

Development and Validation of a Chromatographic Method for the Analysis of Multicomponent Pharmaceutical Preparations

Carola Ferreyra, Cristina Ortiz and M. M. de Bertorello

Depto. de Farmacia, Facultad de Ciencias Químicas, Universidad Nacional de Córdoba. Ciudad Universitaria. 5000 Córdoba, Argentina

Abstract: A reverse phase high performance liquid chromatographic assay was carried out for the simultaneous determination of two out of three active principles present in a pharmaceutical preparation. This method was developed to assess the quality of the product.

Introduction

At present some highly complex pharmaceutical preparations in the pharmacy do not only fail to be analyzed by the traditional chemical methods but also require modern highly selective and sensitive instrumental techniques.

The authorised pharmaceutical product must have certain specific information such as: formulation method, therapeutic prescription, counter effects and quality control. This last procedure involves the performance of assays of the raw material, of the components in the production process, analysis of the final product and stability studies that aim to achieve an efficient medicinal product.

The analytical procedures employed nowadays for the analysis of the pharmaceutical active principles need a chemical substance for reference. Therefore, in the first stage of this study the standard references were prepared for their application to the quality control of a pharmaceutical preparation with commercial use in the Province of Córdoba (Argentina). The two active principles selected for this study were phenylpropanolamine hydrochloride (**I**) and caffeine (**II**).

Experimental

The High Pressure Liquid Chromatography (HPLC) system was equipped with a Konik 500 G pump, a Konik integrator model SP-4290, a variable wavelength UVIS-200 UV detector and a Rheodyne model 7125 injector with a 20 μ L loop. A Supelcosil column LC-18 (250 x 4,6 mm) was operated at a 0.6 mL/min flow, sensitivity 0.02 AUFS, chart speed 0.25 cm/min and wavelength 254 nm. The mobile phase, methanol:water (50:50 v/v) was filtered (0,45 μ m Nylon-66 membrane) and degassed before use.

Results and Discussion

The quality control of the standard references comprised the following steps: sampling, identification technique, IR (ν_{\max} cm^{-1} , KBr) **I**: 3197 (NH_3^+); 3340 (O-H) **II**: 1658 (C=N); 1702 (C=O); Thin-Layer Chromatography (methanol : acetic acid : diethyl ether: benzene - 0.7 : 1.5 : 5 : 10) R_f **I**=0,07; R_f **II**=0,24; UV spectroscopy (λ_{\max} nm-methanol) **I**: 254; **II**: 275; Loss on Drying **I**: 0.50%, **II**: 0.28%; Melting Range **I**: 191-194°C ; **II**: 230–232°C and Purity Degree **I**: 99.53%; **II**: 99.76%. After these studies, the next step involved selection of the appropriate analytical method, its validation and finally quantification of the active principles in a pharmaceutical preparation of wide commercial use in Córdoba (Argentina). The quality parameters determined were: precision (CV) **I** 2.5%; **II** 1.0%, limit of detection (LOD- $\mu\text{g}/\text{mL}$) **I** 0.28; **II** 0.11 and limit of quantification (LOQ- $\mu\text{g}/\text{mL}$) **I** 0.93, **II** 0.36.

The selected and validated HPLC method was applied to the pharmaceutical preparation to assure the quality of the final product. The results were expressed in $\text{mg} \pm \text{CV}$ ($n=5$) for **I** 12.8 ± 2.2 and **II** 39.8 ± 1.6 .

These data indicate that HPLC is an efficient method for simultaneous quantification of the active principles without previous preparation of the sample. Likewise and due to the intensive use of this preparation in the Province of Córdoba and to the lack of quality control, it is planned to continue the analysis of the remaining active principle so as to determine the overall composition of the pharmaceutical preparation.

References and Notes

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