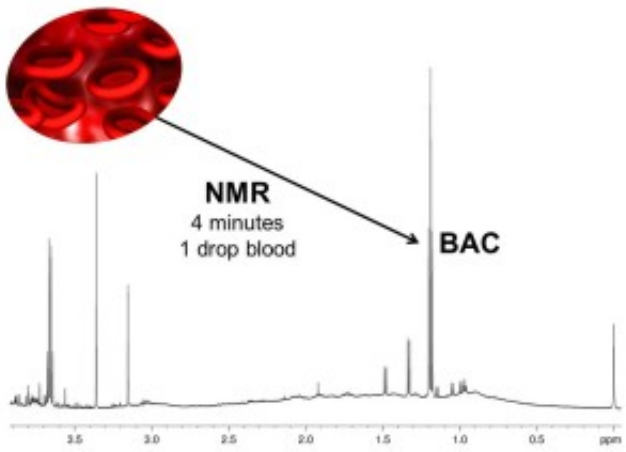
Alternative determination of blood alcohol concentration

Alcohol consumption is a common and legal practice in most of the countries around the world which leads to numerous fatal traffic accidents, and other incidents like leisure injuries, alcohol poisoning and drug facilitated sexual assault. According to official methods in individual countries, BAC is measured with different methods like the classical techniques: headspace gas chromatography (HS-GC) or alcohol dehydrogenase (ADH). Most of the conventional techniques are time-consuming, destructive and expensive due to the numerous calibration steps for each application. This leads to a demand of a precise and rapid methodology for the determination of BAC. In this study the proton nuclear magnetic resonance (^{1}H NMR) spectroscopy is presented as an alternative technique for the BAC determination with several goals: to improve the BAC analysis with regard to decreased sample volume, shorter analysis time, automated processing of large quantities of samples, a non-destructive testing and a possible follow-up examinations or DNA profile analysis after the ^{1}H NMR measurement by different biochemical techniques.



^{1}H NMR spectrum of drunken driver's blood

In the proposed method 20 µL of *whole blood* was taken as the sample material and diluted in D_2O. Only one drop of blood is adequate for the NMR analysis! The prepared blood sample was spectroscopically examined.

The current method was validated in accordance with the Society of Toxicological and Forensic Chemistry (GTFCh) Guidance and Bioanalytical Method Validation in the following terms:

> Specificity and Selectivity: the ability of the developed method to measure and differentiate ethanol (known as BAC) and the internal standard (IS) from other components in human blood.

Precision: the repeatability and reproducibility of BAC measurements obtained from multiple sampling and measuring.
Accuracy: the closeness of the measured concentrations of ethanol to the true concentrations.
Linearity: the ability to obtain NMR signals that are directly proportional to the concentration of ethanol in human blood.
Sensitivity: The lowest amount of ethanol in blood that can be identified (limit of detection, LOD) and quantified (limit of quantification, LOQ) with appropriate precision and accuracy.
Robustness: the capacity to remain unaffected test results by small variations in method parameters.

To show the suitability of the method, real driving under influence (DUI) blood samples were measured with the conventional methods ADH and HS-GC, served as the references and with the ^{1}H NMR spectroscopy, acted as the comparator. The official BAC, passed to the police and the Public Prosecutor's Office, was calculated by the mean value of the double determination by HS-GC and the double determination by ADH. A good correlation between the BAC analyzed by the conventional methods and ^{1}H NMR was suggested. The clinical study with blood samples showed that ^{1}H NMR results were comparable with these from HS-GC and ADH. Therefore, the NMR spectroscopy is a suitable method for a fast and accurate BAC determination.

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Publication

Alternative determination of blood alcohol concentration by (1)H NMR spectroscopy.
Zailer E, Diehl BW.
J Pharm Biomed Anal. 2016 Feb 5