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Photodegradation kinetics of Bilastine in tablets

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Photodegradation is the process by which a chemical substance is broken down through exposure to light, typically ultraviolet (UV) radiation. This process is significant in environmental chemistry, where sunlight can degrade pollutants and materials, influencing their persistence and toxicity. Bilastine is a novel non-sedating histamine H1-receptor antagonist developed for the treatment of allergic rhino conjunctivitis and urticaria. The literature presents some studies on the quantitative determination of bilastine by high-performance liquid chromatography in pharmaceutical forms and biological fluids, however, no data on the photodegradation kinetics of this drug are described. Therefore, the objective of the study was to determine the photodegradation kinetics of the drug bilastine in coated tablets using a liquid chromatography method previously developed and validated by the same research group (unplished data). The study was carried out with methanolic solution containing 100.0 µg mL^{-1} of bilastine drug product exposed to UV-C radiation (254 nm). The irradiation of the samples was done at pre-established times: 0, 15, 30, 60, 120 and 180 minutes. The chromatographic separation was performed in a Shim-pack® RP-18 column; the mobile phase comprising a mixture of 0.3% triethylamine (pH adjusted to 6.0 with 20% formic acid) and acetonitrile (66:34, *v/v*) at a flow-rate of 1.0 mL min^{-1} with isocratic elution. The temperature was set at 25 °C in the column oven. Bilastine was determined by UV detection at 207 nm using photodiode-array. The results demonstrated that the photodegradation kinetics of bilastine in methanolic solution follows the first order of reaction, with a $t_{90\%}$ of 27.11 minutes and degradation rate constant (*k*) of 0.0007 min^{-1}.

Keywords: bilastine, photodegradation, kinetics studies.

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Introduction

Bilastine, chemically 2(2-[4-(2-(4-(1-(2-ethoxyethyl) bencimidazole-2-yl) piperidine-1-yl) ethyl) phenyl]- 2-methylpropanoic acid; Figure 1, is a novel non-sedating histamine H1-receptor antagonist developed for the treatment of allergic rhinoconjunctivitis and urticaria without showing sedative or cardiotoxic effects in clinical trials and in post-marketing experience so far (1). The high selectivity to the receptors was shown to be greater than cetirizine and fexofenadine (2). Bilastine is characterized by rapid onset of action, 30 to 60 min, and a long duration of action, nearly 24 h. Furthermore, the novel drug is not metabolized by liver enzymes and is excreted mostly unchanged in feces (3). The safety and efficacy of bilastine for the relief of allergic rhinitis symptoms have been confirmed in two pivotal clinical trials evaluating over 1,400 patients (4). It has recently been granted marketing authorization for these therapeutic indications in adults and adolescents at a once-daily oral dose of 20 mg in several European countries (1). Although there are some papers describing the determination of bilastine in pharmacokinetic studies by liquid chromatography (LC)-fluorescence detection and liquid chromatography-mass spectrometry- mass spectrometry (LC-MS-MS) (5-7), there are few papers describing quantification methods to bilastine in tablets (8,9). Our research group published methods by ultraviolet (UV) spectrometry (10) and LC using detection by fluorescence (11) and a green-method by LC (12).

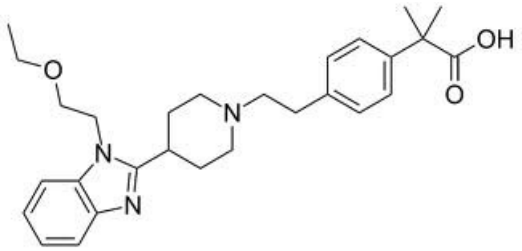

Figure 1. Bilastine chemical structure

Ouarezki and collaborators (2020) developed a stability-indicating method and determined the degradation kinetics under oxidative conditions (13). However, the drug is also very susceptible to photolytic degradation, but these studies are not described in the literature. Therefore, the objective of these studies was to determine the degradation kinetics under photolytic conditions of tablets of bilastine in methanolic solution using a liquid chromatography

method previously developed and validated by the same research group (unplished data).

