

There are several issues that could be happening in this situation.

- When an analyst was using a buffer within two weeks, the **retention** times and tailing fit the limits of the method. When the buffer was stored for longer, (up to three months) the chromatographic values did not meet system suitability criteria for **retention** time %RSD and peak tailing.

Unless there has been validation testing that the mobile phase is stable for longer and no evaporation was shown in their storage, in this situation, the analyst could be experiencing evaporation of the organic. This can then increase the aqueous concentration, causing more retention (non-polar compounds) in reversed phase (subsequently increasing tailing).

- The buffer also contained tetrabutylammonium hydrogen sulfate. Hydrochloric or sulphuric and counter ions have **direct effect on the life of the stationary phase**. Storing buffer for longer than two weeks can cause evaporation and increase the concentration of this salt, putting more strain on the column than expected.
- Lastly, if the mobile phase is degassed manually under vacuum there are chances of volatile solvent (ACN) to evaporate.

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