiazole ethane thionate with 7-NH$_2$-cephalosporinic acid. The product was separated from different of by products such as 2-mercapo benzothiazole. The estimation of cefotaxime sodium synthesized from this procedure was done by derivative spectrophotometric techniques from the combination of products and biproducts mixtures. The method follows the Beer and Lamber's law in the concentration range of 0.005 to 0.080 mg per ml at two hundred seventy six nanometer wavelength maxima. The coefficient of correlation is 0.9995. the procedure is correct and reproducible with the value of relative standard derivation is equal to 0.4 percent & sensitivity index was 1.2 percentage. The validation of method is done for internal work. The recovery of drug from these methods is hundred percentage as obtained.

- **Scanes T et al (2001)** reported development and validation of cefotaxime sodium and its metabolite des acetyl cefotaxime by reverse phase high performance liquid chromatographic technique to estimate simultaneously the drug concentration in serum plasma of human and CSF or cebrospinal fluid. This method includes removal of proteins by deproteinization techniques and further isolation by R.P. high performance liquid chromatograph. The detection wavelength is ($\lambda$$_{max}$ = 262 nm). the R. T. (retention time) for cefotaxime and des acetyl cefotaxime 6.8 minutes and 2.2 minutes. The recoveries for both cefotaxime and des acetyl cefotaxime are seventy eight and eighty eight percentage. Linearity was found to be in the range of 0.58 to
940.0 µg per ml for serum plasma & for cerebrospinal fluid is 0.54 to 148 µg per ml. the method may used regular analysis of drugs in the daily life.

- **Dell D et al (1981)** reported development and validation of third generation cephalosporin cefotaxime sodium and its metabolites des acetyl cefotaxime for determination of concentration from physiological fluid. The deproteinization of drug sample and its metabolites were done with the solvents acetone (CH$_3$COCH$_3$) and chloroform (CHCl$_3$). The supernatant liquid was directly injected to chromatograph after deproteinization of cefotaxime and its metabolite for analysis. The identification of analyte is done on the wavelength maxima of two hundred sixty two nanometers. The urine sample can be analyzed to determine the concentration of cefotaxime and its metabolite desacetyl cefotaxime after deproteinization of urine with centrifugation for other related matters. The limit of detection was found to be 0.5 to 1 µg per ml. for serum plasma and five microgram for urine sample. The method is very useful to find out the concentration of cefotaxime and des acetyl cefotaxime in urine, saliva, serum, plasma, cerebrospinal fluid, infected wound secretions and pus. The two other lactones of cefotaxime metabolites in which beta lactam ring is broken can be identified by this technique. The method is simple and precise for two metabolites analysis containing lactone rings.

- **Dokladalova J et al (1983)** reported method for analysis of microgram amount of cefoperazone in human serum plasma and urine by reverse phase high pressure liquid chromatography using gradient techniques in which mobile phase is contiuously changed after a fixed time of interval. The procedure is utilizing µ Bondpack C-eighteen chromatgraph with two mobile phases gradients. This method is very useful for the separation of degraded products of cefoperazone and other pencillines such as ampicillin, cloxacillin, dicloxacillin, amoxicillin, sodium mehticillin, penicillin potassium G and aminoglycosides such as gentamicin, tobramycin, kanamycin, amikacin, neomycin, framycin may be demostered. The C.V or coefficient of variation is < 7.3 percentage or may be vey low received for cefpodoxime fifty to hundred µg per ml concentration of cefpodoxime for
both urine and serum plasma. The recovery studies was performed on serum plasma and urine sample by adding spiked bulk pure drug and the report was obtained in the range of 97.6 percentage to 98.6 percentage respectively. The lowest dateable amount is 1.0 microgram per milliliter from the sample of urine 0.1 ml and serum plasma 0.1 milliliter. The correlation between assay (percentage of purity) and microbiological assay using test organism micrococcus luteus A.T.T.C. 9342 was established. The antimicrobial method is less specific in the presence of other antibiotic and less time consuming. this method can be used to analyse the drug with the combinations of pencillins and aminoglycoside. The drug can be administered in combination of penicillins and aminoglycosides.

- **Li FS et al (2000)** reported development and validation of cefoperazone and sulbactum for the determination of drugs concentration from the different dosage forms by isocratic reverse phase highpressure liquid chromatography procedure within twelve minutes. The chromatograph was packed with hypersil octadecylsilane C-18 with various dimention of 25 centimeters length, 4.6 milimeter internal diameters and 5 µm particle packing in the column. Prepared by Dalian Elite manufacturing industry. The mobile phase is combination of water (H₂O) adjusted to pH of 4 with one percent phosphoric acid and (CH₃CN) acetonitrile in the ratio of eighty to twenty of each solvents. The detection wavelength was two hundred ten nanometer for the HPLC method and sample size should be 2µl. The linearity of method for sulbactum and cefoperazone is 100 µg per ml to 800 µg per & 100 microgram per ml to 1000 microgm per ml. the correlation coefficients for both are 0.9991 & 0.9997. the method can be applied for routine analysis.

