Gas chromatographical separation of alcohols - temperature effect

Aims of the experiment

- To understand the principle of gas chromatography
- To recognise that also liquid (but volatile) substances can be separated by gas chromatography
- To investigate the influence of temperature on the separation of alcohols by gas chromatography
- To check the purity of the denatured ethanol

Principles

Mixtures of gaseous substances are separated into their components by gas chromatography (GC). In this way, mixtures can be analysed or individual substances can be isolated from mixtures.

The sample, e.g. a gas mixture, is fed into the gas flow (the mobile phase) and carried by this into the stationary phase in a column. The stationary phase consists of a material with which the gaseous substances interact, e.g. by dissolution or adsorption. It is essential that the components of the mixture exhibit different strengths of interaction with the stationary phase. Only then the substances can be separated. Substances which show very little interaction leave the column (elute) more quickly than substances which interact strongly with the column.

The interaction is based on chemical or physical effects. Part of the substance is adsorbed onto the support material. This part is in equilibrium with the non-adsorbed part. Only the non-adsorbed part can be carried further by the gas flow. The larger the proportion of adsorbed material, the slower the substance migrates through the column. The weaker the interaction, the faster the substance migrates.

In this experiment, alcohols are to be separated. These are liquid, but volatile. Here, the vapour phase over the liquid alcohol is analysed (headspace or vapour space method). Alcohols are polar substances which strongly interact with the stationary phase. For this reason, non-polar organic materials, such as polystyrene balls (Poropak™) are used for the separation. Gas chromatographic analysis of alcohols is used in the analysis of blood alcohol.

Even with this optimised stationary phase, the separation process at room temperature is very slow. In gas chromtography we can therefore make use of the characteristic that the adsorption equilibrium is shifted to the side of the non-adsorbed form with increasing temperature. A larger proportion of the substance is therefore located in the mobile phase than in the stationary phase. The substance can then move more quickly through the column.

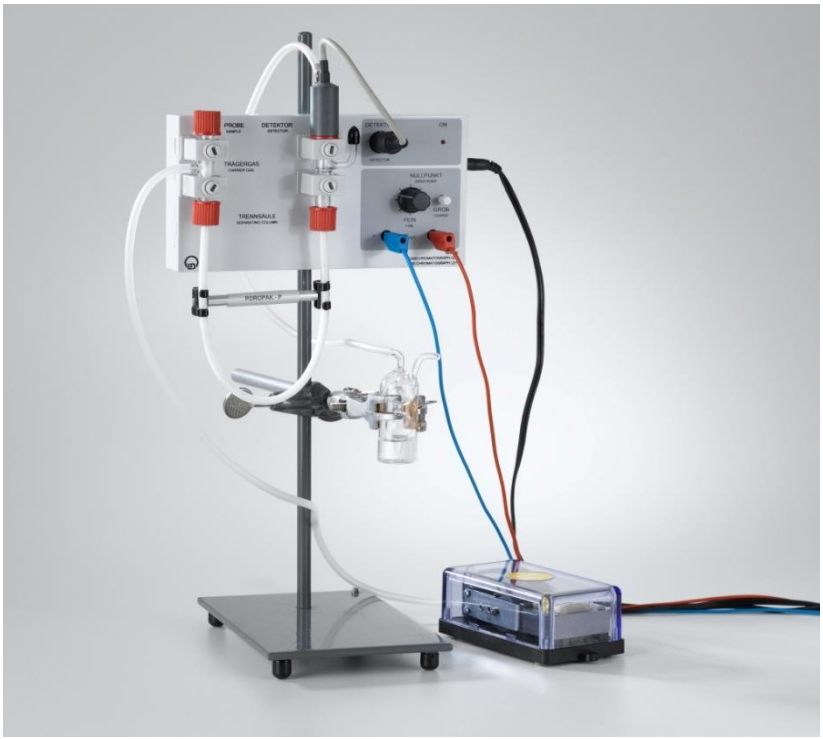

Fig. 1: Set-up of the experiment.

SW-2015-09

Risk assessment

Combustible liquids are used in very small amounts in this experiment. If possible, work in a ventilated room. Wear personal protective equipment when using methanol!

Methanol

Signal word: HAZARD

Hazard statements

H225 Highly flammable liquid and vapour.

H331 Toxic if inhaled.

H311 Toxic in contact with skin.

H301 Toxic if swallowed.

H370 Causes damage to organs.

Precautionary statements

P210 Keep away from heat/sparks/ open flames/hot surfaces. No smoking.

P233 Keep container tightly closed.

P280 Wear protective gloves/protective clothing/eye protection/face protection.

P302+P352 IF ON SKIN: Wash with soap and water.

