Alcohol (Ethanol)
(ADH-UV-Test)

Cat.No.	Package Sizes
104 000	4x10 ml R1/ 4 x 10 ml R2 / Standard
104 016	4x10 ml R1/ 4 x 10 ml R2 / Standard
104 001	1x50 ml R1/ 1 x 50 ml R2 / Standard

METHOD
Enzymatic UV, Kinetic.

PRINCIPLE
Kinetic determination of ethanol, based on the ADH-reaction :
Alcohol is oxidized to Acetaldehyde, while NAD^+ is reduced to NADH. The increase of absorbance of NADH is a measure for the concentration of ethanol .

REAGENTS
Reagents Composition (concentrations in the test):
R1 (Buffer) TRIS-Buffer (pH 8.7) 200 µmol/L

R2 (Enzyme Reagent)
Alkoholdehydrogenase 140 U/L
NAD^+ 1500 µmol/L
Phosphate-Buffer (pH 7.1)

Standard:
Ethanol (see label of the standard vial)

Precautions
- For *in vitro* diagnostic use only.
- Use reagents only until the expiration date as printed on each vial label. Do not use expired reagents.
- Close each vial with its cap after use.
- Reagents contain sodium azide (0,95 g/L) as preservative. Do not swallow, and avoid contact with skin and/or mucous membranes.

Stability
When stored at 2-8° C and protected from light, the reagents are stable until the expiry date printed on the label.

Preparation and Stability of Working Reagents
R1 and R2 are ready for use
Stability after opening :
3 months at 2 - 8°C, when contamination is strictly avoided

SAMPLES
Serum free of hemolysis, heparinized or EDTA plasma, fresh urine
Stability at 2-8°c is minimum 5 days
Note: Ethanol is very volatile, therefore samples, calibrators and controls should be stored well capped and under strict refrigeration.

ASSAY PROCEDURE
The reagent can be used manually (see method below) and on most analyzers. Applications are available on request.

Wavelength : 340 nm
Temperature : 37° C
Cuvette : 1 cm light path
Measure against water (increasing absorbance A)

	STANDARD	SAMPLE
Standard	30 µL	-
Sample	-	30 µL
Reagent R1	250 µL	250 µL
Mix, incubate for 1 min, read A_1		
Reagent R2	250µL	250µL
Mix and incubate, after exactly 3 min read A_2		

Then calculate
$\Delta A = (A_2 - A_1)$ of Sample or Standard

CALCULATION

$$Alcohol\ [mg/dL] = \frac{\Delta A\ Sample}{\Delta A\ Std\ /\ Cal} \times conc.\ Std\ /\ Cal\ [mg/dL]$$

Note: You may also use a fixed factor :
Calculate once

$$F = Concentration_{Standard} / \Delta A_{standard}$$

QUALITY CONTROL
For quality control use Greiner's special Alcohol control .

Greiner Diagnostic GmbH - Unter Gereuth 10 **–** D-79353 Bahlingen – Germany
www.greiner-diagnostic.com

PERFORMANCE DATA

No interference by Bilirubin up to 30 mg/dl, Hemoglobin up to 800 mg/dl and lipemia up to 1000 mg/dl Triglycerides .

- **Linearity**

The test is linear up to a concentration of 600 mg/dl , at higher concentrations the samples have to be diluted e.g. 1+1 with phys. NaCl, the result has to be multiplied by 2.

- Sensitivity/Detection Limit

The lower detection limit is 10 mg/dl.

- Specifity

A number of structural related organic compounds were tested on cross reactivity in the assay .
Aceton, Acetaldehyd, Äthylenglykol, Isopropanol and Methanol, at a concentration of 2 mg/dl, had no effect on the results.

- Precision

Within Run n = 11	Average [mg/dl]	Standard-Deviation [mg/dl]	VC [%]
sample 1	48,6	1,3	2,7
sample 2	100	1,2	1,2
sample 3	290	1,9	0,65

Between Run n = 11	Average [mg/dl]	Standard-Deviation [mg/dl]	VC [%]
sample 1	50,7	2,3	4,5
sample 2	254	6,7	2,6

- Correlation

A comparative study has been performed between the Greiner method (y) and another commercial reagent (x) on 125 human serum samples. The parameters of linear regression are as follows:

y = 1,02 x + 2,05 [mg/dl] r = 0,982

LITERATURE

1. Heise HA. Concentration of alcohol in samples of blood and urine taken at the same time. J For Sci 12, 454 (1967)
2. Beutler HO: Ethanol, Bergmeyer HU, Methods of Enzymatic Analysis, Vol.VI, 3. ed New York: Academic Press, 1984 p 598
3. Mandatory Guidelines for Federal Workplace Drug Testing Program. National Institute on Drug Abuse Federal Register Vol. 53, No 69, pp 11970 (1988)
4. Ellenhorn MJ, and BG Barceloux: Medical Toxicology, New York, Elsevier Science Publ. Company Inc. 1988, pp 525 and 782

SYMBOLS USED

For *in vitro* diagnostic medical use

Batch Code


Use by


Temperature limitation

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