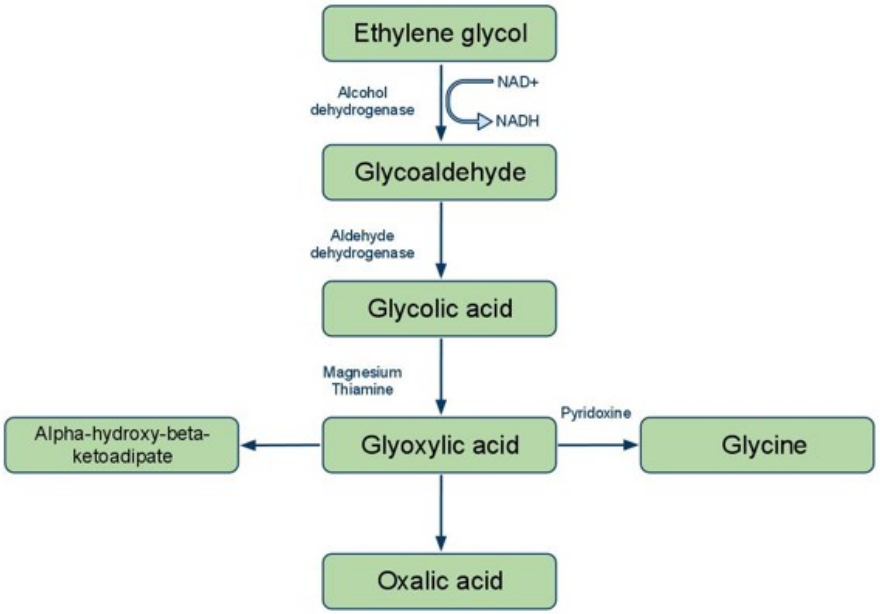
Ethylene Glycol pathway*



Reorder:

UPC	Product #	Description
656930303355	30335	Veti Stx EG 3-Stage Five Pack
65693o30336	30336	Veti Stx EG 3-Stage Ten Pack
656930303256	30325	Reusable 10uL Pipettor
656930303287	30328	Pipette Tips 50 count

KACEY® DIAGNOSTICS

1854 A Hendersonville Rd, Asheville, NC 28803
Ph: 1(828)685-3569 Fax: 1(828)685-3571
www.kaceydiagnostics.com

- Ethylene Glycol pathway chart courtesy of "Critical Care DVM"
https://criticalcaredvm.com/ethylene-glycol-intoxication-antifreeze-kills-pets/?print=print -
- Dr. Louis N. Gotthelf, DVM , Montgomery, AL

KACEY® DIAGNOSTICS

Veti-Stx Antifreeze

EG3 Stage Test

Three (3) Stage, Six Parameter Ethylene Glycol Test Instructions Sheet

KACEY® DIAGNOSTICS

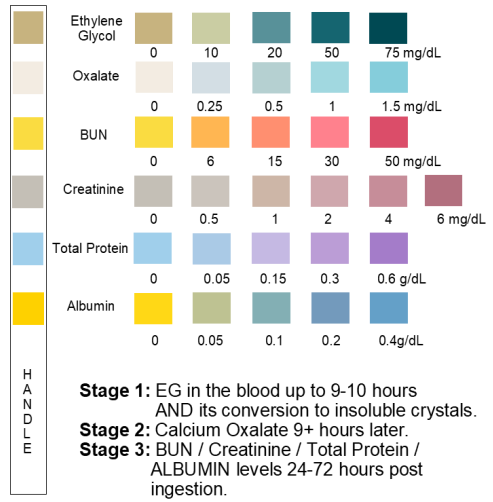
Three Stage Ethylene Glycol Test

Veti-Stx-EG3
6 Parameter E.G. test for Stages 1, 2 & 3

EG, OX, BUN, Creatinine, Total Protein, Albumin

For Invitro Use Only
MUST BE REFRIDGERATED
Veterinary Use Only

Test Parameters
Test Time: 10 minutes
Sample: Plasma or Serum
Sample Size: 10uL



Stage 1: EG in the blood up to 9-10 hours AND its conversion to insoluble crystals.
Stage 2: Calcium Oxalate 9+ hours later.
Stage 3: BUN / Creatinine / Total Protein / ALBUMIN levels 24-72 hours post ingestion.

