

# Qualifying the Ultra-violet detector in a chromatography system

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## Introduction

Liquid chromatography is often used in laboratories to determine the concentration of molecules in a sample. Therefore, qualifying chromatography system is essential to the good process of analysis and the conformity of the results.

Qualification consists in checking each parts of system: the separations module, the detector and thermostatisation in order to verify their suitability.

Here, only the ultra-violet (UV) detector suitability tests are detailed.

## Material and methods

Three tests are realized using a compact system of chromatography.

- Kromasil column (150mm \* 4.6mm, 5 $\mu$ m, C18)
- Temperature column : 30°C
- Mobile phase: 45% ultrapure water / 55% methanol
- Flow: 1.0mL/min
- Volum injected: variable

A standard solution of PHB of methyl which is a UV-absorbent molecule was prepared. Other solutions of different concentrations issued by standard solution were also set.

The first test consisted in measuring the background and the drift of the detector injecting 1 $\mu$ L of standard solution in 80 minutes.

The second test consisted in checking the linearity of the absorbencies injecting 50  $\mu$ L of five solutions prepared with standard solution.

The third test was on the wavelength's exactitude: the spectrum of standard solution was extracted to find the maximum wavelength.

## Results and conclusion

The background and the drift were determined by graphic construction (Fig1) and the values obtained were 55.4  $\mu$ V and 377.5  $\mu$ V respectively. Even if the construction was subjective, the interest of this test was to show that the background and the drift were negligible compared to the peak of PHB of methyl.

Values of linearity differ to expected values less than 2%, the limit of conformity.

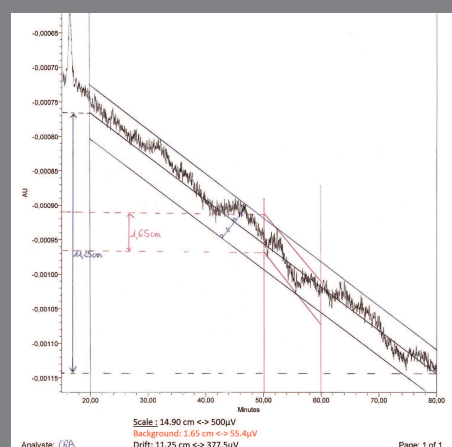
With the third test, the maximum wavelength obtained was 257.1 nm and the theoretical wavelength was 256 nm. This test was in compliance with the accepted limit ( $\pm$  2nm).

To conclude, results of the three tests proved the suitability of the UV detector.

Generally, tests to qualify chromatography system or one part of it are simple to realize. They permit to assure the conformity of results obtained with this system.

To sum up, verifying the suitability of a system is an assurance of reliable results.

Fig1: Graphic determination of the background and the drift on the chromatogram obtained.



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