Forced degradation study for simultaneous quantification of aspirin and omeprazole in pharmaceutical dosage form by RP-HPLC

Abstract

Aims: To study force degradation of aspirin and omeprazole simultaneously by RP-HPLC method

Study design: RP-HPLC method was used to measure % degradation.

Place and duration of study: Study was carried out at center of excellence, G.I.D.C., vapi-396195, Gujarat, India between June 2019 to march 2020.

Methodology: A force degradation study of aspirin and omeprazole was carried out simultaneously. The drugs were subjected to various degradation conditions like hydrolysis by acid and base, Oxidative degradation, and thermal degradation study.

Results: For acidic condition, the degradation was found to be 32.63 % for aspirin and 61.64 % for omeprazole. For basic condition, the degradation was found to be 10.17 % for aspirin and 4.29 % for omeprazole. By oxidative hydrolysis, the aspirin was degraded by 15.48 % and omeprazole was degraded by 26.38 %. By thermal degradation, 0.37 % degradation was observed for aspirin and 4.32 % degradation for omeprazole.

Conclusion: In this proposed method the retention time for drug is less than 8 min, which is less then available method. For omeprazole, strong degradation was observed in acidic conditions and mild in basic hydrolysis conditions. For aspirin, more degradation was observed in basic conditions than acidic hydrolysis. Both drugs were degraded in oxidative conditions using 3% H2O2. Omeprazole degraded more than aspirin by dry heat degradation. The method was successfully applied for the quantitative determination of both Active Pharmaceutical Ingredients.

Keywords: Aspirin, Omeprazole, Acid hydrolysis, Base hydrolysis, Oxidative hydrolysis, Thermal degradation

1. INTRODUCTION

Myocardial Infarction occurs when blood flow stops to a part of the heart causing damage to the heart muscle. [1] Yosprala, a fixed-dose combination is available containing the antiplatelet agent aspirin and the proton pump inhibitor omeprazole. [2-5] Chemical structures of aspirin and omeprazole are given in figure 1. [6-8]

As per the literature survey, no reported method was found for the force degradation study of aspirin and omeprazole simultaneously. There was a method related to esomeprazole and aspirin but there is no actual degradation found simultaneously with aspirin. As per the reported article, there is a difference in the degradation of omeprazole and esomeprazole. The other methods were found for simultaneous estimations are UV [9], and HPLC [10], and some methods for omeprazole and aspirin separately.

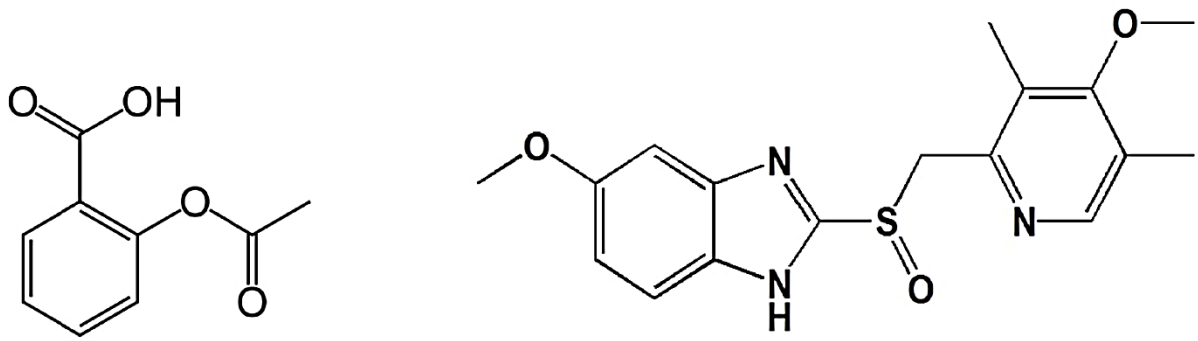


Figure 1: Chemical structure of aspirin and omeprazole

The proposed method is applied for API only, here degradation was done for Hydrolysis by acid and base, Dry heat degradation, and oxidation under given ICH guidelines as formulation can be only applied for the photostability study. [11] Hydrolysis is the most common degradation chemical reaction over a wide range of pH. Hydrolysis is a chemical process that includes the decomposition of chemical compounds via reaction with water. The hydrolytic study involves the catalysis of ionizable functional groups present in a molecule of the drug. Acidic or basic stress testing involves forced degradation of drug substances via exposure to acidic and basic conditions which generates primary degradants in the

desirable range. Hydrogen peroxide is widely used for the oxidation of drug substances in forced degradation studies of other oxidizing agents. The photostability testing of drug substances must be evaluated to demonstrate that light exposure does not result in unacceptable change. Photostability studies were performed for the generation of primary degradants of a drug substance by exposure to UV or fluorescent conditions. Thermal degradation (e.g., dry heat & wet heat) should be carried out at more stress conditions than recommended ICH Q1A accelerated testing conditions. To calculate % degradation the following equation was used.

