Abstract
Ion Chromatography (IC) is a very sensitive technique which requires high quality columns since even very small changes of the packing-material will have a huge impact on the obtained results. In addition, a robust stationary phase is required which delivers proper separations even after a huge number of injections. The reproducibility of Hamilton PRP-X100 anion exchange columns was tested for multiple injections on the same column and for columns from the same as well as from different lots. It was found that the reproducibility was excellent in all cases.

Introduction
Anion chromatography is a well established technique which is very frequently used for monitoring of organic and inorganic anions in tap, waste or environmental water as well as in process chemicals and consumer products [1]. Many industrial countries have guidelines which consider IC as the method of choice for speciation and quantification of ionic components in water (e.g. EPA 300.0, ASTM D4327-11) [2].

An outstanding quality of the anion separation process is required to hit the high standards which are defined in the different directives. Consequently, a robust and easy to handle column with excellent reproducibility is required to fulfill the high standards under real field conditions.

The column quality depends on the physical and chemical properties of the anion exchange resin such as, e.g. pore-size, particle diameter and exchange capacity. In addition, the packing of the resin particles has a huge impact on the separation. For reproducibility purposes all parameters have to be constant over time for columns of the same specification. Since a method is once validated and implemented, the separation on the same as well as on a new column should always give similar results under similar conditions. The separation quality highly depends on different parameters such as, e.g. retention time or peak shape. It is very common to differentiate between three different experiments for covering all aspects of column reproducibility:

- Multiple injection on the same column
- Column-to-column reproducibility
- Lot-to-lot reproducibility

Material & Methods
An extensive reproducibility test was performed on a Hamilton PRP-X100 anion exchange column. A blend of 5 different inorganic anions (standard mixture) was separated isocratic in 4 mM p-hydroxybenzoic acid buffer with 2.8 volume-% methanol (pH = 8.5). The inverse UV detection principle (310 nm) was applied since this method is able to show also compounds which have no or low conductivity and thus it is more sensitive even for small changes in the IC-system. A column with a size of 2.1x250 mm and 5 µm particle diameter was used (PN 79190) because it offers increased resolution. Due to the high residence time the highly retained components will indicate possible derivations in the column packing material in a pronounced manner. The column exhibits a low inner diameter which allows working at low flow rate of 250 µL/min saving a high amount of solvent. Consequently, all separations have been realized with the same eluent which eliminates possible errors caused e.g. by differences in the preparation of the mobile phase. The temperature was kept slightly above ambient at 30°C to further improve the reproducibility. An Agilent 1100 chromatograph equipped with quaternary pump (G1311A), ALS autosampler (G1313A), column heater (G1316A) and DAD detector (G1315B) was used for all experiments. The sample concentration was 40 µg/mL (40 ppm) in water for each component. The injection volume was 20 µL.

Three different experiments were carried out with the aim to cover all aspects of column quality:

- The long term consistency was tested by multiple injection of the same standard mixture.
- The column-to-column consistency was tested by separation of the standard sample mixture on six different columns.
- The batch-to-batch consistency was compared for the separation of the standard mixture on three columns from three different production lots.

Results and Discussion
In Figure 1-3 the results of the reproducibility test are shown. For a better comparison of the different chromatograms...
the relative standard derivations (RSD) of characteristic chromatogram parameters are noted in Table 1 for bromide and nitrite peak.

In Figure 1 the excellent reproducibility of multiple sample injections on the same column is shown. For all injections the retention times and peak shapes are very similar to the first run which is also confirmed by very constant peak parameters in Table 1. It can be assumed that even after a high number of samples no contamination, clogging or degeneration of the stationary phase will occur. The results are good evidence for the increased durability of this column type even under ambitious working conditions.

The results obtained from different columns of the same lot are shown in Figure 2 and Table 1. Even for the comparison of different columns very consistent retention times and peak shapes have been recorded. The proper reproducibility reflects the well controlled polymerization process and surface modification as well as the optimized column packing methods.

Even the results from different lots in Figure 3 are in close agreement with the previous data. The derivations in peak shape are still below 5% and the error of the retention time is below 1% which is in the similar range as it was achieved for the columns from the same lot in Figure 2. Despite the high sensitivity of ion-exchange chromatography to very small changes in the packing material, excellent reproducibility was proven. It can be concluded that the separation efficiency is effectively the same for each column and does not depend on the date of manufacturing.

Conclusion

Hamilton is certified according to ISO 9001. Our advanced quality management system guarantees excellent purity of the raw materials and well controlled production processes resulting in a very constant product quality even for highly sensitive devices, such as the tested IC-columns.

In general, all polymeric LC columns from Hamilton are based on the same kind of PSDVB support material. Due to the high sensitivity of ion chromatography for even very small derivations of the stationary phase the obtained results are also representative for the outstanding quality of the other Hamilton HPLC column lines. Despite the outstanding quality, each Hamilton column is additionally tested under standard conditions prior to shipment. The results of this test are included in a certificate of analysis which is delivered together with the HPLC column providing maximum transparency to the customer.

References