

GUIDELINES FOR THE COLLECTION AND SHIPMENT OF SPECIMENS FOR TOXICOLOGICAL ANALYSIS

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FORENSIC TOXICOLOGY LABORATORY
BEXAR COUNTY MEDICAL EXAMINER'S OFFICE
San Antonio, TX

J. Rod McCutcheon, BS, D-ABFT
Chief Toxicologist

Michael Frontz, MSFS, FTS-ABFT
Quality Assurance Officer

Carolyn Presses, MS, FTS-ABFT
Senior Toxicology Chemist

Tiffany Flowers, MS, MT(ASCP), FTS-ABFT
Senior Toxicology Chemist

Telephone: (210) 335-4030
Fax: (210) 335-4009

World Wide Web: <http://www.co.bexar.tx.us/bcftl>

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Mission:

The Forensic Toxicology Laboratory (FTL), Bexar County Medical Examiner's Office, located in San Antonio, TX is an American Board of Forensic Toxicologists (ABFT) accredited laboratory providing routine forensic toxicology services for a variety of clients. Our primary function is to assist the Bexar County Medical Examiner's Office in the determination of the cause of death in cases under their investigation. This task is accomplished through analytical postmortem toxicology services, the interpretation of analytical results, and case consultations. These services are also available on a fee for service basis to other medical examiners, pathologists, or coroners who require postmortem toxicology support. The FTL also performs a limited number of human performance toxicological evaluations on DUI, DWI, DRE, and sexual assault cases on a fee for service basis. Analytical toxicology services may be provided for hospitals and other agencies for specialty testing on a case-by-case basis; all special requests must be approved before specimens are submitted for analysis. Consultations and expert testimony are also provided to county agencies and other groups (fee for service) on drug-related issues. It is the goal of the staff of the FTL to provide our customers with the highest quality toxicology support services, consultations, and expert testimony available and to provide that service in the timeliest manner possible.

Postmortem Cases:

Collection:

In an effort to furnish the most meaningful toxicological analysis, the following specimens are recommended for submission for toxicology testing:

- Blood:
 - All available up to 30 mL
 - Indicate source on tube
 - Peripheral blood preferred
 - 2 gray top
- Antemortem blood and/or serum as available
- Urine: 50 mL (no preservative)
- Bile: All available
- Vitreous Fluid: All available
- Muscle (thigh): 50 grams
- Brain, Kidney, Lung, Gastric Contents: Optional (at least 20 g if submitted)

Comments:

For decedents who survive and later expire during a hospital stay, it can be extremely beneficial if the contributor can furnish any antemortem samples along with the postmortem samples. Label the source of all samples (e.g., heart blood or peripheral blood) and differentiate antemortem from postmortem specimens. In addition, list the date and time when the specimens were collected. Prompt collection of specimens is essential so that they may be protected from

contamination and degradation. The remains should be refrigerated as soon as possible. No one, under any circumstances, should attempt collection of blood, urine, or bile by needle puncture if an autopsy is to be performed. Such attempts may result in unsuitable toxicology specimens and cause contamination of crucial evidence such as body wounds, markings, and other prominent features. Central blood should never be collected by scooping from the pericardial sack and peripheral blood should be collected from the femoral vessels. Note that due to postmortem redistribution of some drugs, the interpretation of central blood concentrations may be problematic. Excellent specimens can easily be obtained at autopsy. The pathologist should select the toxicology samples during examination of the remains and have his or her assistant(s) prepare properly labeled containers or tubes beforehand. In cases of suspected inhalant use, a blood specimen should be collected in a screw top test tube and completely filled to the top to prevent the loss of any volatile compounds from the specimen. If no fluids or organs can be recovered, 50 grams of muscle can be submitted. In even the most severely burned or fragmented cases, valuable information can often be obtained from only a few grams of dried blood or tissue (esp. spleen). If in doubt, submit as much tissue as is practical. Do NOT submit formalin-fixed tissue for toxicological analysis. All specimens must be labeled with the sample type, decedent's name, and the submitting agency case number. A properly completed request form must be submitted with each fatality, as well as any other documentation pertinent to the case.

