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Degradation Study of Lincomycin by UV Spectroscopy

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Abstract: Lincomycin is obtained from a strain of *Streptomyces lincolnensis.* Lincomycin activity is mainly bateriostatic, though it could be bactericidal. According to ICH guidelines factors which causes forced degradation of a drug product comprise of temperature, time, photo degradation, pH variation (high and low), acid/base Stress testing and/ or with humidity. UV-Vi spectroscopy method was designed to examine and calculate the quantity of drug in the presence of degradation products. According to the USP, the official assay limit of the content should not less than 90% and not more than 120% of labelled amount of lincomycin. From our experiment we can conclude that lincomycin degrades most when exposed to U.V light and heat but do not degrades in basic medium whereas slight degradation occurs in acidic medium.

Key words: Lincomycin · Degradation · UV

INTRODUCTION

Lincomycin is obtained from a strain of *Streptomyces lincolnensis* [1]. Spectrum of activity of lincomycin is limited principally to *staphylococci* (including strains which introduces penicillinase), *pneumococci, C. diptheriae and haemolytic streptococci* [2, 3]. Lincomycin activity is mainly bateriostatic, though it could be bactericidal, depending on the the present concentration of antibiotic and organism [4, 5]. At an alkaline pH lincomycin exhibits maximum activity whereas in the presence of hydrochloric acid lincomycin nevertheless retains its activity, so that gastric acidity does not weaken its activity. With the oral administration of a single dose of 500 mg., 3.5 µg /ml is achieved in blood level within two hrs, declining to 2.2 µg. after 6 hours. So it seems possible to achieve efficient blood levels following oral administration of lincomycin. On parenteral administration of a 600 mg. intramuscular dose of lincomycin produces a peak level of 14.6 µg /ml. within 30 mins, declining to 5.5 µg. in 6 hours and to 2.5 µg. in 12 hours.. Lincomycin can be detected in significant amount in all tissues and also in the cerebrospinal fluid if the meninges are inflamed. Lincomycin excretion takes place in the urine and bile. Side effects of lincomycin are primarily gastrointestinal like vomiting, nausea, soft stools, diarrhea and also mucocutanoeus which includes stomatitis, epistaxis, urticaria vaginitis, pruritus, rash [6].

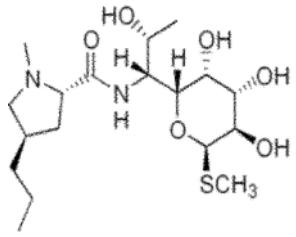

Fig. 1: Structure of Lincomycin

Spectrophotometry is usually prefer over other methods because of less equipment cost and economical maintenance advantage. Spectrophotometry technique is based on measuring the absorption of a monochromatic light in the near ultraviolet region (200-380 nm) by colorless complex. UV spectrophotometry can also be use for stress degradation. According to International Conference of Harmonization (ICH) guideline the active pharmaceutical ingredient is focused to various forced degradation conditions which are acidic, basic and light conditions [7]. During preliminary developmental procedures, forced degradation activities should be performed to make sure that the method is selective to save lot of effort, time, money and to detect the responsible conditions for degradation of drug [8]. Forced degradation is able to demonstrating that the chosen technique is stability indicating that is the technique use to identify the increase in the degradation product and the subsequent loss of active

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components [9]. Our research group has done these type of studies which is very useful for analytical chemist and pharmacist [10-14].

Parameters Involve in Forced Degradation: The distinctive forced degradation studies on drug substance involves acid/base stress testing, photo degradation, temperature and or with humidity, time, pH variation (low and high).

Acid/Base Stress Testing: Acid/base stress testing is used for the evaluation of forced degradation of a drug substance. This test involves degradation of a drug substance by exposure to basic or acidic medium over time to its primary (monomeric) degradation products. Acid/base hydrolysis occur in labile carbonyl functional groups which are amides (lactams), esters (lactones), aryl amines, imides, carbamates, imines and alcohols.

Degradation by UV Light: UV degradation is a main problem in numerous UV-unstable products which are made up of natural and synthetic polymers as they break or disintegrate when exposed to continuous sunlight. As the attack is dependent on the degree and degree of exposure. Nonstop exposure is a more serious problem than intermittent exposure.

Thermal And/Or Humidity Stress Testing: Thermal and/or humidity stress testing is performed by exposing the drug substance to thermal/humidity conditions in due course which causes the substance to degrade forcefully to its main components [8].

Experimental

Lincomycin: For the purpose of degradation study we take the active Lincomycin.

Glass Wares: Pyrex glass wares were used which includes measuring cylinder, beakers, pipette, funnel, stirrer and volumetric flask. For initially washing of glass wares we use chromic acid afterward we use water and finally rinsed with double distilled or DI water (freshly prepared).

Instruments: Ultraviolet Lamp (Serial no. N 045571, Power of 8N, LF-204.LS) '4W-254 and 365 nm', Spectrophotometer with a quartz cuvette (T80 Uv-vi spectrometer) 'PG Instrument', Weighing Balance (Item PA214C): 'Pioneer OHAIUS' and Water Bath 'HH-4' having digital and constant temperature tank.

Reagents: Analytical grade reagents were used which includes 0.1N Sodium hydroxide, 0.1N Hydrochloric acid and de-ionized water or double distilled water.

Preparation of 0.1 N Hydrochloric Acid: For the preparation of 0.1 N hydrochloric acid we take 8.3ml analytical grade hydrochloric acid having 37% purity and 12N normality in a volumetric flask and make up the final volume upto the mark of flask with DI water.