Proudly made in the USA

1854A Hendersonville Rd, Suite 112.Asheville, NC 28803
PH 828-685-3569
www.KaceyDiagnostics.com

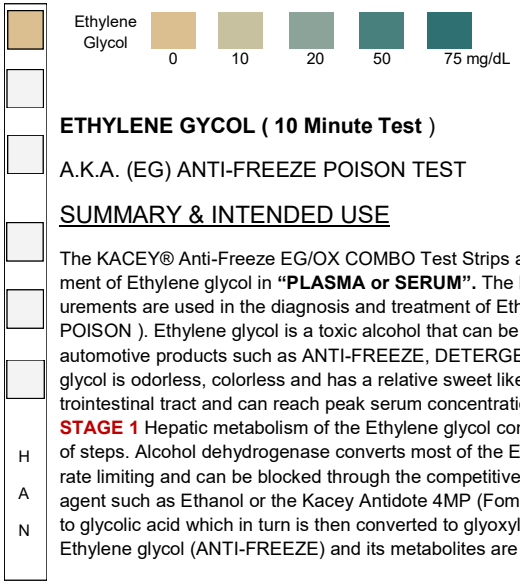
Rev 1 4.6.22

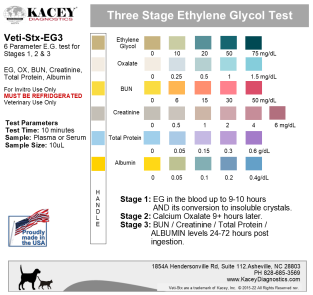
Veti-Stx EG-3 Ethylene Glycol Six Parameter 3 Stage Test Instructions

This multi parameter diagnostic strip is for use as a Veterinary diagnostic test in Ethylene Glycol poisoning. This test is a 3 Stage diagnostic test to better allow the clinician to determine where along the spectrum of EG toxicity the patient may be particularly when etiology and time of ingestion is unknown. This test covers various metrics in three stages to revel the progressive nature of EG in the system:

Stage1: EG in the blood up to 9-10 hours AND its conversion to insoluble crystals.
Stage 2: OX (Calcium Oxalate) 9+ hours post ingestion.
Stage 3: BUN / Creatinine / Total Protein / Albumin levels 24-72 hours post ingestion.

Stage 1 Ethylene Glycol





ETHYLENE GYCOL (10 Minute Test)

A.K.A. (EG) ANTI-FREEZE POISON TEST

SUMMARY & INTENDED USE

The KACEY® Anti-Freeze EG/OX COMBO Test Strips are intended to provide a quantitative measurement of Ethylene glycol in **"PLASMA or SERUM".** The KACEY® Anti-Freeze Combo Test Strips measurements are used in the diagnosis and treatment of Ethylene Glycol Poisoning (A.K.A. ANTI-FREEZE POISON). Ethylene glycol is a toxic alcohol that can be found in many commonly used household and automotive products such as ANTI-FREEZE, DETERGENTS, DEICERS and LACQUERS. Ethylene glycol is odorless, colorless and has a relative sweet like taste. It is absorbed very rapidly from the gastrointestinal tract and can reach peak serum concentration levels within one (1) hour after ingested. **STAGE 1** Hepatic metabolism of the Ethylene glycol converts the parent compound to Oxalate in a series of steps. Alcohol dehydrogenase converts most of the Ethylene glycol to Glycoaldehyde. This first step is rate limiting and can be blocked through the competitive inhibition of alcohol-dehydrogenase with an agent such as Ethanol or the Kacey Antidote 4MP (Fomepizole). **STAGE 2** Gylcoaldehyde is metabolized to glycolic acid which in turn is then converted to glyoxylic acid and finally to Oxalate. Unmetabolized Ethylene glycol (ANTI-FREEZE) and its metabolites are eliminated renally.

The KACEY® Anti-Freeze EG /OX Combo Test Strips measures the presence of both Ethylene glycol and Oxalate in a Plasma sample. A drop of Plasma (10uL) is placed onto each of the six the test pads on the test strip.. After waiting the required 10 minutes the color that forms in the test pad on the strip is compared to a color chart to determine both the presence and concentration of the Ethylene glycol and Oxalate in mg/dl. The Level of Detection (LOD) starts at 10 mg / dl and is Excellent in determining Ethylene Glycol Poisoning in both CATS & DOGS

CHEMICAL COMPOSITION

KACEY® Anti-Freeze Test Strips do not contain any harmful chemicals and is non-hazardous and non caustic

STORAGE & HANDLING (MUST BE REFRIDGERATED)

Store the EG/Ox Combo Vial in the refrigerator
Keep away from heat & direct sunlight
Always replace vial caps immediately after removing a test strip from the bottle.

Plasma or Serum SAMPLE IS REQUIRED – DO NOT USE HEMOLYZED PLASMA FOR THIS TEST

PROCEDURE

1) Place a minimum 300 - 750uL of a whole blood sample into a Green Top Lithium tube. Place the tube in a centrifuge and spin the tube as blood speed for 3 - 5 minutes to separate plasma from the red blood cells.

2) Remove the EG-3 test strip vial from the refrigerator and remove on test strip from the vial, immediately closing the vial and returning to the refrigerator.

3) Place one drop of plasma or serum (10uL) from the centrifuged tube on to each of the six test pads on the strip. 10uL pippetors are available from Kacey®.

4) Wait for 10 minutes. Remove vial with comparison chart from the refrigerator to compare the color of the upper most pad (E.G.) with the color chart on the vial label. (please be sure to place back in refrigerator)

5) DO NOT READ the color test strip after 10 minutes as the color will start to deteriorate and yield false results.

