Laboratory Analysis 5

To analyze samples from clandestine labs, a variety of scientific techniques are employed. These techniques range from simple chemical color tests to the use of x-ray and infrared energy to elicit the compound's chemical fingerprint. The type of test used depends upon the information desired from the sample and the burden of proof required to establish its identity.

In this chapter, the techniques used to analyze evidentiary samples from clandestine labs are specifically addressed. A number of technical issues will be presented in a basic format to provide an understanding of the analytical process for readers. The purpose of this chapter is not to provide a detailed discussion concerning the theory of a particular examination technique. It is simply to present the options available to the analytical chemist. By reading this chapter, the investigator can gain an understanding of what examinations to request when submitting his evidence for examination. Also assisted will be the attorneys involved in the case, by providing them information concerning why certain tests were used as opposed to others.

The laboratory analyses of samples taken from the scene of a clandestine lab are the link between the investigation and the opinions. It provides the scientific proof that corroborates the investigator's theories and is used to justify the opinions rendered in reports, deposition, and testimony. Without complete and thorough laboratory analysis, the case may be unresolved.

The laboratory analysis of evidence is more involved than simply identifying a controlled substance. Identification of the components of the sample matrix may be just as important. A complete analysis is important in establishing the manufacturing method. It is not absolutely necessary. However, if the chemist's analysis is not complete, it may be implied that he is not qualified to perform the analysis or that he has something to hide. The lack of a complete analysis may also affect other aspects of the investigation or prosecution of which the chemist is not aware.

It is not sufficient to say that the clandestine lab operator was using a particular method simply because some or all of the ingredients were found at the site. The presence or absence of a particular precursor or reagent chemical cannot be established beyond a reasonable doubt without laboratory examination. The relabeling or lack of labels on containers at the scene makes the identity of the chemicals at the location questionable.

The same holds true with reaction mixtures. The chemist should identify the ingredients within the reaction mixture. The fact that a chemical or chemical container was located at the scene does not establish its presence in a reaction mixture. It only provides the chemist information he can utilize in developing his analytical scheme.

5.1 The Chemist

The chemist performing the laboratory examinations should specialize in clandestine lab analysis. In bookkeeping, all CPAs are accountants, but not all accountants are CPAs. The same is true with forensic chemists. All clandestine lab chemists are forensic chemists, but not all forensic chemists are clandestine lab chemists. The clandestine lab chemist has additional training in clandestine manufacturing techniques as well as in inorganic analysis. This allows them to expand their analytical scheme to identify all the chemicals used in the manufacturing process. His analytical scheme is geared to identifying the manufacturing process, not just the controlled substance involved.

The chemist's role in a clandestine lab investigation requires a different thought process when approaching his analysis. He approaches each sample as if he has to tell to a jury what components are in the sample and how they fit into the manufacturing process. From an investigative standpoint, his analytical approach is geared toward profiling the sample to provide the investigators information concerning the sample's composition, so the investigators know what components to look for.

There are two schools of thought concerning which forensic clandestine lab chemist analyzes the samples once they enter the laboratory. One school has the chemist who processes the crime scene analyzing the samples, essentially, a "cradle-to-grave" approach. The other school has an independent chemist analyze the samples once they reach the laboratory. This school theorizes that it should not matter who does the analytical work, as long as the person is trained in clandestine lab analysis. Practical applications in Chapter 9 contain examples of actual situations. These applications demonstrate the ramifications that need to be considered when addressing how many chemists should be assigned to process a clandestine lab case.

5.1.1 Single Chemist

Having a single chemist process the scene and subsequently analyze the samples can streamline the analytical process. The scene chemist understands the relationship between samples and the importance of each in the investigation. This broad understanding produces an intuitive prioritization of the samples based upon the direct knowledge of the sample's origin. If a sample's analytical results are consistent with the chemist's on-scene theories, analysis of similar subsequent samples may not be necessary. If they are not theories, analytical schemes and opinions may need to be modified to follow the direction in which the evidence leads.

The scene information is extremely useful to the analytical chemist. He uses this information to devise his analytical scheme. The scene chemist makes mental notes concerning what he believes was the process the operator was using. His sampling scheme is affected by the observations made. Each sample should be geared to address a specific question or questions that will be used to establish that a manufacturing operation was, in fact, taking place at the location. Unless the scene chemist prepares a detailed written report, the information concerning his intuitive impressions of the operation will not be effectively relayed to the analytical chemist.

When the analytical results differ from the on-scene theories, the chemist gains a different perspective of what could have been taking place at the scene. The differing results may address questions the scene chemist had at the scene but could not rectify without a laboratory analysis of the item. The additional knowledge allows the analytic chemist to adapt his analytical scheme and mold his opinions to conform to the new information.

Courtroom presentations should also be considered when addressing how many chemists should be involved. The use of a single chemist provides continuity during courtroom presentations. He can explain the sampling scheme, transition into the laboratory analysis, and finally tie the two together and provide an opinion concerning the operation. All the forensic information can be provided from a single source. The jury receives a less fragmented presentation that walks them through the process. A single chemist addresses what was found at the scene, why samples were taken, and subsequent laboratory results. Finally, as an expert in clandestine manufacturing techniques, he ties all the information together and renders an opinion concerning the totality of the circumstances in the case.

From a case management standpoint, using a single chemist can reduce the overall time necessary to process the samples once they reach the laboratory. As the scene chemist processes the scene, he has subliminally prioritized the samples. Once the samples reach the laboratory, he can analyze only the samples he believes would be necessary to establish the facts of the case. Without a detailed report or specific directions from the scene chemist, the

analytical chemist is compelled to analyze each sample. This may lead to unnecessary analysis and longer turn-around times for the investigator.

5.1.2 Independent Analytic Chemist

The independent analytic chemist does not have specific knowledge concerning the history of the samples from a clandestine lab operation. Philosophically, it is believed that he will provide objective analytical results. Theoretically, he would not be inclined to skew the analysis to meet the opinions formed at the scene.

The independent analytic chemist does not have independent knowledge of the sample history of the case, and he may be obligated to analyze every sample. Unanswered questions may lead to other problems by not doing so. Assumptions concerning the facts of the case can be avoided by providing the analytic chemist with a detailed report and a complete set of the scene photographs. This information should provide an understanding of the thought process used by the scene chemist at the time the samples were taken. Proper scene documentation should convey this information adequately to avoid as many problems as possible.

The case management philosophy of the forensic laboratory will dictate the use of the scene chemist or an independent analytic chemist to analyze clandestine lab evidence. The proper processing of a clandestine lab scene is a time-consuming process. It can remove a chemist from the bench effectively 1 day or more per scene. The skills required to process a clandestine lab scene are different than those required to analyze the samples. Having chemists trained in specific areas of forensic clandestine lab investigation may provide a more efficient flow of the case through the forensic system.

Documentation and the flow of information are essential to the effective forensic investigation of clandestine lab cases. No matter whether a single chemist or multiple chemists are used, communication is critical. A single chemist must document his activities completely to justify his conclusions at any point during the investigation. A qualified chemist should be able to review the scene chemist's documentation and arrive at the same conclusion. Therefore, providing a copy of this documentation to the analytical chemist should provide the information necessary for him to perform a complete evaluation of the evidence.

5.2 Types of Analysis

The analysis of samples from clandestine labs involves a broader range of analytic techniques than are traditionally used by forensic controlled sub-

stance chemists. Many of the same instrumental and wet chemical techniques are used. The differences are the way the techniques are applied and the way information is interpreted. Organic and inorganic examinations can be performed on any individual evidentiary sample. Each type of analysis provides insight into the manufacturing process used by the operator. An individual examination type is necessary to establish the identity of a specific chemical used in the manufacturing process, i.e., organic analysis is used to establish the identities of specific precursor chemicals, or inorganic analysis is used to identify reagent chemicals. A combination of the two types of analysis may be required to establish the manufacturing method used, i.e., using a combination of organic and inorganic analysis to establish the presence of the components of a reaction mixture.

The burden of proof required to identify a particular chemical varies with its role in the manufacturing process. Controlled substances have the highest burden of proof, "beyond a reasonable doubt," because their possession is regulated in some manner. The burden of proof for the presence of precursor chemicals varies with the circumstances. The beyond a reasonable doubt standard may apply if possession of the precursor chemical is illegal under a given set of circumstances (e.g., possession with the intent to manufacture a controlled substance). A preponderance of evidence may be all that is required if the chemical's identity is associative evidence, and thus, the burden of proof may be lessened.

The burden of proof determines the level of testing required. Beyond a reasonable doubt requires specific confirmatory tests that will provide a chemical fingerprint of the substance under examination. These fingerprints can be obtained through the use of mass spectroscopy (MS) or infrared (IR) spectroscopy. Techniques such as nuclear magnetic resonance (NMR) and Raman spectroscopy are considered confirmatory tests, but they are not widely available to the forensic chemist analyzing samples for clandestine labs, and they will not be addressed in this chapter.

A series of nonspecific tests indicating the presence of the chemical in question may be sufficient to meet the burden of proof that requires establishing a preponderance of evidence. These examinations can include one or more chemical color tests, microcrystalline tests, or instrumental examinations that produce nonspecific results. The following is an example of a series of nonspecific tests that can be used to establish the identity of a chemical without using a specific test.

Under low-power microscopic examination, a white powder is found to have granules containing a cubic crystalline structure (Test 1). The granules are water soluble (Test 2). A chemical color test indicates the presence of chloride ions (Test 3). A microcrystalline test indicates the presence of sodium ions (Test 4). When combining the information from these four nonspecific

tests, a chemist could reasonably conclude that the substance is consistent with sodium chloride (NaCl), common table salt.

Techniques such as x-ray diffraction and the use of x-ray detectors could provide specific information concerning the identity of the compound. However, because NaCl is not a controlled substance, the burden of proof does not require that level of detail in the examination. The simple identification of the compound as being "consistent with" NaCl may provide the forensic investigator insight into the manufacturing process used by the operator.

5.2.1 Inorganic Analysis

Many reagent chemicals are considered inorganic (i.e., the molecule does not contain carbon). Their ability to dissolve in water, the resulting pH, and their physical and chemical properties, provide the first insight to their identity. Identifying the inorganic chemicals involved in a clandestine lab or the inorganic components of a reaction and waste mixture enables the clandestine lab chemist to definitively establish the reaction methods that the operator employed or the step in the manufacturing process the operation was in at the time of seizure.

Inorganic analysis is not something routinely performed by the forensic drug chemist. However, many of the same techniques and instruments can be used. The types of tests that can be performed on inorganic compounds include chemical color tests, microscopic examinations, ion chromatography, IR spectroscopy, and the use of x-ray energy.

5.2.1.1 Chemical Color Tests

Chemical color testing is one of the oldest methods of chemical identification. It is a method with which to rapidly establish or exclude the presence of certain categories of compounds or ions. The specificity of the results varies with the test and the ions under examination.

In a chemical color test, a chemical reagent is added to the unknown. The color of the resulting mixture indicates the presence or absence of a group of compounds. For example, a white precipitate resulting from the addition of a 1% solution of silver nitrate to an aqueous solution containing the unknown indicates the presence of chloride ions. Additional testing would be necessary to exclude borate and carbonate ions, which also form a white precipitate with the silver nitrate reagent.