Non-Fatality Cases:

Definition:

Non-fatality cases submitted for toxicological analysis fall into a number of different categories. Each category requires a different analytical profile and provides unique challenges and interpretation of results. The types of cases accepted by the FTL on a fee for service basis are:

- Criminal investigations: questions concerning the use, effects, and toxicity of controlled substances
- DUI/ DWI/ DRE cases: human performance toxicology, usually associated with operating a motor vehicle
- Special Tests: non-routine toxicological analyses not performed by other local laboratories, typically performed on a limited basis upon special request

Comments:

Analyses routinely performed for criminal investigations (primarily sexual assault cases) and DUI/DWI cases include alcohols (ethanol, methanol, acetone, and isopropanol) and drugs of abuse (cocaine, morphine, codeine, and cannabinoids.) If the use of a particular therapeutic drug is known or suspected, e.g., methadone, the drug in question should be identified on the toxicology request form. Note that sexual assault cases in which the use of Rohypnol is suspected require a special benzodiazepine analysis. Rohypnol is not detectable

in blood or urine following a single dose, but its inactive metabolite may be detected in blood for approximately 4 – 6 hours and in urine for approximately 10 – 12 hours after Rohypnol administration. Analysis for DRE cases is determined by the DEC drug category identified on the Toxicology Request Form. The Drug Influence Evaluation sheet should be included with the request form. If the specific drug(s) used by the defendant is known, that information should be included on the Toxicology Request Form and a specific analysis for that drug will be performed. If no categories are listed an immunoassay for the following drugs or drug classes will be performed: cocaine metabolite, opiates, and cannabinoids. Requests for special tests (e.g., ethylene glycol) should be confirmed via telephone before submitting specimens to verify that the analysis can be performed and to determine the most appropriate specimen to be submitted.

Collection:

The following specimens should be collected and submitted:

- Blood: 14 – 20 mL (NaF preservative, gray top tubes)
- Urine: 50 – 70 mL (no preservative)

After collection, hand-mix the blood tubes, and label all specimens with the name and SSAN of the individual, specimen type, date specimens collected, and the initials of the individual collecting the specimens. The chain of custody should be initiated when the specimens are collected.

Packaging and Shipment:

Submission via mail or express carrier service:

Each specimen should be individually wrapped in an absorbent packing material and then placed in a heat-sealed or zip-lock plastic bag; blood and urine should be packaged separately. Next, place all specimens and paperwork (paperwork should also be sealed in a separate plastic bag) from a single individual in another heat sealed or zip-lock plastic bag; do not package different types of specimens together nor package more than one set of patient specimens in each bag. The blood and/or urine should be packed, unfrozen, in a shipping container of sturdy cardboard, plastic, or metal construction, sealed, and then sent by the fastest means possible to the FTL, e.g., FedEx®, U.S. Second-Day Mail, etc. DO NOT send package(s) by Registered, Certified, Air Freight, or "Return Receipt Requested" as this will cause significant delays in the delivery of the specimens. Each individual's set of specimens submitted must have an accompanying Toxicology Request Form and any other documentation pertinent to the case (paperwork should be sealed in a plastic bag). Note that failure to submit a properly completed Toxicology Request Form will delay processing, may result in an incomplete analysis of the submitted specimens, and may cause test results to be returned to the wrong address.

Submission via courier:

Specimens may also be hand-carried to the BCFSC after they are collected. Receiving hours are from 8 AM until 5 PM on regular County workdays. A Toxicology Request Form must also be completed at the time of specimen submission. The form is available online on the toxicology laboratory website and at the BCFSC.

Submission via secure lockbox:

As of January 1, 2009, the Bexar County Medical Examiner's Office will no longer accept specimens that are submitted at any time other than regular County business hours. Refrigerated secure storage is not available for samples that are not received in person by laboratory staff.

Specimen Disposition:

Specimens submitted by police agencies, e.g., SAPD, are maintained in secured, refrigerated and/or frozen storage for two years from the date the final report is issued. During that time period the contributor may make arrangements to either reclaim the evidence submitted to the FTL or to have the evidence destroyed. Any specimens not reclaimed by the contributor at the end of the two year hold period will be destroyed. Other contributors of non-fatality cases may request immediate return of their specimens (please provide a FedEx® account number or a specimen return fee will be assessed), specify a destruction date, or specimens will be destroyed after two years.

Analyses Provided:

The FTL is able to provide a wide variety of analyses; the scope and extent of toxicological analysis is at the discretion of the contributor. Contributors may contact the chief toxicologist in advance of case submission to solicit recommendations concerning the scope, availability, and necessity of testing or choose from the following list of available tests; if the analysis desired is not listed contact the chief toxicologist to determine availability. A fee schedule is attached.