PRINCIPLE OF THE TEST

If Ethylene glycol is present a BLUE GREEN (TEAL) COLOR will appear on the "EG pad. See page 7 for different values pictured at the end of the **Stage 1** Ethylene Glycol (EG) and **Stage 2** Oxalate (OX) section of this insert. These two Stages are grouped together as they overlap on their significance in a toxicity and reading the combination of results may indicate severity of poisoning.. Remember that if no distinct color shift appears past the first ref chart color labeled "Neg" the result is negative for that component.

Caution: The intensity of the color is directly proportional to the concentration of the ethylene glycol. Propylene glycol, glycerol, ethanol, medication products containing alcohol, and synthetic sugars for baking (Ex. Splenda, maniotbol) will give false positive results.

DO TO THE PHENOMENA OF WICKING, DARKER COLORS MAY APPEAR AROUND THE EDGES OF THE TEST PAD. LOOK FOR UNIFORMITY OF COLOR IN THE CENTER OF EACH PAD.

SPECIMEN COLLECTION AND PREPARATION

Collect enough whole blood (with GREEN TOP LITHIUM TUBE) to perform the test.

Blood can be collected from the animal by a direct blood draw using a needle.

PLASMA can be obtained by centrifugation to separate the red blood cells from the PLASMA.

THE TEST MUST BE PERFORMED ONLY ON A PLASMA SAMPLE

QUALITY CONTROL

Good laboratory Procedures for QC (Quality Control) should be followed: EX. ALL Pos. (+) & Neg (-) results should be confirmed by running both Positive & Negative Controls for verification that the test result is indeed positive or negative. KACEY® provides Ethylene Glycol Controls: 0, 10 and 50 mg/dL. These Optional Controls are available separately and can be ordered directly from Kacey®. The user can practice the testing with these controls to get familiar with the test procedure and colors for negative and positive results. . Kacey highly recommends that the tester should always verify all positive / negative results of the patient by comparing the color results with that of the Bi-Level control as a 2nd source of validating the (+) &(-) test results.

PRECAUTIONS

*For in vitro diagnostic use only.
*Out of date or expired strips should never be used to perform a test. Check the vial for the expiration.

*If the sample pad is covered with Plasma, sufficient sample has been applied.
*If an insufficient amount of Plasma is placed on the strip, do not add Plasma to the same strip. Use a new unused test strip.
*Do not agitate the strip after the plasma sample is added.

TEST RESULTS

Results are displayed in "milligrams per deciliter" of ethylene glycol per millgrams per deciliter (mg/dl) and can be converted to (µg/ml). Therefore, a 20 mg /dl is equal to 200 µg / ml.

A FRAGMENTED **BROWNISH-/YELLOWISH COLOR** OR ANY FRAGMENTED PARTS IN THE PAD IS CONSIDERED NEGATIVE**.**

LIMITATION OF THE PROCEDURE

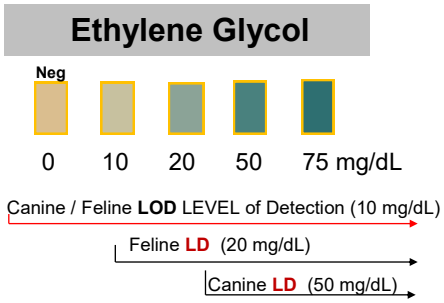
Ethanol interferes with the test and will give False Positive ethylene glycol test results.

Toxiban will interfere and can cause False Positives unless test is performed before Toxiban is administered.

All alcohol or compounds that can be converted to alcohol within the body will result in False Positives. This also include but is not limited to artificial sugars used in baking and cooking.

EXPECTED VALUES - LOD = Level of Detection LD=LETHAL DOSAGE LD FOR A CAT IS 20 mg/dL LD FOR A DOG IS 50 mg/dL

Measuring range: The Kacey® antifreeze test can measure ethylene glycol values starting from 10 mg /dl which is equivalent to 100 µg /ml and above This LOD is low enough to detect Ethylene Glycol Poisoning in Cats and is excellent in early detection of Ethylene Glycol Poisoning in small , medium and large size dogs. The color chart enclosed provides four (5) color blocks in order to measure the ethylene glycol results. Negative, 10 mg/dL, 20 mg /dl, 50 mg /dl, and 75 mg /dl. Cats showing a color greater then or equal to the 10mg /dl color block is positive, dogs showing a result in which the color is greater than or equal to the 10 mg /dl color block would be considered positive. (Note: it is possible for a dog to have a value greater than 10 mg/dl and less than 50 mg/dl. This would suggest that ethylene glycol had been ingested and should be viewed as an ethylene glycol poisoning even if it is not considered to be at a lethal dosage level above the 50 mg /dl threshold mark.



Note: It is "Estimated" that the amount of ½ tsp of anti-freeze (Conc. 95% +) per 1 pound of canine would be a lethal dosage of antifreeze for the average healthy canine. It has been reported that the current recommended standard for a lethal dose of antifreeze (Conc. 95% +) is at a threshold level of 50 mg/dl (Equivalent to 500 µg/ml) and above. The size, weight, age, metabolism, and health condition of the animal plays a major role on the assimilation and toxic response to the amount antifreeze ingested.