Chemical color tests provide a method with which to identify inorganic acids. The use of a series of three chemical color tests can reveal the identity of a clear acidic liquid. These tests have laboratory and field applications. However, caution should be taken when conducting these tests in the field due to the potentially violent nature of the reactions. Nitric acid is a reagent chemical that reacts violently with certain organic acids and has been known

Table 5.1 Acid Test Color Reactions

Acid	Silver Nitrate Reagent*	Silver Nitrate + Nitric Acid	Barium Chloride Reagent*	Barium Chloride + Nitric Acid	Diphenylamine Reagent*
Hydrochloric acid (HCl)	White precipitate	Precipitate remains	No reaction	No reaction	No reaction
Hydriodic acid (HI)	Yellow precipitate	Precipitate remains	No reaction	No reaction	No reaction
Sulfuric acid (H_2SO_4)	White precipitate	Precipitate dissolves	White precipitate	Precipitate dissolves	No reaction
Nitric acid (HNO_3)	No reaction	No reaction	No reaction	No reaction	Blue

* See Appendix J for reagent composition.

to cause ignition when it comes in contact with methamphetamine reaction mixtures containing phosphorus.

Simple chemical color tests can be used to quickly provide presumptive information concerning the identity of acidic liquids. The same color test reagents described in Tables 4.4 and 4.5 can be used for the examination of acidic solution under the controlled conditions in a laboratory setting. Correlated in Table 5.1 are the various color reactions with the inorganic acids commonly encountered in clandestine lab operations.

In some laboratories, a simple chemical color test is the only test available to establish the presence of some inorganic compounds (Table 5.2). For example, the reaction between hydrolyzed starch solution and iodide ion produces the characteristic blue color seen in an elementary school science experiment. This may not be a specific identification of iodine. However, to a trained forensic chemist, the color reaction is characteristic enough to

Table 5.2 Color Test Reactions for Inorganic Compounds

Reagent*	Color	Indication
Silver nitrate	White	Cl^-, CO_3^{-2}, SO_3^{-2}
	Crème	Br^-
	Yellow	I^-, PO_4^{-3}
	Brown	OH^-
	Black	S^-
Barium chloride	White	CO_3^{-2}, SO_3^{-2}, SO_4^{-2}, PO_4^{-3}
Diphenylamine	Blue	NO_3^-, ClO_3^-, ClO_4^-, nitro compounds, oxidizers
Thymol	Green	NO_3^-
	Brown	ClO_3^-
Nesslers	Orange	NH_4^+
Starch	Blue	I^-
Sulfuric acid	Yellow orange	ClO_4^-

* See Appendix J for reagent composition.

establish the preponderance of evidence of its existence. The chemist cannot make a statement concerning the existence of iodine in a sample without testing to support his conclusion. This simple color test provides that support for those situations in which the laboratory does not have access to the instrumentation that can establish the presence of iodine beyond a reasonable doubt.

5.2.1.2 Microscopic Techniques

Microscopic examinations of inorganic compounds are the second type of testing that can be performed on inorganic compounds. As in chemical color testing, the specificity of the results depends upon the compounds being examined and the tests being performed.

The three types of microscopic examination involve observation of the compound's basic optical properties, recrystallizations, and microcrystal examinations. Each type of microscopic examination requires different levels of microscopic expertise. Each method requires practice on the part of the examiner to be able to recognize crystal structures as specific to a given ion.

Observing the optical properties of pure compounds under the microscope can be used to identify them (Table 5.3). Information concerning the compound's color, crystal form, and index of refraction can be used to make a specific identification. Contained in Appendix G is a table of the optical properties of inorganic compounds found in clandestine laboratories. The physical structures and optical properties of a compound or a mixture of compounds can be observed by placing the unknown in a drop of nonvolatile organic liquid, such as mineral oil, or the Cargile liquids that are used to establish the refractive index. The use of polarized light and optical filters can assist the chemist in visualizing the various crystals as well as can provide information concerning their birefringence and other optical properties.

Recrystallization is a method in which the inorganic components of pyrotechnics have been identified. The component is dissolved in a minimal

Table 5.3 Microcrystal Development Techniques

- Add a few crystals of the unknown to a liquid in which the substance IS NOT soluble, and observe the crystalline structure and optical properties.
- Dissolve a few crystals of the unknown in a liquid, and observe the crystals that develop as the liquid evaporates.
- A drop of reagent solution is caused to flow into the test drop.
- A drop of the reagent is added directly to the test drop at the center (or vice versa).
- The reagent and test drop mixture is scratched or mixed to induce crystal formation.
- Reactions take place in a capillary tube.
- A fragment of solid reagent is added to the test drop.
- A drop of the reagent is suspended over a test drop (or vice versa).
- A drop of acid, base, or solvent may be added to the test drop to assist in the volatilization.

amount of deionized water and is placed under a microscope. As the water evaporates, the solution reaches the saturation point, and the compound begins to crystallize around the edge of the drop. The shape of the resulting crystals is characteristic of the compounds in the sample. As with other microscopic techniques, the use of a polarized light microscope is beneficial but not absolutely necessary.

Microcrystal tests are conducted in a manner similar to how chemical color tests are conducted. A chemical reagent is added to the substance under examination. Instead of observing the resulting color, the examiner looks for the formation of characteristic crystals under the microscope. The use of a polarized light microscope is not necessary but can be beneficial. Caution should be exercised, because a number of anions may produce similar crystals using the same chemical reagent. However, results of a series of microcrystal tests can be considered specific identification for an anion. Listed in Appendix H are the various reagents used for inorganic microcrystal examinations.

Microcrystal tests can be used on pure compounds as well as mixtures. There is a limitation. A single test will only identify one half of the inorganic compound. Only the cation (Y^+) or the anion (X^-) component is identified using a given reagent. Additional tests need to be undertaken to identify the other half of the compound. In mixtures, this can be problematic if there is more than one set of cations and anions present. The burden is on the examiner to establish which cation is paired with which anion. Assumptions can be made using information from the scene. However, if the only information the analytic chemist has is the sample in front of him, he may be hard pressed to "prove beyond a reasonable doubt" what the cation and anion pairing was originally.

The use of microscopic techniques as a means of identification can be problematic if the testing is not documented. All testing used to make identifications should be documented in such a manner as to allow an independent expert to evaluate the results. Instrumental techniques, such as MS and IR spectroscopy, provide a paper record of the results of the examination. This demonstrates that the analytic chemist actually performed the test, documents the results, and provides a means of independent evaluation if that becomes necessary.

When the analytic chemist utilizes microscopic techniques as a means of identification, he should follow the same protocol of documenting his test results, as he is obligated to do when utilizing instrumental techniques. It is recommended that the results of microscopic examinations used to identify compounds be documented through the use of photomicrographs. This provides a record of examination used to identify the compound. This also provides a means for an independent evaluation of the results. If he cannot use photomicrographs to document his examination, he should sketch and

describe the crystal forms he observed and used to make his identification (Table 5.4). Each photomicrograph or sketch should have documentation correlating the resulting crystal form to the sample preparation technique used.

5.2.1.3 Infrared Spectroscopy

Infrared (IR) spectroscopy has long been used as a method of positively identifying organic compounds. A compound's IR spectrum has been called its chemical fingerprint. For the identification of inorganic compounds, it has not been extensively used in the forensic arena. This may be due to the broad bands and lack of detail in the spectra. Even with these handicaps, IR spectroscopy can be used to identify inorganic compounds.

The two keys to using IR for inorganic compound identification include sample preparation and peak identification. As with any analytical technique, sample preparation is the key to obtaining a usable spectrum. The exact locations of peaks in the IR spectrum are critical in identifying the salt form of an inorganic compound.

The broad absorbance bands of inorganic IR spectra make it difficult to identify the maximum absorbance of the peak. The sample concentration should be diluted until a definite peak is observed in the primary absorbance band. This will allow the examiner to identify the maximum absorbance of each peak of the spectrum.

Determination of the maximum absorbance of each peak in the spectrum is critical. A shift in the maximum absorbance of 10 wave numbers can be the difference between the sodium and potassium salt of a compound. These minor shifts may seem insignificant, but if they are reproducible in properly prepared samples, they provide the specificity required for identification purposes.

Sample preparation is critical when using IR to identify and differentiate inorganic compounds. Many inorganic compounds are efficient absorbers of IR radiation and easily overload the test sample. It is essential that the primary absorbance bands of the resulting IR spectra have well-defined peaks with resolvable maximum absorbance values, as opposed to broad nondescript bands with rounded or flat maximum absorbance areas. Broad rounded absorbance bands do not show subtle absorbance shifts needed to differentiate between salt forms of an inorganic compound.

Many anions have characteristic absorbance bands in the IR spectrum. These can be used as a screening tool to classify the type of inorganic compound with which the examiner is dealing. The presence of particular anion absorbance bands in a mixture can provide information that can be used to categorize the types of compounds that may be present. Appendix I contains a table of anions and their corresponding IR absorption wavelengths.

Table 5.4 Crystal Descriptions

Crystal	Shape	Description
Blade		Broad needle
Bunch / Bundle		Cluster with the majority of the crystals lying in one direction
Burr / Hedgehog		Rosette, which is so dense that only the tops of the needles show
Cluster		Loose complex of crystals
Cross		Single cruciform crystal
Dendrites		Multibrachiate branching crystals
Grains		Small lenticular crystals
Needles		Long thin crystals with pointed ends
Plates		Crystals with the length and width that are of the same magnitude
Prisms		Thick tablet
Rod		Long thin crystals with square cut ends
Rosette		Collection of crystals radiating from a single point
Sheaf		Double tuff
Splinters		Small irregular rods and needles
Star		Rosette with 4 or 6 components
Tablet		Plates with appreciable thickness
Tuff / Fan		Sector of a rosette

In some instances, the results from IR analysis cannot distinguish between salt forms of a given compound. In these situations, the analytic chemist may be able to use the results of other techniques to make a specific identification. For example, the IR spectra of sodium and potassium cyanide are almost indistinguishable. However, sodium and potassium are easily distinguishable using microcrystal techniques. Combining the results of these examinations provides the analytic chemist with the information necessary to render an informed opinion.

5.2.1.4 Ion Chromatography

Ion chromatography (IC) is an instrumental method that allows the chemist to identify the anion and the cation of an inorganic substance or mixture. The analysis is a three-step process. First, the cation is determined. Then, the anion is determined. Finally, the results are combined, and the compound is determined. (e.g., $Na^+ + Cl^- \rightarrow NaCl$). Additional testing may be necessary to establish the hydration state of a compound that contains multiple hydration states.

Ion chromatography is effective in separating and identifying cations and anions. However, when there are multiple components in a solution, the IC cannot distinguish which cation is associated with what anion, or what were the forms of the compounds originally placed into the mixture. Here is where the chemist uses his knowledge of clandestine manufacturing methods, along with the chemical inventory from the clandestine lab scene, to establish the most probable combination of chemicals that would produce the results obtained from an IC run on a complex mixture.

An example of how ion chromatography can be used to propose a reaction mechanism would be the analysis of a basic aqueous solution from a clandestine lab that contained a trace of methamphetamine. Anion analysis revealed the presence of iodide with small amounts of chloride and carbonate present. Cation analysis revealed the presence of sodium. From this information, the chemist proposed that the iodide originated from HI, and the chloride originated from the HCl salt of the ephedrine precursor. The sodium came from sodium hydroxide that was used to neutralize the HI. The odd trace of carbonate was a result of the sodium hydroxide reacting with the carbon dioxide in the air to produce sodium bicarbonate.

5.2.1.5 X-Ray Analysis

The use of the x-ray detector on various instruments provides the analytic chemist with a method for identifying the elemental composition of a compound or mixture. This is accomplished by recording the energy emitted from the substance that has been exposed to a beam of electrons. Each element releases a characteristic wavelength(s) of x-ray energy, as it releases

the energy it absorbed from the electron beam. The instrument also calculates the percentage of each element in a sample. This information can be used to determine the molecular formulas of most inorganic compounds.