Area of the standard peak – Area of degradant peak / Area of standard peak * 100

2. MATERIALS AND METHODS

A standard sample of Aspirin was given as a gift sample from Sidmak, Valsad, India, and Omeprazole was given as a gift sample from Mangalam drugs, Vapi. Methanol HPLC-grade, water HPLC-grade, and Disodium hydrogen phosphate, Hydrochloric acid, Sodium hydroxide, Hydrogen Peroxide has been purchased from Rankem, RFCL Limited, New Delhi, India

2.1 Instruments

HPLC: A LC-2010 AHT HPLC of Shimadzu corporation equipped with LC P-100 pump, a PDA detector, a high-pressure gradient mixer of 1500 µL, a loop injector of 20 µL capacity, and Class-VP software was used for the analysis.

Sonicator: A digital ultrasonic cleaner (Equitron) was used for mixing

Heating water bath: A digital water bath (Equitron) was used for heating solutions for degradation study.

2.2 Chromatographic conditions:

The chromatographic column used was a C18 column (250mm x 4.6mm, 5 µm) all obtained from Waters Corp. (Milford, MA, USA). The LC method consists of 0.05 M Disodium hydrogen phosphate buffer as mobile phase A, and Methanol as a mobile phase B in a ratio of 68: 32 v/v, pH was adjusted to 4.5 by using phosphoric acid. The column temperature was maintained at 25°C and the detection was monitored at a wavelength of 280 nm. The injection volume was 20µL. The chromatogram of standard aspirin and omeprazole is given in figure 2.

