

Production and stability of a ready-to-use hydroxocobalamin solution for paediatric parenteral use

R. Moudry, S.Hornstein, M. Fehr-Bigger, S. Guyer
Kantonsspital Graubünden, Institute for Hospital Pharmacy, Chur, Switzerland

Background

A paediatric patient in our hospital suffers from a rare, hereditary transcobalamin II deficiency resulting in an intracellular vitamin B12 deficiency. Specific treatment consists of massive parenteral intake of vitamin B12. Parenteral vitamin B12 solution, suitable for young children, is not commercially available in Switzerland.

Our aim was to produce a preservative-free, sterile hydroxocobalamin (vitamin B12) solution at a concentration of 10 mg/2 ml for intramuscular use, and to develop a stability indicating analytical method in order to determine the shelf life of the solution.



Methods

The entire manufacturing process occurred under aseptic conditions. Hydroxocobalamin hydrochloride was dissolved in NaCl 0.9% to give a concentration of 10 mg/2 ml. The pH was adjusted to 4.3 - 4.5 using hydrochloric acid. The solution was 0.2 µm filtered and a 2.4 ml solution was aseptically filled in sterilised brown glass vials. The filter integrity was tested using the bubble point method. The vials were kept in the refrigerator. Alternatively, the solution was autoclaved at 121 ° C for 15 min and assessed by high performance liquid chromatography (HPLC) for degradation products.

Results

The autoclaved solution showed degradation products and the hydroxocobalamin content was decreased by 20% (see figure 1).

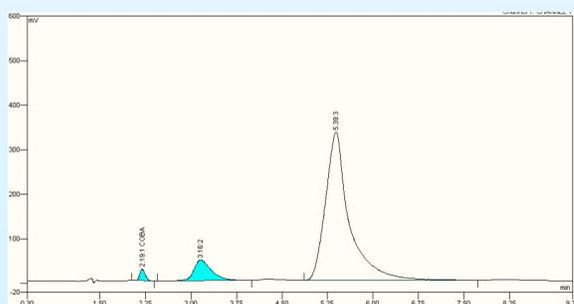


figure 1: chromatogram of autoclaved hydroxocobalamin hydrochloride

The following HPLC procedure was used:

Column: Nucleodur® 100-5C,18ec (Macherey-Nagel)
Eluent: Acetonitrile/0.5mM Sodium 1-octanesulfonate pH 3.0 , 15:85 (v/v)
Standard: Hydroxocobalamin hydrochloride (300 µg – 600 µg/ml)
Flow rate: 1ml/min
Detection: UV, 351 nm
Retention time of hydroxocobalamin: approximately 5.4 min.
Retention time of heat degradation products: 2-3 min.

In contrast, the sterile filtered solution showed no degradation products and no loss in the hydroxocobalamin content was observed after storage for 24 months at 2 – 8 ° C (see figure 2 and figure 3).

In addition, the solution was stable when stored for one month at room temperature and even exposure to 56° C for 2 days did not cause the product to degrade.

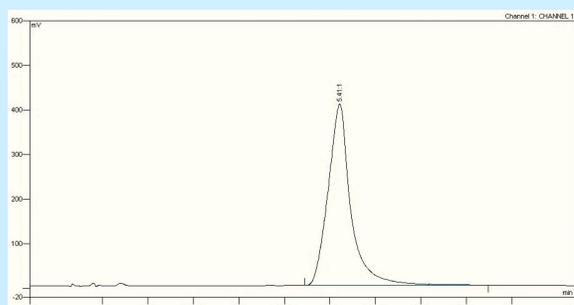


figure 2: chromatogram of freshly dissolved hydroxocobalamin hydrochloride (500 µg/ml)

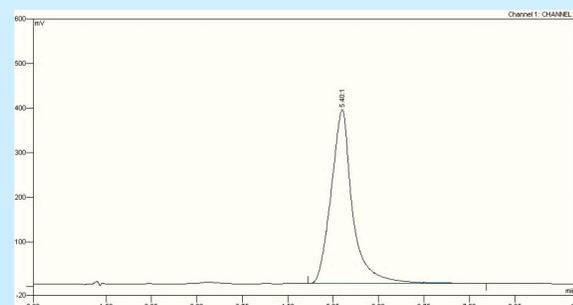


figure 3: chromatogram of hydroxocobalamin hydrochloride (10 mg/2 ml), stored for 24 months at 2 – 8 ° C.

Conclusions

We produced a hydroxocobalamin solution for intramuscular use with a shelf-life of at least 24 months if refrigerated. The treatment of our patient with this solution, administered as an intramuscular injection once a week, has been extremely successful for more than 3 years.