Experimental section

Chemicals

Bilastine chemical reference standard (CRS) (99.0%) was acquired by Toronto Research Chemicals (Toronto, Ontario, Canada). Alektos® (Takeda, Brazil), composed of a 20-mg bilastine, was purchased in the market. The excipients contained in the dosage form (microcrystalline cellulose, sodium starch glycolate, magnesium stearate and colloidal silicon dioxide) were all pharmaceutical grades and were acquired from different suppliers. All chemicals used were of analytical grade and all solvents were of LC grade. Methanol and acetonitrile were purchased from Tedia (Fairfield, USA). Phosphoric acid and triethylamine were purchased from Merck (Darmstadt, Germany). Purified water was prepared using Milli-Q Plus® (Millipore, Bedford, USA).

Apparatus

A Shimadzu Prominence® liquid chromatograph (Kyoto, Japan) equipped with a model LC-20AD quaternary pump, SIL-20AC HT auto sampler, CTO-20AC column oven, SPD-M20A photodiode-array detector and LC Solution V. 1.24 SP1 manager system software was used. Photodegradation studies were carried out in a photostability UV chamber (1.0 x 0.17 x 0.17 m) with mirrors and equipped with UV-C lamp (Ecolume ZW®, 254 nm, 30W).

Chromatographic conditions of the LC determination

The chromatographic separation was performed in a Shim-pack® RP-18 column (150 x 4.6 mm I.D., 5 µm, Shimadzu, Kyoto, Japan). The mobile phase comprising a mixture of 0.3% triethylamine (pH adjusted to 6.0 with 20% formic acid) and acetonitrile (66:34, *v/v*) at a flow-rate of 1.0 mL min^{-1} with isocratic elution. The injection volume was 20.0 µL for both reference substance and drug product solutions and the run time was 5.0 min. The temperature was set at 25 °C in the column oven. Bilastine was determined by UV detection at 207 nm using photodiode-array. The quantitation was performed using the absolute area of the peak.

Preparation of bilastine Chemical Reference Substance (CRS) and sample stock solutions

Bilastine CRS was accurately weighed and dissolved in a volumetric flask with methanol to obtain a concentration of 200.0 $\mu g.mL^{-1}$. This solution was diluted appropriately in the same diluents to yield a final concentration of 20.0 $\mu g.mL^{-1}$. To prepare a sample stock solution, twenty tablets of Alektos® were weighed and finely powdered. A quantity equivalent to 5.0 mg of the bilastine was transferred into a 25 mL volumetric flask with 15 mL of methanol and kept in an ultrasonic bath for 30 min. The volume was completed with the same diluent to yield a final concentration of 200.0 $\mu g.mL^{-1}$.

Bilastine photodegradation kinetics

The effect of light in the photodegradation of the bilastine product drug was studied exposing the pharmaceutical formulation (tablets) solutions to the UVC irradiation (254 nm). Samples of the stock solution of bilastine tablets in methanol at 200 $\mu g.mL^{-1}$ were inserted into quartz cuvettes (1 cm) and positioned horizontally at 10 cm from the lamp to provide maximum area of radiation exposure. The sample solutions were exposed to the UVC irradiation in the following time intervals: 0, 15, 30, 60, 120 and 180 minutes and three samples (n=3) were analyzed for each time interval. The influence of the temperature also was evaluated through analyzed protected samples, wrapped in aluminum foil (dark controls). After the required time, aliquots of 1.0 mL were transferred to a 10 mL volumetric flask and diluted with methanol to 20 $\mu g.mL^{-1}$ for the LC determination, employing the developed and validated stability-indicating method. The standard solution was prepared in methanol at the concentration of 20 $\mu g.mL^{-1}$ for the quantitation of the bilastine. The solutions were filtered through a 0.45 µm membrane filter before injection and peak purity tests were performed by the photodiode array. The influence of the formulation excipients was evaluated by irradiating a placebo solution in methanol under the same experimental conditions.

Chemical Reaction Kinetics

Chemical reaction kinetics is the study of the rates at which chemical reactions occur and how they are affected by various factors such as concentration, temperature, and pressure. The reaction order is a key concept in kinetics, reflecting how the rate of reaction depends on the concentration of reactants. The reaction kinetics can be:

Zero-Order Reactions: in zero-order reactions, the rate of reaction is constant and independent of the concentration of the reactants.