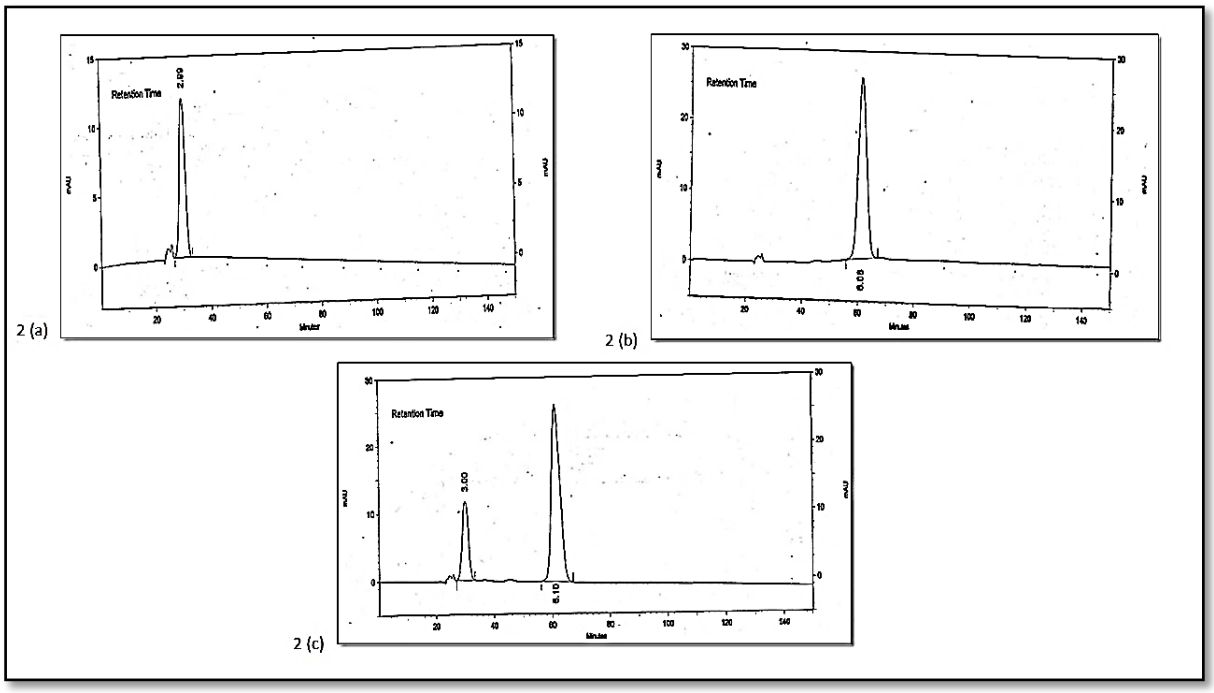


Figure 2: chromatogram of standard aspirin (2a), chromatogram of standard omeprazole (2b), chromatogram of standard aspirin and omeprazole (2c).

2.3 Preparation of solutions:

A degradation study was performed by preparing 4 µg/mL solution for aspirin and 2 µg/mL of omeprazole. For acid hydrolysis, 0.1 N HCl is used, and the solution was heated on a water bath at 60 ◦c for an hour. For base hydrolysis, 0.1 N NaOH was used by heating a solution on a water bath at 60 ◦c for an hour, after that by using 0.5 N NaOH heating a solution on water bath at 60 ◦c for different duration of time. For oxidative degradation 3% H2O2 was used and solutions with Hydrogen peroxide were kept at room temperature for 3 hours. Thermal degradation was performed by solid drug sample using the dry heating method for 5 hours under sunlight.

3. RESULT AND DISCUSSION

3.1 Acid hydrolysis

Aspirin and omeprazole were heated on a heating water bath for an hour with 0.1 N HCl at 60˚c temperature, 61.64 % omeprazole was degraded (Fig. 3b) and in aspirin, 4.24 % degradation has been

found. (Fig. 3a). No new peaks of degradation products were obtained, so this study was stopped after 1 hour of heating because of the strong degradation of omeprazole.