- **Vikas Pareek, Santosh Tambe et al (2010)** studied the development and validation of spectrophotometric analytical techniques for the determination of cefoperazone in different dosage form by using different hydrotropic agents like sodium benzoate 2 molar, urea ten molar, sodium citrate 1.25 molar, sodium acetate 4 molar, potassium acetate 4 molar etc. the hydrotropic solubilization technique was applied to solubilize the drugs with these hydrotropic agent that work on salting in phenomenon on large
concentration of these hydrotropes. These hydrotropic agents (sod. Citrate, sod. Acetate, pot. Acetate, potassium citrate & urea) do not indicate any type of absorption above two hundred forty five nano meter so there is no interference with medicament cefoperazone that absorb at two hundred eighty nanometer wavelength maxima ($\lambda_{\text{max}}$) of the pure drug. The drug was mixed with these hydrotropic agents slowly at higher concentrations and agitated continuously with mechanical shekhar of Khera Company. The recovery studies was performed by each agents by adding spiked drug and recovery studies showed results between 87.19 % to 100.93 percent by method first and ninty six to 100.97 percent for method second. The reprodiblity of each method was performed and found to be precise for every procedures for the determination of cefoperazone with each hydrotropic solvents as sod. Acetate & citrate, potassium acetate & citrate and urea.the results of each method of cefoperazone was validated as per united states of pharmacopoeia. The AUC or area under curve techniques was observed more sensitive than conventional spectroscopic technique for estimation of cefoperazone. Cefoperazone was more soluble in urea and the LOD & LOQ was found more suitable for potassium citrate as comparision with other hydrotropic agents.

- **Abdollah Iravani et al (1991)** do Comparison of Cefperazone and Cefaclor for Treatment of Acute Urinary Tract Infections in Women. The one hundred and eight women that are suffered from acute urinary tract infections were selected from a college and treated with cefoperazone and cefclor cephalosporin antibiotics. 500 mg of cefoperazone B.M.Y. 28100-03-800 was administered once a day or OD and 250 mg of cefclor administerd thrice a day (TDS) for the treatment. The cure rate was ninty three to four percent for cefoperazone & 94 to 94 percentage for cefclore in one week treatment. Both the drugs cefoperazone and cefclor was found to be safe.

- **Hartmut Load et al (1992)** Christine Muller,Klaus Borner,Karl-Eric Nord and Petter Koepee studed the Multiple-“Dose pharmacokinetic of Cefperazone and its impact on intestinal flora of volunteers.
• **W.C. Shyu, R.B. Wilber et al (1992)** studied the effect on antacid on the bioavailability of Cefoperazone. Two way cross over study was designed to observe the effect of antacid on bioavailability of drug. Cefoperazone 500 mg drug was administered to eight healthy male subjects that may be contain or not thirty milliter antacid suspension of magnesium hydroxide \([\text{Mg(OH)}_2]\) or aluminum hydroxide \([\text{Al(OH)}_3]\). The ratio of \(Z\) cis or \(E\) tran isomers of cefoperazone was nearly ninety to ten. The drug is administered alone in the treatment schedule of A and the ratio of cis and trans isomers are 9.2 to 1.2 in the formulation. When cefoperazone is administered with antacids of magnesium hydroxide and aluminium hydroxide then the ratio of cis and trans isomers are 8.3 and 1.3 of each respectively. The result of analysis indicates that biavailability of cefoperazone does not effect with antacids.

• **Charlotta Idlund et al (2000)** studied effect on normal human microflora of oral antibiotics for treatment of urinary track infection. In eight volunteers who received 500 mg Cefperazone bd for 8 days there were minor increase in the number of enterococci, staphylococci and bacteroides in the intestinal in the intestinal microflora and a moderate decrease in enterobacteriaceae during the administration of Cefperazone. Three volunteers harboured C. difficile strains.

• **Nomeeta Gupta et al (2004)** Pediatric tonsillopharyngitis – An Evaluation of Cefperazone in Indian Patients. The emergence of penicillin resistant strain and the presence of co-pathogens have made the treatment of bacterial infection in children a challenge. Streptococcal tonsillopharyngitis which is a common infection has been treated with Cefperazone, a novel second generation cephalosporin. The aim of the present study was to evaluate Cefperazone in Pediatric tonsillopharyngitis.

• **Anna Jeli Ska, Beata Medenecka et. al. (2008)** performed the stability studies of Cefperazone in peroral suspension cefzil. Stress stability test was performed to study the stability of oral suspension of cefoperazone or CEFZIL. Reverse phase high performance liquid chromatographic method was utilized for the evaluation of degraded product of oral suspension using ultraviolet detector of wavelength of 280
The degradation of cefaperazone was occurred mainly through increased temperature and air wet humidity that may destroy the products. Parallel and reversible consecutive reactions causes product degradation. The second types of reactions are ten times more faster than second type of reaction. The first order rate constant was determined at the relative humidity of 76.4 percent at temperature of 333ºC, 338ºC, 343ºC, 348ºC and 353ºC.

- **Rajiv Dua, Suman Shrivastava et al (2011)** studied the Pharmacological Significance of Synthetic Heterocycles Scaffold.

- **S. Sharma, M.C.Sharma et al (2011)** studied the visible spectroscopic technique for the estimation of Cefperazone utilizing methyl orange. Simplest, rapid, accurate precise, low cost & precise (repeatable) Ultra Violet spectroscopic technique were being prepared for the simultaneous determination of Cefperazone in pure drug & solid tablet dosage form. The 1st procedure was on the basis the simultaneously equaticn and second upon estimation of Q value.cefproz! has absorption maxima at 373 nm. Beer’s law obeyed in concentration range of 5-35 µg/ml Cefperazone. The recovery studied from tablet are indicative of accuracy of method and are found in between 99.97-100.66% at 3 levels of standard addition.