Rate Law: Rate=k, here, k is the zero-order rate constant.

Integrated Rate Law: the concentration of the reactant decreases linearly over time: $[A]_t=[A]_0-kt$, where $[A]_t$ is the concentration at time t, $[A]_0$ is the initial concentration, and k is the zero-order rate constant.

First-Order Reactions: in first-order reactions, the rate of reaction is directly proportional to the concentration of one reactant. Doubling the concentration of the reactant results in a doubling of the reaction rate.

Rate Law: Rate=k[A], where k is the first-order rate constant and [A] is the concentration of the reactant.

Integrated Rate Law: the concentration of the reactant follows a logarithmic decay: $\ln[A]_t=\ln[A]_0-kt$, where $[A]_t$ is the concentration at time t, [A] is the initial concentration, and k is the first-order rate constant.

Second-Order Reactions: for second-order reactions, the rate depends on the square of the concentration of one reactant or the product of the concentrations of two reactants.

Rate Law: for a reaction involving one reactant: $Rate=k[A]^2$ and for reactions involving two reactants: Rate=k[A][B], where k is the second-order rate constant.

Integrated Rate Law: for a reaction with one reactant: $1/[A]_t=1/[A]_0+kt$, where $[A]_t$ is the concentration at time t,

$[A]_0$ is the initial concentration, and k is the second-order rate constant.
Understanding the reaction order and the corresponding equations allows chemists to predict how changes in concentration affect the rate of a reaction and to design experiments or processes accordingly. Each reaction order provides insight into the mechanistic details of the reaction and is crucial for both academic research and industrial applications (14-16).

Results and Discussion

Photolytic degradation kinetics in solution involves the study of how compounds degrade when exposed to light, typically in aqueous environments. This area of study is critical in various fields such as environmental chemistry, pharmacology, and materials science, due to its implications for the stability, safety, and environmental impact of chemical substances. The main fundamentals of photolytic degradation are photolysis and the formation of free radicals. Photolysis refers to the chemical breakdown of compounds induced by the absorption of light. When a molecule absorbs photons, it can reach an excited state, leading to the formation of reactive free radicals or other reactive species. These radicals can further interact with the surrounding molecules, resulting in the degradation of the original compound (17-18). The objective of photostability testing is to provide evidence on how the quality of a drug varies with the time under the influence of the light. Furthermore, in forced degradation studies, photodegradation is a tool used to evaluate the stability of a substance under stress conditions and the selectivity of the analytical method (19, 20). In our study, the photodecomposition of bilastine was carried out through the employment of a stress condition during different periods of time using UV-C radiation. Many factors can affect the stability of a pharmaceutical product, such as the stability of the active ingredient, the manufacturing process, the environmental conditions (heat, light, and moisture during storage), as well as some chemical reactions like oxidation, reduction, hydrolysis, and racemization that might occur. Besides that, the photodecomposition is an important factor of the pharmaceutical's stability. Since UV radiation has high energy, it can be the cause of many degradation reactions (21-23). Due to the instability of the drug in photolytic conditions during the stability-indicating studies, previously carried out, this condition was used to determine the drug photodegradation kinetics. Bilastine methanolic solutions were exposed to UV-C radiation (254 nm) at different time intervals (15, 30, 60, 120, 180 and 240 min). The chromatograms obtained revealed that the bilastine peak area was reduced over time and it was also possible to verify that degradation products were not formed from the excipients in the formulation (Figure 2). The results of photodegradation kinetics of bilastine pharmaceutical formulation solutions exposed to UVC radiation are showed in the Table 1 and the plots of concentration, log concentration and reciprocal of concentration of the remaining drug versus time are shown in Figure 3. Through the evaluation of the correlation coefficient, it can be demonstrated that the bilastine photodegradation process in methanolic solution can be described by first-order kinetics, under the experimental conditions applied in this study and the values of degradation rate constant (*k)* and t_{90} were 0.0007 min^{-1} and 27.11 min, respectively. The temperature in the chamber was always below 32 °C and the concentration of the thermal control solution remained stable during the studies. Therefore, it was found that the only factor that influenced the degradation of the drug bilastine was UV-C radiation. The photodegradation of a chemical substance can be caused by oxidation or by the breakdown of certain weak chemical bond. Both phenomena are energy related, and consequently, they are be preceded by photons with specified wavelengths. On the other hand, the rate of photodegradation reaction is dependent on light intensity, so higher light intensity provides faster reaction rates. In the chemical structure of bilastine is possible verify the presence of chromophores groups, such as conjugated double bonds, nitro and amide, which can be susceptible to photodegradation by UV radiation (254 nm). The International Conference on Harmonization (ICH) guideline requires that stress testing should be carried out to elucidate the inherent stability characteristics of the active substance in a pharmaceutical preparation. The light testing should be an integral part of stress testing (24-25).