Estimated detection @10mg/dL is approximately 30-45 minutes after ingestion with a window of opportunity up to 9-10 hours: See Note above in the section for Expected values

ACCURACY

The ANTIFREEZE POISON TEST STRIPS were used against a commercially available test kit, Ethylene Glycol Test Kit by Allelic Biosystems, confirmed by spiked SERUM samples with known E.G. concentrations.

NOTE: IT IS RECOMMENDED AS A GOOD LABORATORY TESTING PROCEDURE TO RUN CONTROLS ON THIS ETHYLENE GLYCOL TEST TO CONFIRM AND OR VALIDATE ALL POSITIVE AND NEGATIVE RESULTS. THE KACEY COMPANY PROVIDES BI-LEVEL CONTROLS WHICH CAN BE PURCHASED TO VALIDATE THE RESULTS OF THE EG TEST. TO ORDER EG BI-LEVEL CONTROLS PART # 30304 EMIAL YOUR ORDER TO: sales@kaceyinc.org

THE KACEY® ETHYLENE GLYCOL (ANTIFREEZE) POSION TEST STRIPS COMPARES WELL TO THE REFERENCE METHOD.

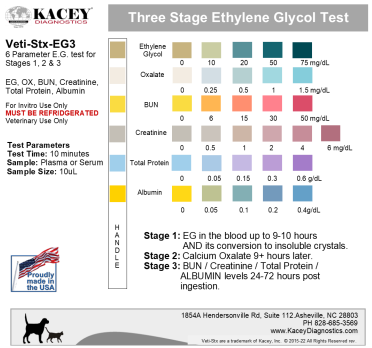
RE-ORDER PART# 30335 (FIVE TESTS PER KIT) or 30336 (TEN TESTS PER KIT)

PART# 30325 -10 µL PIPETTOR & PART# 30328 (50 –PIPET TIPS)

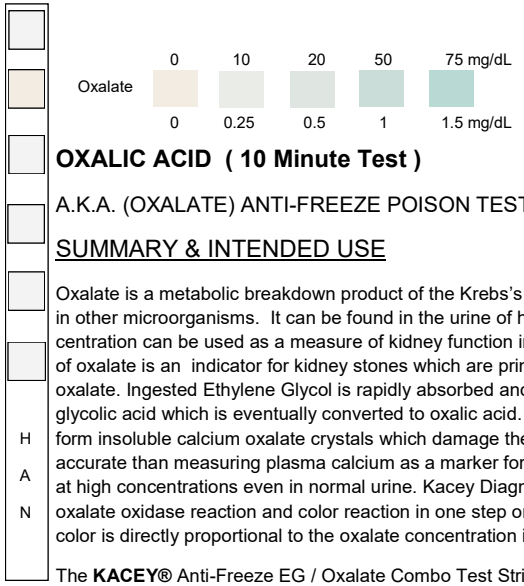
1. Data on file, Kacey® Asheville, N.C. 28801

2. Clinical Diagnostics and Management by Laboratory Methods, eighteenth Edition, John Bernard Henry -Editor. W.B. Saunders Company, Philadelphia 991

3.Young, D.L. et. Al, Effects of Drugs on Clinical Laboratory tests, AACC Press Wash., D.C. 1990.
Rev 5 –JUL 2015



Stage 2 Oxalate (OX)



OXALIC ACID (10 Minute Test)

A.K.A. (OXALATE) ANTI-FREEZE POISON TEST

SUMMARY & INTENDED USE

Oxalate is a metabolic breakdown product of the Krebs's Cycle in eukaryotes, and the glyoxylate cycle in other microorganisms. It can be found in the urine of humans and other mammals. Oxalate concentration can be used as a measure of kidney function in chronic kidney disease where a high level of oxalate is an indicator for kidney stones which are primarily made of the insoluble salt calcium oxalate. Ingested Ethylene Glycol is rapidly absorbed and metabolized in the liver over 8-10 hours to glycolic acid which is eventually converted to oxalic acid. Oxalic acid then combines with calcium to form insoluble calcium oxalate crystals which damage the kidneys. Measuring plasma oxalate is more accurate than measuring plasma calcium as a marker for kidney stones because calcium is excreted at high concentrations even in normal urine. Kacey Diagnostics' Oxalate Test Strips combine the oxalate oxidase reaction and color reaction in one step on a plasma sample. The intensity of product color is directly proportional to the oxalate concentration in the sample.