Alkaline Drug Screen: (GC, GC/MS)

General screen for many therapeutic drugs (e.g., antidepressants, antihistamines, analgesics, sedative-hypnotics, amphetamines, some cardiac drugs, etc.) and some drugs of abuse (e.g., MDMA, methamphetamine, cocaine, etc.) The detection limit for most basic drugs with this method is approximately 50 ng/mL. Very potent drugs, such as fentanyl and many of the benzodiazepines (esp. Rohypnol), may be detected by this method only in lethal concentrations. If the use of such drugs is suspected an analysis for that drug should be specifically requested.

Acid/Neutral Drug Screen: (GC, GC/MS)

General screen for a limited number of therapeutic drugs (most drugs in this category have fallen into disuse.) The drugs most commonly found using this screen are: barbiturates (esp. phenobarbital and butalbital), acetaminophen, salicylates, carisoprodol, meprobamate, phenytoin, valproic acid, and theophylline.

Amphetamines: (Immunoassay, GC/MS)

Although most members of this drug class (with the exception of phenylpropanolamine, an OTC decongestant) are detected by the Alkaline Drug Screen, the GC/MS method is more specific and sensitive for those drugs. If the use of amphetamine, methamphetamine, MDMA (ecstasy), MDA, phenylpropanolamine, pseudoephedrine, ephedrine, phentermine, fenfluramine, or phenmetrazine is suspected, this assay should be requested. The immunoassay screen provides a rapid means of detecting the presence of amphetamine, methamphetamine, MDA, and MDMA in urine; positive results are confirmed by GC/MS.

Benzodiazepines: (Immunoassay, LC/MS/MS)

Although the members of this drug class are also detectable with the Alkaline Drug Screen, many of the newer benzodiazepines have therapeutic and toxic concentrations near or below the detection limit of that screen, e.g., midazolam, flunitrazepam (Rohypnol,) alprazolam, and lorazepam. If the use of a benzodiazepine is suspected, the drug should be noted and this assay requested.

Cocaine/Opiates: (Immunoassay; GC/MS)

This procedure is designed to identify cocaine, benzoylecgonine (inactive cocaine metabolite), cocaethylene, morphine, codeine, 6-monoacetylmorphine (an indicator of heroin use), hydrocodone, and hydromorphone. If urine is submitted and the use of cocaine and/or heroin is suspected, an immunoassay screen for benzoylecgonine (a marker of cocaine use), morphine, and codeine may be substituted for the more comprehensive GC/MS procedure. The immunoassay screen may be employed first in such cases because it is atypical to detect these drugs in blood and not in urine. Positive results are confirmed by GC/MS.

Alcohols: (GC, Enzymatic Assay)

The GC procedure screens for ethanol, acetone, isopropanol, and methanol. Blood, preferably collected in a gray top tube, is the best specimen for interpreting the behavioral effects of alcohols, but this analysis can be performed on any specimen. It is recommended that two different specimens (esp. blood and vitreous fluid) be used for alcohol analysis in postmortem cases to facilitate the differentiation of antemortem consumption from postmortem production of alcohols (vitreous fluid and urine are the specimens least susceptible to the

formation of alcohol as the body decomposes.) The presence of ethanol is confirmed using an enzymatic assay.

Volatiles: (GC; GC/MS)

This procedure primarily screens for butane, methane, Freons, toluene, benzene, trichloroethane, and trichloroethylene. If inhalant abuse is suspected, blood should be collected in a Teflon-lined, screw top, test tube and the tube completely filled to prevent the loss of these volatile agents. Because these agents are extremely volatile they are rapidly lost through exhalation during life and also rapidly lost due to “off-gassing” after death. Specimens must be collected quickly and properly if the volatile agents are to be identified. Urine may be submitted for volatile analysis along with blood, but due to the extensive metabolism of these compounds they are typically not detectable in urine. This laboratory does not currently perform an analysis for nitrous oxide.