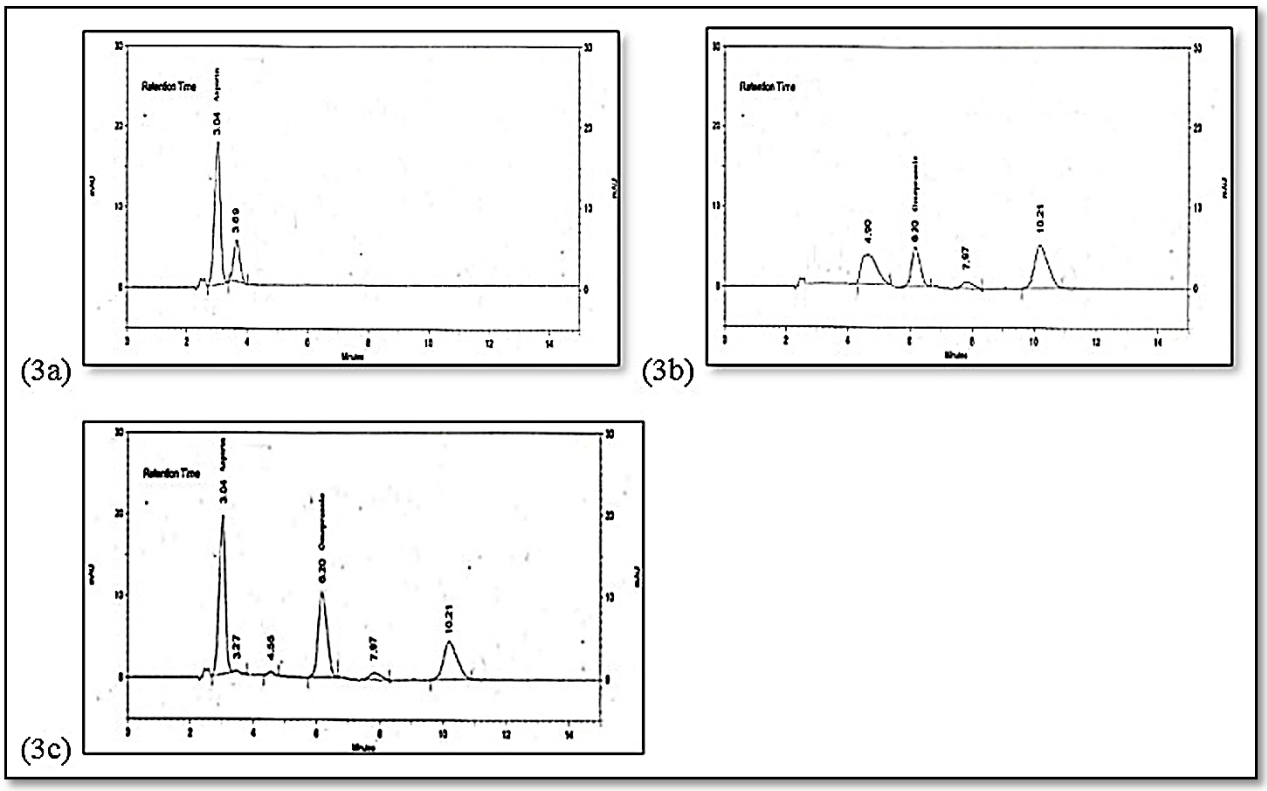
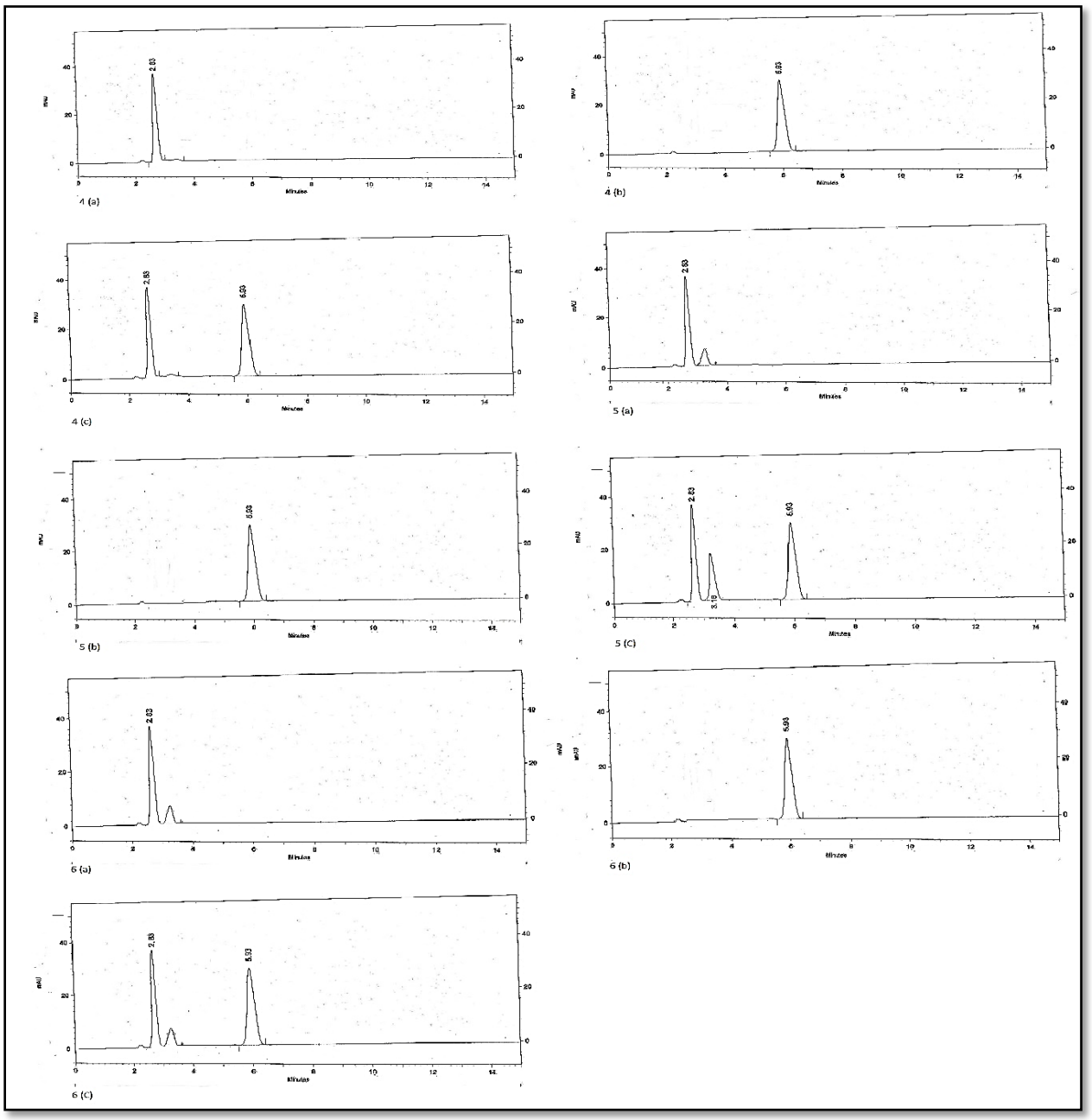


