

The method is based on quantitative determination of volatile compounds in beer, that is, in beer distillate, by applying the method of external calibration.

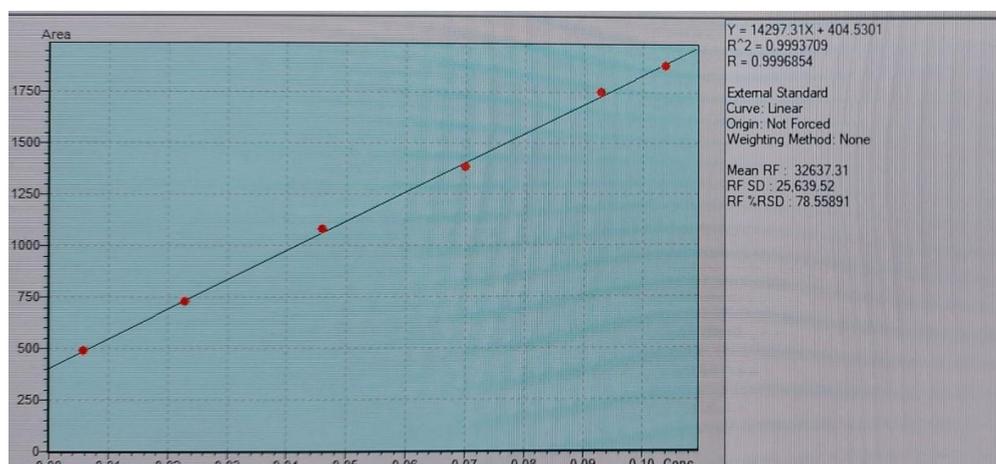
This is a method used for determination of quantitative relationship between the analyte concentration in the sample and responding signals generated during analysis.

Calibration curve was done using the following standards:

| Chemical   | CAS number      | Producer                   |
|--|-----------------|----------------------------|
| Acetaldehyde, C <sub>2</sub> H <sub>4</sub> O                      | 75-07-0         | 99.5%, Sigma-Aldrich       |
| Ethyl acetate, C <sub>4</sub> H <sub>8</sub> O                     | <u>141-78-6</u> | 99.5%, Sigma-Aldrich       |
| 1-propanol, C <sub>3</sub> H <sub>7</sub> OH                       | 71-23-8         | 99.5%, Sigma-Aldrich       |
| izo-butanol, C <sub>4</sub> H <sub>9</sub> OH,                     | 78-83-1         | > 99%, Fluka               |
| izo-amil acetate, C <sub>7</sub> H <sub>14</sub> O <sub>2</sub>    | 123-92-2        | ρ=0.87 g/mL, Sigma-Aldrich |
| 3-metilbutan-1-ol  | 123-51-3        | ρ=0.81 g/mL, Sigma-Aldrich |
| 2-feniletil acetat, C <sub>10</sub> H <sub>12</sub> O <sub>2</sub> | 103-45-7        | 99%, Sigma-Aldrich         |
| 2-feniletanol, C <sub>8</sub> H <sub>9</sub> OH,                   | 60-12-8         | 99%, Sigma-Aldrich         |
| Dimethyl sulfide (DMS), C <sub>2</sub> H <sub>6</sub> S            | 75-18-3         | 99%, Sigma-Aldrich         |
| butan-2,3-dion, C <sub>4</sub> H <sub>6</sub> O <sub>2</sub>       | 431-03-8        | > 99%, Merck-Schuchardt    |
| pentan-2,3-dion  | <u>600-14-6</u> | > 99%, Merck-Schuchardt    |

**Standard solutions preparation:** standard solutions of different concentrations are prepared using known concentrations of standard solution and 5% ethanol solution. The prepared solutions of standards cover the expected spectrum of each single component in real samples. The prepared standard solutions are injected into gas chromatograph, one after another, using the autosampler (AOC-6000 Plus, Shimadzu). A responding signals generated during analysis are then being noted for each standard solution.

**Calibration curve:** for each standard, the generated signal, or more precisely, the area underneath the peaks is being determined depending on the known standard solution concentration. This results with calibration curve which shows the relationship between the area and analyte concentration.



Linearity has been checked by measuring different concentrations of standard solutions through the entire working area. It is important to make sure that the area (A) is growing linearly with concentrations ( $\gamma$ ). The calibration curve is linear if  $R^2 \geq 0.99$ .

**Sample analysis:** samples of analyte of unknown concentrations have been injected into GC, and their signals have been measured during analysis. Based on the measured signal and calibration curve, the analyte concentration in the sample has been determined.