Preparation of 0.1 N Sodium Hydroxide: For the preparation of 0.1 N Sodium hydroxide we take 4 grams of sodium hydroxide and transfer it in 100ml volumetric flask and dissolve it in small quantity of water and finally make up the volume up to the mark of the flask with de ionized water.

Preparation of Lincomycin Solution: For the preparation of 200 ppm lincomycin solution we weigh the active Lincomycin 0.020 gm in a teared beaker then add small quantity of water and dissolve it. Transfer this solution in a 100 ml volumetric flask finally make the volume up to the mark with water.

To determine the absorbance spectrophotometer was used in which solution of 200 ppm were transferred to cuvette and absorbance were determined at wavelength max 243 nm.

Procedure for Degradation Studies:

For Acid: To determine the effect of acid on lincomycin, transfer 5 ml of 200 ppm solution of lincomycin in a test tube then add 5 ml of 0.1 N hydrochloric acid and left it for 30 minutes. After 30 minutes determine the absorbance of the solution by spectrophotometer at wavelength max 243nm.

For Base: To determine the effect of base on lincomycin, transfer 5 ml of 200 ppm solution of lincomycin in a test tube then add 5 ml of 0.1 N sodium hydroxide and left it for 30 minutes. After 30 minutes determine the absorbance of the solution by spectrophotometer at wavelength max 243nm.

For UV Light: To determine the effect of UV light on lincomycin, transfer 5 ml of 200 ppm solution of lincomycin in a test tube then add 5 ml of deionized water and left it for 30 minutes in UV light of 320 nm. After 30 minutes determine the absorbance of the solution by spectrophotometer at wavelength max 243nm.

For Heat: To determine the effect of heat on lincomycin, transfer 5 ml of 200 ppm solution of lincomycin in a test tube then add 5 ml of deionized water and left it for 30 minutes in water bath at 50°C. After 30 minutes determine the absorbance of the solution by spectrophotometer at wavelength max 243nm.

RESULTS

We study degradation parameters on active of lincomycin (Table 2 Figure 3). When lincomycin subjected to 0.1 N Hcl, lincomycin showed deccreased availability (80.625 %). In the same way lincomycin when subjected to 0.1N NaOH, lincomycin do not showed significant changes in terms of availability (110.213%). When lincomycin exposed to heat for 30 minutes, deccreased availability observed (70.082%) and when exposed to U.V light at wavelength max 243nm, lincomycin also showed decreased availability (69.994%).

From our results we can conclude that lincomycin when introduced in acidic medium i.e. 0.1N HCl it degrade to a little extend (80.625%) but do not degrade when subjected to basic medium i.e. 0.1N NaOH (110.213%). When lincomycin exposed to U.V light and heat for 30 minutes it degrade to a larger extent (70.082% and 69.994%) respectively.

Table 1: Absorbance Of Lincomycin

Degradation Parameters	Lincomycin 1	2	3	Average
Before	2.023	2.024	2.023	2.023
After acid	1.632	1.630	1.632	1.631
After base	2.216	2.237	2.237	2.23
After heat	1.420	1.412	1.414	1.415
After U.V	1.395	1.396	1.397	1.396

Table 2: Degradation Pattern In Terms Of Percentage Of Lincomycin

Degradation Parameters	Lincomycin 1	2	3	Average
Before	100.00%	100.049%	100.00%	100.016%
After acid	80.672%	80.533%	80.672%	80.625%
After base	109.540%	110.523%	110.578%	110.213%
After heat	70.192%	70.158%	69.896%	70.082%
After U.V	68.956%	68.972%	69.055%	69.994%

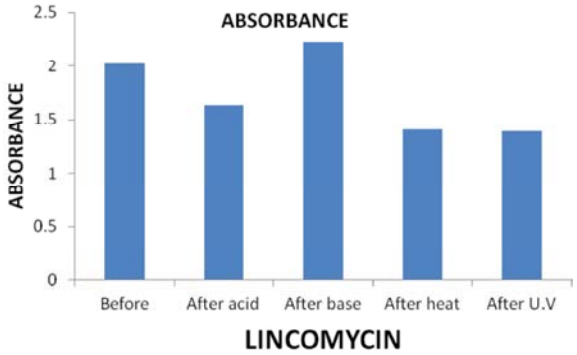

Fig 2: Absorbance Of Lincomycin

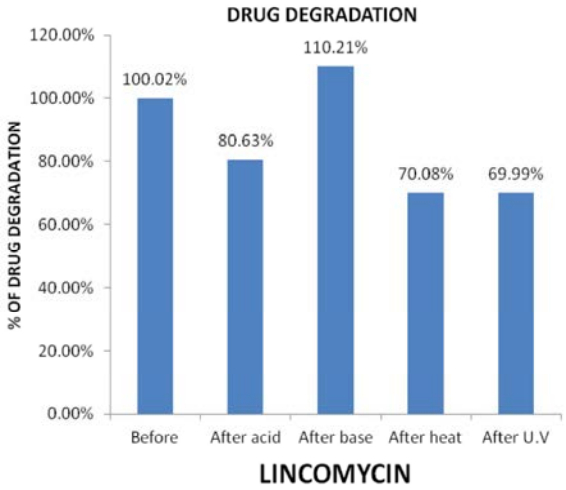

Fig. 3: Degradation Pattern

CONCLUSION

According to United States Pharmacopoeia (USP) specification, the official assay limit of the content should not less than 90% and not more than 120% of labeled amount. From our experiment we can conclude that lincomycin degrades most when exposed to U.V light and heat but do not degrades in basic medium whereas slight degradation occurs in acidic medium.

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