

Forensic Toxicology

Andrea Grippo

An Article from forensic biology, 2001

In order to understand forensic toxicology it is logical that you must first know the definition of toxicology. Funk and Wagnall's says that it is "the science that treats of the origin, nature, and properties of poisons." Forensic toxicology is the application of toxicology for the purpose of law.

Forensic Chemistry and Toxicology generally concerns the detection and characterization of poisons or toxins exhibiting adverse physiological effects. Laboratory methods used in chemical toxicological analysis cover a wide range and may be broadly classified as follows: (1) physical tests (2) crystal tests (3) chemical spot tests (4) spectrophotometric tests (5) chromatographic tests.

Toxicology is the study of substances that are harmful to human beings. Toxicologists have the responsibility of detecting and identifying the presence of drugs and poisons in fluids, tissues and organs. Their services are not just required in crime laboratories and medical examiner's offices; they reach into hospital laboratories where the identification of an overdose can mean the difference between life and death. The work of a toxicologist generally falls into three main categories:

- ◆ Routine testing for alcohol in blood or urine samples following a "breathalyzer" test. However with the advent of new technology much of the testing can be carried out by police with fewer cases being passed on to the forensic laboratory.
- ◆ Identification of drugs such as heroin, cocaine, cannabis, etc.
- ◆ Detection of drugs and poisons in body fluids, tissues and organs.

Measuring Alcohol in the Blood

The most publicized area of work in recent years, even though it is not one of the most important, has been the routine testing for alcohol. Alcohol (C_2H_5OH) is a drug which affects the nervous system and even in small amounts (less than the legal limit) can reduce judgement and reduce driving skills. On average, about one third of all driving accidents are connected with alcohol.

There are many ways to test blood alcohol content (BAC) but in the interests of efficiency and safety, breath analysis is the preferred method over both blood and urine analysis. Over the years methods of testing BAC have advanced from the old "tube and bag" type of breathalyzer with follow up blood and urine tests to the newer fuel cell variety followed by infrared breath analyzer.

The "Tube and Bag" Type Breathalyzer

This was by no means an accurate test but it gave police a rough idea as to the driver's state. The breathalyzer, as the name suggests, measure the amount of alcohol in the breath. According to Henry's Law the amount of alcohol in a person's breath is dependent on the concentration of alcohol in the bloodstream, making breathalyzing a reliable way to measure BAC. In this particular test, drivers were asked to blow through a tube into a plastic bag. The tube contained crystals of a yellow/orange chemical called potassium dichromate ($K_2Cr_2O_7$). When this chemical comes into contact with alcohol it undergoes a color change too. The more alcohol in the breath, the more crystals change color. On the tube a line was marked which represented the legal limit. If the crystals changed to green on both sides of the line then the driver was taken asked to supply a blood or urine sample for a more accurate test to be carried out in a forensic laboratory.

Gas Chromatography

Gas chromatography was, and still is, used to identify exactly how much alcohol is present in the blood or urine of a suspect driver. Because it is very accurate, results can be used as evidence in a court of law. It's applications are not only limited to the measuring of BAC, gas chromatography is also ideal for the detection of many other drugs and substances. This technique works by separating gases and liquids into their elemental components. The process is relatively quick, but in the case of BAC measurement, is still a tedious and routine job.

The sample is injected into a heated reaction chamber where it will vaporize (if it is not already a gas). Then the sample is moved by an inert carrier gas through the stationary phase column that is a long tube made of either stainless steel or glass that has been tightly wound into a coil. This column may be up to six meters (19.7ft) long and only 3mm (0.12 in) in diameter! The elements in the sample will separate out during the passage through the tube and at the end of the column, will pass through a detection device. Each component generates its own individual electrical impulses that are recorded as a chromatogram of response (vertical axis) versus time (horizontal axis). So a chromatogram usually turns out as a number of uneven peaks which can be identified by comparing the shape and time they came out with a reference chromatogram or a control sample.

Electronic Breathalyzer

The breathalyzer that is most commonly used today uses a fuel cell that can convert chemical energy combined with oxygen straight into electrical energy. The fuel required by the cell is provided

by the alcohol present in the driver's breath, and the voltage produced corresponds directly to the amount of alcohol in the breath.

This voltage is fed into an electronic amplifier that shows whether the driver is above or below the legal limit by displaying the results via "pass" and "fail" lights. Three additional lights tell the police when the device is ready, when the person is blowing hard enough and when enough of a sample has been taken to reach an accurate result. The procedure is quick, taking around one minute and the instrument can provide up to 3000 tests before the fuel cell needs to be replaced. For the sake of hygiene the mouthpiece is replaced after each test. With respect to re-usability, the electronic version of the breathalyzer has obvious advantages.

Infrared Detection

This device provides a fast, accurate and tamper-proof measurement of a driver's breath-alcohol concentration, restricting the need for blood and urine samples to be taken and tediously analyzed. The basic principle is that when infrared light is shone onto chemical compounds, certain wavelengths of light are absorbed by certain substances. By shining infrared light of different wavelengths onto the substance and recording how much light is absorbed, a spectrum for that particular chemical is produced.