The drawback to this method is that many x-ray detectors cannot detect hydrogen, carbon, nitrogen, and oxygen. This limits their use in organic analysis and in the determination of hydrogen, oxygen, nitrogen, and certain low-molecular-weight metals, e.g., lithium that can be used in the Birch reduction method is outside the detectable range of most x-ray detectors.

Mineral acids are an example of compounds that cannot be directly identified using x-ray technology. Besides the acid being corrosive and detrimental to internal instrument parts, the detector cannot detect hydrogen. However, derivatization techniques can be used to compensate for this problem. The chemist replaces the undetectable H with a detectable element, like potassium or sodium, by reacting the mineral acid with a strong base. The water is evaporated, and the remaining solid is analyzed (e.g., HI + NaOH $\rightarrow$ NaI + HOH).

X-ray techniques can be used to analyze solid inorganic waste to determine the elemental makeup of the mixture. The dried waste solids are essentially the same inert inorganic salts that are created when a mineral acid is neutralized with a strong base. However, these waste solids can contain other inorganic by-products that may complicate data interpretation. A Practical Example in Chapter 9 demonstrates the utilization of this technique.

5.2.2 Organic Analysis

The examination of organic compounds is the analysis that is most familiar to the forensic chemist who analyzes controlled substances. The methodology and instrumentation used to analyze drugs of abuse and explosives are routinely used in the analysis of organic compounds. Gas chromatography (GC), IR spectroscopy, MS, chemical color, and microscopic techniques can be used to analyze organic compounds. As in inorganic analysis, each technique has advantages and limitations.

5.2.2.1 Test Specificity

The analysis of individual organic chemicals is relatively straightforward. The process begins with a visual examination, followed by presumptive testing, and concluding with a confirmatory examination. However, before this process can commence, the analytic chemist must decide what degree of certainty is required for the identification of this exhibit. The degree of specificity needed to identify a solvent may be different than that needed to identify a precursor chemical or a controlled substance. The degree of specificity needed will determine the type of examination sequence used to identify the compound.

Solvents are an example of chemicals that have a low degree of specificity needed for their identification. Positive identification of the type of solvent used is generally unnecessary to establish the elements of a manufacturing charge. The solvent's identity only assists the chemist in formulating his opinion on the manufacturing method being used. Generally, the scene chemist can establish the probable identity of a solvent by comparing a container's contents to the label. If the contents look, feel, smell, taste, and act like the solvent on the label, generally, that will be sufficient to establish the probable identity of the solvent. If it does not react as expected, the scene chemist should perform presumptive tests on the substance to confirm or refute the label, and possibly sample the substance for laboratory analysis.

Clear liquid solvents present a unique problem. Laboratory analysis is the only way to determine whether a controlled substance is dissolved in them. What appears to be an unused liquid may, in fact, contain the final product or other components that would give light as to the manufacturing process used. Therefore, at a minimum, the analytic chemist should perform a screening examination on unknown solvents and not disregard them just because the liquid sample appears to be unaltered.

Reagents are an example of chemicals that require a moderate degree of certainty in establishing their identity. Reagents are not controlled, but they are used to create a controlled substance. Their identification may help in identifying the manufacturing route. An example of using a reagent chemical's identity to establish a manufacturing method would be the use of acetates to manufacture phenylacetone. Sodium and lead acetate can be used to manufacture phenylacetone. Each is used in a specific synthesis route, and they are not interchangeable. A simple microcrystal test for the presence of sodium or lead, along with a chemical color test for the presence of an acetate, can be used to identify the type of acetate, which helps identify the manufacturing route the operator probably used.

Nonspecific presumptive tests include chemical color and microscopic techniques, ultraviolet (UV) absorbance, GC retention time, index of refraction, or density. Each test has its own degree of specificity. A combination of these techniques is necessary to rule out other compounds.

Precursor chemicals and the controlled substances they form, as a rule, need to be specifically identified. This usually involves using instrumental analysis to confirm any presumptive test that may have been done. The possession of precursor chemicals may not be controlled. However, the necessity of their positive identification increases when they are possessed in conjunction with the appropriate reagent chemicals or equipment, which creates the potential ability to produce a controlled substance. In that situation, the identity of the precursor chemical should be specifically established, as well as the identity of any controlled substance.

The specific tests used in most forensic laboratories include IR spectroscopy and MS. NMR and Raman spectroscopy are also considered specific tests; however, this book will not address those techniques. Each technique is considered specific for the identification of a compound, but each has its limitations. For example, the salt form of a compound cannot be determined by using MS. With MS, there may also be problems differentiating between stereo- and geometric isomers. The resulting ion patterns can be almost indistinguishable. Without retention time data or derivatization before analysis, identification is not completely possible with MS alone. IR spectroscopy cannot distinguish between optical isomers.

With a significant portion of the samples submitted for laboratory analysis, it is required that organic compounds be identified. The composition of the samples may vary, but the procedure remains the same. Each sample requires a screening step, an extraction or sample preparation step, and a confirmatory step. These steps can be subdivided into wet chemical or instrumental procedures. Wet chemical procedures are used as screening methods or for sample preparation. Instrumental procedures are used for screening or as a confirmation tool.

5.2.3 Wet Chemical Procedures

Wet chemical procedures are used in the initial stages of the organic chemical identification process. These nonspecific tests provide a method with which to quickly indicate whether a controlled substance is present within a sample. These procedures can also be used to isolate controlled substances for confirmatory testing using instrumental techniques. Wet chemical procedures consist of chemical color tests, microscopic techniques, thin layer chromatography, and various extraction techniques. A series of these tests can be used to deductively identify a compound or mixture.

5.2.3.1 Chemical Color Tests

Chemical color tests are chemical reactions that provide information regarding the structure of the substance being tested. Certain compounds or classes of compounds produce distinct colors when brought into contact with various chemical reagents. (See Appendix J for a list of color test reagents and their compositions.) These simple reactions can indicate the presence of generic classes of compounds.

Chemical color tests are generally conducted by transferring a small amount of the substance being tested to the well of a spot plate or into a test tube. The test reagent is added to the substance. Some tests may be conducted in a sequential fashion utilizing multiple reagents. The results of each step in the sequence are observed and noted. Positive and negative controls should be run on a regular basis to ensure the reliability of the testing reagents.

There is a certain amount of subjectivity when a color is reported. It is not uncommon for two people to describe the same color differently. The colors produced can also be influenced by the concentration of the sample, the presence of diluents and adulterants, and by the age of the reagent. The length of time the reaction is observed may also influence the color reported. Color transitions and instabilities are not unusual. Allowances should be made for these differences.

5.2.3.2 Microscopic Techniques

Microscopic techniques are used as a screening tool to confirm a diagnosis made using other testing methods. Many of the same microscopic techniques used for inorganic analysis have organic applications as well. They are fast, simple to administer, and can be highly specific. There is a debate as to whether they are specific enough to be used as a confirmatory test.

The microscopic crystal structures of a compound can be used to tentatively identify components within a solid mixture. The examiner can obtain a profile of the various components within the mixture by placing a sample into a liquid test drop in which most, if not all, of the components are insoluble. (Mineral oil works well for this type of analysis.) The component's physical and optical characteristics are then observed under plain or polarized light. Commonly encountered components can be tentatively identified and quantitated when using this technique.

Microcrystal tests involve observing the crystals formed when the questioned sample is reacted with a test reagent. The test reagent and the sample can be combined using any of methods described in Table 5.3. A reaction between the component of interest and the test reagent forms a solid compound that is not soluble in the test drop. This solid forms uniquely shaped crystals that can be observed with a microscope (Table 5.4).

Microcrystal tests can also be used to determine the optical isomer of a compound. Single isomer compounds (*d* or *l*) produce different crystal forms than a racemic mixture (*d* and *l*) of the same compound. Single isomer crystals will form if a substance with the same isomer is added to the test solution prior to addition of the test reagent. Racemic crystals will form if the opposite isomer configuration is added to the test solution prior to analysis.

Mixed crystal examinations can give insight into a compound's optical orientation. They are performed by seeding a sample with a known isomer of the substance under examination. The crystals that result from the addition of reference material with the same optical orientation will result in single isomer crystals. Racemic crystals are formed if the optical orientation of the two compounds is different.

Microcrystal identification relies on the comparison of the crystals formed by the unknown with those formed by a reference standard using

the same reagent. Difficulties obtaining a match between the crystals of the unknown and those of the reference sample may arise. Impurities in the unknown sample may lead to the formation of deformed, irregular, or unusual crystals. These problems can be overcome by utilizing a cleanup procedure, such as TLC, extractions, or particle picking, prior to microcrystal analysis.

Polymorphism can occasionally be a source of trouble. Sample concentration and reagent age can lead to the creation of different microcrystalline forms. This reemphasizes the comparative nature of microcrystal identification. The comparison should be done using the same sample concentration with the same crystal reagent.

Differences in crystal appearance can arise from the concentration of the solution. The crystals in highly concentrated test drops develop rapidly, resulting in a distortion of the classic crystal shapes. Concentrated test drops should be diluted to a concentration that produces classic crystal forms that are conducive to comparison and identification.

The reagent's age will also affect crystal development. Therefore, unknown and reference samples should be run using the same reagents, under the same conditions, and at approximately the same concentrations. Reagent should also be checked on a regular basis to ensure not only that it will produce crystals with reference standards, but also that the crystals produced are consistent with the accepted crystal form for the reaction between the reagent and the substance in question.

5.2.3.3 Thin-Layer Chromatography

Thin-layer chromatography (TLC) is a wet chemical test used to screen for the presence of drugs and explosives. It is a separation technique that utilizes molecular mobility and solvent compatibility to separate and distinguish compounds within a mixture. In other words, the way a component dissolves in the TLC solvent and how it reacts with the coating on the thin-layer plate as the solvent travels over it affects the separation. Compounds are separated by their size, shape, and reactivity with the solvent, similar to rocks flowing down a river. Small compact molecules will travel across the TLC plate at different rates than large rambling molecules.

In the typical TLC procedure, a sample of the unknown is placed toward the bottom of a glass plate containing a thin layer of silica gel. A sample of a reference compound is placed the same distance from the bottom of the plate. The TLC plate is placed into a tank containing a solvent (or mixture of solvents). As the solvent travels up the TLC plate, the various components within the sample are separated. When the solvent migration is stopped, the TLC plate is removed from the tank, and the solvent is allowed to evaporate. The compound movement is then visualized through observation under UV

light or through development with a chemical color reagent designed to react with various compounds.

The *Rf* value is used to establish the identity of the spots on the TLC plate. The use of *Rf* values for a known solvent system only provides a generic insight as to the identity of the unknown spot. They should not be relied upon for confirmation of unknowns. A known reference sample, run on the same TLC plate, should be used for comparison.

Rf values can be affected by many factors. The adsorbent uniformity on the thin-layer plate, sample concentration (spotting is too weak or strong), room temperature during the mobile phase, and development distance of the solvent during the mobile phase, all will affect the results. Care should be taken to eliminate variances in the method caused by any of these factors. Placing a reference sample containing the suspected compound on the TLC plate with the questioned sample reduces the variables involved in TLC comparisons.

5.2.3.4 Extractions

Extractions are not a screening test per se. However, the fact that the compound was isolated as a result of the extraction indicates that the compound had certain chemical characteristics. These are class characteristics the can be used to deductively support the confirmatory test.

Extractions are used to separate the compound of interest from the rest of the sample. The type of extraction used will depend upon the compound of interest and the matrix in which the compound is located. In some cases, multiple extraction techniques are necessary to separate the substance of interest from the remainder of the sample. In other instances, instrumental analysis is the only way to separate compounds with similar chemical properties for confirmation.

