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Short communication

Forced degradation study of a finished drug product containing an active component from the cephalosporin group

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Introduction

Forced degradation is a degradation of drug substance and drug product at conditions more severe than accelerated conditions. It is required to demonstrate specificity of stability indicating methods and also to provide an insight into degradation pathways and degradation products of the drug substance. Forced degradation studies show the chemical behavior of the molecule which in turn helps in the development of formulation and package selection.

The ICH guideline entitled "Stability Testing of New Drug Substances and Products" requires that stress testing of the drug substance can help identify the likely degradation products, which can in turn help establish the degradation pathways and the intrinsic stability of the molecule and validate the stability indicating power of the analytical procedures used. (ICH Q1A (R2), 1993). The results obtained from this study should demonstrate the specificity and the stability indicating power of the analytical procedures for determination of related and degradation products and assay.

Materials and Methods

Materials

A certified secondary reference standard of the active component was used. Ammonium acetate ($NH_4CH_3CO_2$ p.a.), acetonitrile (ACN), methanol (MeOH) and ethanol (EtOH) (Merck, HPLC grade) are used as reagents and were obtained from Sigma-Aldrich. The diluent was prepared by mixing acetonitrile and water (1:2, % *v/v*).

Methods

Chromatographic analysis was carried out using HPLC under two methods to assess both the assay and related and degradation products.

The assay method used a mobile phase of 96% ethanol and water (44:56, *v/v*). Conditions: Zorbax Eclipse Plus Phenyl Hexyl RR column (150×4.6 mm, 3.5 µm), column temperature : 40 °C, flow: 1.0 mL/min, detection at 235 nm, and 15 min run time.

The related and degradation products method used a gradient of ammonium acetate buffer (pH 4.0) and acetonitrile. Conditions: Zorbax Extend-C18 RR column (250 × 4.6 mm, 3.5 µm), column temperature : 30 °C, flow: 0.8 mL/min, detection at 254 nm, and 125 min run time. For both methods, the autosampler temperature was 8 °C and the injection volume was 20 µL.

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Forced degradation study conditions

In order to determine whether the analytical methods for related and degradation products and assay are stability indicating, the finished drug product was stressed under various conditions to induce degradation.

Thermal degradation

Samples and placebo were directly exposed (in an open petri dish) to thermal degradation by keeping them at 60ºC for 7 days and 10 days. After 7 days and 10 days of exposure, the samples were analysed as per the proposed methods

Thermal/Humidity degradation

Samples and placebo were directly exposed (in an open petri dish) at 40ºC/75% RH, for 1 month. After 1 month of exposure, samples were analysed for assay and related and degradation products.

Photolytic degradation

Photostability study was conducted according to ICH guideline Q1B" Photostability Testing of New Drug Substances and Products"- Option 1. (ICH. Q1B, 1996). The samples and the placebo were placed in quartz petri dishes, alongside protected samples and placebo (wrapped in aluminium foil) used as a dark control and were exposed to light five times the normal irradiance dose for 35h. Samples were analysed as per the proposed methods.

Acid hydrolysis

Samples and placebo were treated with 1mL 0.5M HCl for 10 minutes on magnetic stirrer and after that were neutralized with 1mL 0.5M NaOH and analysed as per the proposed method.

Base hydrolysis

Samples and placebo were treated with 2.5mL 0.04M NaOH for 15 minutes on magnetic stirrer and after that were neutralized with 2.5mL 0.04M HCl and analysed as per the proposed method.

Oxidative degradation

Samples and placebo were treated with 10mL 5% Hydrogen peroxide solution for 15 minutes on magnetic stirrer and analysed as per the proposed methods.

Results and discussion

The specificity of the analytical procedure for assay was established by peak purity determination using UV- DAD detection. Values with purity match greater than 950 indicate that the spectra are very similar and the peak can be considered as pure. All the obtained values exceeded 950, indicating high spectral similarity and confirming peak purity. This demonstrates the absence of interference and supports the specificity of the method for stability analysis.

From the results for related and degradation products, several trends in the impurity profile were observed. Elevated temperature and humidity conditions led to increased levels of Impurities C and H, along with several unknown impurities. Photolytic exposure mainly increased Impurity D. Acid and base hydrolytic conditions caused notable formation of multiple unknown impurities and a significant increase of Impurity C. Oxidative degradation also resulted in increases in several unknown impurities, with minor increases in Impurities A and H. From all the results obtained in this study it can be concluded that the method for related and degradation products has the ability to successfully separate, detect, quantify and identify degradation products.

Conclusion

The forced degradation study confirms the importance of evaluating drug degradation under stress conditions to predict possible degradation pathways and assess the stability profile of the drug substance. The developed analytical methods for assay and related substances were found to be reliable and efficient, thereby confirming their suitability as stability-indicating methods.

References

ICH. Q1A(R2): Stability Testing of New Drug Substances and Products. Step 4 version, 6 February 2003. International Council for Harmonisation; 2003

ICH. Q1B: Photostability Testing of New Drug Substances and Products. Step 4 version, 6 November 1996. International Council for Harmonisation; 1996.