The **KACEY®** Anti-Freeze EG / Oxalate Combo Test Strips are intended to provide a quantitative measurement of Oxalate in "PLASMA" 8-10 hours post ingestion. The **KACEY®** Anti-Freeze Oxalate Test Strip measurements are used in the diagnosis and treatment of Ethylene Glycol Poisoning (A.K.A. ANTI-FREEZE POISON). Ethylene glycol is a toxic alcohol that can be found in many commonly used household and automotive products such as ANTI-FREEZE, DETERGENTS, DEICERS and LACQUERS. Ethylene glycol is odorless, colorless and has a relatively sweet like taste, but even when a bitterant is added it seems to attract animals who invariably ingest it. It is absorbed very rapidly from the gastrointestinal tract and can reach peak serum concentration levels within hours of ingestion. Hepatic metabolism of the Ethylene glycol converts the parent compound, Glycolic acid to Oxalate in a series of steps. During this process there is a window of opportunity from 0-8/10 hours in which the Kacey Ethylene Glycol test (EGT) strips can determine exposure. However, after this time depending on weight, species and amount of ingestion, Ethylene Glycol converts to an Oxalate and the Kacey EGT strips may indicate a false negative on exposure.

The Kacey Oxalate test will effectively measure the presence of oxalate in the secondary phase when the glycolic acid has been converted to oxalate. Knowing if there is oxalate present or not with a positive Ethylene Glycol test allows for the correct detoxification modality to be employed. (SEE CHART BELOW) Often pets are not presented until many hours post ingestion and this second phase test from Kacey becomes critical. The detox modality for each of these two phases is different and therefore it is critical that if possible both tests be administered concurrently to determine exposure time. Alcohol dehydrogenase converts most of the Ethylene glycol to Glycol aldehyde. This first step is rate limiting and can be blocked through the competitive inhibition of alcohol-dehydrogenase with an agent the Kacey Antidote 4MP (Fomepizole) however, after the 8-10 hour mark, Glycol aldehyde is metabolized to glycolic acid which in turn is then converted to glyoxylic acid and finally to an Oxalate. This second phase **(stage 2)** when combined with calcium produces Calcium Oxalate crystals which precipitate in the kidneys leading to severe renal tubular damage and potential acute renal failure.

TEST RESULT POSSIBILITIES FOR PLASMA ETHYLENE GLYCOL TEST (EGT) AND PLASMA OXALATE TEST

EGT NEGATIVE	OXALATE NEGATIVE	NO EXPOSURE	
EGT POSITIVE	OXALATE NEGATIVE	EARLY EXPOSURE	4MP
EGT POSITIVE	OXALATE POSITIVE	LATER EXPOSURE	4MP and IV fluids
EGT NEGATIVE	OXALATE POSITIVE	POOR PROGNOSIS	IV Fluids(4MP ineffective at this stage)

Ethylene glycol toxicosis causes acute renal failure!! Fluid diuresis and correction of acidosis are important parts of therapy. Prognosis is good with early aggressive treatment (<8 hours of ingestion) with 4MP, but poor or grave with prolonged untreated exposure. The total volume of intravenous fluids needed can be calculated using the equation: Maintenance (40 - 60 ml/kg/day) plus Deficit in L (% dehydration * kg body weight) plus insensible losses from vomiting and/or diarrhea. Deficit amounts can be infused rapidly, but so that fluids have time to equilibrate between the vascular, interstitial, and cellular compartments, maintenance fluids must be divided and administered over 8–12 hours. A balanced electrolyte solution can be used (LRS, Normosol); however, if hyperkalemia exists, 0.9% saline should be substituted. Due to the reduced ability of the kidneys to regulate fluids, Ethylene Glycol renal failure patients are at a high risk for overload and death. If oliguric renal failure occurs, Furosemide, a loop diuretic, has experimentally been shown to increase urine production, dislodge tubular obstructions, and induce vasodilatation. Often it is given as a bolus (2–4 mg/kg), and if urine production increases, a CRI is started (0.25–1.0 mg/kg/h). An indwelling urinary catheter should be aseptically placed to measure urine output. Hourly urine output is then used to determine "maintenance" fluid rate. Metabolic Acidosis occurs about 12 hours after EG ingestion and lasts up to 24 hours. Severe metabolic acidosis leads to compensatory respiratory alkalosis. Laboratory abnormalities include increased anion gap, decreased serum bicarbonate, and hyperglycemia. Generally, it is not treated unless serum bicarbonate is <14 mEq/L or pH <7.2. A conservative bicarbonate dose can be calculated: (bodyweight in kg x 0.3) x (16 - measured bicarbonate mEq/L). One-third this dose is given over 30 minutes while another one-third of the dose can be given with intravenous fluids every 4–6 h.

The KACEY® Oxalate test in the Anti-Freeze Combo Test Strips measures the presence of Oxalate in a PLASMA sample after the 8-10 hour mark depending on species, weight and quantity of ingested material. A drop of plasma is placed on the strip pad. After waiting the required 10 minutes the color that forms on the pad of the strip is compared to a color chart to determine both the presence and concentration of an Oxalate in mg/dl Level of Detection (LOD) starts at 0.25 mg / dl (see Expected Value Section Below) Kacey® Oxalate test is Excellent in determining both exposure to Ethylene Glycol Poisoning and respective change to an Oxalate in both CATS & DOGS.

CHEMICAL COMPOSITION

KACEY® Oxalate Anti-Freeze Test Strips do not contain any harmful chemicals and is non-hazardous and non caustic.