- **Rajendra Kumar Sharma et al (2011)** done Correlation Studies of Topological Indices for Cephalosporin Type Antibiotic Drug. In present work most of the available Cephalosporin derivatives are treated as a series and examined to their suitability for QSAR and QSPR. For this purpose 51 Cephalosporin derivative compounds are selected and various Topological and Geometric indices are calculated with the help of DRAGON software. Then obtained indices are inter correlated with the help of Microsoft Office Excel 2003. Correlation studies gives the result that in all selected indices for study viz. Wiener Index, Randic Connectivity Index, Balban Index (J), Detour Index (W), Harary Index (H), T (N-N), T (N-S), T (N-O), the inter correlation of Wiener index with Schultz Molecular Topological Indices (R2= 0.998) and Detour index (W) with Schultz
Molecular Topological Indices (SMTI) shows strong result (R2 value = 0.993) as compared to other indices. The other correlation like Wiener-Detour, Weiner–Harary, Weiner–Schultz Molecular Topological Indices, Detour-Harary, Detour-SMTI, Harary-SMTI also shows strong correlation (R2 >0.9802). So these can be used for QSAR and QSPR.

- **Dr. Sapna Patil, Dr. Kumar T.N et al (2011)** reported analysis of application of single and in combination to head and neck onco surgeries. This is cost effective type of analysis.

- **Baxter Healthcare Corporation (2014)** emphasized following intramuscular injection oto unit five hundred milligram or one gram dose of cefotaxime inj. for simple human being volunteers plasma serum mean peak concentration were attained within 30 minutes (eleven point seven & twenty & half) µgram per milliliter consequently. Nearly sixty percent of the administered dosage had been excreted from urinal at the time of first six hrs after the beginning of the drip. Nearly twenty to thirty six percent of an Intra Venous dosage of cephotoxime sod. is eliminated through renal flow as without change cephotoxime & fifteen to twenty five percentage as the des acetyl metabolites. The des acetyl metabolites also have been found show the activity of anti-bacterial type. Cefotaximes two other lactone ring containg metabolites have the concentration of twenty to twenty five percentage have no antibacterial type of activity. The cefotaxime is third generation parental cephalosporin antibiotic. The optimum adult dose should not more than twelve micro gram per ml.

- **Yiqi Huang et al (2014)** determined the effect of release of cefotaxime sodium from modified starch-g-polylactic acid and the release increased with raise in pH of buffer.

- **Aphios Corporation USPC424499 (2013)** invented the nature of dosage form, method of preparation of micro particulates consisting proteins or derivatives enclosed in polysaccharides or derivatives and applications of the formulation in animals and humans to produce immunization.
• **Le Shin Chang** USPC424499 *(2013)* invented the processes of encapsulation of growth factor in a polysaccharide micro-particle.

• **Murugesh, S. et al** *(2013)* reported the nature of grafting and cross linking of Chitosan-acryl amide and polyethylene glycol to extend the release of Cefotaxime from hydrogels based delivery system.

• **David W. Grainger et al.,** *(2012)* reviewed that hard tissue disorders and diseases are the main causes of physical disability. He indicate that novel drug delivery devices for combination device applications intra-operatively, efficiently undergoing drug therapies on implanted hard tissue fixation devices.

• **Govind Asane et al.,** *(2012)* fabricated gastro retentive sustained release microparticles containing hydroxy propyl methyl cellulose derivative and chitosan as retardant material. The study demonstrates that the drug release from the formulation was found to be extended. It indicates that the increase in concentration of both the polymers shown to sustain the release of active ingredient.

• **Shagufta Khan et al.,** *(2012)* stated microspheres take much consideration in the area of long time relay, and useful for the treatment of cancer by these microspheres.

• **Stefania racovita et al.,** *(2012)* determined the absorption kinetics and equilibrium of cefotaxime sodium salt on chitosan-polybetaine complexes. This study carried out as a preformulation study in the development of oral drug delivery system.

• **Bhatt et al.,** *(2011)* studied that the processing variables effect in preparation and growth of biodegradable microparticles. The principal method of encapsulation is by emulsion solvent evaporation technique includes 2 principal processing steps, Growth of droplets with stability containing drug polymer combinations with organic solvents that is removed very fastly by evaporation techniques.

Itai Cohen et al., (2011) fabricated microparticles using selective withdrawal of one solvent so that coating of small particles with polymer films takes place. By using a single tube he determined that 10,000 particles can be generated per hour.

Prasanth V.V et al., (2011) emphasized different types of microspheres bioadhesive microspheres, magnetic microspheres, floating microspheres, radioactive microspheres, and polymeric microspheres further divided into biodegradable polymeric microspheres, and synthetic polymeric microspheres.

Vijaya Ramesh et al., (2011) used different polymers for the development of microparticles for controlled release of antibiotic drug. Microspheres were prepared by emulsion solvent evaporation technique. Attempts are also made to increase the entrapment efficiency by changing experimental variables.

Shekhar, K et al., (2011) investigated the release of cefotaxime sodium from microparticles using ethylcellulose as retardant polymer.

Adrian Raiche USPC528354 (2010) invented the processes of manufacturing microparticles by emulsification method using solvent and salt in a continuous medium.