Table 1. Photodegradation kinetics of bilastine pharmaceutical formulation solutions exposed to UVC radiation

Time (minutes)	Concentration (µg.mL^{-1}) *	Log of concentration	1/ concentration
0	20.01	1.30103	0.04999999
15	18.90	1.276527	0.05290211
30	17.52	1.243497	0.05708244
60	15.78	1.198197	0.06335826
120	10.67	1.028172	0.09371905
180	8.31	0.919735	0.1202997

* Data expressed as the mean of three determinations.

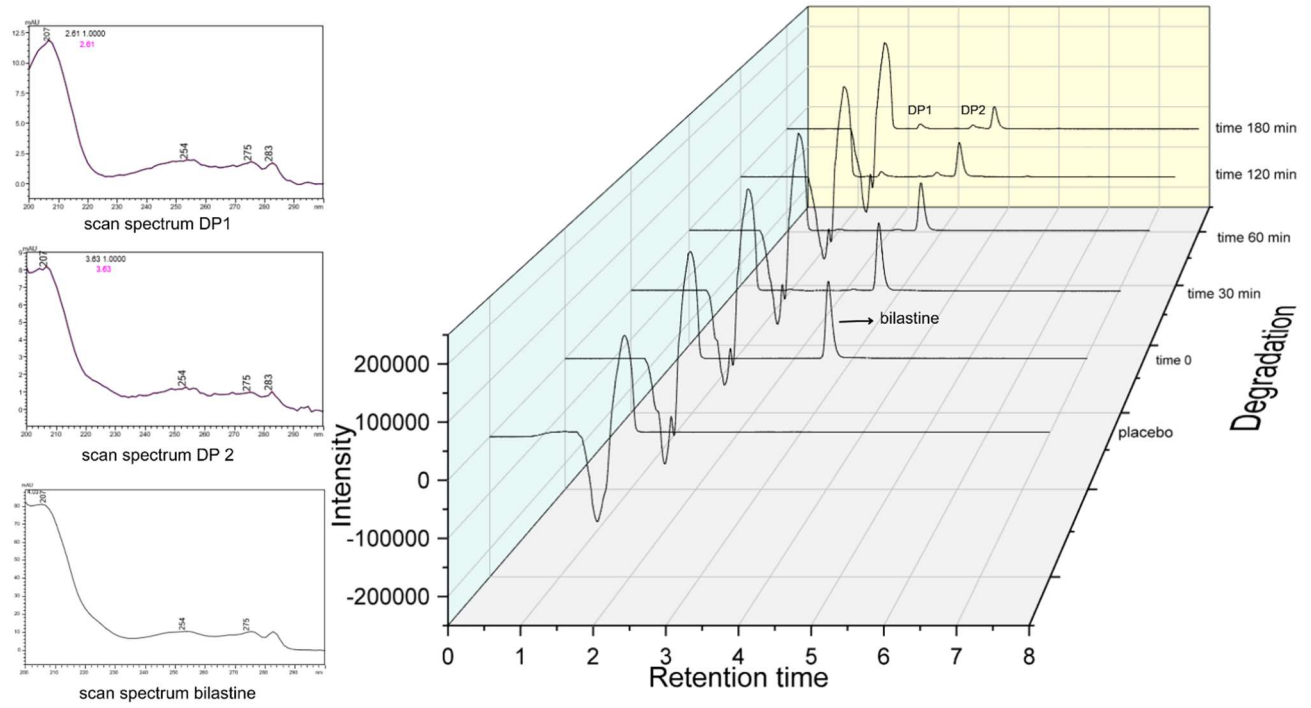

Figure 2. Chromatograms obtained for bilastine (20.0 µg.mL^{-1}) after photodegradation process under UVC radiation for different analysis times: A) zero; B) 30 min; C) 60 min; D)120 min; E) 180 min; Chromatographic conditions: Shim-pack® RP-18 column (150 x 4.6 mm I.D., 5 µm); mobile phase: mixture of 0.3% triethylamine (pH adjusted to 6.0 with 20% formic acid) and acetonitrile (66:34, *v/v*) at a flow-rate of 1.0 mL min^{-1} with isocratic elution; UV detection at 207 nm.