STORAGE & HANDLING (must be refrigerated at 4 C or 39.2 F)

- Store in the Refrigerated (Do Not Freeze)
- Keep away from heat & direct sunlight
- Always replace vial caps immediately after removing a test strip from the bottle.

A PLASMA SAMPLE IS REQUIRED – DO NOT USE SERUM OR HEMOLYZED SERUM FOR THIS TEST

PRINCIPLE OF THE TEST

If Oxalate is present a LIGHT GREEN TO TEAL COLOR will appear on the pad of the test strip. Match that color in the test strip to one of the color blocks on the color chart on the bottle. If NO DISTINCT LIGHT GREEN TO TEAL COLOR appears THROUGHOUT THE ENTIRE PAD, the test is negative (FRAGMENTS -SEE TEST RESULT SECTION). The intensity of the color is directly proportional to the concentration of the Oxalate.

SPECIMEN COLLECTION AND PREPARATION

Collect enough whole blood (with GREEN TOP TUBE LITHIUM HEPARIN Anticoagulant) to perform the test.

Blood can be collected from the animal by a direct blood draw using a needle.

Plasma can be obtained by centrifugation to separate the red blood cells from the plasma.

PROCEDURE

- PLACE A MINIMUM 300 – 750 µL OF A WHOLE BLOOD SAMPLE INTO A GREEN TOP LITHIUM HEPARIN TUBE
- PLACE THE HEAPARIN TUBE IN A CENTRIFUGE AND SPIN THE TUBE AT BLOOD SPEED FOR 3-5 MINUTES TO SEPARATE PLASMA FROM THE RED BLOOD CELLS.
- PLACE THE COMBO TEST STRIP (EG&OX) TEST STRIP ON A FLAT SURFACE OR PAPER TOWEL.
- INNOCULATE WITH ONE DROP OF PLASMA FROM THE ABOVE CENTRIFUGED TUBE USING THE KACEY 10uL PIPETTOR (PART #30329) ONTO EACH OF THE TWO (2) TEST PADS ON THE TEST STRIP. ENCOMPASS THE ENTIRE SQUARE PAD WITH THE PLASMA. THE 10µL PIPETTOR AND PIPET TIPS ARE AVAILABLE FROM KACEY ®.
- WAIT FOR 10 MINUTES AND COMPARE THE COLOR ON EACH OF THE TWO PADS OF THE TEST STRIP TO THE COLOR CHART PROVIDED ON THE BOTTLE. (see illustrations on back page) DO NOT READ THE COLOR STRIP AFTER 10 MINUTES AS THE COLOR WILL START TO DETERIORATE AND YIELD ERRONIOUS RESULTS.

TEST RESULTS

Results are displayed in “milligrams per deciliter” of ethylene glycol per milliliter (mg/dl) and can be converted to (µg/ml). Therefore, a 20 mg /dl is equal to 200 µg / ml. A FRAGMENTED BROWNISH-/YELLOWISH COLOR CONSIDERED NEGATIVE. A DARKER COLOR GREEN MAY APPEAR AROUND THE OUTER EDGES BUT THIS IS ATTRRIBUTED TO WHAT WE CALL THE PHENOMENA OF A WICKING EFFECT WHERE A LIQUID WILL TRY TO SEEP TO THE OUTER EDGES OF THE PAD. . A TRUE TOTAL BLUE /GREEN COLOR MUST ENCOMPASS THE ENTIRE SQUARE FOR A EG POSITIVE.

EXPECTED VALUES LETHAL OXALATE VALUE FOR A CAT IS 0.25 mg/dL AND 0.25 mg/dL VALUE FOR A DOG

Measuring range: The Kacey® antifreeze Combo test can measure **OXALATE .0.25** mg /dl which is equivalent to 2.5 µg /ml and above. This LOD is also low enough to detect Ethylene Glycol Poisoning in Cats and is excellent in DETERMINING POST 8-10 HOUR POISONING OF of Ethylene Glycol in small, medium and large size dogs. The color chart enclosed provides **five (5)** color blocks in order to measure the OXALATE glycol results. Negative (0), 0.25 mg/dL, 0.5 mg /dL, 1.0 mg /dL, and 1.5 mg /dL. Cats & DOGS showing a color greater than or equal to the **0.25 mg /dl** color block is positive FOR EXPOSURE TO BE GREATER THEN 8-10 HOURS

QUALITY CONTROL

Good laboratory Procedures for QC (Quality Control) should be followed: EX. ALL Pos. (+) & Neg (-) results should be confirmed by running both Positive & Negative Controls for verification that the test result is indeed positive or negative. KACEY® provides Oxalate Controls: 0, 20 and 80 µg/ml. These Optional Controls are available separately and can be ordered directly from Kacey®. The user can practice the testing with these controls to get familiar with the test procedure and colors for negative and positive results. Kacey highly recommends that the tester should always verify all positive/ negative results of the patient by comparing the color results with that of the Bi-Level control as a 2nd source of validating the (+) & (-) test results.

