A Chromatographic Determination of the Stability of Solutions of Amlodipine Benazepril & Amlodipine Besilate

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Introduction

Drug substances (or actives) and drug products can be susceptible to chemical degradation by different mechanisms such as hydrolysis, and oxidation, leading to loss of potency and formation of degradation products. Several factors, such as temperature, storage containers, excipients and oxygen affect the stability of actives and drug products. Stability testing is carried out to investigate how different factors affect the quality of an active or a drug product with time (Tsong Y., 2003) and to determine a time interval over which the potency of the drug remains within a certain limit. Stability of an active relates to its ability to remain within a certain acceptance criteria so as to ensure its strength, purity, quality and identity during a defined period of time (Tsong Y., 2003).

The two products investigated during this study were amlodipine besilate tablets and amlodipine benazepril capsules. Amlodipine is the active present in amlodipine besilate whereas in amlodipine benazepril two actives are present which are amlodipine and benazepril hydrochloride.The objectives of this project were to investigate the stability of sample solutions and standard solutions of both products stored under different conditions so as to determine whether degradation occurred over time, as measured by the loss in percentage assay of the active, to determine whether different storage conditions affected the degree of degradation of these products, and to determine whether there was a time interval between the preparation of solutions and their analysis during which they were still stable, where stability of the active was related to its ability to remain within the limits defined in the acceptance criteria.

Methodology

For each product, stability testing was carried out by preparing a stock standard solution, a working standard solution and a sample solution which was obtained by carrying out a dissolution test of the product. The incubated working standard solutions and sample solutions, and a working standard solution freshly prepared from the incubated stock standard solution on the day of analysis were analyzed against a freshly prepared working standard solution on days 0, 1, 2, 4 and 7 by HPLC on a Waters Alliance system. The data obtained was assessed against the acceptance criteria for both mean percentage assay and coefficient of variation using statistical analysis. Forced degradation studies were also carried out so as to determine whether the placebo or degradants of the placebo and sample interfered with the peaks of the actives and also to determine the identity of the unknown peaks present in sample chromatograms. Evaporation studies were carried out so as to investigate the degree of evaporation of diluent during the time period studied. This was carried out by storing the sample diluent and the standard diluents under the different sets of storage conditions that sample solutions and standard solutions were stored respectively. The loss in weight of diluent with time was recorded at every time interval.

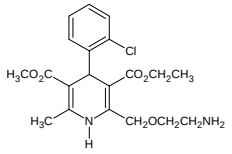

Figure 1: Amlodipine

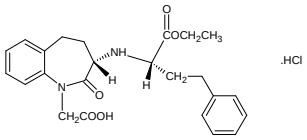

Figure 2: Benazepril hydrochloride

Results

Amlodipine degraded significantly with time under all investigated storage conditions in amlodipine besilate sample solutions. A kinetic model was fitted to the mean percentage assay of amlodipine in sample solutions. When stored under all sets of conditions except in syringes in the refrigerator, degradation followed zero-order or approximations to zero-order kinetics. In syringes with and without cap in the refrigerator, degradation followed first-order or approximations to first-order kinetics. Amlodipine besilate standard solutions were considered as being stable over the time period studied. Amlodipine also degraded significantly with time under all investigated storage conditions in amlodipine benazepril sample solutions. A kinetic model was fitted to the mean percentage assay of amlodipine in amlodipine benazepril sample solutions. When stored under all sets of conditions, degradation followed zero-order or approximations to zero-order kinetics.

In the case of amlodipine benazepril standard solutions, one or both actives degraded when the solutions were stored under certain conditions. A kinetic model was fitted to the mean percentage assay of active/s which degraded with time. When stored in glass vials at room temperature, degradation of amlodipine followed first-order or approximations to first-order kinetics while degradation of benazepril HCl followed zero-order or approximations to zero-order kinetics. When stored in plastic vials at room temperature and in glass vials in the refrigerator, degradation of benazepril HCl followed zero-order or approximations to zero-order kinetics.

In both products, the peaks of degradants and placebo did not interfere with the peaks of the actives, therefore, the method for both products can be considered as being specific and it can be used in stability studies. The unknown peak in the chromatograms of amlodipine besilate sample solutions was Impurity D. The unknown peaks in the chromatograms of amlodipine benazepril sample solutions were Impurity C and Impurity D.

Evaporation of standard diluent occurred to a larger extent than sample diluent in both products. Evaporation occurred when 40% methanol and 65% acetonitrile were stored in volumetric flasks which were capped, sealed and from which 5 mL diluent was removed at every time interval. This shows that evaporation occurred during stability analysis.

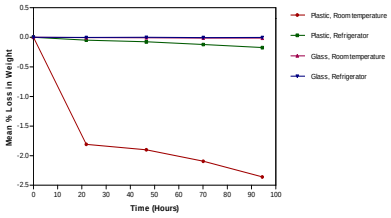

Figure 3: Evaporation of sample diluent in vials

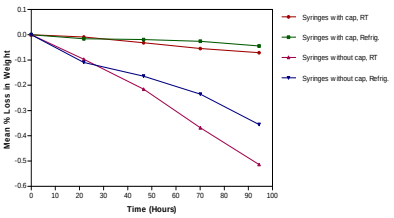

Figure 4: Evaporation of sample diluent in syringes

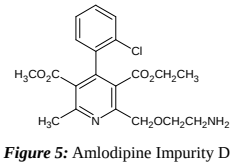

Figure 5: Amlodipine Impurity D

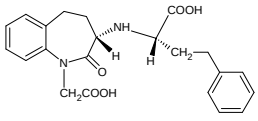

Figure 6: Benazepril HCl Impurity C

Conclusions

Amlodipine in both amlodipine besilate sample solutions and amlodipine benazepril sample solutions stored under all sets of storage conditions was unstable till the first time interval had passed. Therefore, a time interval during which sample solutions of the products were stable could not be determined. However, amlodipine in amlodipine besilate standard solutions was considered as being stable over the time period studied. In the case of amlodipine benazepril standard solutions, a time interval during which the actives were considered as stable when stored under certain sets of conditions was determined. Amlodipine benazepril standard solutions stored in glass vials at room temperature and in the refrigerator were determined to be stable up to 43.5 hours while working standard solutions prepared by dilution on the day of analysis from stock standard solutions stored at room temperature and in the refrigerator were determined to be stable up to 24.6 hours. However, these results have to be interpreted with caution since according to the evaporation studies carried out, evaporation of diluent occurred in all sets of storage conditions both in the case of sample solutions and standard solutions of both products to different extents thus decreasing the accuracy of the results obtained.

References

1. Tsong Y. (2003), "Recent Issues in Stability Study" *Journal of Biopharmaceutical Statistics*, Vol. 13 No. 3 pp 7 – 9

Acknowledgements

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