Jaromir Hubalek et al., (2010) determine the possibility of magnetic nanoparticles for drug delivery and drug therapy is to carry the active drug to the specific site of action
and thereby treat it knowingly, without affecting other areas on the body. Increasing the magnetic property is beneficial to facilitate modification in drug delivery designs. He listed most commonly used as source of magnetization materials and some others. It is evident that only maghemite and magnetite suitable for biouse.

- **Ketie Saralidze et al., (2010)** fabricated polymeric microspheres for range of applications in therapeutics. The components of the microparticles changes with the site of application and therefore different materials has been utilized to produce microparticles. Alteration of the surface with constituents of the extra-cellular matrix, would prompt adhesion of cells, and therefore, stronger fixing of the microparticles at the injection site.

- **Khan et al., (2010)** determined the floating behavior of micro particles utilizing viscosity of low grade H.P.M.C. or hydroxyl propyl methyl cellulose. He concluded that using analytical techniques the co acervation is a preferable procedure to formulate micro particles of floating type applying addition of non solvent and viscosity of low grade type of cellulose polymers.

- **Ye M. et al., (2010)** shown that biodegradable microparticles can be applied in long-term protein delivery. The conventional way of delivering a protein drug needs daily, sometimes multiple, injections to achieve its therapeutic effectiveness. To perfect patient compliance and ease, sustained release dosage forms have been developed. This section examines the properties of protein-loaded microparticles, in specific, protein loading and release characteristics from polymeric microparticles.

- **Dalmoro et al., (2009)** referred enteric microparticles for controlled and made drug delivery applications through different ways of microencapsulation (namely single emulsions: water in oil-W/O; oil in water-O/W; or double emulsions: water-in oil-in water-W/O/W) and their impact on final properties of the product. Microcapsules or microspheres can be designed to progressively release active ingredients. A coating
may also be given to open in specific areas of body “smart polymers” which are perfect candidates for advancing self regulated delivery systems.

- **Alagusundaram M. et al., (2009)** described micro spheres were typical powders of free flowing nature enclosing synthetic polymers such as HPMC, methyl & ethyl cellulose & proteins that found to be bio degradable in nature. This was safe equipment to bring the medicament to the hitting target site to definite, though transformed, & to record required concentra
tion at site of good place by unfavorable results.

- **Anderson D. G. et al., (2009)** developed microparticles for controlled drug delivery using a microfluidic flow-focusing device. He formulated biodegradable drug-loaded microparticles by uniting the formation of droplets in a microfluidic flow-focusing producer with rapid evaporation of solvent from the droplets.

- **Maria Letizia Manca., (2009)** developed chitosan microspheres by precipitation method containing rifampicin. He concluded that the PLGA polymer is superior that chitosan, for the formation of microparticles.

- **Naikwade S. et al., (2009)** studied the pulmonary delivery of budesonide microparticles formulation and *in vitro* determination by spray drying. Prepared Microparticles were spherical in shape and they are characterized by smooth surface with low-density particles. Formulations shown extended *in vitro* drug release for hours thus use of microparticles possibility offers sustained release profile along with increase delivery of drug to the pulmonary tract.

- **Ravi Kumar Reddy J. et al., (2009)** investigated the delayed release microparticles prepared from different polymers by emulsion-solvent evaporation method and examined the physico-chemical characters. The mechanism of drug release was set up to be erosion as it was caused by (1-Mt/M) 1/3 versus time plots. Relative drug release study allow that the formulated product have more sustained effect than the marketed product.
• Roy S. et al., (2009) prepared mefenamic acid microsperes by cross linking chitosan with gluteraldehyde. The in vitro release pattern was found to follow zero order release as the dissolution exponent come nearer to 1.

• Sree Harsha et al., (2009) demonstrated the possibility of site-specific targeting albumin microsperes to deliver drug to the organ without affecting other areas of the body. Following intravenous administration the drug concentration of microparticles group in organ of mice after 15 min when compared to that of controlled.

• Vasiliu S. et al., (2009) designed microparticles based on acrylic ion exchange resin as delivery system. Resin microparticles were prepared by suspension polymerization technique and then core-shell microparticles are prepared by immersing into polysaccharides aqueous solutions containing cefotaxime.

• Beata Chertok et al., (2008) determine the possibility of magnetically controlled nanoparticles for the delivery of drugs to brain cancer using iron oxide. In vivo study of magnetic targeting reveals that the nanoparticle gets accumulated in cancers of rats was identified with images of MRI.

• Lu et al., (2008) formulated microparticles with an ability to enter ovarian carcinoma using PLG polymers. These microparticles were prepared by solvent evaporation method. The present study provided several findings that may be applied to improving intraperitoneal therapy.

• Parthiban, K. (2008) determined the invitro release of niosomes by diffusion model using dialisys membrane tide to open cylinder inserted into a medium containing buffer.

• Ajay Kumar Gupta et al., (2007) determine the magnetic nanoparticles ability to deliver drugs, proteins and antibodies to cell, tissue or tumors. He also reviewed magnetic
particles applications for early diagnosis of chronic diseases such as cancer, atherosclerosis and diabetes.

- **Daniel S. Kohane.**, (2006) studied that generally, microparticles have the inability to cross most biological barriers, and they should be delivered directly to the site of action. Micro- and nanoparticles for drug delivery has become the tool in area of research and, growth, in clinical practice, food, cosmetics and other industries.

