UTILITY OF PECTROHOTOMETRIC TECHNIQUE FOR MICRODETERMINATION OF RECENTLY DRUGS IN PURE AND IN HARMACEUTICAL FORMULATIONS

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This thesis consists of three main chapters:The first chapter (the introduction)Represents short notes about the structure and action of the three drugs under study (cefatoxime, ceftazidime and cefepime). It also includes a literature survey on the previous works carried out on the different techniques for the determination of these drugs.The second chapter (the experimental)Describes the procedures used throughout the study so as to get the optimum conditions favoring colored complex formation between the drug and reagent molecules by ion – pair mechanism. This chapter also describes the instruments used, how to prepare different solutions and the suggested procedure for determination of drugs either in pure or in dosage forms.The third chapter (results and discussion)Includes the results obtained throughout the work and their discussion, it is subdivided to three parts:Part I: Presents optimum conditions that favor the spectrophotometric determination of cefotaxime using the four reagents eosin bluish (EB), eosin yellowish (EY), bromocresole purple (BCP) and orange G (OG). These conditions are summarized as:i- Britton – Rhobinson universal buffer solution was found to be the best media for complexation process. This series of buffer solutions has the advantages of wide range of pH (2 – 12) and that its components do not interfere with the drug or reagents. It was found that, maximum tendency for complex formation takes place at pH 3.35, 3.35, 12.0 and 12.0 for EB, EY,BCP and OG respectively.ii- An evidence for complex formation between cefotaxime and the reagents is observed by determination of the maximum wave-length (λmax) of the colored complexes. It was found that complexes of cefotaxime with EB, EY,BCP and OG absorb maximally at 544, 538, 626 and 536 nm respectively.iii- Study on the effect of time and temperature showed that the complexes are formed within 5 minutes and remain stable for about 6 hours. Also, the obtained complexes are stable to heating up to 50oC.iv- The sequence of addition was found to be of significance importance. The best sequence of addition is reagent – buffer – drug. Thus, it can be concluded that buffered media are required to maintain the reagent molecule in the suitable form for complex formation.v- The stoichiometry of the complexes formed in solution was detected using the mole ratio and continuous variation methods. It was found that, all complexes are of 1:1 stoichiometric ratio. The stability constants of the formed complexes were calculated from spectral data of the two methods which indicate that these complexes are fairly stable.vi- The

optimum concentrations of cefotaxime which can be successfully determined by the reagents under study were detected by Beer's law. from the data obtained, it was found that cefotaxime was successfully determined up to 9.6, 5.8, 10.5 and 7.1 µg/ml on using EB, EY,BCP and OG respectively. The values of molar absorptivity (ε) lie within the range 6.36 – 4.03 x104 l mol-1 cm-1 and Sandell sensitivity in the range (0.048 – 0.088 µg/cm2). Such high values reflect the sensitivity of the proposed method. Regression analysis for the results were carried out using least square method. In all cases, Beer's law plots were linear with very small intercepts (-0.0084 - 0.0113) and good correlation coefficients (0.9884 -0.9989).vii- Another way for detecting the lower and higher limits of concentration was determined using Ringbom method where a satisfactory agreement between Beer's law and Ringbom methods was observed.viii- The accuracy and precision of the proposed method was determined by analyzing 6 replicate samples within the concentration range obtained from Beer's law and Ringbom methods. At these concentrations, the relative standard deviation (RSD) values are in the range 0.231 - 0.552, the detection limits are in the range 0.36 – 1.87 µg/ml and the quantification limits are within the range 4.05 – 8.4 µg/ml.ix- As an application of the proposed method, the content of cefotaxime in some local samples was determined. The results obtained agreed with the label claim and with those of the reference method. The performance of the proposed method was judged further by the Student's t-test for -accuracy and F-test for precision. At 95% confidence level, the calculated t- and F values did not exceed the tabulated values (t = 2.57 and F = 5.05) suggesting the accuracy and precision of the method.x- The accuracy and validity of the proposed method were further ascertained by performing recovery studies from standard addition technique using the four reagents EB, EY,BCP and OG. The pre-analyzed ampoule solutions (cefotaxime amp. And claforn) were spiked with pure cefotaxime at three levels and the total was found by the proposed method. Each determination was repeated three times. The results reveal good recoveries of pure drug added.Parts II presents the results obtained when the factors affecting the complexation of ceftazidime with the four reagents; eosin bluish (EB), orange G (OG), bromocresole purple (BCP) and arzanazo I (ARZ I) were studied:i- The optimum pH values required for complex formation are: 3.35, 7.81, 12.0 and 12.0 for EB, OG, BCP and ARZ I respectively.the λmax (nm) at which each complex absorbs are 537, 513, 626 and 565 nm for EB, OG, BCP and ARZ I respectively.ii- 2.0 ml of 1x10-3 M of reagent and 3.0 ml of buffer solution were found to be sufficient for complex formation.,iii- Complexes were formed within few minutes and unaffected by temperature up to 50oC time,iv- All the formed complexes are of 1:1 stoichiometric ratio as gathered from mole ratio and continuous variation methods.v- Using Beer's law, it was found that ceftazidime is successfully determined up to 15.40, 14.13, 12.32 and 12.11 µg/ml on using EB, OG, BCP and ARZ I respectively. The values of molar absorptivity (ε) lie within the range 3.43 - 6.31 x104 l mol-1 cm-1 and Sandell sensitivity in the range (0.036 – 0.072 µg/cm2). In all cases, Beer's law plots were linear with very small intercepts (-0.0017-0.063) with good correlation coefficients (0.9984 - 0.9993).vi- The accuracy and precision of the proposed method was determined by analyzing 6 replicate samples within the concentration range obtained from Beer's law and Ringbom methods. At these