Figure 3: chromatogram of aspirin with 0.1 N HCl (3a), chromatogram of omeprazole with 0.1 N HCl (3b), chromatogram of aspirin and omeprazole with 0.1 N HCl (3c).

3.2 Base hydrolysis

On heating with 0.1 N NaOH for 1 hour 4.37 % degradation of aspirin was observed (Fig. 4a) and a little amount of (1.91%) omeprazole was degraded (Fig. 4b). After 3 hours 7.01 % and 2.45 % degradation



were observed for aspirin (Fig. 5a) and omeprazole (Fig. 5b) respectively. Then the concentration of NaOH was increased and after 1 hour of heating with 0.5 N NaOH at 60˚c temperature, around 10.17 % degradation was observed in aspirin (Fig. 6a) and 4.29 % degradation was observed in omeprazole. (Fig. 6b) So, in alkaline conditions, more degradation of aspirin was observed than omeprazole.

Figure 4 to 6: chromatogram of aspirin an hour after heating with 0.1 N NaOH (4a), chromatogram of omeprazole an hour after heating with 0.1 N NaOH (4b), chromatogram of aspirin and omeprazole an hour after heating with 0.1 N NaOH (4c), chromatogram of aspirin 3 hours after heating with 0.1 N NaOH (5a), chromatogram of omeprazole 3 hours after heating with 0.1 N NaOH (5b), chromatogram of aspirin and omeprazole 3 hours after heating with 0.1 N NaOH (5c), chromatogram of aspirin an hour after heating with 0.5 N NaOH (6a), chromatogram of omeprazole an hour after heating with 0.5 N NaOH (6b), chromatogram of aspirin and omeprazole an hour after heating with 0.5 N NaOH (6c)

3.3 Oxidative hydrolysis

Here almost 8% degradation was obtained for aspirin (Fig. 7a) and 8 % for omeprazole (Fig. 7b) with 3% H2O2 at room temperature after 1 hour and 15% and 27% degradation were obtained for aspirin (Fig. 8a) and omeprazole (Fig. 8b) respectively after 3 hours at room temperature.