- **Siepmann, J. et al.**, (2006) envisaged that microparticles offer an effectual defence of active agents that are encapsulated after degradation (ii) chance release rate is precisely controlled by incorporation of medicament above times of hrs to months, & (iii) route of administration should be very easy.

- **Kevin et al.**, (2006) emphasized that controlled release drug delivery systems are being evolved to address many of the difficulties connect with conventional methods of administration. Controlled release drug delivery utilize devices—such as polymer-based disks, rods, pellets, or microparticles—that incorporate drug and release it at controlled rates for comparatively long periods of time.

- **Rouholamini najafabadi et al.**, (2006) studied the cause of subtle lactose as an excipient on aerosolization of cefotaxime as dry powder formulations. He determined the deposition profile of a drug, cefotaxime, using coarse and fine carriers.

- **Ajay Kumar Gupta et al.**, (2005) reported that micro and micromolecules such as, enzymes, proteins, antibodies, or nucleotides and drugs can be targeted to the specific site to an organ, tissue, or tumour by binding these substances to polymeric magnetic nanoparticles under the influence of an external magnetic field.

- **Desai et al.**, (2005) demonstrated drug release that when the amount of polymer increased in microparticles. The highly important variable use to be the crystallinity of
the drug, volume of polymer solution added, and molecular weight of polymer, significantly changes particle morphology and release rate.

- **Kinam Park et al., (2005)** evidenced that the drug delivery has grown increasingly significant mainly due to knowledge of problems found to be present in various types of old and new medicaments. Of the numerous polymeric drug delivery systems, biodegradable polymers have been used broadly as medicament release process due to biodegradable and biocompatible behavior of these polymers.

- **Vinod Labhasetwar et al., (2005)** developed iron oxide nanoparticles which is capable of sustained and controlled intracellular delivery of anticancer agents. He also emphasized that the formulation may be used as a delivery device for systemic administration of hydrophobic drugs while at the same time permitting magnetic targeting and/or imaging.

- **Sathesh Kumar S. et al., (2004)** formulated and carried out physico-chemical evaluation of polystyrene nanoparticles containing sodium salt of cefotaxime. Preparation was made by emulsion polymerization and the graphical representation indicates that the release of the drug from the nanoparticles followed zero order kinetics.

- **V. R. Sinha et al., (2004)** reviewed the possibility of using biodegradable and biocompatible natural polymer with improved dissolution and serves as a carrier for hydrophobic drugs. The author also considered the factors that affect the incorporation efficiency and release of drugs from chitosan microparticles.

- **Yeo, Y. et al., (2004)** revealed initial burst is ordinarily unwanted because the drug released in this time is not accessible for prolonged release, and, more significantly, it can effect in toxic side effects. In order to inhibit the initial burst and gain effective control over the release rate, it is needful to realize possible causes of the initial release and relevant formulation variables.
• **Arul, B. et al (2003)** determined the *invitro* release of microspheres by diffusion model using dialysis bag suspended in a medium containing buffer.

• **Pascal Le Corre et al., (2002)** formulated bupivacaine incorporated microparticles using spray-drying method and reported that the prepared microparticles were able to control the release of the drug. He reported that the release pattern shows a zero-order absorption profile for 24 hrs.

• **Jong eun lee et al., (2001)** studied the preparation and evaluation of microparticles formulated from natural polymer hyaluronan. In this study the quality of hyaluronan as a carrier system for sulfadiazine was evaluated and their physiochemical properties were decided.

• **M Tuncay et al., (2000)** fabricated microparticles for parenteral delivery of diclofenac sodium and the release rate is controlled by poly (lactide-co-glycolide) polymers. The designed drug delivery systems were formulated for intra-articular administration in patients with severe inflammatory disease.

• **Dubernet C et al., (1999)** compared two ethylcellulose forms as raw material and microsphere using thermal analysis study. Ethylcellulose microspheres were prepared by the emulsion solvent evaporation procedure. Author had determined that the major physicochemical properties of the polymer remain unchanged.

• **M Guyot et al., (1998)** optimized the effect of nifedipine/ethylcellulose/hydroxypropyl cellulose viscosity, or ethylcellulose/hydroxypropylmethylcellulose viscosity on the physical properties of microparticles like particle size, drug content and release kinetics.

• **Dabbagh M.A. et al., (1996)** determined the release rate of anti-hypertensive drug, from matrices containing ethylcellulose can be transformed using smaller particle sizes and a lower viscosity grade of cellulosic polymers. Cellulose appeared to alleviate the penetration of water into the wafers consist of HPMC: ethylcellulose.
• **R J. Ko et al., (1991)** investigated the nature of cefotaxime and its metabolite in patients with chronic parenchymal liver disease. Toxicity is indicated in patients chronic liver abnormalities due to high therapeutic index of the drug, and dosing adjustment may not be required.

• **Ulf, D. et al., (2007)** determined the possibility of Superparamagnetic iron oxide nanoparticles as a promising tool to diagnose the tumours identified by MRI scanning.

**Pharmacopeia methods of cephalosporins**

- **Cefotaxime Sodium (U. S. P. 30/N. F. 25)** describes assay method of cefotaxime sodium not less than 916 µg and not more than 964 µg of \((C_{16}H_{17}N_{5}O_{7}S_{2})\).

  **Assay:** (0.05 M/L Phosphate buffer) – add 7.1 gram anhydrous dibasic sod. Phosphate \((Na_3PO_4)\) in 1000 milliliter of \(H_2O\).

