Research Article

# Kinetic Study on the Degradation of Phenobarbitone in Oxidative Condition by UV-Vis Spectrophotometric Method

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**ABSTRACT**

The aim of the present work is to detect kinetic rate of reaction of Phenobarbitone in bulk. Determination of rate of degradation, half life of Phenobarbitone when subjected to the oxidative degradation condition. A kinetic investigation of the oxidative degradation of Phenobarbitone was carried out in solution of Hydrogen Peroxide 6%,7%,8% by monitoring the parent compound itself. The method development was carried out using 0.1 N NaOH solution.

The linearity range was found to be 2-20µg/ml. the method showed high sensitivity with good linearity. The reaction order of followed pseudo-first order kinetics. Energy of activation was found to be 6.71 kilo joule $mol^{-1}$.

**Keywords:** Phenobarbitone, kinetic study of the degradation, UV-Vis Spectrophotometric Method.

## INTRODUCTION

The quality of a drug substance or drug product varies with time under the influence of a variety of environmental factors such as temperature, humidity and light. The two main aspects of drug product that play an important role in shelf life determination are assay of active drug, and degradants generated, during the stability study.[1,2]

Phenobarbital is called a barbiturate that acts by slowing down the activity of the brain. It has sedative and hypnotic properties, which will help patients to relax before surgery or help to sleep. It also reduces or controls seizures or convulsions, except for absence (petit mal) seizures. Generic Phenobarbital oral elixir is available containing a high amount of alcohol which can increase possible unpleasant effects.[3,4]

The present work aimed is to carry out this study that the determination of the degradation rate or kinetics of Phenobarbitone. The no work is carried out in the determination of the degradation rate or kinetics of Phenobarbitone. In MSDS sheet of Phenobarbitone it is mentioned that it is sensitive towards the strong oxidizing agent. So we mainly focus on the study of the oxidative degradation rate or kinetics of Phenobarbitone.

Chemically Phenobarbitone is 5-ethyl-5-phenylbarbituric acid or 5-ethyl-5-phenyl-2,4,6(1H,3H,5H) pyrimidinetrione or Phenobarbituric acid.[5] (fig. 1)

## MATERIALS AND METHODS[6-9]

### MATERIALS

Phenobarbitone sample was obtained from Abbott Healthcare, Mumbai. The solvent used distilled water (AR grade), NaOH (AR grade) and $H_2O_2$ (AR grade). These chemicals were purchased from Merck Chemicals (Mumbai, India).

### Equipment

The instrument used for the present study was Shimadzu Corporation, UV-Vis double beam ( Model UV-1800 240V) high speed scanning spectrophotometer.

### Kinetic studies[6,7]

**Preparation of stock solution:** Standard stock solution of Phenobarbitone was prepared by dissolving 10 mg of Phenobarbitone in sufficient quantity of 0.1 N NaOH and make up the volume upto 100 ml, which gives 100µg/ml solution.

### Preparation of working solution

From the above stock solution 1 ml was transferred into 10 ml volumetric flask and volume make up with 0.1N NaOH to give 10µg/ml. Then sample was scanned with

UV-Vis spectrophotometer in the range 200-400 nm and the wavelength corresponding to maximum absorbance was noted (λmax 232 nm).

**For studying the effect of concentrations of hydrogen peroxide at Temperature 30°c**

In 10 ml of volumetric flask, 1ml of stock solution were added and in that 6%, 7%,8% hydrogen peroxide was added respectively, volume was make up to the mark. Kept this solution at 30°c. after 3,6, 9 hrs. the absorbance of corresponding solution is taken. And calculate the % drug remaning is calculated by comparing the standard solution absorbance. Log of % drug remaining vs time in hrs. is plotted. (fig. 2)

**A. Studying effect of temperature on 6% hydrogen peroxide**

In 10 ml of volumetric flask, 1ml of stock solution were added and in that 6% hydrogen peroxide was added, volume was make up to the mark. Kept this solution at 30°c, 40°c, 50°c. after 3,6, 9 hrs. the absorbance of corresponding solution is taken. And calculate the % drug remaning is calculated by comparing the standard solution absorbance. Log of % drug remaining vs time in hrs. is plotted. (fig. 3)