The screening techniques used should be designed to identify as many of the components of the sample matrix as possible. This allows the examiner to select the extraction technique that efficiently and effectively isolates the component of interest from the rest of the compounds. Misidentified or unidentified components within a sample mixture may lead to the selection of an inappropriate extraction technique, which in turn, may affect the results of the confirmatory test.

5.2.3.5 Wet Chemical Documentation

Wet chemical tests are generally nondocumentable techniques. There is no independent record of the performance of the test. The test documentation solely rests on the examiner's handwritten notes. Therefore, the chemist should describe his observations as completely as possible. A (+) or (–) notation next to a test name does not provide a peer reviewer insight as to the examiner's observations during the performance of the test.

The colors or transition of colors that were observed during the course of a chemical color test should be described. Photographing a chemical color test may or may not be a solution to the documentation issue. Photography demonstrates the color that was observed during the examination. However, it may only preserve a portion of the test. Many chemical color tests have a transition of colors from the beginning of the test to the end. Photographs do not adequately reflect the examiner's total observations.

No supporting documentation is generally generated with microcrystal examinations. Therefore, the examiner's description of his observations should be as complete and accurate as possible. When definite crystals are formed, their forms and habits should be noted (described, sketched, or photographed). Listed in Table 5.4 are descriptive terms with diagrams that can be used to describe the observed crystals.

Lack of supporting documentation may be less significant if microcrystal tests are used as a screening tool. However, if they are to be used as a tool to specifically establish a compound's identity or isomer configuration, steps should be taken to provide reliable documentation concerning the examiner's observations. Photomicrographs should be taken of the microcrystals that were used to make identifications. The photomicrographs should be included in the examiner's notes for peer review, when necessary.

As with chemical color and microcrystal examinations, no supporting documentation is generally generated using TLC. Accurate notes regarding the solvent system used should be included in case notes, along with the *Rf* calculations used for compound identification. Any deviations from the referenced method or unusual occurrences should also be documented. The examiner should thoroughly describe the observations used to make his conclusions, including the colors and patterns observed on the TLC plates as well as any observations made under UV light.

Photography of TLC plates is an option. Photographs can document the examiner's observations of the colors and positions of the sample spots. If the photograph is scaled properly, a peer reviewer or independent examiner can calculate *Rf* values.

The extraction phase of the analysis is not used for preliminary or confirmatory identification purposes. However, it is a means to those ends. As such, it should be documented. Peer reviewers should be able to evaluate the extraction technique used to prepare the sample for any subsequent testing.

5.2.4 Instrumental Examinations

Instrumental examinations are documentable testing methods. This point is key to the confirmation process. It is not enough for the examiner to be able to say the compound had the same chemical fingerprint as the substance in question. He has to be able to demonstrate it beyond a reasonable doubt.

This includes subjecting the examination to peer review. Instrumental examinations provide the vehicle for this review.

There are four basic instruments routinely utilized by forensic chemists analyzing clandestine lab samples. The UV spectrophotometer and the GC are used as screening and quantitative tools. Liquid chromatography is utilized as a screening tool but not as widely. The IR spectrometer and the mass spectrometer are instruments used to confirm the identity of unknowns. As stated previously, NMR and Raman spectroscopy are used as confirmatory tools, but they do not have as broad a base of use.

5.2.4.1 Ultraviolet Spectroscopy

Ultraviolet (UV) spectroscopy is an instrumental technique that provides compound classification. It is a screening tool and not a confirmatory test. Although some compounds exhibit unique UV spectra, the spectra are considered class characteristics and do not contain sufficient detail (individual characteristics) to be considered a compound's chemical fingerprint.

The two general uses for UV spectroscopy in the controlled substances unit are general screening and quantitation. The shape of the spectrum provides insight into the identity of the compound. The amount of UV light absorbed can correlate to the amount of substance in the sample.

UV spectroscopy is a useful tool for single-component analysis of samples with known or suspected composition, such as pharmaceuticals. The UV spectrum can confirm or rebuff the composition of the preparation under examination. However, if compound identification is required, it should be done using a specific test such as IR or MS.

Mixtures of compounds capable of absorbing UV energy can present an analytical problem. Compounds have differing capacities to absorb UV light. A strong UV-absorbing substance mixed with a controlled substance that is a weak UV absorber, may result in a UV spectrum that does not reflect the presence of the controlled substance.

Quantitation is another venue in which UV spectroscopy is useful. To be most effective, the sample should contain a single UV-absorbing component. If there are multiple UV absorbers in the sample, the component of interest should have a distinct resolvable absorption band. The quantitation procedure can be as simple as comparing the concentration of the suspected tampered sample with that of a known unaltered sample. The UV absorbances should be the same if the concentrations and compositions of the samples are identical. An in-depth analysis can determine the actual concentration of the substance in question. The absorbance value of the test sample is compared to the absorbances of a series of known solutions. The concentration of the test sample can be taken from the graph of concentration versus absorbance values of the reference samples.

5.2.4.2 Gas Chromatography

Gas chromatography (GC) is a documentable chromatography form that can be used in lieu of TLC. It is not a specific confirmatory test for controlled substances. However, dual-column techniques and the evaluation of alkaloid peak patterns can be used for identification purposes. The GC is also used as a separation device for confirmatory examinations, such as MS and Fourier transform IR spectroscopy (FTIR).

The GC separates compounds by their size, shape, and reactivity with the chemical coating of the GC column, in a manner similar to rocks flowing down a river. The carrier gas acts as the water, and the column coating acts as the riverbed. The small molecules travel through the chromatographic column more rapidly than larger molecules. Their shapes and their reactivities with the column's coating separate molecules of the same size.

Chromatograms from GCs are used to identify unknowns based on the retention time or relative retention time of a peak under certain operating conditions. The retention time (*Rt*) is the time it takes a compound to travel from the injection port of the GC to the detector. The relative retention time (*RRt*) is the ratio of the retention time of the substance to the retention time of an internal standard placed into the sample.

The *RRt* is considered a more reliable value. The use of an internal standard provides a reference point with which to calculate *RRt* values. It also demonstrates the precision and accuracy of the instrument. The internal standard eluting at the proper time indicates that the gas flow and oven conditions are operating properly. The size of its peak indicates proper operation of the detector, if the concentration of the internal standard is known.

The GC can be used to differentiate geometric isomers. An example of the use of GC retention times to differentiate between isomers is the identification of the *cis*- and *trans*-phenylaziradines that are by-products of the HI reduction of ephedrine to methamphetamine. Even though these compounds have essentially the same mass spectrum, the GC retention times are significantly different. On a nonpolar GC column, the *cis*- isomer has a retention time noticeably less than the *trans*- isomer. Baseline resolution of the three isomers of the explosive compound dinitrotoluene is another example.

Analysis by GC alone is not generally considered confirmation of a controlled substance. More than one compound could possibly have a given *Rt* or *RRt*. Therefore, with conventional detectors (i.e., flame ionization, electron capture, nitrogen/phosphorus, etc.), the chemist cannot definitively tell what compound elutes at a given *Rt* or *RRt*. The specificity increases with the specificity of the detector. For example, the use of a nitrogen/phosphorus detector will only detect compounds containing nitrogen or phosphorus, thus, narrowing the field of potential organic compounds. Fortunately, this group is the one to which many drugs and explosives belong.

Dual-column GC has been used as a confirmatory test. A single sample is injected into a GC that divides the sample into two chromatographic columns. Each column contains a different liquid phase (the interior coating that causes compound separation). A compound is considered identified if it has the proper *Rt* or *RRt* values on both columns.

Commonly, GCs are used as separation tools for the confirmatory tests of MS and FTIR. The GC separates the compounds, and the MS or the FTIR provides information concerning the chemical properties of each of the compounds as they elute from the chromatographic column.

Quantitation is another use for the GC. This can be accomplished by analyzing a series of diluted samples using a method similar to that used in UV analysis. The other method uses the relative response of the item in question to that of an internal standard.

As a quantitation tool, GC has an advantage over UV. The effects of multiple components within the sample are reduced or eliminated, because the GC separates the components of the sample during the analysis. The compound's UV absorbtivity also does not affect the analysis. Each component has a similar detectability range with a given detector.

5.2.4.3 Mass Spectroscopy

Mass spectroscopy (MS) is the workhorse instrument used by the forensic chemist. It uses the pattern of molecular pieces (ions) produced when a molecule breaks apart after it is exposed to a beam of electrons as a means of identification. The resulting characteristic pattern is called the mass spectrum. It is considered one of a compound's chemical fingerprints.

The mass spectrometer exposes the compound under analysis to a beam of high-energy electrons that shatters the molecules. The mass spectrometer then sorts and counts the resulting pieces (ions) and produces a pattern, the mass spectrum. When the energy of the electron beam remains constant, the molecule will produce the same mass spectrum, which is considered one of the compound's chemical fingerprints.

MS has its limitations. It cannot differentiate among certain types of isomers. Stereoisomers and geometric isomers may produce mass spectra that are essentially identical. Stereoisomers (molecules that are mirror images of each other) have identical mass spectra. Without additional information, i.e., GC retention time data, the chemist may not be able to say the compound was one isomer or the other. Ephedrine and pseudoephedrine are examples of two compounds that have essentially the same GC retention times and mass spectra.

Geometric or positional isomers will also produce similar, if not the same, mass spectra. Many times, the compounds can be differentiated by their chromatographic retention times. Other times, there are one or two clusters

of ions that have ratios specific to a particular isomer. Methamphetamine and phentermine are two geometric isomers that can be differentiated through the use of MS.

The mass spectrometer generally cannot distinguish between the salt- and freebase form of a drug. The salt portion of the compound is generally outside the detection range of the MS. The detector only "sees" the freebase portion of the compound.

The information obtained from the mass spectrometer can be used to establish the synthesis route used to manufacture the controlled substance. Each reaction produces by-products. In some instances, the by-products produced are specific to a particular manufacturing method. Even if the detected by-products are not specific to a reaction, their presence can be used to corroborate other information, i.e., notes, chemicals on hand, etc., as to the method of manufacture. Shown in Appendix K is a five-peak table of drug precursor chemicals, controlled substances, and by-products. Also included is the reaction indicated by the presence of these compounds.

There are a number of mass spectra libraries available to assist in the identification of unknowns. The spectra in these libraries can provide insight into the identity of numerous components that can potentially be within these mixtures. However, final confirmation is only accomplished by comparing the mass spectra of the unknown to the mass spectra of a traceable reference standard. The reference spectra should be obtained on the same instrument, under the same operating conditions. The burden of proof required in a given situation will dictate how the information from these libraries should be used.

Library spectra should not be used for proof beyond a reasonable doubt. The variations in operating parameters between the instrument used to obtain the sample's spectrum and the one used to obtain the library spectra will differ. These deviations may be subtle, but they can be significant enough to eliminate the compound as the source.

Library spectra provide a preponderance of evidence concerning the identity of a compound. Many of the by-products of clandestinely produced controlled substances do not have traceable primary standards that can be used for positive identification purposes. However, their probable identity, established through a mass spectral library search, can be used as associative evidence to render opinions concerning the manufacturing methods used in the operation.

5.2.4.4 Infrared Spectroscopy

Infrared (IR) spectroscopy has been the traditional method used for confirming the identity of a controlled substance. Traditionally, the sample went through a series of screening tests to establish the compound's suspected

identity, and the identity of any adulterants or diluents were determined. The controlled substance was then extracted and purified. Finally, an IR spectrum was obtained. With the instrumentation of modern technology, an IR spectrum can be obtained from a single particle or from a peak in a GC run. This has reduced the need for the nonspecific tests used as screening tools and the extractions necessary to isolate the compound of interest.

