PRODUCT INSTRUCTIONS

Q.E.D.®

SALIVA ALCOHOL TEST

A150

Product Number 31150

FOR THE QUANTITATIVE DETERMINATION OF ALCOHOL IN SALIVA FOR *IN VITRO* DIAGNOSTIC USE

INTENDED USE

The OraSure Technologies' Q.E.D.® Saliva Alcohol Test is intended for the rapid, accurate quantitative determination of alcohol in saliva.

These products are recommended for professional use in the evaluation of persons suspected of being intoxicated and as an aid in the management of alcoholism.

SUMMARY

Ethanol is the most common toxic substance encountered in medical cases. Not only is it lethal in its own right, but is commonly a contributory factor in accidents of all types. In the case of a patient brought to the hospital in a coma, the effect of alcohol, if any, must be ruled out in a differential diagnosis of the cause of coma.[1]

The distribution of alcohol in saliva and blood is well established with the concentration of ethanol in saliva being approximately 1.07 times higher than the corresponding blood alcohol level.[1,2] While there is good correlation between blood and saliva alcohol levels, normal physiological variability in these levels does exist. In situations where exact blood alcohol concentrations must be known, follow-up testing of patients with positive Q.E.D.® test readings should be performed. Appropriate follow-up tests include whole blood alcohol determinations using gas chromatography.

PRINCIPLE

The Q.E.D.® Saliva Alcohol Test uses alcohol dehydrogenase to catalyze the oxidation of ethanol to acetaldehyde, with the simultaneous reduction of nicotinamide adenine dinucleotide (NAD).[3] An alkaline pH and an acetaldehyde trapping agent force the reaction to generate one mole of NADH for each mole of alcohol present.

In the presence of an oxidizing agent (the Electron Sink), diaphorase and a tetrazolium salt, all of which are incorporated into the solid phase, NADH is oxidized and a colored end product is formed. The length of the resulting colored bar is directly proportional to the concentration of ethanol in the specimen.

Chemical Reaction Sequence

Fig. 1

$$\text{ETHANOL} + \text{NAD} \xrightarrow{\text{ALCOHOL DEHYDROGENASE}} \text{ACETALDEHYDE} + \text{NADH}$$

$$\text{ACETALDEHYDE} + \text{TRAPPING AGENT} \xrightarrow{\text{ALKALINE pH}} \text{COMPLEX}$$

$$\text{NADH} + \text{ELECTRON SINK (ES)} \xrightarrow{\text{DIAPHORASE}} \text{NAD} + \text{REDUCED (ES)}$$

$$\text{NADH} + \text{TS} \xrightarrow{\text{DIAPHORASE}} \text{NAD} + \text{FORMAZAN DYE}$$

TS = TETRAZOLIUM SALT

REAGENTS

Each Q.E.D.® Saliva Alcohol Test contains alcohol dehydrogenase, diaphorase, NAD, an oxidizing reagent and a tetrazolium salt, all of which are immobilized on a solid substrate.

PRECAUTIONS

1. For *in vitro* diagnostic use.
2. Patient specimens and used tests contain potentially infectious, human saliva; handle with appropriate care.
3. Because this test is visually read, it should not be interpreted by readers who are color-blind or visually impaired.

REAGENT PREPARATION AND STORAGE

Q.E.D.® Saliva Alcohol Tests are ready-to-use. No additional preparation is required. The storage and usage of Q.E.D.® tests at room temperature 15-30°C (59-86°F) is recommended.

An unopened Q.E.D.® test is stable until the date printed on the pouch when stored as recommended. Once the pouch has been opened, the Q.E.D.® test must be used immediately.

SPECIMEN COLLECTION

NOTE: Accurate alcohol level determination requires that samples be collected at least 10 minutes after eating or drinking anything-especially alcohol-containing substances.[4]

1. Open the foil pouch immediately prior to use and remove the Q.E.D.® Saliva Alcohol Test and desiccant packet.
2. Discard the desiccant packet. The central stripe of the device and QA Spot™ at the closed end of the Q.E.D.® device should not be purple. Discard any device in which these areas are purple.
3. Place the cotton-tipped end of the collector in the mouth and swab around the cheeks, gums, and under the tongue for 30-60 seconds or until cotton is thoroughly wet (Fig. 2).
4. Complete the remainder of the test at once.

CALIBRATION

The Q.E.D.® Saliva Alcohol Tests are precalibrated. No additional calibration is required.

TEST PROCEDURE

Materials Provided:

Q.E.D.® A150 Saliva Alcohol Test (Product Number 31150)
10 Ready-to-use Q.E.D.® A150 Tests (0-145 mg/dL) and Specimen Collection Swabs
1 Product Instructions

Materials Not Provided:

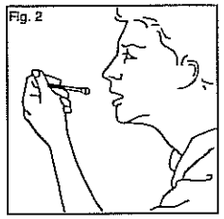
Quality Control Material: Q.E.D.® Ethanol Controls (Product Numbers 31050S, 31150S)
Timing device capable of accurately measuring 2, 5, and 10 minutes

Test Procedure:

Note: It is recommended that all tests be run at room temperature 15-30°C (59-86°F).