**B. Studying effect of temperature on 7% hydrogen peroxide**

In 10 ml of volumetric flask, 1ml of stock solution were added and in that 7% hydrogen peroxide was added, volume was make up to the mark. Kept this solution at 30°c, 40°c, 50°c. after 3,6, 9 hrs. The absorbance of corresponding solution is taken. And calculate the % drug remaning is calculated by comparing the standard solution absorbance. Log of % drug remaining vs time in hrs. is plotted. (fig. 4)

**C. Studying effect of temperature on 8% hydrogen peroxide**

In 10 ml of volumetric flask, 1ml of stock solution were added and 3ml in that 8% hydrogen peroxide was added, volume was make up to the mark. Kept this solution at 30°c, 40°c, 50°c. after 3,6, 9 hrs. the absorbance of corresponding solution is taken. And calculate the % drug remaining is calculated by comparing the standard solution absorbance. Log of % drug remaining Vs Time in hrs. is plotted. (fig. 5)

**Method validation**

**Linearity**

Various aliquots were prepared from the stock solution (100µg/ml) ranging from 2-20µg/ml. The samples were scanned in UV-Vis Spectrophotometer against 0.1N NaOH as blank. It was found that the selected drug shows linearity between the ranges of 2-20µg/ml. (fig. 6)

**Precision**

Precision of the method was demonstrated by intraday and interday variation studies. In intraday variation study six different solutions of same concentration 12 µg/ml were analyzed three times in a day i.e. from morning, afternoon and evening. In the interday variation studies, solution of same concentration 12 µg/ml were analyzed three times for the three consecutive days and the absorbance result mean, standard deviation and % RSD was calculated and result shown in Table1,2,3.

Robustness, Ruggedness, LOD, LOQ was performed and summary of result shown in Table 4.

**RESULT**

**Kinetics of degradation**

Different parameters that affect the rate of the reaction were studied. The effect of temperature was studied by conducting the reaction at different temperature using different concentrations of hydrogen peroxide solution. (fig. 3,4,5)

At each temperature the rate constant and $t_{1/2}$ were calculated and then the log of the rate constant was plotted against the reciprocal of the temperature in Kelvin units. (Arrhenius plot (fig. 7) to demonstrate the effect of temperature on the rate constant.

It was conclude that as the temperature increased the rate of oxidation increased with decrease in the $t_{1/2}$ (Table 5).

Also, the energy of activation was determined by calculating the rate constant from the following equation.

$$\log \frac{k2}{k1} = \frac{Ea}{2.303R}\left(\frac{T2-T1}{T1T2}\right)$$

Where,

**Ea**- Activation energy

**$T_1$, $T_2$** – Temperatures degree in Kelvin

**R**- Gas constant

**$K_1$,$K_2$**- Rate constant at two temperature.

The calculated Ea was found to be 6.71 kilo joule $mol^{-1.}$

## CONCLUSION

In conclusion, the oxidation of Phenobarbitone was found to follow a pseudo first order reaction rate. Also the reaction rate increase in the temperature and the strength of the oxidative solution. The proposed method provides a simple, sensitive method sutaible for the quality control analysis of Phenobarbitone in the bulk .

**Table 1: Precision results showing repeatability of Phenobarbitone**

| Concentration (µg/ml) | Absorbance | Statistical analysis |
|---|---|---|
| 12 | 0.941 | Mean=0.940833<br>SD= ±0.00075277<br>%RSD=0.0800 |
| 12 | 0.942 | |
| 12 | 0.941 | |
| 12 | 0.94 | |
| 12 | 0.941 | |
| 12 | 0.94 | |

**Table 2: Intra-assay precision**

| Concentration (µg/ml) | Absorbance 1 | Absorbance 2 | Absorbance 3 | Average %RSD |
|---|---|---|---|---|
| 12 | 0.941 | 0.939 | 0.941 | 0.1136 |
| 12 | 0.939 | 0.939 | 0.941 | |
| 12 | 0.939 | 0.941 | 0.939 | |
| 12 | 0.941 | 0.941 | 0.939 | |
| 12 | 0.94 | 0.94 | 0.938 | |
| 12 | 0.939 | 0.94 | 0.941 | |
| %RSD | 0.1045 | 0.0951 | 0.1414 | |