IR spectroscopy uses a compound's ability to absorb IR light as a means of identification. The bond of each of the molecule's functional groups will absorb specific wavelengths of IR radiation. The exact wavelength will depend on the arrangement of the functional groups on the molecule. The pattern that results from charting the absorbance or transmittance of IR light that is pasted through (or reflected from) a sample is considered a chemical fingerprint.

The ability to differentiate between isomers is a benefit of using IR as a confirmation tool. Compounds with isomers that are indistinguishable by MS may be differentiated through the use of IR. The position of the functional groups on the molecule dictates how they will vibrate, which affects the wavelength of IR radiation that is absorbed. The stereoisomer of the compound may allow or hinder the vibration of a particular functional group. This allows the chemist to differentiate between stereoisomers such as ephedrine and pseudoephedrine (Figure 5.1). However, optical isomers, i.e., *d*-ephedrine and *l*-ephedrine, do not exhibit significant differences in their IR spectra.

In some jurisdictions, a compound's salt form may be important in determining the sentence after a conviction is obtained. IR spectroscopy can be used to identify a compound's salt form. In Figure 5.2, a differentiation between freebase cocaine (crack) and cocaine hydrochloride is made. The specific salt form can be used to establish a manufacturing method. Shown in Figure 5.3 are the IR spectra of the HI and HCl forms of methamphetamine. In both examples, the most obvious difference is demonstrated in the spectra's front portion (4000 cm^{-1} to 2000 cm^{-1}).

IR spectroscopy is also useful in differentiating structural and geometric isomers that the MS cannot without derivatization, retention time data, or both. Changing the position of a functional group on an aromatic ring will change the IR spectrum enough to allow easy identification. Figure 5.4 is an example of the IR differences of the three structural isomers of dinitrotoluene. The only difference is the position of one nitro ($-NO_2$) group on the aromatic ring.

Traditionally, IR confirmation has been limited to compounds that have gone through some type of extraction to produce a pure compound prior to analysis. Analysis time could be lengthy, depending on the resolution the analyst desired. Advances in technology have reduced the time required for

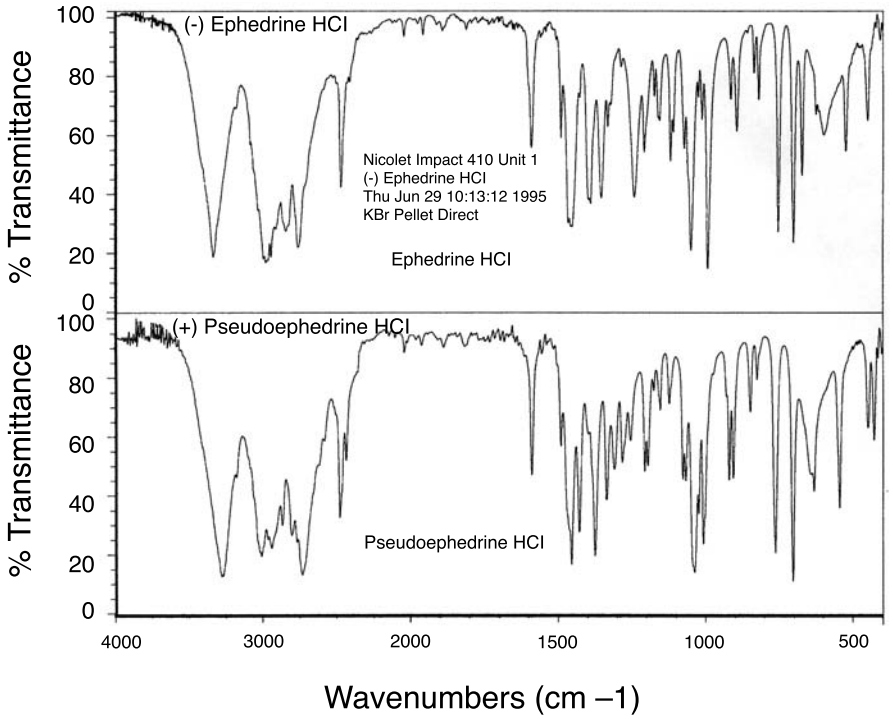


Figure 5.1 Ephedrine/Pseudoephedrine IRs.

sample preparation and analysis. With the advent of FTIR analysis, time has gone from minutes to seconds. The ability to obtain an IR spectrum instantaneously has allowed FTIR detectors to be used in conjunction with GCs. This allows the chemist to identify the components of a mixture by IR without first separating each of the components. Additionally, the micro-FTIR can isolate and obtain IR spectra of individual particles within a mixture.

Sample preparation is a key element in IR examinations. The physical state of the sample will significantly affect the resulting spectra. For example, the spectra obtained from the GC/FTIR will be in the vapor phase. These spectra will be different than the liquid- or solid-phase IR spectra a chemist traditionally uses for identification purposes. Pellet spectra of solid samples will vary from those produced using the thin-film technique. Transmission spectra and reflectance spectra of the same compound will have variations. There can even be significant variation between thin-film spectra of the same compound that are a result of polymorphism when the compound crystallizes. Each sample preparation technique produces a unique reproducible result that can be used for identification purposes. However, the analytical chemist must be sure to compare "apples to apples" when making an identification.

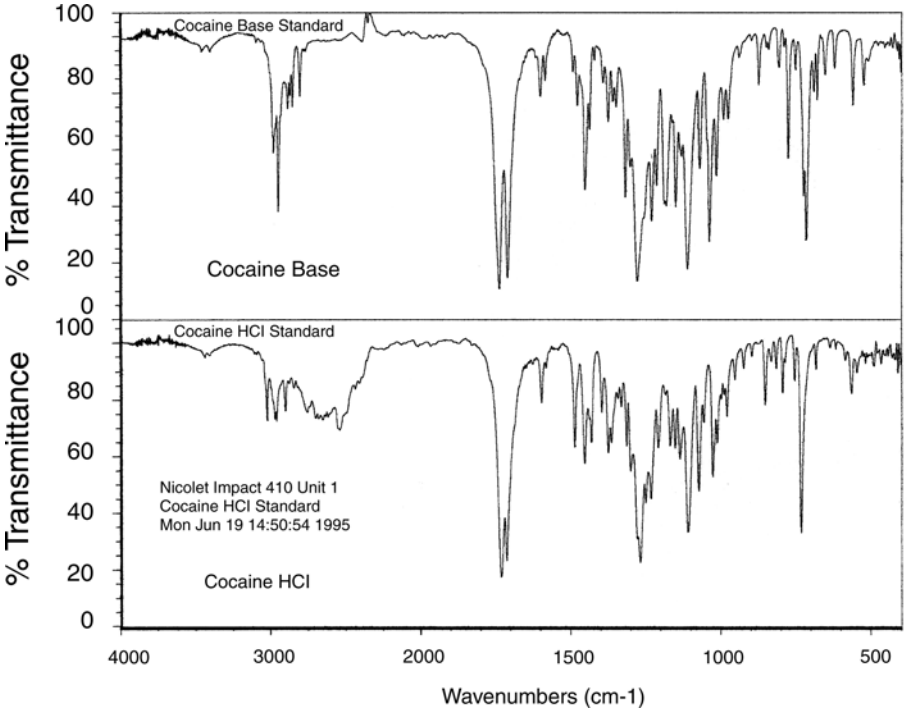


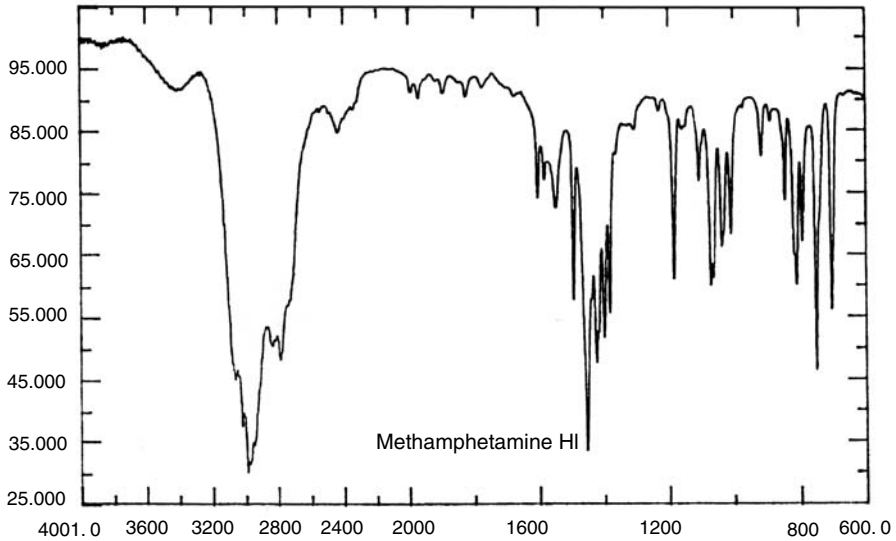
Figure 5.2 Cocaine Base/Cocaine HCl IR comparison.

With computerization, we now have the ability to compare the IR spectra of an unknown to those in various libraries. As with the MS library searches, the results should not be used for identification purposes for compounds that require proof beyond a reasonable doubt. The compound's physical state, type of detector used, and sample preparation techniques will affect the spectra obtained and the results of a computerized library search. Identification should only be made by comparing the spectra from the questioned sample to the spectra of a sample from a traceable reference that was prepared under the same conditions, using the same instrument.

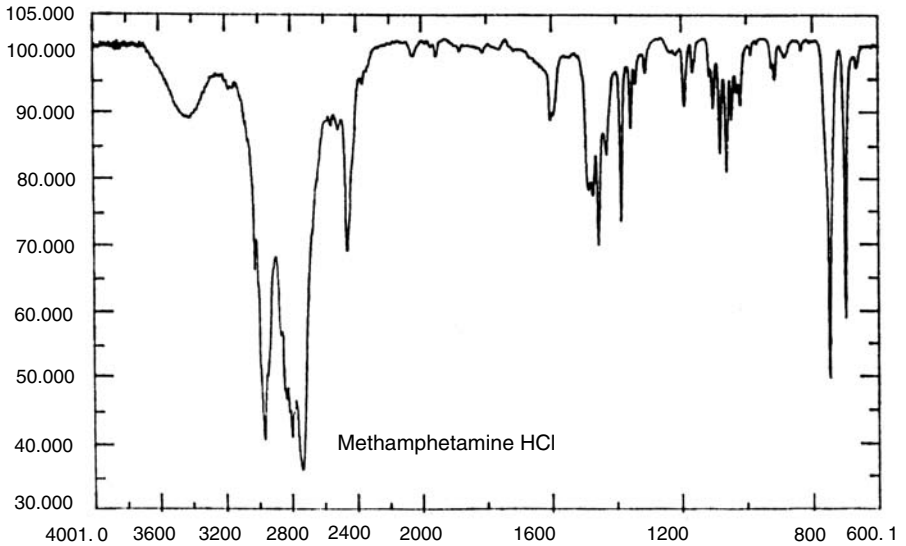
5.2.4.5 Documentation

Instrumental techniques are documentable in that they generate analytical data in a form that demonstrates that the analysis was performed. The data are objective and can be subjected to peer review as part of a quality assurance program or independent evaluation at a later date. Interpretation of this data is less subjective than in other areas of the forensic laboratory. However, it is still subject to interpretation.