Alcohol will always produce the spectrum with peaks at around the same wavelength, however it is the height of the peaks that determine the exact quantity of alcohol in the sample of breath.

Usually such a device prints out its results as a permanent record along with the driver's identification details that are entered by the police. Additionally, neither the results nor the date can be tampered with and the machine will perform control tests before and after the driver's breath has been analyzed to insure that the results are completely accurate. These instruments have decreased in size over the years and are often found in vans near random breath testing stations on the road.

Identification of Drugs

The identification of drugs is very difficult work with the extensive list of drugs and their hybrids increasing by the month. The substance in question must be subjected to rigorous tests with no margin for error. There exists a huge range of tests for the detection of certain substances but to conduct all of these tests to determine the identity of the substance would be time consuming, so "presumptive

testing" is done. This type of testing does not positively identify the substance but it does sufficiently narrow down the many thousands of possibilities so that more specific tests can later be conducted.

Color Testing

Color tests are usually the first technique used to narrow down the type of drug present. When a drug is put into contact with a specific chemical reagent the reagent will change color.

Name of test	Testing for:	Reagent color change for positive results
Dillie-Koppanyi Test	Barbiturates	blue-violet
Duquenois-Levine Test	Marijuana	(3 part reagent test) final color is purple. Three Part Test for Cocaine: Solution A is composed of 2% cobalt thiocyanate in one part each of water and glycerine; Solution B is concentrated hydrochloric acid; Solution C, chloroform. Solution A is added to the suspect substance which, if it contains cocaine, will turn it blue. Confirmation is provided by the addition of Solution B that will turn the blue color pink, and with the addition of Solution C the blue color will reappear in the chloroform layer.
Marquis Test	Opium derivatives	purple
	Opiates (amphetamines)	orange-brown
Scott Test	Cocaine	3 part test
Van Urk Test	LSD	blue-purple

Microcrystalline Testing

The substance in question is brought into contact with a drop of chemical reagent on a glass microscope slide and left until a chemical reaction produces a crystalline precipitate. The crystalline structure for a wide range of drugs is specific and can be viewed through a microscope.

Microcrystalline testing is more specific than color testing and there are a few hundred individual crystal tests available.

Chromatography

It is common to perform preliminary color or microcrystalline tests before using chromatography to identify a substance as chromatography yields somewhat inconclusive results. However, both thin layer and gas chromatography in conjunction with the preliminary tests is well suited for drug analysis.

Thin Layer Chromatography

In this technique a glass slide is coated with a thin layer of stationary phase (usually silica gel), then small amounts of sample are dotted near the bottom of the slide. The slide is then placed in a liquid (that will act as a suitable solvent for the sample) so that the level of liquid reaches just below the dots on the slide. Then through capillary action the liquid climbs the slide separating the sample as it goes. The separation is usually colorless so in order to view the results the slide must be placed under ultra-violet light or be sprayed with a reagent that will cause the "trail" to become colored. The sample can be identified by comparison with other reference samples or a control.

Gas Chromatography

Gas chromatography is also very well suited for the identification of drugs.

Spectrophotometry

In this technique, EMR (electro-magnetic radiation) is directed at a sample where certain wavelengths of the light will be absorbed. The intensity of the light transmitted is measured and recorded by the spectrophotometer and printed on a graph, or absorption spectrum. Which wavelengths are absorbed, and to what degree they are absorbed depends on the chemical structure of the sample. While still largely inconclusive in the ultra-violet spectrum, it is specific enough to narrow down the possibilities considerably. However it is possible to identify a substance positively in the infrared spectrum as each pattern is unique. It is customary to screen the sample using ultra-violet, then use infrared to positively identify the suspect substance. There are drawbacks to this seemingly ideal technique for analysis. The sample must be in as pure a state as possible, which usually involves the sample having to go through extensive purification.

Analysis of Organs, Tissues and Body Fluids

This area of forensic toxicology involves the analysis of organs, tissues and body fluids in such circumstances as sudden deaths and suspected poisonings. A post mortem is performed by a pathologist who sends specimens of various body tissues and fluids to the forensic toxicologist for examination. Many techniques are used in this area of work including chromatography and scanning electron microscopes, emission spectrographs and atomic absorption interments for the detection of heavy metals such as mercury and lead common in poisoning cases. One of the most important and accurate devices used in this area is the mass spectrometer.

Mass Spectrometry

By itself, a gas chromatograph is not conclusive enough to positively identify a substance. However if connected to a mass spectrometer, when a substance emerges from the chromatograph and enters the spectrometer, it is bombarded with high-energy electrons which break up the molecules. The principle is that no two substances break up in quite the same way. Each has its own unique pattern, so every substance tested can be positively identified. The mass spectrometer system is capable of rapid screening for a wide range of drugs and poisons in quantities as small as 10^{-12} g.