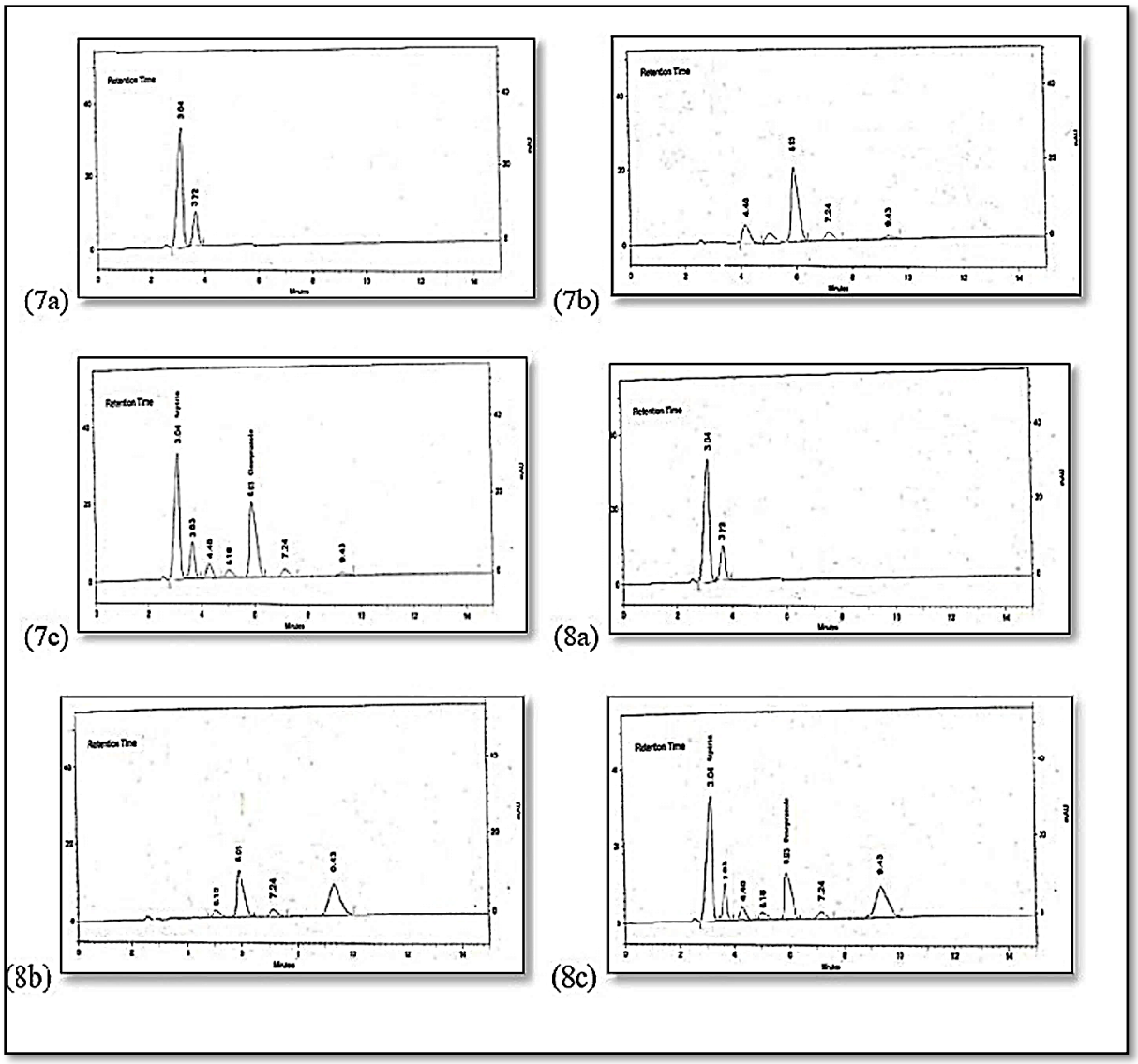


Figure 7-8: chromatogram of aspirin an hour after 1 hour with 3% H_2O_2 (7a), chromatogram of omeprazole an hour after 1 hour with 3% H_2O_2 (7b), chromatogram of aspirin and omeprazole after an hour with 3% H_2O_2 (7c), chromatogram of aspirin after 3 hours with 3% H_2O_2 (8a), chromatogram of omeprazole after 3 hours with 3% H_2O_2 (8b), chromatogram of aspirin and omeprazole after 3 hours with 3% H_2O_2 (8c).

3.4 Thermal degradation (Dry heat degradation)

Dry heat degradation was carried out by heating both the drugs in oven at 110℃ for 5 hours. Around 5% degradation was obtained for omeprazole (Fig. 9b). Negligible degradation of aspirin was seen (Fig. 9a). All results are described in Table 1.