For peer review purposes, case notes or instrument printouts should include the operating conditions of the instrument during the analysis. This



FN : C TITLE : METHAMPHETAMINE HI
DS1 : HOH EXT / ETHER WASH / CHCI 3 EXT / ACETONE EXT
DS2 : THIN FILM ON KBR PLATE
X =WN Y=%T DATE = Tue Mar 02 06:02:23 1993 MODEL = Analect RFX – 40
SCNS = 16/16 GAIN = 8. 00 / 8. 00 RESD = 4. 000 APOD = NB - M DET = TGSC 0



FN : D TITLE : METHAMPHETAMINE HCl
DS1 : NEAT SAMPLE
DS2 : THIN FILM ON KBR PLATE
Y =WN Y=%T DATE = Tue Mar 02 06:30:33 1993 MODEL = Analect RFX – 40

Figure 5.3 Methamphetamine HI/HCl comparison.

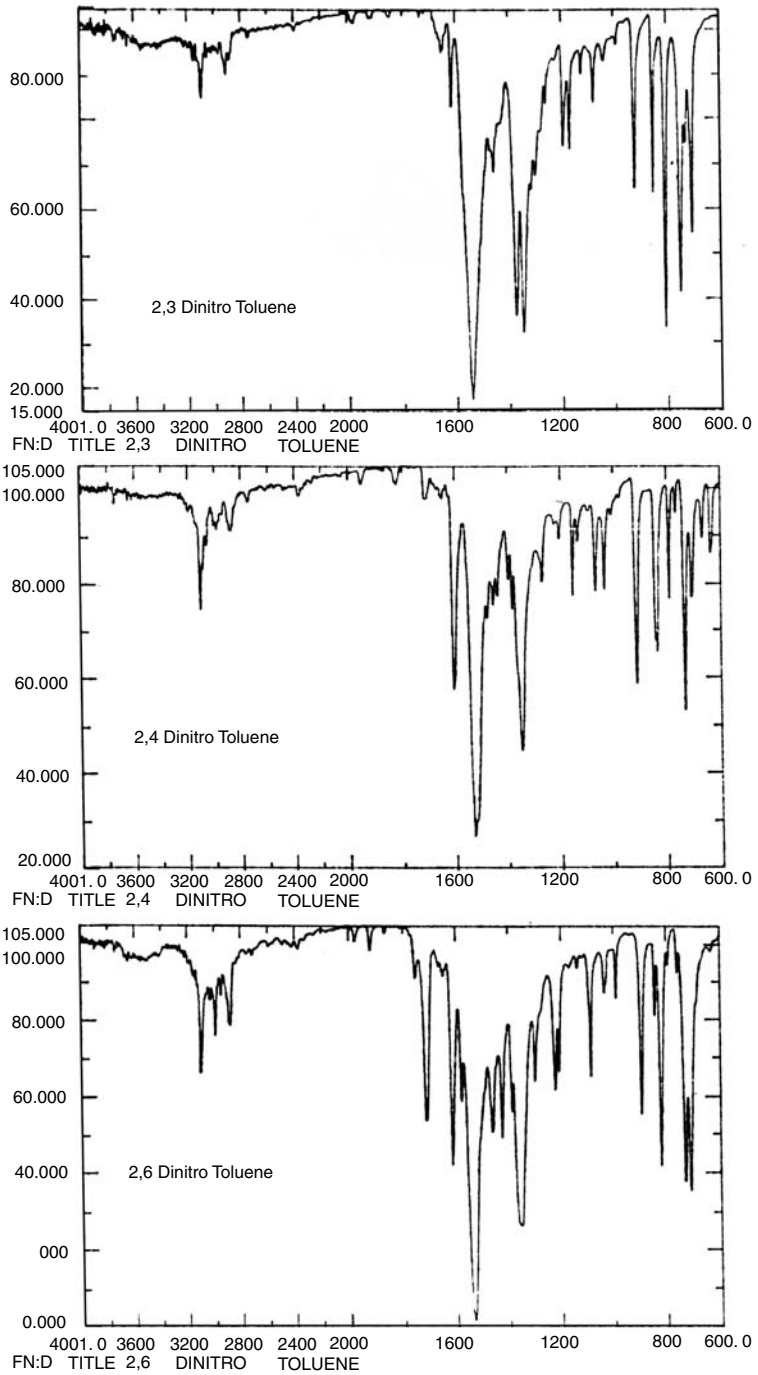


Figure 5.4 Dinitrotoluene isomer comparison.

allows the reviewer to evaluate whether instrumental results are consistent with analytical conditions. If necessary, an independent examiner should be able to achieve the same results under the same test conditions. All data should contain, at a minimum, the examiner's initials, case number, exhibit number, solvent information, and date of analysis. The examiner should have the instrument print this information on the spectra at the time of analysis, if the instrument has the capacity to do so. For GC analysis, the calculated *RRt* value should be on the chromatogram or on the printout of the peak retention times. The divisions of the mass value axes on MS data should be such that the examiner can easily determine the mass value of each of the ions of the spectra. The wave number of the significant peaks of an IR spectrum should be labeled or should be easily determined by a peer reviewer. The examiner should have the instrument print this information at the time of analysis, if the instrument has the capacity to do so.

5.2.5 Analytical Schemes

The analytical schemes used to examine clandestine lab samples can be divided into solid and liquid schemes. Solids are usually precursors, reagents, or controlled substances and should be treated as unknown controlled substances. Liquid samples can be organic, aqueous, or a mixture of the two. They can be pure chemicals, reaction mixtures, or waste products and should be treated as if they contain a controlled substance.

It is common practice for operators to remove the labels from chemical containers and repackage chemicals into different containers. They place waste or finished product into empty chemical containers. Therefore, a chemist may find a container's label little more than insight into what chemical the operator possessed at one time. A container labeled ethyl ether may just as well contain a brown liquid with a chlorinated solvent odor, as the clear volatile liquid it is supposed to be. The chemist will need to modify his analytical scheme to identify the unknown mixture in this situation, as opposed to confirming the identity of a chemical from a labeled container.

All samples should be screened for the presence of controlled substances. The screening method used is up to the chemist and the capability of his laboratory. The screening method should not only detect the presence of controlled substances but should also include techniques that would tentatively or positively identify the presence of precursor chemicals, reagent chemicals, or reaction by-products commonly used or encountered in the manufacture of the controlled substances. If a controlled substance is detected, its identity should be confirmed. If the substance appears to be a precursor, reagent, or solvent, its identity should be established to the degree of certainty dictated by the circumstances.

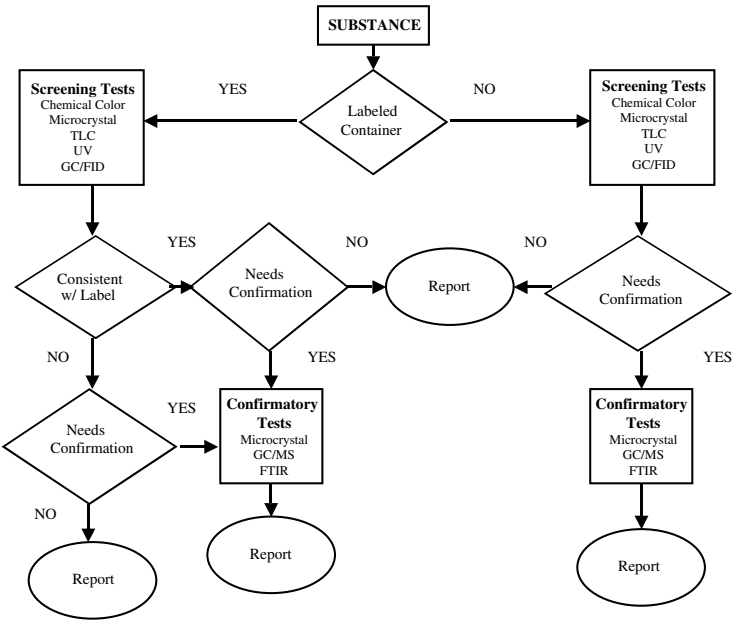


Figure 5.5 Unknown solid scheme.

5.2.5.1 Solid Samples

The analysis of solid samples follows the same analytical scheme as a controlled substance unknown. The same systematic analytical approach is used, as if the sample was an unknown and even if the identity of the substance is suspected. Sample analytical schemes should include screening for the presence of controlled substances, confirming the identity of controlled substances that have a high burden of proof, and confirming the identity of other substances to the extent their burden of proof dictates (Figure 5.5).

The sampling schemes for solid samples from labeled containers and loose solid materials are basically the same. Initially, the samples are screened using a combination of nonspecific tests. At this point, a decision is made. In the case of known samples, the following questions are asked: Is the sample consistent with the label? Does the sample contain a compound that needs a confirmatory examination? In the case of unknown samples: What does the sample contain? Does it need a confirmatory examination? The answers to these questions and the levels of specificity needed for particular identifications will guide the flow for the balance of the testing.

5.2.5.2 Liquid Mixtures

Liquid samples from clandestine labs deviate from the sample form normally encountered by the forensic chemist who deals with controlled substances.

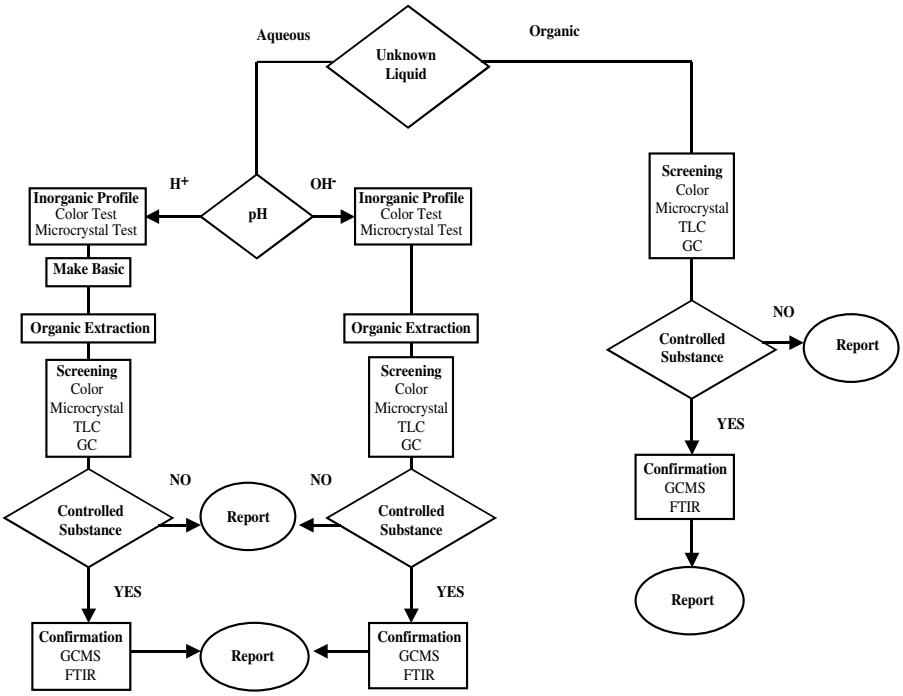


Figure 5.6 Unknown liquid scheme.

These samples, however, can give the chemist the most complete picture of the type of synthesis the operator was using. An in-depth analysis of these liquids can produce information about the product and by-products of the reaction as well as the precursor and reagent chemicals that were used in the synthesis. By evaluating the information that can be obtained from these liquid samples, the chemist can also determine at what step in the process the operation was at the time of seizure.

Liquid samples come in organic and aqueous forms (Figure 5.6). Both forms can be analyzed using the same analytic tools that were utilized for solid sample analysis. Organic samples are usually extraction solvents that may or may not contain a controlled substance. They are treated simply as a controlled substance exhibit in a liquid substrate. The analysis of unknown aqueous liquids requires a combination of organic and inorganic analytical techniques.