  **Sol^n A:** prepare a combined solution of MeOH (Methanol) and phosphate in the ratio of fourteen to eighty six. 0.05 mol per lit. Filter using porous media aid of .05 micron or less. For degassing sonicate the mobile phase in sonicator before use.

  **Sol^n B:** prepare a combined solution of CH3OH (Methanol) and phosphate in the ratio of forty to sixty Filter through filter using porous filter aid of .05 micron or less. For degassing sonicate the mobile phase in sonicator before use.

  **M. phase**: utilize different combination of sol^n A & sol^n B instructed to mobile phase preparation.

  **Standard solution & method**: transfer forty miligram cefotaxime sod. weigh to 50 ml volum. flask add forty mililiter of sol^n A. after that liquid chromatograph is used with dimensions of \((3.9 \text{ mm} \times 15 \text{ cm} \text{ with packed size of } 5\mu \text{m} \text{ at } 30^\circ \text{ C. }\) the wavelength maxima is taken 235 nm in detector. The sample and standard solutions are respectively \((20 \mu l)\). the retention time for desacetyl ceftoxime and 14 minutes for cefotaxime sodium. The formula for calculating drug content

  \[
  = 50 \times C \frac{P}{W} \times \left(\frac{r_u}{r_s}\right)
  \]
British Pharmacopoeia (2009) reported method of analysis of cefotaxime sod. By reverse phase liq. Chromatography to find out the concentration of the drug by this procedure. The mobile phase was taken as a combination of mixture 0.05 mol per litre phosphate buffer and CH3OH or methanol in the ratio of eighty six to fourteen of each solvent. The drug concentration was measure at wavelength $\lambda_{\text{max}}$ of 234 nanometer with the retention of drug was found to be thirteen minutes. Calculate the percent purity of drug sample by multiplying the factor 1.048 and the flow rate was one ml per minute.

Indian Pharmacopoeia (2007) reports the analytical procedure of the determination of percentage purity of cefotaxime sodium to find out the concentration of drug with the mobile phase of forty gram potassium di hydrogen phosfate (KH$_2$PO$_4$) & 1.2 gram of di sodium hydrogen phosfate (Na$_2$HPO$_4$) in thousand milliliter of H$_2$O & combining to one twenty milliliter MeOH. Chromatograph is made of stainless steel with the length of thirty centimeter. F. R. was 1.5 ml/min of mobile phase. Spectrophotometer reading was measured at two hundred fifty four nanometer. The vol$^m$ of injector loop had been 20 µl. inject the sample and reference solution and find the concentration. The method is not correct till the relative standard deviation (R.S.D.) for replicate injection is < 2.0 %, inject the reference sol$^n$ & test sol$^n$. measurement the amount of C$_{16}$H$_{17}$N$_5$O$_7$S$_2$. The same method is used for cefotaxime sodium injection.

Cefaclor (U. S. P. 30/N. F. 25) reports assay method of cefaclor by using the equivalent of not less than 90% and not more than 120% the labeled content of C15H14ClN3O4S.

Cefaclor has a potential of greater than equal to 950 µg and less than 1020 µg of C15H14ClN3O4S per miligram, anhydrous form by calculation. The wavelength maxima $\lambda_{\text{max}} = 220$ nm detector is set by spectrophotometer.

Assay:- mobile phase water (780 ml) with 1 g of sodium 1-pentainesulfonate + 10 ml triethylamine) + 220 ml methanol. Adjust pH = 2.4 to 2.6 by H$_3$PO$_4$.

Standarard preparation:- fifteen gram of cefaclor drug in fifty milliliter of volum. flask holding mobil phase. Next step is dilution of solution with mobil phase. Sonicate the sol$^n$ for degassing. Liquid colmn is joint to 265 nanometer wavelength maxima
instrument and (twenty five cm long chromatograph including 5 micron packing x 4.6 milimiter radius of column). Rate of movement of mobile phase is 1.5 ml/min. retention time is 0.8 minute to cefaclor.

**Method:** [note:- peak responses are used as peak area] inject the same amount of standard drug cefclor twenty microliter of standard sample into the chromatograph. The HPLC is run by using the mobile phase and chromatogram is observed. The responses are major and the amount of the drug is measured by the given formula in milligram as compared to standard cefclor(C_{15}H_{14}ClN_{3}O_{4}S).

\[
= \left( \frac{W_s}{W_v} \right) \times (P) \times \left( \frac{r_v}{r_s} \right)
\]

Where \(W_s\) & \(W_v\) are the weight in miligram of United States Pharmacopoeia Cephaclor RS and Cephaclor carried for preparatn standard preparation and assay preparation respectively. \(P\) is desired strength in µgram of cephaclor (C_{15}H_{14}ClN_{3}O_{4}S) / miligram of United States Pharmacopoeia cefaclor RS and \(r_s\) and \(r_v\) are highest observations of cefaclor(C_{15}H_{14}ClN_{3}O_{4}S) peak received from the assay preparatn & the standard preparatn frequently.

**Capsules of Cephaclor** (C_{15}H_{14}ClN_{3}O_{4}S):- same procedure is taken only weighing of twenty capsule for assay is different & then powder in paste and mortar than take the required quantity of the medicament and proceed in same manner as in cephaclor medicament. The strength of capsule was measured and determined by the formula as mentioned above.