Cannabinoids: (Immunoassay; LC/MS/MS)

Delta-9-tetrahydrocannabinol, the psychoactive component of marijuana, is rapidly converted to 11-OH-delta-9-tetrahydrocannabinol (also psychoactive, 11-OH-THC), which in turn is rapidly converted to 11-nor-9-carboxy-delta-9-tetrahydrocannabinol (THC-COOH). These compounds are most easily identified in urine using an immunoassay screen followed by LC/MS/MS confirmation. Detection of all three is possible in the blood, but more difficult due to their rapid metabolism and clearance from the blood. No interpretation concerning impairment may be made from the detection of cannabinoids in the urine. Analysis for cannabinoids is usually of significance only in a limited number of postmortem cases (e.g., MVA driver) since their presence provides no information concerning the cause of death.

Carbon Monoxide: (Spectrophotometry)

Exposure to carbon monoxide is determined by the measurement of the percent carboxyhemoglobin (%COHb). This analysis is preferentially performed on EDTA preserved blood (purple top), but can be performed on any hemoglobin-containing (red) fluid, especially from the spleen, provided sufficient hemoglobin is present; such “tissue fluid” must be accurately identified as to its source. Although carbon monoxide is a significant factor in fire-related deaths and faulty gas and propane heaters and stoves, the advent of catalytic converters on automobiles has made CO intoxication less of an issue in this type of exposure.

Other Analyses:

On a case-by-case basis and by specific request the following tests are available:

- Heavy Metals: mercury, bismuth, antimony, and arsenic (Reinsch test, colorimetric)
- Ethylene Glycol (GC/MS)
- Gabapentin (Neurontin) (GC/MS)

Communication:

J. Rod McCutcheon, B.S., D-ABFT
Chief Toxicologist
Telephone: 210-335-4040
FAX: 210-335-4009
E-mail: rmccutcheon@bexar.org

Michael Frontz, MSFS, D-ABC, FTS-ABFT
Quality Assurance Officer
Phone: 210-335-4031
Fax: 210-335-4009
E-mail: mefron@bexar.org

Carolyn Presses, MS, FTS-ABFT
Senior Toxicology Chemist
Telephone: 210-335-4035
FAX: 210-335-4009
E-mail: cpresses@bexar.org

Tiffany Flowers, MS, MT (ASCP), FTS-ABFT
Senior Toxicology Chemist
Telephone: 210-335-4032
FAX: 210-335-4009
E-mail: tflowers@bexar.org

Forensic Toxicology Laboratory:
Main phone: 210-335-4030

Mailing Address:
Bexar County Medical Examiner's Office
Attn: Forensic Toxicology Laboratory
Bexar County Forensic Science Center
7337 Louis Pasteur
San Antonio, TX 78229-4565

World Wide Web (WWW) address:
<http://www.co.bexar.tx.us/bcftl>

Paperwork:

The Toxicology Request Form is the contributor's mechanism for communicating with the toxicology laboratory. This form should be used to:

- Identify the contributing agency (include telephone numbers, point of contact, and e-mail address if available)
- Identify the address that the final report and the bill for services should be submitted to

- Inform the laboratory concerning the disposition of submitted specimens after the analysis is complete
- Provide any relevant clinical history, e.g., circumstances surrounding the death, pending or suspected cause of death, incidents surrounding the arrest, DUI/DWI/DRE/sexual assault case, etc.
- Provide information concerning known or suspected drug history or medications found in the possession of the decedent or defendant
- Establish a chain of custody for the specimens from the time of collection through the submission process.

Specimens submitted by mail or courier should be signed over to “Mail” or “FedEx®” on the chain of custody form by the individual releasing the specimens. Specimens hand carried to the Forensic Science Center should be signed over to the individual receiving the specimens at that time. The importance of chain of custody documentation cannot be overemphasized. The chain of custody document provides a history of the specimen from collection to laboratory. Any individual who takes possession of the specimens must sign the chain of custody documentation. Providing complete historical information and any applicable facts can assist the toxicologist in selecting special procedures to supplement routine analysis. Paperwork should be carefully printed or typed. Correctly completed paperwork must be submitted with each case or significant delays will occur.

Final Notes:

Please call for information or clarification concerning collection and shipment policies if you are unsure of what to do. It is better to temporarily delay shipment of specimens than to send specimens improperly collected, labeled, packaged, and shipped or to submit cases without the correct paperwork. Interpretation of results is also available by telephone or by written report if requested. Complete history, any relevant drug information, and chain of custody are all critical aspects of case submission. Please fill-out all paperwork completely and provide as much information as possible. The judicious selection of tests requested can decrease the laboratories turn-around-time by reducing the number of unnecessary tests and, thereby, reducing your costs/case.

Thank you for the opportunity to serve you.