5.2.5.2.1 Organic Liquids. Organic liquids from clandestine labs can be reaction mixtures; extraction solvents (ether, Freon, chloroform, or petroleum products) that contain the final product; extraction solvents from which the final product has been removed, leaving only trace quantities of the final product; wash solvents that contain reaction by-products with traces

of the final product; clean unused solvents, reagents, and precursors; or the final product.

The only way for the chemist to definitively say a clear liquid contains more than a single component is to analyze its contents. Just because a liquid looks, smells, feels, and tastes like Freon, does not mean there is not something dissolved in it. In almost every case, if the final product came in contact with the organic liquid, there will be a detectable amount of that substance in the liquid. It is the chemist's challenge to find it, if it is there.

The analysis scheme of organic liquids mirrors that of organic solid samples. Using chromatographic techniques, the sample is screened to establish whether or not it contains a controlled substance. A profile of the sample's contents is used to establish the liquid's place in the manufacturing process. Confirmatory tests are performed on compounds for which burden of proof is required. Finally, a report is generated.

5.2.5.2.2 Aqueous Liquids. Aqueous liquids can be reaction mixtures or waste products. Each type of liquid contains a wealth of information concerning the manufacturing process and will require organic and inorganic examinations. The inorganic profile of the liquid will guide the analysis.

Determining the pH of an aqueous liquid is the initial analytical step that provides the analytical chemist information concerning the liquid and the compounds that it may contain. As a general rule, acidic liquids are reaction mixtures, and basic liquids are waste material. Each type of liquid will have characteristic organic and inorganic compositions.

Once the pH of the liquid is established, the chemist must determine what type of information he desires from the sample. He should ask himself: Does he simply want to isolate and identify any controlled substances from the sample? Does he want to extract all of the organic constituents from the solution and try to establish a synthesis route? Does he want to identify the inorganic components of the aqueous solution? Or, does he want to forego analyzing the sample, because it is similar to 12 other samples from the same location? The answers to these questions will determine the analytical sequence as well as any extraction techniques that may be used.

Establishing the inorganic profile of an aqueous sample is the next segment of the analysis. This can be accomplished by using the same methods described in the inorganic analysis section. The chemist is looking for the type of acid or base that was used in the reaction as well as any inorganic reagents that may indicate a particular reaction route. A series of chemical color tests and microcrystal examinations can provide the chemist with a sense of what the sample contains and where it fits into the manufacturing process. Three drops of sample may be all that is necessary to provide the chemist with a complete inorganic profile of the aqueous liquid.

As a general rule, the chemist should screen all aqueous liquids for their organic component content. Acidic liquids are generally reaction mixtures, and potentially, they can contain large quantities of the controlled substance. Basic liquids are generally waste products that will contain reaction by-products characteristic of the manufacturing process used as well as a detectable amount of the controlled substance that was manufactured. The fact that there were no organic components detected in the sample is significant information.

Most controlled substances and precursors are soluble in acidic aqueous liquids and may be visually undetectable. Thus, the analytic chemist should expect high concentrations of these chemicals in acidic solutions. To remove the controlled substance from the aqueous solution, he should change the pH of the solution and extract it with an organic solvent. This will remove the basic and neutral organic compounds from the aqueous solution. The organic extract can then be analyzed as if it were an unknown organic liquid.

There may be instances in which the acidic and neutral compounds that the aqueous liquid may contain will be of significance. In those situations, the acidic liquid should be extracted with an organic solvent prior to making the aqueous solution basic to extract any controlled substances that may be there. Again, the organic extract is analyzed as if it were an unknown organic liquid.

The analysis of the organic extracts of aqueous samples mirrors that of organic liquid unknowns. Chromatographic techniques are used to screen the sample to establish if the sample contains a controlled substance. The chromatographic profile of the sample's contents is used to establish the liquid's place in the manufacturing process. Confirmatory tests are performed on samples that contain compounds for which burden of proof is required. Finally, a report is generated.

5.2.5.3 Chromatographic Screening

Chromatographic techniques include gas and liquid chromatography. They are useful tools for analyzing organic liquid samples. These techniques allow the chemist to obtain a profile of the organic makeup of the unknown with a single test. The chemist can then determine whether a liquid contains one component, a mixture of a dozen, or none. Using retention time data, the chemicals and reaction by-products associated with various clandestine manufacturing methods can be identified. The peak areas from the chromatograms can be used to establish concentration ratios leading to quantitative estimates.

With chromatographic analysis, the chemist can quickly establish the number and probable identity of the components in an organic mixture. Under general screening parameters, the unknown's solvent will elute from the chromatography column with the solvent the chemist uses to prepare his sample for analysis. If the analyte is a clean solvent, the resulting chromatogram will appear to be blank. The identity of the analyte solvent can be

determined by modifying the chromatograph's acquisition parameters to enable separation of the low-boiling-point solvents.

The significance of the peaks in the chromatogram is determined by the analytical chemist's interpretation of data. The relative amounts of the compounds will depend on the sample preparation technique used. The chemist should use an established sample preparation scheme so that the results from all of the samples of a case can be compared. For example, a sample of an extraction solvent will contain a large amount of final product, possibly overloading the chromatographic column. If the sample is a waste solvent, there will be only trace amounts of product present. If the chromatographic peaks of samples that were prepared identically produce different peak areas for a peak that elutes at the same time as methamphetamine (e.g., Sample 1 area is 500 counts; Sample 2 area is 500,000 counts), the analytical chemist could infer that Sample 1 was a waste solvent, and Sample 2 was an extraction solvent containing the product.

The symmetry of a chromatographic peak can provide information concerning the sample. In some instances, it can be used as a presumptive test to establish whether the compound in a solution is the freebase or is in a salt form. Generally, the peaks of freebase and neutral compounds produce sharp symmetrical GC peaks when using nonpolar columns commonly used in drug and ignitable liquid analyses. Sulfate salts and HCl salts of low-molecular-weight compounds chromatograph poorly, producing asymmetrical peaks that can tail badly. High-molecular-weight salt compounds do not demonstrate this tendency.

If there is an indication of a controlled substance, its identity must be confirmed by a documentable technique. The type of information desired will determine the confirmatory route taken. If only the identity of the compound is desired, purification extraction prior to confirmation may be desirable. If the identity of all of the components of the mixture were desired, a simple dilution and analysis would be appropriate.

The chromatograms from similarly prepared samples can be used to establish a common origin or manufacturing technique. The pattern and ratio of product, precursor, and by-product peaks can be used to determine common origins of samples or positions in the reaction sequence. Samples from the same case can be compared in a manner similar to the way a chemist compares ignitable liquids and the residues extracted from fire debris.

5.2.6 Extractions

Extractions are used to separate the compound of interest from the rest of the sample. The type of extraction scheme used will depend upon the compound of interest, the sample matrix, and the information desired from the resulting analysis. In some cases, multiple extraction techniques are necessary

for separating the substance of interest from the remainder of the sample. In other instances, instrumental analysis is the only way to separate compounds with similar chemical properties for confirmation.

In devising an extraction scheme, the chemist must decide what he wants to isolate and the form it will be in after it is separated. The answers to these questions are in the statutes under which the chemist is working. Some statutes require only the presence of the controlled substance without regard to its salt form, isomer status, or purity. Other statues are specific when it comes to identifying a controlled substance by its salt form, structure, isomer form, or purity. In these cases, be sure that extraction does not alter the form.

The basic types of extractions include physical extractions, dry washes, dry extractions, and liquid/liquid extractions. In this section, the generic applications of the different types of extraction will be described. Specific extraction procedures are described in Appendix L.

5.2.6.1 Physical Extraction

Physical extractions are the simplest. They involve physically removing the substance of interest from the balance of the sample. The isolated substance is then analyzed by the technique the examiner deems appropriate.

Physical extraction is appropriate when the examiner observes particles of different sizes, shades, and consistencies within the sample. The particles are physically or manually separated from the bulk sample by using stereomicroscopes, tweezers, sieves, or other devices designed to physically isolate particles of different sizes.

5.2.6.2 Dry Wash/Extraction

Dry washes and dry extractions are different versions of the same process. The only difference is the substance that is removed from the sample matrix. With a dry wash, a solvent is used to dissolve and remove adulterants and diluents from the sample matrix, leaving the compound of interest. With a dry extraction, a solvent is used to dissolve and remove the compound of interest from the sample matrix.

5.2.6.3 Liquid/Liquid Extractions

The ability of a substance to dissolve in a liquid can change with the liquid environment. Liquid extractions utilize these solubility characteristics to separate a substance from a mixture. Listed in Appendix L are the general solubility rules used for liquid/liquid extractions.

During a liquid/liquid extraction, the sample is initially mixed into an aqueous solution. The aqueous liquid is washed with an organic solvent in which the compound of interest is not soluble but the diluents and adulterants are soluble. The organic liquid is separated, and the pH of the water is

changed in such a way that the compound of interest is made insoluble in the water solution. An organic solvent is used to separate the substance from the water.

Care must be taken when selecting the acidic environment and the organic solvent used in liquid/liquid extractions. Some drugs are subject to ion pairing. This means that the hydrochloride salt form of the drug is soluble in chlorinated solvents (i.e., chloroform) and will choose the chlorinated solvent over an acidic environment with a high chloride concentration (i.e., HCl).

Ion pairing can be used to the examiner's advantage when there are multiple basic drugs within a matrix that need to be isolated. If one of those drugs is subject to ion pairing, it can be isolated from the other drugs that, under normal circumstances, could not be separated.

In some instances, the compound of interest cannot be isolated, because the sample matrix contains multiple drugs of the same salt type. In these instances, a combination of techniques may be necessary to isolate the component of interest. An example of a combination extraction would be performing a TLC separation of the final extract of a liquid/liquid extraction. The silica gel around the spot corresponding to the compound of interest is physically removed from the TLC plate. A dry extraction or another liquid/liquid extraction is performed to isolate the substance from the silica gel.

5.2.7 Isomer Determination

Once the identity of a substance has been confirmed, the analysis is usually complete. Most statutes are written to include isomers, salts, and salts of isomers when defining a controlled substance. However, there are instances when the statute is specific in defining the controlled substance. They specifically define the structural configuration or the optical isomer of the compound that is controlled. In these instances, additional work may be necessary to satisfy the statutory definitions.

"Isomer" is a generic term that can encompass a number of different meanings. Isomers are compounds that have the same molecular formula but a different structural formula. The differences can be obvious, as in the case of structural isomers, or subtle, as with stereoisomers.

Structural isomers are compounds that have the same molecular formula but a different structural formula. Examples of structural isomers are ethyl ether and ethanol. Each compound has the molecular formula of C_2H_6O. However, the structural formulas are CH_3OCH_3 and CH_3CH_2OH, respectively. Their different structures give them different chemical and physical properties that allow them to be differentiated through various instrumental techniques.

Geometric isomers are isomers that result from the positioning of two different functional groups attached to different ends of a double bond. The double bond prevents rotation, creating a *cis* (functional groups on the same side of the double bond) and *trans* (functional groups on opposite sides of the double bond) configuration. Geometric isomers have similar chemical properties but different physical properties. GC and IR can be used to differentiate them through analysis.

Optical isomers are compounds that have the same structural formula. The only difference is the arrangement of functional groups around a chiral (asymmetric) carbon. This difference affects the rotation of plane polarized light. One configuration will rotate light to the right (*d*, dextrorotatory) and the other to the left (*l*, levorotatory). Otherwise, the chemical and physical properties of these isomers are identical. Microcrystalline tests and instrumental analyses of the derivatized compound are two methods available to forensic labs to use to differentiate optical isomers.