1. Open the foil pouch and remove the Q.E.D.® test immediately prior to use.
   Discard any test in which the desiccant pack indicator has turned pink.
   Discard any test in which the central stripe of the device or QA Spot™ is purple.

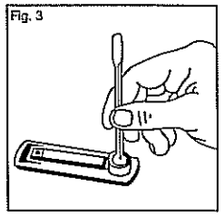
Fig. 2



2. Actively swab around the cheeks, gums and under the tongue for **30-60 seconds or until the cotton swab is thoroughly wet.**

   The cotton must be saturated.

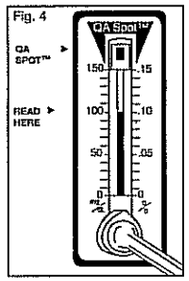
Fig. 3



3. Place the Q.E.D.® test on a flat surface. Insert the collector into the entry port; then apply gentle, steady pressure until the pink fluid flows past the QA Spot™ at the end of the device.

   With the filling method, the key is to apply gentle, steady pressure and to watch the capillary fill. The background color should appear pink after the capillary is filled.

Fig. 4



After the test is complete, examine the QA Spot™ located at the closed end of the device. The QA Spot™ should be purple. Any purple color across the QA Spot™ area is considered acceptable. Read the alcohol concentration from point on the scale where the purple bar stops. If bubbles appear in the device, read the alcohol concentration from the highest point on the scale where the purple bar stops.

The test is invalid if the QA Spot™ is not purple after 5 minutes.

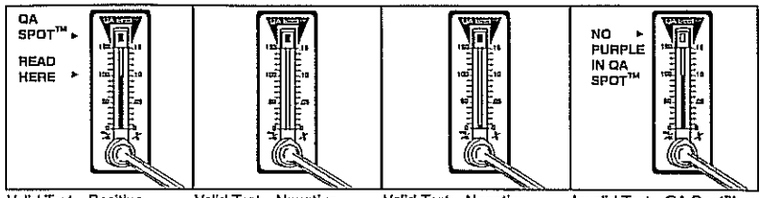
4. Do not pick up the Q.E.D.® device until the test is complete.
   The Q.E.D.® A150 test is complete in 2 minutes.

NOTE: Do not remove the swab after it has been inserted. Removal and reinsertion of the swab may cause bubbles to form in the device and may make the test result difficult to read.

IMPORTANT TEST PROCEDURE TIPS

1. Be sure to actively swab around the mouth until the cotton is **thoroughly** wet. The QA Spot™ will not turn purple unless activated by the sample. Repeat any test in which the QA Spot™ does not turn purple.
2. The red dye in the filter colors the saliva pink to aid in visualization of the capillary filling. The background color should appear pink after the capillary has been filled. The QA Spot™ will turn purple to indicate that the test procedure was performed correctly.

Interpreting Q.E.D.® A150 Test Results



Valid Test - Positive Reading: 100 mg/dL (.1%)	Valid Test - Negative Reading: 0 mg/dL (0%)	Valid Test - Negative Reading: Less than 10 mg/dL (.01%)	Invalid Test - QA Spot™ did not turn purple. Repeat Test.

QUALITY CONTROL

The functionality and stability of the Q.E.D.® test can be determined by examination of the QA Spot™ located at the closed end of the device. The QA Spot™ will turn purple **within 5 minutes** after the device has been completely filled with saliva. The purple color indicates that the device has been properly filled and that the chemical reagents contained in the device are fully functional.

Normal and Ethanol Controls

Quality Control materials are useful in determining assay reliability and performance. OraSure Technologies recommends that both normal and ethanol controls be run upon receipt of each new lot. Because ethanol is not normally present in the body in measurable quantities, normal saliva contains essentially no ethanol. Use distilled or deionized water for the normal control. Use the Q.E.D.® Ethanol Control for the positive control.

Substitute control materials for the patient sample. For ethanol controls, compare the Q.E.D.® test result to the range of expected values published in the OraSure Technologies' Q.E.D.® Ethanol Control product instructions.

Failure to achieve expected control values may be an indication of problems with user technique or assay reliability. Contact OraSure Technical Service if you require assistance 800-869-3538.

RESULTS

Test results are read directly from the Q.E.D.® test device designated by the end of the colored bar. The length of the bar is directly proportional to the concentration of ethanol in the sample. The Q.E.D.® test device has concentration scales in both mg/dL and %.

The Q.E.D.® Saliva Alcohol Test results are directly correlated to whole blood alcohol concentrations. There is no need to correct the Q.E.D.® test result to account for the 1.075:1 distribution between alcohol levels in saliva and blood.

