ELSEVIER

Journal of Chromatography A, 746 (1996) 286–288

JOURNAL OF CHROMATOGRAPHY A

Short communication

Gas chromatography–mass spectrometry study of isosorbide 5-mononitrate stability

Valentina D. Marinković[a], Branislav Gudžić[a], Smiljana S. Milojković[b], Jovan M. Nedeljković[b], Jožef J. Čomor[b,*]

[a]*Zdravlje Pharmaceutical and Chemical Industry, Quality Control Sector, 16000 Leskovac, Yugoslavia*
[b]*Vinča Institute of Nuclear Sciences, P.O. Box 522, 11001 Belgrade, Yugoslavia*

Received 4 January 1996; revised 29 March 1996; accepted 29 March 1996

Abstract

The stability of isosorbide 5-mononitrate (5-ISMN) was investigated at 40°C under dry and wet air (75% relative humidity). The degradation products were identified and quantified by means of capillary GC–MS preceded by drying and silylation of the samples. The only degradation products identified were the two diastereoisomers of isosorbide. From the initial slope of degradation curves the maximal degradation rates were estimated to be 0.17 and 2.44% year^{-1} under dry and wet conditions, respectively.

Keywords: Pharmaceutical analysis; Stability studies; Isosorbide; Isosorbide mononitrate

1. Introduction

Isosorbide 5-mononitrate (5-ISMN), usually synthesized from isosorbide (producing the acetate ester as an intermediate step; usually the monoacetate), is a common active component in various coronary vasodilator dosage formulations. During its synthesis some side reactions take place and the final product can be contaminated with up to 8 different compounds [1]. Therefore, numerous analytical procedures based on HPLC [2], GC [3], TLC [4], or polarography [5] were developed for controlling the purity of 5-ISMN and its formulations. On the other hand, except a study of hydrolysis in aqueous solution [6], no work has been done considering the stability and degradation pathways of solid 5-ISMN. The degradation rate of pharmaceuticals is of primary importance for their licensing and dosage formulation packaging, therefore in this paper we present a GC–MS study of the 5-ISMN degradation during its ageing under constant temperature and humidity conditions.

2. Experimental

2.1. Ageing of the samples

5-ISMN (Kali Chemie Pharma) was used in experiments as received, without further purification or drying. The samples, 2.0 g each distributed in a thin layer (<1 mm) on a small Petri dish, were kept in two glass desiccators maintained at constant temperature (40±0.1°C) using a water thermostat (Lauda R52). The humidity in desiccators was controlled either by molecular sieves (4A, Ferak)

*Corresponding author.

0021-9673/96/$15.00
PII S0021-9673(96)00296-8

previously dried at 400°C overnight to obtain dry air, or by a concentrated aqueous NaCl solution ensuring a constant relative humidity of 75% at 40°C. The samples were taken out from the desiccator every 7 days and analyzed immediately.

2.2. GC–MS analyses

The 5-ISMN samples were dissolved in acetone (Merck, analytical-reagent grade) and dried by anhydrous Na_2SO_4 for 2 h. The dry solution of 5-ISMN was separated from the solid and evaporated in vacuum at room temperature. A small portion of the dried 5-ISMN was weighed on a micro-balance, dissolved in the stock solution of internal standard (2-chlorobenzoic acid in diethyl ether, both analytical-reagent grade from Merck) and an aliquot of 50 μl was transferred to a micro-reaction vessel. Before sealing the vessel, the solvent was evaporated applying a slow stream of dry nitrogen to the surface of the solution. For silylation 50 μl of N,O-bis-(trimethylsilyl)trifluoroacetamide (BSTFA, Sigma product) was added through the vessels' septa and the reaction was carried out at 100°C for 10 min [7].

A HP5890/II Plus (Hewlett–Packard) gas chromatograph equipped with a HP5971 mass spectrometer as a detector and a SPB5 (Supelco) capillary column (30 m×0.32 mm I.D., 0.25 μm) was used for the chromatographic measurements using helium as a carrier gas. The injector, operated in splitless mode, was held at 175°C and the temperature program was started at 100°C, held for 2 min, than ramped at 8°C min^{-1} to 300°C. All the measurements were repeated five times and the average peak areas were used for quantitative determinations.

Calibration standards and diagrams were used for the quantification of the measurements. A primary standard of isosorbide was prepared, from previously dried pure isosorbide (99.9%, Kali Chemie Pharma), by dissolving it in the stock solution of internal standard. Using series of different concentrations of this standard, the detector signal (TIC), normalized to the internal standard, was calibrated for isosorbide determination. Since the obtained 5-ISMN initially contained more than 2.2% of isosorbide (as impurity), certain amount of 5-ISMN was dried, dissolved in the stock solution of internal standard, and the isosorbide concentration was determined by GC–MS analysis using the primary standard of isosorbide. From the mass balance the 5-ISMN content was determined and this solution was further used as a stock solution for preparing secondary standards for both isosorbide and 5-ISMN determinations. Standard solutions of 5-ISMN, with higher concentrations of isosorbide than the initial, were prepared by mixing the primary standard of isosorbide with the secondary standard solution of 5-ISMN.

The detector linearity was approved in the range 2–200 mg dm^{-3} for isosorbide and 10–500 mg dm^{-3} for 5-ISMN.

