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NEW SPECTROPHOTOMETRIC METHODS FOR THE DETERMINATION OF ZIDOVUDINE

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ABSTRACT

Two Simple spectrophotometric methods [A and B] have been developed for the determination of zidovudine in pure and its Pharmaceutical formulations. Methods 'A' is based on the formation of red coloured complex with ferric chloride and 2, 2^1-bipyridine having absorption maximum at 266 nm. Where as method 'B' is based on the oxidation of the drug with a know excess of oxidant, potassium permanganate. The excess permanganate is determined using the dye. Fast Green FCF at 355 nm. Beer's Law is obeyed at the range of 1-10 ug/ml for method 'A' and 2-12 ug/ml for method 'B'. The results obtained are reproducible and are statistically validated.

KEYWORDS : *Zidovudine, spectrophotometery, formulation.*

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INTRODUCTION

3-Azido -3,2 deoxythymidine, azidothymidine, AZT [Retrovir]. This nucleoside was synthesized in 1978 by Lin and Prusoff as an intermediate in the preparation of amino acid analogues of thymidine.

The authors have developed two simple sensitive and reproducible spectrophotometric methods [A and B] for the determination of AZT. In method A, AZT react with ferric chloride [$Fecl_3$] and 2. 2^1 bipyridine and forms a red coloured complex having absorption maximum at 266 nm. Where as in method it 'B' reacts with a know excess of oxidant, potassium permanganate [$KMno_4$]. The excess permanganate is determined using the dye, Fast green FCF [FGFCF] at 355 nm. The consumed potassium permanganate is proportional to the concentration of AZT present in simple.

Zidovudine is active against retroviruses a group of RNA viruses responsible for AIDS and certain types of leukemia, Retroviruses process a reverse transcriptases, or RNA- directed DNA polymerase are that directs the synthesis of a DNA copy [Proviral DNA] of the viral RNA genome that is duplicatal, circularized and incorporated in to the

DNA of infected cell. Nucleoside reverse transciptase inhibitiors (NRTT) after Phosphorylation in the host cell Zidovudine triphosphate selectively inhibits viral reverse transcriptase (RNA-depend DNA polymerase) in preference to cellular DNA polymerase.

Single stranded viral RNA

↓ Virus directed reverse transcriptase

[inhibited by zidovudine triphaphate]

Double – stranded viral DNA

Zidovudine two derivatives are [Stavudine d4T], [amivuadine 3TC], common side effects of AZT include nausea, headache change in body fat and discolouration of fingernails and toenails more severe side effects include anemia and bone marrow suppression.

Present work deals with the new spectrophotometric methods for the determination of zidovudine in pharmaceutical formulation.

EXPERIMENTAL

Material and methods :-

All the chemicals used were of analytical grade ferric chloride [3.3 x 10^{-3}M], 2, 2^1 bipyridine [1.0 x10^{-2} M] and [Na_2 So_4] [1.0m]were prepared in distilled water $kMno_4$ [2.0 x10^{-3} M] and 2.0 H_2so_4 and FGFCF [1.23 x 10^{-4} M] in 1.0 M. H_2SO_4 were prepared. The commercially available tablets were procured from the local market. Spectral and absorbance measurements were made on Systronics , UV-visible spectrophotometer model 117 with 10 mm matched quartz cells.

Standard and Sample Solution :-

About 100 mg of zidovudine was accurately weighed and transferred to a clean dry 100 ml calibrated standard flask and dissolved in methanol.

ASSAY PROCEDURE:-

Method A :–

Standard solution of AZT ranging from 0.5 to 2 ml [1ml = 100 ug] were transferred in to a services of 10 ml volumetric flasks. To that 0.5 ml of $Fecl_3$ [0.0033 M] and 2.0 ml of 2.2^1 biphyridine [0.01 M] were successively added and the final volume was made to 10 ml with distilled water. Then the flask were set aside for five minutes for complete colour developments. The absorbance of the real coloured species formed was measured at 266 nm against reagent blank and the amount of AZT present in the sample solution was computed from its calibration curve.

Method B :–

To a series of 25 ml graduated test tube aliquot sample of working standard solution of AZT ranging from 0.5 to 3 ml [1 ml = 100ug] were transferred 0.5 ml of $kMno_4$ solution was added and the total volume in each was brought to 10 ml with distilled water and kept aside for 15 minutes at room temperature. Then 4.0 ml of FGFCF solution a 4 ml of sodium sulfate were added successively after 10 minutes the volume was made up the mark with distilled water. A blank experiment was carried out in similar manner omitting the drug. The decree absorbance corresponding to the drug was obtained by subtracting of the blank from that of the standard solution the amount of drug present in the sample solution was computed from the standard calibration graph.

RESULTS AND DISCUSSION :-

The optical characteristics such as beers law limits sandell's sensitivity molar extinction coefficient percent relative standard deviation [calculated from the eight measurement containing ¾th of the amount of the upper Beer's law limits of AZT] % range of error [0.05 to 0.01 confidence limits] were calculated for both methods and the results are summarized in table-1.

Table 1 :- Optical characteristics and precision

Parameters	Method A	Method B
Beer's Law limit [ug/ml]	1-10	1-12
Sandell's Sensitivity [ug/cm^2 / 0.001 absorbence limit	0.0390	0.0140

Molar extinction coefficient [1 mole^{-1},cm^{-1}]	5.312x 10^3	1.7440x 10^4
% Range of error 0.05 confidence limits 0.01 confidence limits	± 0.0371 ± 0.5111	± 0.4738 ± 0.6036
Correlation coefficient	0.9999	0.9999
Regression equation (Y*)		
Slope (a)	0.0102	0.0558
Intercept (b)	0.0016	0.0014

Y*= b+ ac, where "c" is Concentration in ug/ml and 'Y' is absorbance limit.

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