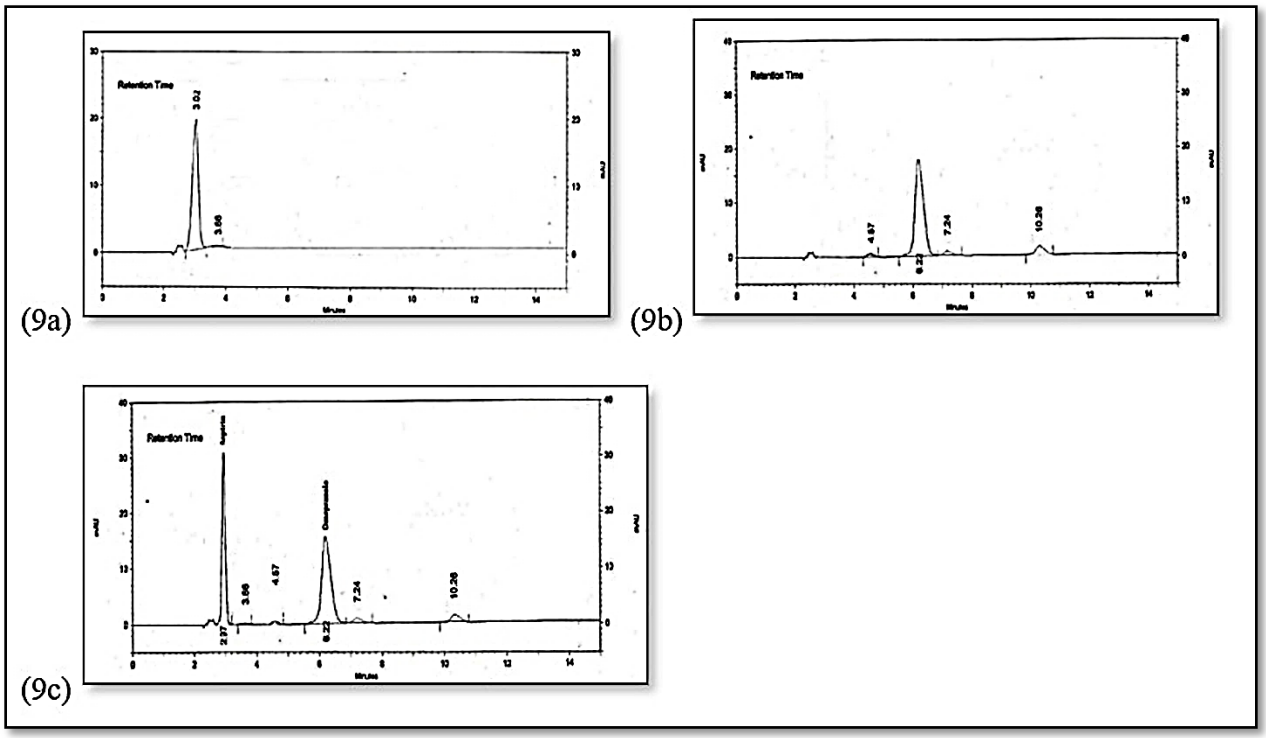


Figure 9: chromatogram of aspirin an after 5 hours of heating at 110 ̊c (9a), chromatogram of omeprazole after 5 hours heating at 110 c (9b), chromatogram of aspirin and omeprazole after 5 hours heating at 110 c (9c)

Table 1: Result of degradation study

Stress Condition	Parameter of stress condition	% degradation of API	
		Aspirin	Omeprazole
Acid hydrolysis	Without heating with 0.1 N HCl	0.97	32.63
	1 hour heating with 0.1 N HCl	4.21	61.64

Base hydrolysis	1 hour heating with 0.1 N NaOH	4.37	1.91
	3 hour heating with 0.1 N NaOH	7.01	2.45
	1 hour heating with 0.5 N NaOH	10.17	4.29
Oxidative hydrolysis	1 hour heating with 3% H_2O_2	8.17	15.44
	3 hour heating with 3 % H_2O_2	15.48	26.38
Thermal degradation	Sunlight for 5 hours at 110℃	0.37	4.32

3.5 Linearity study:

The linearity of the proposed method was evaluated by analyzing six different concentrations of the standard solutions of aspirin and omeprazole in the range of 10 - 60 µg/mL and 5-30 µg/mL respectively. For linearity studies, six solutions with different concentrations for aspirin (10 µg/mL, 20 µg/mL, 30 µg/mL, 40 µg/mL, 50 µg/mL and 60 µg/mL) and omeprazole (5 µg/mL, 10 µg/mL, 15 µg/mL, 20 µg/mL, 25 µg/mL and 30 µg/mL) were prepared by diluting 1,2,3,4,5 and 6 ml and 0.5, 1.0, 1.5, 2.0 and 3.0 ml of working standard solutions (100 µg/mL) of aspirin and omeprazole respectively up to 10 ml with mobile phase. The resulting solutions were injected in triplicate and the area was measured. A plot of average area vs concentration was plotted and regression coefficient (R2) was calculated. (Fig. 10) The linearity equation for aspirin and omeprazole was obtained by linear regression analysis.

4. CONCLUSION

In this proposed method Aspirin and omeprazole were stressed under ICH guidelines. Method was applied for active pharmaceutical ingredients. Here for aspirin more degradation was seen in base than acid hydrolysis, it was degraded in oxidative conditions but approximately stable in thermal degradation. Omeprazole was strongly degraded in acidic condition and oxidative hydrolysis. Mild degradation was observed in alkaline hydrolysis and it was degraded by dry heat degradation.

5. COMPETING INTERESTS:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use

these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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