3. Results and discussions

Fig. 1 presents the enlarged parts of the chromatograms of initial (A) and the most aged, 70 days

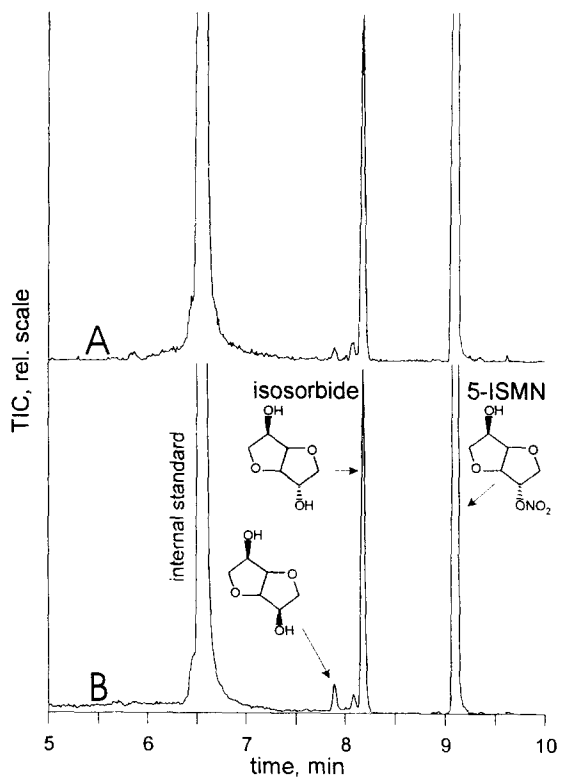

Fig. 1. Chromatograms of initial (A) and the most aged, 70 days at 40°C, 75% rel. humidity, (B) 5-ISMN. The actual compounds detected by the MS are the trimethylsilyl derivatives of the indicated structures.

at 40°C, 75% rel. humidity, (B) 5-ISMN. The chromatograms are normalized to the isosorbide peaks, the peaks of the internal standard and 5-ISMN are shown off-scale. No degradation products other than the two diastereoisomers of isosorbide could be detected (identified by mass spectra and retention time in case of isosorbide), even when the injected amount of 5-ISMN was increased to the upper limit of the detector. The ratio between the two diastereoisomers of isosorbide was not constant during the ageing process, indicating that they are formed through different reaction mechanisms. We had only isosorbide as reference material, therefore the other diastereoisomer could not be determined quantitatively. Assuming the same detector response to both diastereoisomers, the amount of this other diastereoisomer was estimated to be less than 0.1% in all samples.

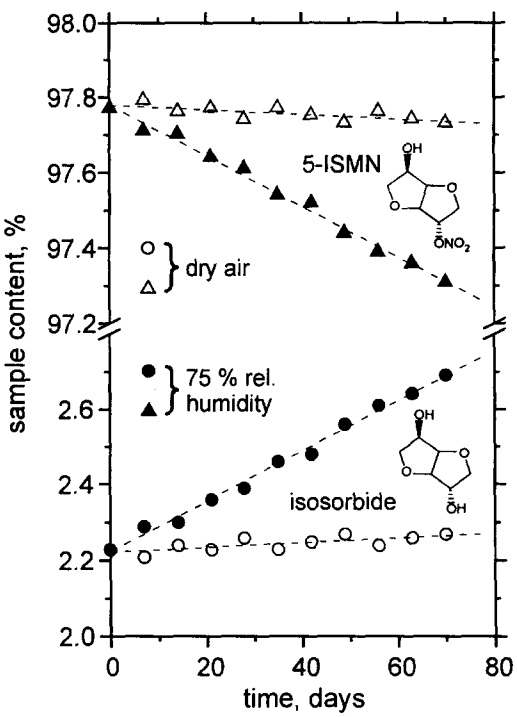

Fig. 2. The kinetics of 5-ISMN degradation.

The kinetics of 5-ISMN degradation is illustrated in Fig. 2. It is evident that the reaction rate is practically constant (under constant temperature and humidity) within the limits of the experimental error, when the total amount of degraded 5-ISMN is less than 3%. This finding allowed us to calculate the maximal degradation rates (from the initial slope of kinetical curves) to be 0.17 and 2.44% year^{-1} under dry and wet conditions, respectively. The fact that the degradation rate in wet air is more than 14 times higher than in dry air indicates that humidity is an important factor in storing the dosage formulations based on 5-ISMN.

References

[1] N. Russeva, N. Dimova, G. Spyrov and M. Jurovska, J. Chromatogr., 295 (1984) 255.
[2] Mizuno, Ch. Shimizu, E. Morita, D. Shinkuma and Y. Yamanaka, J. Chromatogr., 264 (1983) 159.
[3] E. Doyle, L.F. Chasseand and T. Taylor, Biopharm. Drug Dispos., 1 (1980) 141.
[4] M. Carlson and R. Thompson, J. Chromatogr., 368 (1986) 472.
[5] B. Persson and L. Rosen, Anal. Chem., 123 (1981) 115.
[6] N. Mizuno, C. Shimizu, E. Morita, D. Shinkuma and Y. Yamanaka, J. Chromatogr., 264 (1983) 159.
[7] Alan E. Pierce: "Silylation of Organic Compounds", Pierce Rockford, IL, 1968.