PRECAUTIONS

*For in vitro diagnostic use only.

*Out of date or expired strips should never be used to perform a test. Check the vial for the expiration date.

*If the sample circle is covered with Plasma, sufficient sample has been applied.

*If an insufficient amount of Plasma is placed on the strip, do not add Plasma to the same strip. Use a new unused test strip.

*Do not agitate the strip after the plasma sample is added.

CONTROLS

It is recommended as a good laboratory testing procedure to run controls on both the EG & Oxalate test to confirm and/or validate all positive and negative results. Kacey Diagnostics provides by-level controls which can be purchased to validate the results of the EG & Oxalate Test. To order contact : sales@kaceyinc.org
Ethylene Glycol , Oxalate Control Bi-Level control part # 30346

LIMITATION OF THE PROCEDURE

Propylene glycol, glycerol, ethanol, medication products containing alcohol, and synthetic sugars for baking (Ex. Splenda, mannitol) will give false positive results. All alcohol or compounds that can be converted into alcohol within the body will results in False positives. This also can include but NOT limited to artificial sugars used in baking or cooking. Toxiban ® will interfere and cause False Positives unless test is performed before Toxiban is administered.

Note: It is "Estimated" that the amount of ½ tsp of anti-freeze (Conc. 95% +) per 1 pound of canine would be a lethal dosage of antifreeze for the average healthy canine. It has been reported that the current recommended standard for a lethal dose of antifreeze (Conc. 95% +) is at a threshold level of 50 mg/dl (Equivalent to 500 µg/ml) and above. The size, weight, age, metabolism, and health condition of the animal plays a major role on the assimilation and toxic response to the amount antifreeze ingested. Estimated LEVEL OF Detection (LOD) of Oxalate 0.25 mg/dL l Or 2.5 µg/ml is approximately 8-10 hrs after ingestion: See Note above

ACCURACY

The ANTIFREEZE POISON TEST STRIPS were used against a commercially available test kit, Ethylene Glycol Test Kit by Allelic Biosystems, confirmed by spiked PLASMA samples with known ethylene glycol concentrations. The Kacey ® Ethylene Glycol Poison Test for Oxalate compares well to the reference method.

1. Data on file, Kacey® Asheville, N.C. 28801
2. Clinical Diagnostics and Management by Laboratory Methods, eighteenth Edition,

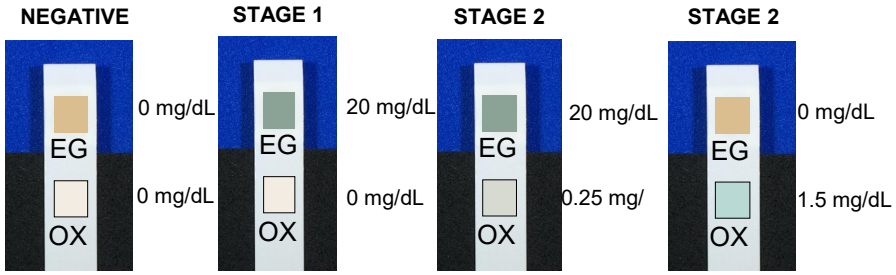
   John Bernard Henry -

   Editor. W.B. Saunders Company, Philadelphia 991

3. Young, D.L. et. Al, Effects of Drugs on Clinical Laboratory tests, AACC Press Wash., D.C. 1990. Rev 5 –JUL 2015

CORRELATION BETWEEN ETHYLENE GLYCOL AND OXALATE

Shown is the new combination EG and OX on one test strip which accurately tracks the progression of Ethylene Glycol through the two stages after exposure. As you can see in end Stage 2 Ethylene Glycol detection drops to zero and Oxalate levels signifying a poor prognosis necessitating the use of Oxalate detection.



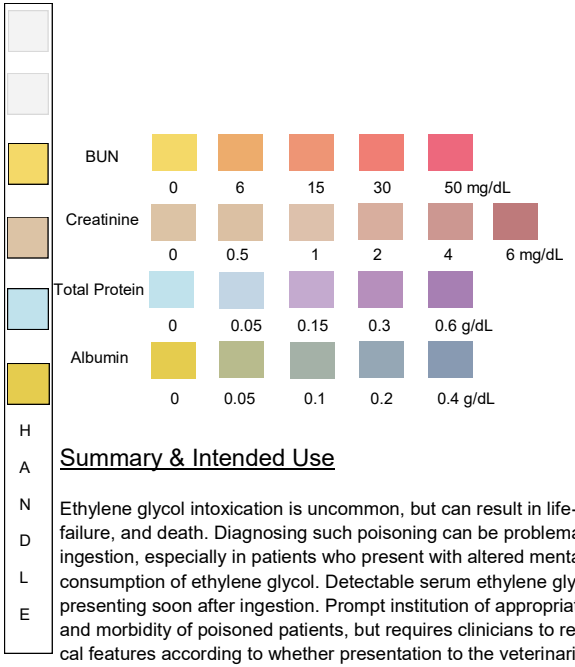
EG Neg, OX Neg
No Exposure.