Alcohol levels of less than 0.01% (10 mg/dL) should be reported as negative for alcohol.[4]

The Q.E.D.® Saliva Alcohol Test is a precise and accurate means of measuring whole blood alcohol levels through saliva alcohol determinations. However, in situations where exact blood alcohol concentrations must be known, follow-up testing of patients with positive Q.E.D.® readings should be performed. Appropriate follow-up testing includes whole blood alcohol determinations using gas chromatography.

LIMITATIONS

Accurate alcohol level determination requires that samples be collected at least 10 minutes after eating or drinking anything - especially alcohol-containing substances.

DYNAMIC RANGE

The Q.E.D.® A150 Saliva Alcohol Test is accurate from 10 - 145 mg/dL (0.01 - 0.145%).

Should quantitation of alcohol levels in excess of 0.145% (145 mg/dL) be necessary, another method should be employed.

EXPECTED VALUES

Alcohol is not normally present in saliva in measurable quantities, i.e., less than 10 mg/dL.[4]

PERFORMANCE CHARACTERISTICS[5]

Precision

Using the procedure outlined above, repetitive analysis with OraSure Technologies' Q.E.D.® Saliva Alcohol Test resulted in the following precision:

Q.E.D.® A150 Test

Within Run Level	Replicates	Mean mg/dL	S.D. mg/dL	C.V. %
Moderate	n=20	66.1	2.2	3.4
Elevated	n=20	107.6	4.0	3.7

Day-to-Day (5 day Period) Level	Replicates	Mean mg/dL	S.D. mg/dL	C.V. %
Moderate	n=20/day	65.6	2.1	3.2
Elevated	n=20/day	109.4	3.6	3.3

Accuracy

The accuracy of OraSure Technologies' Q.E.D.® product was evaluated by comparison to another enzymatic method. The Q.E.D.® test was also compared to gas chromatography using whole blood.[5]

A summary of the results of these comparisons appears below:

Q.E.D.® A150 Test

Method	Sample	Number	Correlation Coefficient	Regression Equation
GC	whole blood	n=168	r=0.98	y=1.03x - 2.4
enzymatic	saliva	n=71	r=0.98	y=0.94x - 2.0
enzymatic	serum*	n=54	r=0.97	y=0.83x + 5.0

*Serum alcohol levels are reported to be approximately 1.2 times (1/0.83) higher than blood levels.[1]
The correlation between saliva and blood alcohol levels is reported to be 0.96-0.97.[2,5]

Specificity

The following substances have been evaluated and do not interfere with the Q.E.D.® Saliva Alcohol Test at the concentration indicated.

Compound	Concentration (mg/dL)	Compound	Concentration(mg/dL)
Ethylene Glycol	17.5	1-Propanol	7
Acetone	70	2-Propanol	35
Methanol	70	1-Pentanol	7
1-Butanol	7	Ascorbic Acid	3.5
2-Butanol	35		

Using alcohol-containing products such as mouthwash, cough syrup, breath spray, or chewing tobacco may cause elevated results. Refer to limitations section.

REFERENCES

1. Tietz, R.W., Fundamentals of Clinical Chemistry, W.B. Saunders Company, Philadelphia, PA, 1976.
2. Jones, A.W., Inter- and Intra-Individual Variations in the Saliva/Blood Alcohol during Ethanol Metabolism in Man, J. Clinical Chemistry, (1979) 25, pp. 1394-1398.
3. Bergmeyer, H.U., Methods of Enzymatic Analysis, Academic Press, New York, (1965), p. 285.
4. Dubowski, K.M., Alcohol Analysis: Clinical Laboratory Aspects, Part II, Laboratory Management, April,1982, p. 27.
5. Data on file, OraSure Technologies, Inc., Bethlehem, PA.
6. Jones, A.W., Distribution of Ethanol Between Saliva and Blood in Man, Clinical, and Experimental Physiology, 6, (1979), pp. 53-59.

PRODUCT AVAILABILITY

Product #	Description
31150B	Q.E.D.® A150, case of 30 tests
31150C	Q.E.D.® A150, case of 100 tests
31050S	Q.E.D.® DoT Ethanol Control
31150S	Q.E.D.® A150 Ethanol Control
31000V	Q.E.D.® Alcohol Procedure Video
31150T	DoT STT Training Kit
31150X	DoT STT Extra Student Kits

The Q.E.D.® Test is a single use alcohol screening device to be administered by a certified Screening Test Technician or trained professional. IN NO EVENT SHALL ORASURE BE LIABLE FOR ANY DIRECT OR INDIRECT CONSEQUENTIAL, INCIDENTIAL OR SPECIAL DAMAGES.

TECHNICAL ASSISTANCE

For further information and technical assistance, contact OraSure Technical Service:**1-800-869-3538**

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