



Ethanol (Liquid Ready Reagents) (K-ETOHLQR) Procedure for ChemWell®-T Auto-Analyser

Requirements:

- Ethanol Assay Kit (Liquid Ready Reagents) (K-ETOHLQR) (provides ~ 600 assays).
- K-ETOHLQR ChemWell®-T assay file and the K-ETOHLQR % ChemWell®-T indices file.
- Use in association with the Ethanol Assay Kit (Liquid Ready Reagents) (K-ETOHLQR) product data booklet.

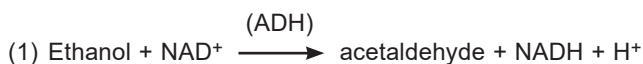
Use:

For the specific measurement of ethanol especially in wines, fruit juices, beverages and food products.

For specific sample preparation methods refer to the Ethanol Assay Kit (Liquid Ready Reagents) (K-ETOHLQR) data booklet.

Assay Principle:

Conversion of ethanol via the following reactions is directly proportional to the coupled formation of NADH:



Procedure:

Prepare the assay reagents, calibrators and use with the K-ETOHLQR ChemWell®-T assay file. To generate values as % (v/v) use the K-ETOHLQR % ChemWell®-T indices file.

Ethanol Assay Kit Components:

Bottle 1: Reagent 1 (120 mL)

Contains sodium azide (0.02% w/v) as a preservative. Ready to use.
Stable for > 1 year at 4°C.

Bottle 2: Reagent 2 (30 mL)

Contains sodium azide (0.02% w/v) as a preservative. Ready to use.
Stable for > 1 year at 4°C.

Bottle 3: Ethanol Standard (5 mL, 5 mg/mL)

Stable in a well-sealed container (as supplied)
for > 2 years at 4°C.

Preparation of Assay Reagents:

- 1 & 2.** Use the contents of bottles 1 & 2 as supplied.
Stable for > 1 year at 4°C.
- 3.** To prepare the K-ETOHLQR 4 calibrator (0.3 g/L) dilute 0.5 mL of the contents of bottle 3 to 16.6 mL with distilled water. Store in a well-sealed Duran® bottle. When diluted, this solution is stable for 2 days at 4°C.
Perform 2-fold serial dilutions of K-ETOHLQR 4 (0.3 g/L) in distilled water to prepare K-ETOHLQR 3 (0.15 g/L) and K-ETOHLQR 2 (0.075 g/L).

Calibrators:

- K-ETOHLQR 1: 0 g/L (use distilled water)
- K-ETOHLQR 2: 0.075 g/L ethanol
- K-ETOHLQR 3: 0.15 g/L ethanol
- K-ETOHLQR 4: 0.3 g/L ethanol

Assay Parameters:

- Assay volumes: Reagent 1: 0.200 mL
Sample: 0.010 mL
Reagent 2: 0.05 mL
- Calibrators: 0, 0.075, 0.15, 0.3 g/L ethanol
- Reaction time: ~ 7 min at 25°C or 37°C
- Wavelength: 340 nm
- Assay type: endpoint
- Reaction direction: increase
- Linearity: up to 0.3 g/L of ethanol

Example Method:

Pipette into cuvettes/wells	Sample
Reagent 1	0.20 mL
sample	0.01 mL
Mix, read the absorbances of the solutions (A_1) at 340 nm after 3 min and start the reactions by addition of:	
Reagent 2	0.05 mL
Mix and read the absorbance of the solutions (A_2) at 340 nm after 7 min.	

