STUDIES OF STRESS DEGRADATION AND IMPURITY PROFILES OF SOME 5-HT_3 ANTAGONISTS

A

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CERTIFICATE OF DECLARATION

This is to certify that the thesis entitled "Studies of Stress Degradation and Impurity Profiles of some 5-HT_3 Antagonists" submitted by Md. Mokaram Hossain, Reg. no.101, session 2015-2016, Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Dhaka, for the complete fulfillment of Degree of Philosophy, is a record of bona fide work carried out by him under my direct supervision, as per the code of academic and research ethics of University of Dhaka.

The contents of this report have not been submitted and will not be submitted either in part or in full, for the award of any other degree of diploma. The thesis fulfills the requirements and regulations of the University of Dhaka and in my opinion meets the necessary standards for submission.

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DECLARATION

I do hereby declare that the materials embodied in this thesis entitled "Studies of Stress Degradation and Impurity Profiles of some 5-HT_3 Antagonists" prepared for submission to the University of Dhaka, Dhaka-1000, Bangladesh for the Degree of Doctor of Philosophy in Pharmaceutical Chemistry are the original research work of mine and have not been previously submitted for the award of any degree of diploma.

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CONTENTS

List of Figures

List of Tables

ABSTRCT

Three issues of fundamental importance in drug therapy are safety, efficacy and stability. This thesis paper has reports the scientific data of five 5-HT_3 antagonists in the field of stress degradation, degradation kinetics and impurity profiles. The stress conditions are aqueous, acid, base, oxidation and photodegradation. Ramosetron HCl, ondansetron HCl, granisetron HCl, tropisetron HCl and palonosetron HCl are the five 5-HT_3 antagonists that were selected for the studies. Extensive information derived from studies of stress degradation, degradation kinetics and impurity profiling expanded the scientific thought further to ensure the achievement of intended quality of drug substances and drug products available in the market. Stress degradation screening of 5-HT_3 antagonists in aqueous conditions, different strengths of acid-base conditions, oxidative condition and photo degradation at different time point and temperature revealed the pH dependent stability, oxidative and photo sensitivity.

From stressed degradation conditions, it was evident that ramosetron HCl and palonosetron HCl are more stable in aqueous conditions. However, ondansetron HCl, granisetron HCl and tropisetron HCl produced degradants in aqueous conditions. On the other hand, four antagonists except ondansetron HCl produced degradants and growing impurities in acid stressed conditions. Each 5-HT_3 antagonist out of five was evident to produce degradants and growing impurities.

Degradation kinetic studies were conducted for four 5-HT_3 antagonists, ramosetron HCl, ondansetron HCl, granisetron HCl, and tropisetron HCl, to observe the effect of temperature and also to calculate activation energy (E_a). Degradation kinetics of ramosetron HCl was studied in 0.1N NaOH and ondansetron HCl, granisetron HCl, and tropisetron HCl in 2.0N NaOH at 60°C, 70°C and 80°C at different time interval. The calculated activation energy (E_a) was found as 10.05 $kcalmol^{-1}$, 7.57 $kcalmol^{-1}$, 16.98 $kcalmol^{-1}$, and 16.86 $kcalmol^{-1}$ for ramosetron HCl, ondansetron HCl, granisetron HCl and tropisetron HCl, respectively.

Degradants and growing impurities were reported after evaluation of HPLC and MS data taking consideration of the relative retention time (RRT), mass spectroscopy (MS) intensity and molecular weights of each 5-HT_3 antagonist generated.

In acidic conditions, ramosetron HCl produced some potent degradants with relative retention time (RRT) of 0.79, 0.89, 2.18, 2.33 and 3.50. In basic conditions, ramosetron hydrochloride also produceed potent degradants with relative retention time (RRT) of 0.38, 0.51, 0.56, 0.64, 0.67 and 0.70. Potent degradants of ramosetron HCl with relative retention time (RRT) of 0.65, 0.76, 0.94 and molecular weight of 113.9, 141.1 and 361.2 Da were evident in oxidative conditions.

Ondansetron HCl increased the content of two growing impurities with relative retention time (RRT) of 0.51, 0.65 and molecular weight of 82.1 and 211.26 Da. Growing impurities of ondansetron HCl with relative retention time (RRT) of 0.44, 0.47, 0.49 and molecular weight of 604.77, 211.26 and 279.34 Da were evident in oxidative conditions. On the other hand, potent degradants of ondansetron HCl with relative retention time (RRT) of 0.51, 0.56, 0.65, and 1.39 was also observed in oxidative conditions. Ondansetron hydrochloride is light sensitive and increased the concentration of a growing impurity with relative retention time (RRT) of 0.25 and molecular weight of 256.2 Da when exposed to 3.6 million lux fluorescence light and 600 watts hour/m^2 UV light.

Granisetron hydrochloride was observed to produce one degradant with relative retention time (RRT) of 0.85 in aqueous condition. In acidic conditions, granisetron HCl produced some potent degradants with relative retention time (RRT) of 0.24, 0.30, 0.58, and 1.22. In basic conditions, granisetron hydrochloride was seen to increase the concentration of a growing impurity with molecular weight of 335.1 Da. It was found to be stable in 10.0% hydrogen peroxide. It showed no photosensitivity.

Tropisetron hydrochloride produced two degradants with relative retention time (RRT) of 0.35 and 0.40 in aqueous condition. In acidic conditions, tropisetron HCl produced some potent degradants with relative retention time (RRT) of 0.17, 0.21, 0.24, 0.40 and 0.59. In basic conditions, two growing impurities with relative retention time (RRT) of 0.51, 0.65 and molecular weight of 82.1 and 211.26 Da were evident for tropisetron HCl. Potent degradants of tropisetron HCl with relative retention time (RRT) of 0.23, 0.24, 0.38 and 0.43 were produced in oxidative conditions.

In acidic conditions, palonosetron HCl produced a potent degradant with relative retention time (RRT) of 0.13. In basic conditions, palonosetron HCl also produced a potent degradant with

relative retention time (RRT) of 0.13. One known growing impurity with molecular weight of 310.4 Da and three unknown potent degradats with molecular weight of 314.4, 328.4 and 344.4 Da were evident for palonosetron HCl in oxidative conditions.

Therefore it is clearly evident that more attention should be given during formulation development, process validation and stability testing to minimize or control these growing impurities and potential degradants.