**Table 3: Inter-assay precision**

| Concentration (µg/ml) | %RSD | | | Average %RSD |
|---|---|---|---|---|
| | Day1 | Day2 | Day3 | |
| 12 | 0.0864 | 0.1081 | 0.1203 | **0.104933** |

**Table 4: Summary of validation**

| Parameter | Result |
|---|---|
| Linearity indicated by correlation coefficient | 0.996 |
| Limit of Detection | 0.87µg/ml |
| Limit of Quantification | 2.65µg/ml |
| Range | 2-20µg/ml |
| Linear regression equation | 0.074x + 0.038 |
| Linear regression equation | 0.074x + 0.038 |

**Table 5: kinetic data of Phenobarbitone**

| Concentration of $H_2O_2$ | Temperature | K in hrs. | $t_{1/2}$ in hrs. |
|---|---|---|---|
| 6% | 30 ◦C | 0.075 | 9.24 |
| | 40 ◦C | 0.096 | 7.21 |
| | 50 ◦C | 0.133 | 5.21 |
| 7% | 30 ◦C | 0.0921 | 7.52 |
| | 40 ◦C | 0.0890 | 7.00 |
| | 50 ◦C | 0.1266 | 5.47 |
| 8% | 30 ◦C | 0.0921 | 7.52 |
| | 40 ◦C | 0.1289 | 5.37 |
| | 50 ◦C | 0.1469 | 4.71 |