- **Ceftazidime (B. P. 2009)** reports assay method of Ceftazidime by using mobile phase of buffer pH 7 adjusted with ammonia (Ammonium dihydrogen phosphate) : acetonitile :water (8 : 24 : 68) with sampling of 10 µl in HPLC. The HPLC Column was 015 m long 4.6 internal diameters and 5 micrometer particle size of hexadecylsilica gel. The wavelength maxima (\(\lambda_{max}=245\) nanometer) was detected. The R. T. or retention T had been nearly 5 minutes. Calculate the percentage content of drug by multiplying with 1.048 factor. The movement of mobile phase flow was 2 mililiter per minute. the content of C_{22}H_{22}N_{6}O_{7}S_{2} was calculated.
• **Indian Pharmacopia (2007)** gives techniques of analysis for the determination of concentration of ceftazidime pure drug by reverse phase liquid chromatography. The chromatograph was made of stainless steel & ODS octadecyl silane and thirty centimeter length with 139 mm of radius. The mobile phase was prepared with the addition of potassium dihydrogen phosphate and disodium hydrogen phosphate buffer and methanol. The mobile phase was run and chromatogram is obtained and the amount of drug was calculated.

• **Ceftazidime (U. S. P. 30/N. F. 25)** reports assay method of ceftazidime by using the equivalent of not less than 95% and not more than 102% the labeled content of \( \text{C}_{22}\text{H}_{22}\text{N}_6\text{O}_7\text{S}_2 \).

Ceftazidime had a strength of > 950 µgram and < 1020 µg of \( \text{C}_{22}\text{H}_{22}\text{N}_6\text{O}_7\text{S}_2 \) per milligram, calculated on the basis of anhydrous or dry. The wavelength maxima \( \lambda_{\text{max}} = 220 \text{ nm} \) detector is set by spectrophotometer.

**Assay:**– mobile phase acetonitrile (40 ml) + 200 ml buffer. Adjust pH = 7 (Prepare with 42.59 g sodium phosphate & 27.22 potassium phosphate.

Standard preparation:– fifteen mg of ceftazidime in 50 ml of volume flask containing mobile phase. Further make dilution with mobile phase. Sonicate the solution for degassing. Liq. chromatograph is equipped with 254 nanomet wavelength maxima detector and (2.3 m.m.radius x 250 mm column containing 5 µm packing). The FR is 1.5 milliliter per minute. retention time is 0.8 minute for ceftazidime.

**Process:** [Note: utilize optimum area where optimum results are given]. Injection was given separately (nearly 20 µl) of standard preparation and the drug is administered into the chromatograph. Note the response in the form of chromatograms & record the results for the major peaks. Determine the strength in milig per mgram of ceftazidime \( \text{C}_{22}\text{H}_{22}\text{N}_6\text{O}_7\text{S}_2 \) taken by formula

\[
= (C/V) \times (r_v/r_s)
\]

Where \( W_s \) & \( W_v \) are the weight in mg of USP Ceftazidine RS and Cefperazone taken standard preparation and assay preparation respectively. \( P \) is designed strength in micro gram of ceftazidime \( \text{C}_{22}\text{H}_{22}\text{N}_6\text{O}_7\text{S}_2 \) / mg of U.S.P. ceftazidine R.S. & \( r_s \) and
are peak results of ceftazidine peak carried out from the assay prepara
and the stand. Preparat
respectively.
Ceftazidine capsule:- other conditions are same except we weigh 20 capsule for assay and then powder in pastle & mortar than carry out the required amount of the drug and proceed same procedure as in ceftazidine bulk drug.

- **Ceftriaxone Sodium (U. S. P. 30/N. F. 25)** describes assay method of cefotaxime sodium not less than 795 µg and not more than 964 µg of (C\textsubscript{18}H\textsubscript{18}N\textsubscript{8}O\textsubscript{7}S\textsubscript{3}).

**Assay:-** (0.05 Mol L\textsuperscript{-1} H\textsubscript{3}PO\textsubscript{4} buffer) – dissolution of four gram anhydrous dibasic Na\textsubscript{3}PO\textsubscript{4} in one liter of aqua solution.

**Sol\textsuperscript{A}:-** prepare a mixture of 13.6 g Dibasic Pot. phosphate & 4.0 g of Mono basic Potassium phosphate (pH= 7). Filter through filter using porosity of 0.05 µm or less. For degassing sonicate the mobile phase in sonicator before use.

**Sol\textsuperscript{B}:-** prepare a mixture of 25.8 g of sod. Citrate and citric acid (pH=5). Filter through filter using porosity of 0.05 µm or less. For degassing sonicate the mobile phase in sonicator before use.

**Movable phase:** combine mixture of sol\textsuperscript{A} (44 ml) & sol\textsuperscript{B} (4 ml) + 400 ml actonitrile as directed for mobile phase preparation.

**Standard solution & method:-** transfer four zero mili gm of U.S.P. ceftixaijone sod. And calculated volume of 40 ml of sol\textsuperscript{A} was added to volumetric flak after that liquid chromatograph is used with dimensions of (4.0 mm × 15 cm with packed size of 5µm at 30º C. the wavelength maxima is taken 270 nm in detector. The sample and standard solutions are respectively (20 µl) . the retention time for ceftriaxone sodium is 3.2 minutes.