P309 + P310 IF exposed or you feel unwell: Immediately call a POISON CENTER or doctor/physician.

1-Propanol

Signal word: HAZARD

Hazard statements

H225 Highly flammable liquid and vapour.

H318 Causes serious eye damage.

H336 May cause drowsiness or dizziness.

Precautionary statements

P210 Keep away from heat/sparks/ open flames/hot surfaces. No smoking.

P233 Keep container tightly closed.

P280 Wear eye protection.

P305+P351+P338 IF IN EYES: Rinse continuously with water for several minutes. Remove contact lenses if present and easy to do. Continue rinsing.

P313 Get medical advice/attention.

Ethanol (absolute or denatured)

Signal word: HAZARD

Hazard statements

H225 Highly flammable liquid and vapour.

Precautionary statements

P210 Keep away from heat/sparks/ open flames/hot surfaces. No smoking.

2-Propanol

Signal word: HAZARD

Hazard statements

H225 Highly flammable liquid and vapour.

H318 Causes serious eye damage.

H336 May cause drowsiness or dizziness.

Precautionary statements

P210 Keep away from heat/sparks/open flames/hot surfaces. No smoking.

P233 Keep container tightly closed.

P305+P351+P338 IF IN EYES: Rinse continuously with water for several minutes. Remove contact lenses if present and easy to do. Continue rinsing.

Equipment and chemicals

For setting up with stand materials:

1 Base plate for Bunsen stand, 13 x 21 cm 666 503
1 Stand rod 450 mm, 12 mm diam., M10 ..... 666 523
1 Universal clamp 0...80 mm 666 555
1 Bosshead S 301 09

For setting up in CPS

1 Panel frame C50, two-level, for CPS 666 425
1 Base panel for gas chromatograph LD1 ..... 665 588
1 Pedestal, CPS 666 441
1 Blank panel 200 mm, CPS 666 467
1 Blank panel 300 mm, CPS 666 468

For both versions

1 Gas chromatograph LD 1 ..... 665 580
1 Hydrocarbon sensor 665 582
1 Separation column with Porapak P 665 584
1 Pocket-CASSY 2 Bluetooth 524 018
1 CASSY Lab 2 ..... 524 220
1 UIP Sensor S 524 0621
1 Aquarium pump, 100 L/h 662 2861
2 Beaker, Boro 3.3, 800 mL, tall form 602 013
1 Laborboy II (laboratory jack-stand) 300 76
1 Magnetic stirrer with hot plate, square 607 5025
1 Stirring magnets 25 mm x 6 mm diam. 666 851
1 Stirring thermometer, -10...+110 °C/1 K 204 110
1 Bubble counter with flash-back protection 664 814
1 Disposable syringe 1 mL 665 957
1 Cannula 0.45 mm diam., set of 10 ..... 665 960
1 Connecting leads 19 A, 50 cm, R/B, pair 501 45
1 Septa, set of 10 ..... 665 589
1 Silicone tubing 4 mm diam., 1 m 667 197
1 Methanol, 250 mL 673 2700
1 Ethanol, absolute, 250 mL 671 9700
1 Ethanol, denatured 250 mL 671 9740
1 1-Propanol, 250 mL 674 4310
1 2-Propanol, 250 mL 674 4400

Also required:

Computer with Windows XP/Vista/7/8

Also necessary for wireless measurement:

1 Battery for Pocket-CASSY 2 Bluetooth 524 019
1 Bluetooth dongle 524 0031

Set-up and preparation of the experiment

Set-up of the gas chromatograph LD 1

Set-up with stand materials

Firmly screw the gas chromatograph LD 1 to the Bunsen stand (see Fig. 1) and connect to the power. Attach the bubble counter beneath the gas chromatograph with a universal clamp.

Set-up in CPS

Mount the base panel for the gas chromatograph LD1 in the upper section of the panel frame and fix the gas chromatograph on the panel. Lock the bubble counter in place in the position provided on the base panel. Also, insert the smaller blank panel. Place the pedestal and the larger blank panel into the lower section. Place the aquarium pump and the Pocket CASSY onto the pedestal.

Set-up for both versions

Fill the bubble counter with water so that the inner glass tube is just immersed. Connect the pump to the inlet of the chromatograph with a piece of tubing. Use the other tubing to connect the outlet of the chromatograph to the bubble counter.

Fit the hydrocarbon sensor to the gas chromatograph as shown in Fig. 1.

Screw the column into the chromatograph using the GL fittings so that the marking on the column faces forwards. If necessary, push the inlet of the column in the chromatograph upwards so that it is about 5 mm below the septum.