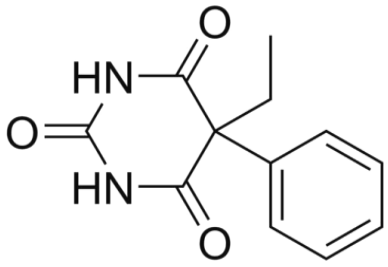


**Fig. 1: Representating the structure of Phenobarbitone**

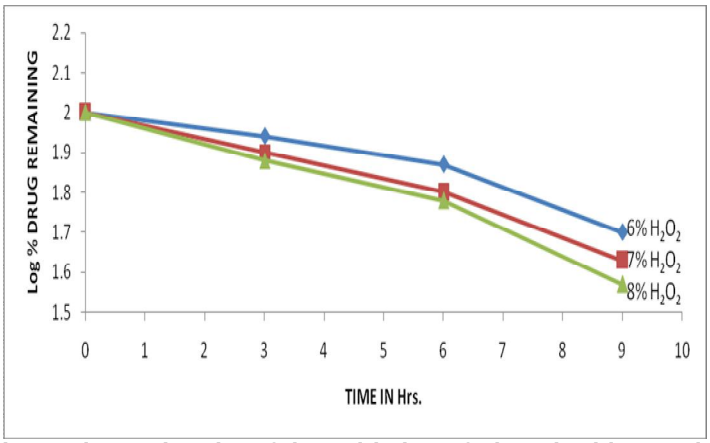


**Fig. 2: First order plot of the oxidation of Phenobarbitone with different concentrations of hydrogen peroxide**

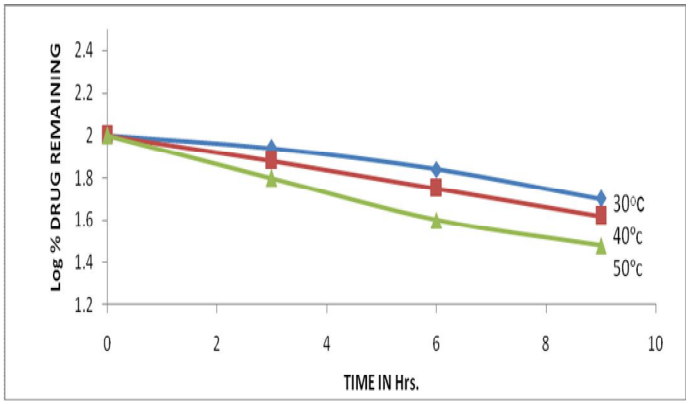


**Fig. 3: First order plot of the oxidation of Phenobarbitone with 6% hydrogen peroxide at different temperature**

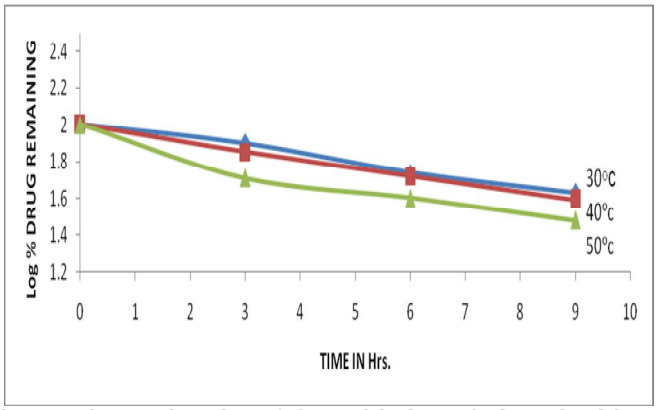


**Fig. 4 : First order plot of the oxidation of Phenobarbitone with 7% hydrogen peroxide at different temperature**

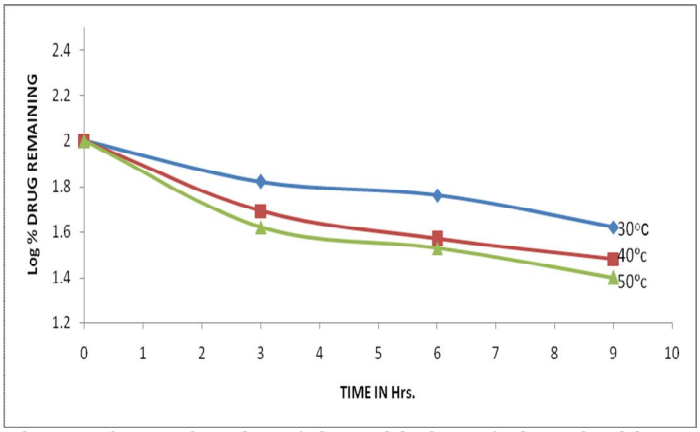


**Fig. 5 : First order plot of the oxidation of Phenobarbitone with 8% hydrogen peroxide at different temperature**

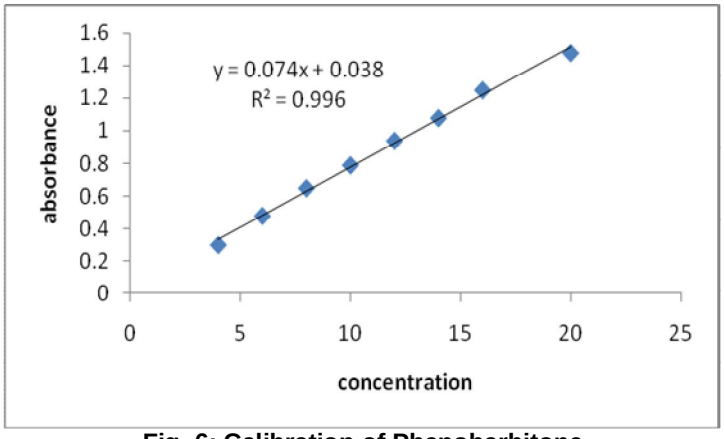


**Fig. 6: Calibration of Phenobarbitone**

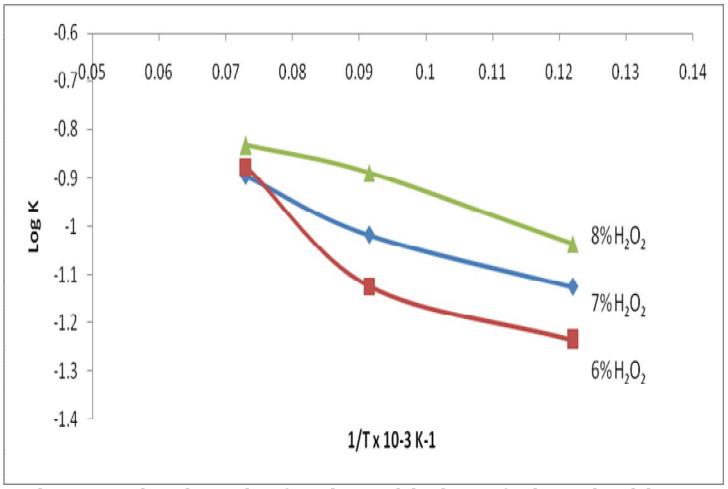


**Fig. 7: Arrhenius plot for the oxidation of Phenobarbitone with 6%,7%,8% hydrogen peroxide**

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