concentrations, the relative standard deviation (RSD) values are in the range 0.035 – 0.132, the detection limits are in the range 0.29 – 9.78 µg/ml and the quantification limits are within the range 0.053 – 0.97 µg/ml.vii- As an application of the proposed method, the content of ceftazidime in some local samples (fourtum and fortaz) was determined. The results obtained agreed with the label claim and with those of the reference method. The performance of the proposed method was judged further by the Student's t-test for accuracy and F-test for precision. At 95% confidence level, the calculated t- and F-values did not exceed the tabulated values (t=2.57 and F= 5.05) suggesting the accuracy and precision of the method.Part III: Presents the optimum conditions that favor the spectro-photometric determination of cefepime using the four reagents, eosin yellowish (EY), eosin bluish (EB), orange G (OG) and arzanazo I (ARZ I). These conditions are summarized as:i- Studying the effect of pH on the complex formation between cefepime and the four reagents it was found that, maximum tendency for complex formation takes place at pH 5.35, 4.52, 12.30 and 10.21 for EY, EB, OG and ARZ I respectively.ii- The complexes of cefepime with EY, EB, OG and ARZ I absorb maximally at 622, 531, 531 and 563 nm respectively.iii- The complexes are formed within 5 minutes and remain stable for about 6 hours. Also, the obtained complexes are stable to heating up to 50oC.iv- The best sequence of addition is reagent – buffer – drug, indicating that buffered media are required to maintain the reagent molecule in the suitable form for complex formation.v- All complexes are of 1:1 stoichiometric ratio as shown from the data of mole ratio and continuous variation methods. The stability constants, calculated from spectral data, indicate that these complexes are fairly stable.vi- Using Beer's law, it was found that cefepime is successfully determined up to 10.42, 11.51, 8.78 and 9.76 µg/ml on using EY, EB, OG and ARZ I respectively. The values of molar absorptivity (ε) lie within the range 3.52 – 4.81 x104 l mol-1 cm-1 and Sandell sensitivity in the range (0.066 – 0.081 µg/cm2). In all cases, Beer's law plots were linear with very small intercepts (-0.011 – 0.018) with good correlation coefficients (0.9978 - 0.9998).vii- The detection of the lower and higher limits of concentration was determined using Ringbom method where a satisfactory agreement between Beer's law and Ringbom methods was observed.viii- The accuracy and precision of the proposed method was determined by analyzing 6 replicate samples within the concentration range obtained from Beer's law and Ringbom methods. At these concentrations, the relative standard deviation (RSD) values are in the range 0.040 – 0.247, the detection limits are in the range 0.02 – 0.064 µg/ml and the quantification limits are within the range 0.02 – 2.12 µg/ml.