The formula for calculating drug content

\[
= 200 \times \frac{C}{P \times W} \times \left(\frac{r_u}{r_s}\right)
\]

- **British Pharmacopoeia (2009)** mentions method of ceftriaxone sodium to find out strength or percentage purity by utilizing movable phase of phosphate buffer Potassium dihydrogen phosphate(KH\textsubscript{2}PO\textsubscript{4}) : methanol(CH\textsubscript{3}OH) (14 : 86) with sampling of 10 µ lit in HPLC. The wavelength maxima was selected to two thirty four.
Time of retention of drug by mobile phase was around thirteen minute. Calculate the percentage content of drug by multiplying with 1.048 factor. Flow rate was 1 ml/min.

- **Indian Pharmacopoeia (2007)** mentions technique for determining the concentration of cephtriazone sodium by taking chromatograph of S.S. (thirty centimeter × 3.9 m.m) packed with octa decyl silyl (ODS) silica gel (three to ten micron). The movable phase was a solut^n formulated by adding sixty m.g. of pot.

- **B. P. 2009 (Cefixime)** provides procedure to determine cefixime with mob. phase of phosphate buffer Potassium dihydrogen phosphate : methanol or MeOH (14 : 86) with sampling of 10 µl in HPLC. The absorbance is measure with Ultravilet detector at the wavelength of two hudred sixty four nano meter. The retention time was nearly 13 minutes. Calculate the percentage content of drug by multiplying with 1.048 factor. Flow rate was 1 ml/min.

- **I. P. 2007 (Cefixime)** gives analytical method to estimate cefixime by revers phase high performance liquid chromatography. 30 cm in length & 3.9 m.m. in diameter were packed with ODS silica G (3 - 10 µ.m). the movable phase was taken as 40 gram of potassium di hydrogen phosphate and methanol solution in the ratio of eighty six to fourty. This was run and wavelength maxima is selected for the measurement and calculation of cefixme.

- **Cefixime (U. S. P. 30/N. F. 25)** explains development of assay method for Cefixime in the range of 950 µg to 1050 µg of (C_{16}H_{17}N_{5}O_{5}S) anhydrous cefixime..
   Assay:- (pH 5 buffer) – two liter solution in water was prepared by dissolving 13.6 gram of phosphate buffer. pH was corrected to 5 by 10 N potassium hydroxide and mix well.
   Mob. Phase:- one liter solution was prepared by adding 960 ml of acetonitrile and 40 ml of phosphor buffer of pH=5 further this solution was filtered from a filter aid of fine porus . Do adjustment if compulsory. Increasing acetonitrile content of mobile phase
decreases retention time of cefadroxil & decreasing the acetonitrile contents increase the retention time.

Preparation of Standard:- standard solution was prepared by dissolving the weighed quantity of drug cefixime and the buffer of pH=3 to get a solution of keeping a standard conc. of 1.06 m.g. per m.l. the solution contain 1000 µg equivalent of Cefixime (C16H17N3O5S)/ml. use the solution for same day otherwise discard.

Procedure:- two hundred twelve gram of cefixime transferred to correctly weighed volumetric flask of twenty mililter. This solution was dilute with pH 5 to require vol & stirred by mechanical stirrer for five minutes. The high performance liquid chromatography of reverse phase is attached with ultraviolet detector and its detection wavelength was adjusted to two thirty nano meter for measuring concentration of drug. The movable phase was composed of buffer and methanol or acetonitrile. The chromatograph was taken with the length of twenty five centimeter and internal radium of 2.3 m. m. the flow of movable phase was taken one ml per minute. The capacity factor is between two to 3.5 and the column efficiency was found to be less than 2. The relative standard deviation was less than two percentage.

Method:- The 2 micro ml sample of the solution was injected directly to the column and the resulting chromatogram was shown with the peak under area curve. The concentration of cefixime was calculated and estimated from the curve.

\[ C = 200 \left( \frac{CP}{W} \right) \times \left( \frac{r_u}{r_s} \right) \]

Where C is concentr. of medicament and E is equivalent sample medicament. W is weight of medicament in mg (milligram) \( r_u \) & \( r_s \) are the realative peaks to determine strength of sample medicament & standard medicament respectively.

Analytical techniques for assay of Cefixime oral suspension was found to be same, in the case of Cefixime caps. & Cefixime tab. we weight the 20 capsules or tablets correctly and than taken the mentioned amount of medicament after making powder of the drug samples. Further processing is same as mentioned in the technique.

- **British Pharma. (2009) Ceftazidime** reported development and validation method to analyze Ceftazidime by taking m phase of phosphor buffer Potassium di hydrogen phosphate KH2PO4 : methanol (CH3OH) (fourty : eighty six) with sampling of tem µl
in High Pressure Liquid Chromatography of reverse phase. The drug was injected and its absorbance is measure at the $\lambda_{\text{max}} = 246$ nanometer. The measure concentration of drug was produce by chromatogram. The drug is retained for seven minute in the column and exited from the chromatogram to analyze it.

- **Ceftazidime (IP - 2007)** the drug is analyze by the high pressure chromatographic technique. The developed method utilized the chromatogram of standard size mean 25 to 30 centimeter in length and 2.3 mm in radius with totally porous five micron particle of silica G. the mobile phase is selected on the basis of hit and trial. The detector of determining the concentration of drug was selected ultraviolet type and this type of detector is best for measurement of concentration of drug with selected wavelength that is 254 nano meter with the flow rate of one ml per minute.