Connect the voltage output socket of the chromatograph to the voltage input socket of the UIP sensor of the Pocket CASSY using the connecting leads. Connect the Pocket CASSY to the computer.

Performing the experiment

Blank run

First perform a blank run. In this case, no substances are place on the column.

1. Connect the pump to the power. It then runs automatically. Bubbles should now rise in the bubble counter so quickly that they are difficult to count.
2. Load CASSY Lab settings
3. Initially set the voltage U_1 on the GC to approximately 0 with the coarse control. Re-adjust with the fine control. The calculated voltage U will be displayed during the experiment. CASSY Lab automatically subtracts the measured voltage at the start of the measurements from all further measured values. This ensures that all measurements start at 0.
4. Start the measurement by clicking on the symbol .
5. Stop the blank run after around 10 to 15 minutes.

Note: The blank run is needed so that the column is ready to use and constant values can be expected.

The influence of temperature on the retention time of ethanol

1. Heat water in a beaker on the stirrer hot plate to around 70 °C.
2. Meanwhile, perform the first run at room temperature. For this, shake the ethanol bottle in order to saturate the vapour space with ethanol.
3. Fill the syringe with a sample (0.2 mL) of gaseous ethanol from the vapour space. Place a thin cannula onto the syringe.

Caution! Never apply the liquid alcohol, but rather the gaseous part on the column!

4. Insert the cannula into the septum on the GC.
5. Start the measurement in CASSY Lab. After 5 seconds (visible in the field "measuring time"), press the entire contents of the syringe quickly into the GC.
6. Once the signal has appeared, end the run.
7. For the next run, the column will be warmed in a column bath (beaker). For the run at 45°C, mix hot water with cold water so that it is around 45 - 50°C. Locate the column bath using the laboratory jack-stand such that the column hangs completely in the beaker. Add enough warm water to completely immerse the column. Quickly perform the run at 45°C analogously to the run at room temperature.
8. Heat the water again to around 70°C.
9. Perform a run at 65 - 70°C in the same way.
10.

Separation characteristics of various alcohols on the GC

Analogously to the above, the various alcohols methanol, ethanol, 1-propanol and 2-propanol will be separated at one temperature on the GC column. The gas volumes to be injected are compiled in Table 1. As 1- and 2-propanol have very long retention times at room temperature, it is recommended to perform the analysis at around 65°C. Make sure that the water has a similar starting temperature, i.e. repeat the heating.

Tab. 1: Gas volumes of the individual substances to be injected onto the column.

Substance	Volume
Methanol	0.1 mL
Ethanol	0.2 mL
1-Propanol	1 mL
2-Propanol	1 mL

Analysis of the purity of denatured ethanol

The purity of alcohols can be checked using gas chromatography. For this, ethanol of denatured quality is used here as an example.

1. Prepare a column bath at around 45 to 50 °C.
2. As described above, remove a sample from the vapour space of the denatured ethanol bottle and separate it on the gas chromatograph.
3. Compare the retention time of the resulting signals with the runs performed previously at 45°C.

Observation

Alcohols can also be separated using gas chromatography.

If the GC column is heated to different temperatures from outside by a water bath, the retention time of the substance to be separated, here ethanol, changes distinctly (see Fig. 2).

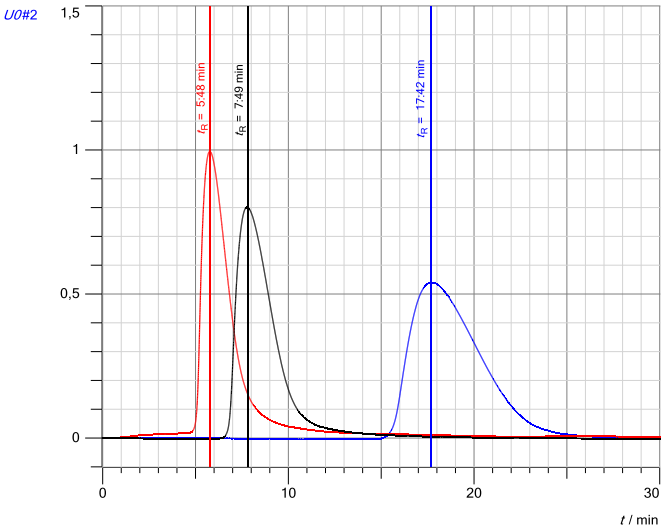

Fig. 2: The influence of temperature on the retention times of ethanol. Temperatures: red: 65 °C, black: 45 °C, blue: 25 °C.

The alcohols being investigated elute in the following order: methanol – ethanol – propanol (see Fig. 3).