EG Pos, OX Neg
Early exposure.
Use Fomepizole (4MP)

EG Pos, OX Pos
Later exposure.
Use Fomepizole (4MP) and fluid therapy

EG Neg, OX Pos
Poor prognosis.
Use IV fluids (4MP is ineffective at this stage

Stage 3: BUN, Creatinine, Total Protein, Albumin



Summary & Intended Use

Ethylene glycol intoxication is uncommon, but can result in life-threatening metabolic acidosis, kidney failure, and death. Diagnosing such poisoning can be problematic in the absence of a clear history of ingestion, especially in patients who present with altered mental status or those whose owners deny consumption of ethylene glycol. Detectable serum ethylene glycol level is characteristic in patients presenting soon after ingestion. Prompt institution of appropriate treatment can reduce the mortality and morbidity of poisoned patients, but requires clinicians to recognize these characteristic biochemical features according to whether presentation to the veterinarian is early or late after ethylene glycol ingestion. In cases of unexplained kidney failure, kidney biopsy may prompt the diagnosis by showing intratubular and intracellular calcium oxalate crystal deposition.

In this Third Stage of EG toxicity there are four key chemistries that determine this stage, prospects for survival, and course of treatment. In general metrics for these four chemistries may indicate a third stage toxicity or "Renal Stage":

Increased BUN

Increased Creatinine

Decreased Total Protein

Decreased Serum Albumin

This Third Stage is covered by the VetiStx EG-3 with results in 10 minutes. When compared to the metrics for EG and Oxalate a better overall picture of post ingestion can be seen. Additional metrics can help further determine toxicity:

Increased Lactate

Decreased pH

Decreased Bicarbonate

Increased Anion Gap

Further indicators can be derived from urinary abnormalities:

Low specific gravity

Proteinuria

Microhematuria

Pyuria

Chemical Composition

KACEY® BUN, Creatinine, T Protein and Albumin Test Strips do not contain any harmful chemicals and are non-hazardous and non caustic

Storage & Handling

- Store in the Refrigerated (Do Not Freeze)
- Keep away from heat & direct sunlight
- Always replace vial caps immediately after removing a test strip from the bottle .

A PLASMA SAMPLE IS REQUIRED – DO NOT USE SERUM OR HEMOLYZED SERUM FOR THIS TEST

Principle of the Test

The mechanism of acute kidney Injury (AKI) in EG toxicity is believed to be caused by glycolate-induced tubular cell necrosis with minimal glomerular damage. The characteristic histopathologic feature of ethylene glycol poisoning is the presence of intratubular and intracellular calcium oxalate crystals, with degeneration of tubular epithelium. Recent studies in rat models showed the accumulation of oxalate, the final metabolite product of hepatic ethylene glycol oxidation, in the form of calcium oxalate monohydrate crystals that were internalized by the proximal tubular cells, producing mitochondrial damage that resulted in cell death and tubular necrosis.

The use of the VetiStx EG –3 cover all three stages with the last being a grouping of chemistries whose values when taken will indicate how far along the patient is on the spectrum of EG toxicity. This 10 minute test of plasma is a comprehensive diagnostic too as it relates to potentially end stage EG poisoning via systemic organ failure especially of the renal system.

Specimen Collection and Preparation

Collect enough whole blood and place in a GREEN TOP LITHIUM TUBE or SSP TUBE.

Blood can be collected from the animal by a direct blood draw using a needle.

Plasma can be obtained by centrifugation to separate the red blood cells from the plasma.

Procedure

- Place a minimum of 300 - 750 uL of whole blood into either a Green top Lithium Heparin tube or an SSP tube.
- Place the tube in a centrifuge and spin at blood speed for 3-5 minutes to separate plasma from the red blood cells. Place the VetiStx EG-3 strip on a flat surface or paper towel. .
- Inoculate with one drop of Plasma from the centrifuged tube using the Kacey ® 10 uL pipette tips(part # 30328) and pipettor (part # 30325) on to each of the pads on the test strip. One drop per pad. .
- Wait for 10 minutes and compare the color of each tests to the color chart on the vial. Do not read the color strip after 10 minuets as the color will start to deteriorate and yield erroneous results.

Test Results

Should be used in conjunction with the prior two test (EG & OX) In addition to other tests cited in this stage.

Expected Values

See color chart for normal values & Abnormal values.

Quality Control

Both QC & QA Procedures and GMS Standards are used in the manufacturing of the antifreeze test strips.

Precaution

PPE is preferred method for standard laboratory procedure when handling samples and Chemistry reagents UNDER BOTH THE CLIA & COLA FED ACTS.

Controls

Spiked controls used for al of the parameter values listed on the color chart for each chemistry test.

Limitation of the Procedure

Sugars, and compound containing alcohol derivatives found in food & drugs can render FALSE POSITIVES results.

Accuracy

96% accurate Coefficient of variation of 4 %. Tested on spike samples for Stage 3 tests for Normal and Abnormal values as listed on color chart.