



## Phosphatidylethanol (PEth) Overview

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Traditional alcohol detection has been conducted by either blood or breath analysis and used primarily for assessing impairment while driving. However, these tests are only acceptable for alcohol use within a few hours which made them unsuitable for abstinence monitoring, evaluating past alcohol abuse, or predicting future medical issues (i.e. cirrhosis). Traditional blood biological markers, gamma-glutamyltransferase (GGT) and carbohydrate deficient transferrin (CDT), can be monitored as early predictors of toxicity which may be attributed to alcohol abuse. But hepatic damage may have already occurred from years of alcohol abuse. This limitation led researchers to look for alcohol use biomarkers in the urine and blood for past use.

### Urine EtG/EtS

Ethyl Glucuronide (EtG) and Ethyl Sulfate (EtS) are alcohol metabolites which can detect alcohol use in the urine for up to 3 days post drinking and became a standard test for alcohol abstinence programs in professional monitoring programs. The EtG/EtS test is very sensitive, but the interpretation of a positive result does not reveal whether the alcohol positive is from a single drink 3-4 hours earlier or from a binge drinking episode 2-3 days earlier. The test is performed consistent with traditional urine drug testing and is screened using immunoassay and LC-MS/MS confirmation testing with multiple cutoffs available for enhanced sensitivity. However, since many products such as hand sanitizers, beverages, or foods contain alcohol, interpretation of low levels can be difficult. Standard cutoffs are typically established at 500 ng/mL to eliminate alternative sources other than beverage alcohol.

### Blood PEth

An additional test was needed to detect alcohol use within the past two or more weeks. Researchers identified phosphatidylethanol (PEth) as a blood biomarker as a clinical tool to identify past alcohol consumption. This test provides the best view into past alcohol use and has the ability to determine alcohol use patterns over several weeks, not just within 2-3 days as with EtG/EtS, or within hours as with blood/breath alcohol. PEth provides additional information which is needed to objectively evaluate past alcohol use as self-disclosure of alcohol consumption is vastly under-reported.

PEth is a phospholipid which is formed on the surface of red blood cells by a reaction caused by phospholipase D in the presence of alcohol and phosphatidylcholine. There are several homologues which contain the same glyceryophospholipid central chain with two side chains of long-chain carboxylic acids. PEth nomenclature for the primary molecule is identified as 16:0/18:1 which contains



palmitic acid (16 carbon atoms and no double bonds) and oleic acid (18 carbons and 1 double bond). A second common molecule is 16:0/18:2 which reflects the presence of two double bonds on the 18-carbon chain. It is these two homologues which are evaluated for past alcohol use and identified using LC-MS/MS instrumentation. PEth can be detected within 1-2 hours post drinking and peaks approximately 8 hours post ingestion. Once it is formed it slowly degrades over 12 days.

A standard drink contains 14 grams of alcohol, meaning one 12 oz beer (5% alcohol), one 5 oz glass of wine (12%), or one 1.5 oz of hard liquor (40% or 80 proof) are each one drink. Social drinking has been defined as  $\leq 60$  grams of alcohol per day (4.2 drinks) and heavy drinking as  $> 60$  grams per day.

As a measure for evaluating alcohol abstinence, professional monitoring programs have established the abstinence cutoff at 20 ng/mL for PEth 16:0/18:1. Other industries have not established cutoffs; however, a review of scientific literature is summarized into the following table.

<b>PEth Cutoff (ng/mL)</b>	<b>Alcohol Use Level</b>
$< 20$ ng/mL	Abstinence or low use
$\geq 20$ ng/mL	Low/occasional use
$\geq 80$ ng/mL	4 drinks per day
$\geq 221$ ng/mL	Chronic and excessive
$\geq 400$ ng/mL	Severe misuse
$\geq 700$ ng/mL	Severe misuse, DUI drivers

As with all tests, PEth has become a very useful to assess the potential for long term alcohol abuse, but it should be evaluated with other clinical tests and evaluations by professionals.

A minimal amount of blood is necessary for testing and is performed from blood collected in grey top tubes. Excess blood remaining from clinical testing can be used to perform this testing and a second blood tube is not required. Tests can be performed with as little as 0.1 mL of blood or from Dried Blood Spots (DBS).

For more information on testing, please contact your CRL sales representative.



## References

William Ulwelling, Kim Smith. The PEth Blood Test in the Security Environment: What it is; Why it is important; and Interpretative Guidelines. *J Forensic Sci.* November 2018, Vol 63, No. 6 Pages 1634-1640.

Guido Viel, et. al. Phosphatidylethanol in Blood as a Marker of Chronic Alcohol Use: A Systemic Review and Meta-Analysis. *Int J Mol Sci.* 2012, Vol 13, Pages 14788-14812.

Natalie Kummer, et.al. Quantification of Phosphatidylethanol 16:0/18:1, 18:1/18:1, and 16:0/16:0 in venous blood and venous and capillary dried blood spots from patients in alcohol withdrawal and control volunteers. *Anal Bioanal Chem.* 2016, Vol 48, Pages 825-838.

Ragnhild Bergene Skrastad, et al. Stability of Phosphatidylethanol 16:0/18:1 in Freshly Drawn, Authentic Samples from Healthy Volunteers. *J Anal Toxicol.* 2021, Vol 45, Pages 417-421.