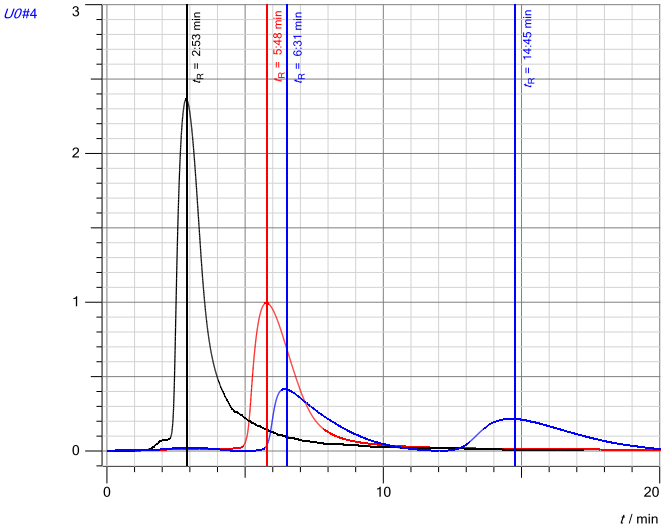

Fig. 3: Chromatograms of the various alcohols. Temperature: 65 °C. Black: methanol, red: ethanol, blue, 1st signal: 2-propanol, blue, 2nd signal: 1-propanol.

Evaluation

Determine the retention times t_R in CASSY Lab as follows.

1. Mark with a vertical line the maximum of each signal.
2. Using drag-and-drop, drag the retention time t_R from the status bar into the diagram next to the vertical line.
3. Proceed in this way with all signals.

Results

Separation characteristics of various alcohols on the GC

Volatile liquids can also be separated using gas chromatography. The separation depends on various factors.

1. The larger the molecule, the later it elutes from the column. Therefore, methanol elutes before ethanol and the propanols (see Fig. 3).
2. The more polar a substance, the later it elutes from the column. 2-propanol elutes from the column before 1-propanol. Whilst 1-propanol possesses a terminal hydroxyl group, this is arranged centrally in the case of 2-propanol. As a result, 2-propanol is considerably less polar than 1-propanol. The more complex the separated substances are, the more the separation depends on the structure and not on the size of the molecule.
3. The higher the temperature, the earlier a substance elutes from the column and the more peaked the signal (see Fig. 2 as an example for ethanol, and Table 2 for all alcohols investigated). At higher temperatures, the equilibria responsible for the separation set in more quickly, so that diffusion processes no longer play a large role.

Tab. 2: Retention times of selected alcohols in dependence on the column temperature.

Column-temperature	Methanol	Ethanol	1-Propanol	2-Propanol
25 °C	5:41 min	17:40 min	73:31 min	28:54 min
45 °C	02:53 min	07:49 min	--	--
65 °C	02:45 min	05:48 min	14:45 min	06:31 min

Analysis of the purity of denatured ethanol

"Denatured" grade ethanol is denatured with a bitter substance and is also characterised by a lower purity. The purity of ethanol can be investigated using GC. For this purpose, a comparison is made with ethanol and methanol (see Fig. 4). Even though the retention times are not identical, it is clear that the denatured ethanol contains small amounts of methanol.

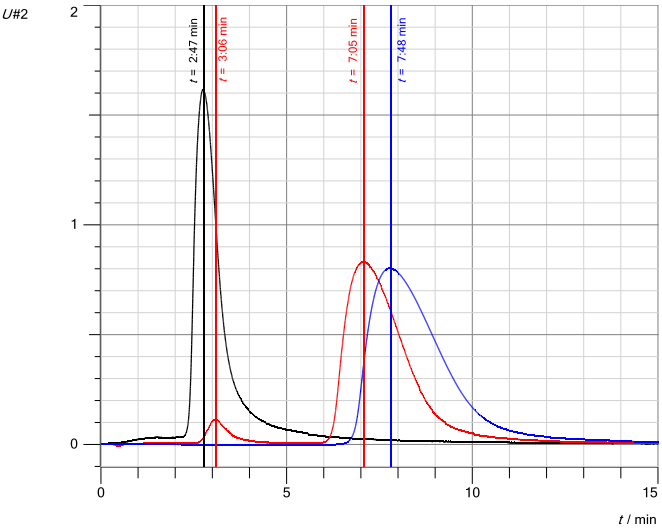

Fig. 4: Investigation of ethanol of "denatured" quality. Temperature: 45 °C. Red: denatured ethanol, black: methanol, blue: ethanol, pure.

Cleaning and disposal

After the last run, flush the column out with air for around 20 minutes to ensure that it is free from substances. Remove from the gas chromatograph and close the ends with the black caps. Store in a dry, dark place.