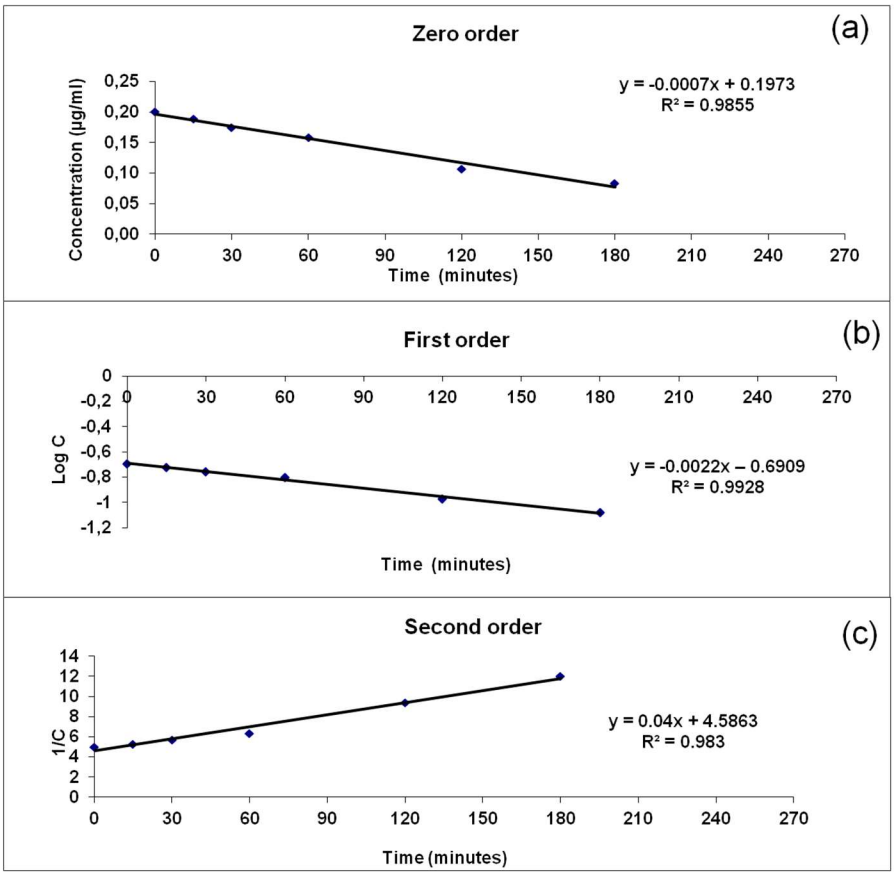

Figure 3. Plots of concentration, log concentration and reciprocal of concentration of the remaining bilastine versus time. Graphic (a): zero order; (b) first order, (c) second order.

Conclusions

The bilastine photodegradation kinetic studies in methanol solutions were determined in the tablets. The degradation of this drug in tablets during photodegradation in UV-C radiation follows first-order reaction kinetics, that is, the rate of degradation of the drug is directly proportional to its concentration at the beginning of the reaction. The values of degradation rate constant (*k)* and t_{90} were 0.0007 min^{-1} and 27.11 min, respectively.

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Conflict of interest

The authors declare no conflicts of interest.

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