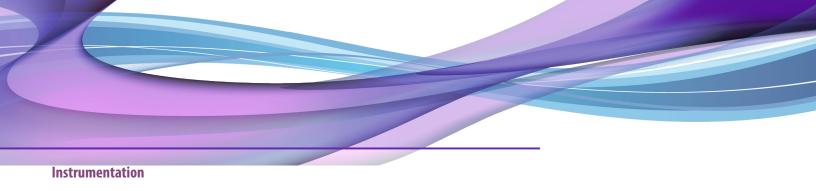


Introduction

The analysis of ethanol in a complex matrix such as blood is a quite difficult process in itself and the laboratories are therefore in the need of tools that can provide reliable, repeatable and fast analyses. The most widely used technique for analyzing the blood alcohol content in forensic laboratories is the headspace - gas chromatography (HS-GC).

The headspace extraction allows a better quantification, a reduction in sample preparation and an increased throughput. The Gas Chromatograph provides the greatest flexibility and reliability to analyze the ethanol content.

In this application note, the DANI HSS 86.50 Plus Static Headspace Sampler interfaced to a DANI Master GC Gas Chromatograph is used for the determination of ethanol in blood.



The analysis was performed by using the DANI HSS 86.50 Plus Static Headspace Sampler with the 44 seat sample tray and the DANI Master GC Gas Chromatograph equipped with a split/Splitless injector and dual FID detectors.

HSS 86.50 Plus		
Oven temperature	70° C	
Manifold temperature	90° C	
Transfer Line temperature	90° C	
Sample Volume	0.5 mL	
Vial Equilibration Time	15 min	
Shaking	FAST	

Table 1: HSS 86.50 Plus Analysis Conditions

Master GC	
Oven	30°C isothermal for 4 minutes
Injector	SL/IN at 150°C
Flow	8mL/min
Split flow	40 mL/min
FID	300°C
Retention Gap	10 cm x 0.53 mm
Columns	
Column1	DN-ALC1 30m x 0.32 mm x 1.8 μm
Column2	DN-ALC2 30m x 0.32 mm x 1.2 μm

Table 2: Master GC Analysis Conditions



RESULTS

Ethanol and Methanol were analyzed, confirmed by the dual columns chromatography and n-propanol was used as internal standard.

The linear response has been evaluated with three repetitions for each 10 concentration level, from 50 to 3000ppm.

DN-ALC1	Methanol	Ethanol
R ²	0.9995013	0.9996408
DN-ALC2	Methanol	Ethanol

Table 3: Linear Response



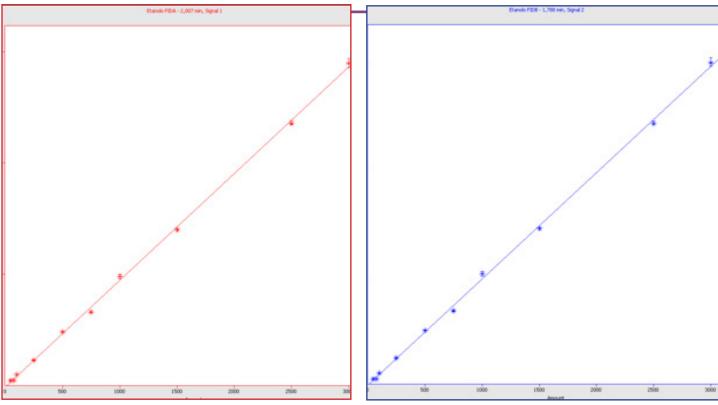


Figure 1: Calibration Curve of Ethanol (DN-ALC1)

Figure 2: Calibration Curve of Ethanol (DN-ALC2)

The repeatability has been evaluated on 10 consecutive injections obtaining the following results:

DN-ALC2	Methanol	Ethanol
RSD%	0.6%	1.3%
DN-ALC1	Methanol	Ethanol

Table 4: RSD% Response

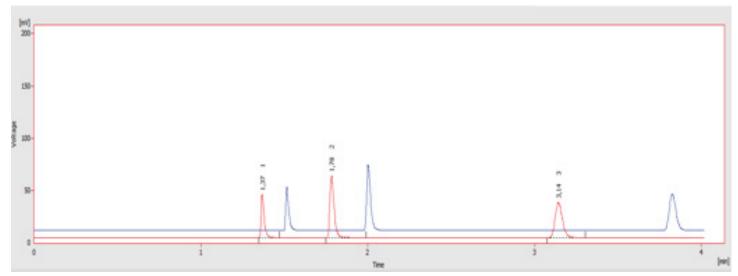
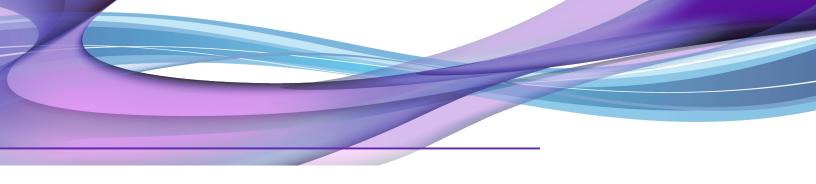


Figure 3 : Overlay Chromatogram of BAC Analysis.

Red Chromatogram DN-ALC 1 (Methanol-Ethanol-n-Propanol)

Blue Chromatogram DN-ALC 2 (Methanol-Ethanol-n-Propanol)



CONCLUSION

The DANI HSS 86.50 Plus coupled with the DANI Master GC Gas Chromatograph as tested permits BAC analysis offering a reliable and economical solution to the analysis of ethanol in biological fluids.

The DANI HSS 86.50 Plus needs a minimal operator experience due to complete automation of all process steps. Method parameter can be easily set up through the easy-to-use keypad.

The DANI DN-ALC1 and DN-ALC2 columns show excellent precision and reproducibility for the determination BAC concentration from a complex blood matrix.

The combination between dual columns configuration and internal standard analysis allows accurate qualiquantitative results.





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