5.2.7.1 Microcrystal Examination

Microcrystal examinations used to determine the orientation of optical isomers are rapid analytical methods that require only a microscope and the necessary reagent chemicals. The compound in question reacts with an inorganic reagent chemical to form a complex that is insoluble in the test solution. The resulting complex has a characteristic crystal shape that can be observed under the microscope. Racemic mixtures (mixtures that contain both optical isomers) produce different microcrystals than single isomer compounds. The microcrystals of a single optical isomer generally cannot be distinguished from the microcrystals of the other optical isomer.

If the chemist needs to know the optical orientation of a compound, he can perform a mixed crystal test if the microcrystals of a single optical isomer were observed. A mixed crystal test involves placing an equal amount of a compound with a known optical orientation with the unknown sample (e.g., a small amount of known *d*-methamphetamine is combined with the same amount of unknown single isomer methamphetamine). The microcrystal test is performed on the known/unknown mixture. If the resulting crystals are single isomer crystals, the unknown has the same optical orientation as the known. If the resulting crystals are the crystals obtained from a racemic mixture, the unknown is of the opposite optical orientation.

5.2.7.2 Derivatization

The other method of determining the optical orientation of a compound is through derivatization. In this technique, the derivatizing reagent reacts with the compound at a reactive on the molecule, usually at a nitrogen site or at a hydroxyl group. The addition of the derivatization agent to the molecular

structure of the compound alters the chromatographic properties of the compound to such an extent that optical and stereoisomers can now be chromatographically separated.

Derivatization not only alters the chromatographic properties of the derivatized compound, but also alters the resulting ion patterns of the mass spectra between the derivatized isomers, making them differentiable. Shown in Table 5.5 are the eight most prominent peaks of the *n*-trifluoroacetyl-(S)-prolyl chloride (TFAP) derivative of the amines commonly encountered at clandestine drug labs or in the controlled substance samples. Not only are the mass spectra differentiable, but also, each is distinguishable chromatographically.

5.3 Quantitation

Once the identity of the controlled substance has been established, it may become necessary to determine the exact amount of that substance that is the sample. This may be necessary for a number of reasons. The governing statutes may require that the exact amount of controlled substance be determined. The percentage of the sample that is a controlled substance may influence the chemist's opinion as to whether the substance is finished product, waste material, or something in between. Or, the chemist might just want to know.

As a general rule, there is no statutory requirement to perform a quantitative examination on controlled substance samples. Quantitation is used as an investigative tool or is done as part of a laboratory's internal security policy. With a few exceptions, criminal statutes regulate only the possession of a given substance. The concentration of a sample does not affect guilt or innocence.

The concentration of a sample may become an issue during the sentencing phase of a trial. Some statutes provide enhanced penalties for possession of a substance over a given quantity. The words "possession of *X* grams of compound *Y*" are distinctly different than "*X* grams of substance containing compound *Y*." This wording may affect whether a quantitative exam is required to establish a sentence of 1 year or 10.

There are numerous quantitative techniques that the chemist can use to determine the concentration of a substance in a sample. Before the chemist can begin his quantitative analysis, he must determine the type of information he is trying to obtain. Does he want to accurately know how much controlled substance is in a given sample? Or, does he want his analysis to reflect the amount of substance the operator could obtain from the sample? The answer to these questions will determine the type of quantitation method used.

Table 5.5 TFAP Derivative MS Table*

Isomer	PK1	PK2	PK3	PK4	PK5	PK6	PK7	PK8	Mol. Ion
l-Ephedrine	58	166	251	252	69	42	167	41	None
d-Ephedrine	58	166	252	251	69	42	167	77	None
l-Pseudoephedrine	58	166	251	252	42	77	167	43	None
d-Pseudoephedrine	58	166	252	251	69	42	77	41	None
(1) Propylhexadrine	166	58	69	125	55	41	182	167	348
(2) Propylhexadrine	166	58	69	41	125	55	182	167	348
l-Amphetamine	166	237	194	91	118	69	44	167	None
d-Amphetamine	166	237	194	91	118	44	69	41	None
l-Methamphetamine	58	166	251	91	42	41	69	96	None
d-Methamphetamine	166	58	251	91	69	41	42	119	None
(1) *p*-Methoxyamphetamine	148	166	194	121	44	69	41	167	358
(2) *p*-Methoxyamphetamine	148	166	121	194	149	44	69	41	358
(1) 3,4,5-Trimethoxyamphetamine	208	166	418	194	181	193	209	167	418
(2) 3,4,5-Trimethoxyamphetamine	208	166	418	194	181	193	209	167	418
(1) 2,4,6-Trimethoxyamphetamine	181	208	166	182	44	121	209	69	418
(2) 2,4,6-Trimethoxyamphetamine	181	208	166	182	209	120	69	44	418
(1) 4 Bromo 2,5 dimethoxtamphetamine	166	285	256	194	44	237	468	69	467
(2) 4 Bromo 2,5 dimethoxtamphetamine	166	285	256	194	44	237	69	468	467
(1) 4 Methyl 2,5 dimethoxtamphetamine	192	166	402	194	193	165	69	44	402
(2) 4 Methyl 2,5 dimethoxtamphetamine	192	166	402	194	193	165	69	44	402
(1) Methylenedioxyamphetamine	162	166	194	135	372	163	69	44	372
(2) Methylenedioxyamphetamine	162	166	194	135	44	77	69	163	372
(1) Methylenedioxymethamphetamine	166	58	162	251	163	69	135	77	386
(2) Methylenedioxymethamphetamine	58	166	162	69	163	135	251	96	386

Source: From McKibben, T., *J. Clandestine Lab. Investigating Chemists Assoc.*, 2, 1, 13, January, 1992. With permission.

The four basic methods of quantitating the amount of controlled substance in a sample are microscopic examination, gravimetric comparison, UV analysis, and GC analysis. Below are the generic descriptions of the various quantitation methods.

5.3.1 Microscopic Examination

The quickest and most subjective solid sample quantitative method is accomplished through microscopic examination. In this technique, a sample is placed on a microscope slide and diluted with a solvent to which the components are insoluble. The examiner estimates the percentages of crystals of the various substances in the sample under observation. This is the most subjective, least precise, and least accurate method. It is subject to the examiner's ability to recognize the microscopic crystalline form of the controlled substance under consideration. The uniformity of the bulk sample also affects the accuracy and reproducibility of the results.

5.3.2 Gravimetric Techniques

Gravimetric analysis provides a rapid means with which to determine the approximate amount of controlled substance in a sample. This technique can be used on organic and aqueous samples. This technique also mimics the method operators use to extract the final product from reaction mixtures or extraction solvent. Therefore, it provides a practical approximation of how much of the final product the operator could expect to recover from the sample.

Gravimetric techniques can be performed in conjunction with the extraction phase of an analysis. The examiner weighs or measures the volume of the sample to be extracted prior to the extraction process. He obtains a weight of the extracted substance prior to any confirmatory tests being performed. The ratio of the postextraction weight to the preextraction weight provides the percentage of the item that is the controlled substance.

A limiting factor to the precision and accuracy of this technique is the efficiency of the extraction solvents. If they do not effectively remove the diluents and adulterants, the calculated controlled substance percentage will be high. If the solvents do not efficiently and completely isolate the controlled substance, the percentage will be low.

An advantage of gravimetric techniques is that the identity and composition of the final extract can be confirmed. If all the diluents and adulterants have been removed from the matrix, the resulting residue can be analyzed for purity and then identified.

The examiner must be aware of the salt form the controlled substance is in before and after the extraction process. This will affect the percentage

calculated, because the molecular weights of the salt form differ from the molecular weights of the freebase. For example, a 100% pure sample of cocaine hydrochloride contains 89.38% by weight freebase cocaine. The examiner must take into account the mass of the salt when calculating the percentage of controlled substance in the sample or must qualify the conclusion by stating the salt form of the substance identified.

5.3.3 UV Techniques

The use of UV light provides an effective method to quantitate a sample, if it has a single UV absorber. If the sample has components with overlapping UV absorbances, the instrument cannot determine which compound is contributing to the absorbance. Compounds also absorb UV radiation at different rates. Therefore, UV methods are not conducive to quantitating mixtures.

Simple "yes" and "no" concentration comparisons can be accomplished with the use of UV techniques. These comparisons are conducted in association with a product tampering case, in which the product in question may have been diluted or altered. Comparison of the UV spectra of the item in question to a known reference sample can indicate if the unknown has been diluted or altered. The compositions of both samples should also be confirmed through separate examinations.

A detailed examination can determine the concentration of the substance in question. To accomplish this, the examiner obtains the UV spectra for a series of solutions with a known concentration of the substance in question. The absorbance values are placed on a concentration versus absorbance graph. A solution of the unknown is prepared and analyzed. The absorbance value is placed on the graph to determine the concentration of the substance in the solution. This value is then used to calculate the percentage of substance in the unknown.

UV techniques done properly are precise. However, the accuracy of the results for multicomponent mixtures may be in question because of the interference of the UV absorbance of other compounds in the sample.

5.3.4 GC Technique

The use of GC for quantitation provides the most accurate and precise results compared to the other analytical techniques discussed. This technique provides the examiner the ability to isolate and quantitate a specific compound in a single method. The identity of the chromatographic peak can be confirmed at the time of the analysis or by analyzing the test solution with a GCMS. The same chromatographic conditions should be used during the confirmation test so that a direct correlation between the two techniques can be made.

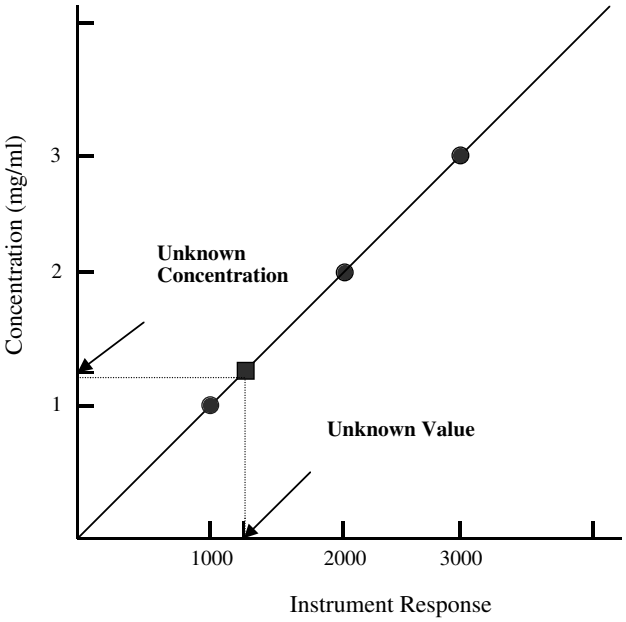


Figure 5.7 Beer's law plot.

Traditionally GC quantitation uses a concentration versus peak area plot to establish the concentration of an unknown solution (Figure 5.7). If the peak areas of the serial dilutions of a substance are charted, the concentration of an unknown solution can be determined from its instrumental responses. This method uses the relationship between the concentration of a sample and the instrumental data to calculate the concentration (i.e., doubling the sample concentration will double the GC peak area). A series of diluted samples is prepared and analyzed on the GC. The resulting peak areas are plotted on an *X*/*Y* graph with their corresponding solution concentrations. The unknown solution is analyzed in the same manner. Its concentration is obtained by using the graph generated by the known solutions.

The increase in the precision and accuracy of modern instrumentation has allowed the analytical chemist to reduce the number of reference samples necessary for GC quantitation. The relative response GC method of determining sample concentration uses the ratio of the compound's and internal standard's peak areas, known sample concentrations, and algebra. Two GC injections using this method can provide the same results as multiple injections using the serial dilution method. This procedure is based on the predictable relationship between sample concentration and the peak area of a chromatogram, i.e., doubling the sample concentration will double the resulting peak area of the chromatogram.

The use of the area concentration ratio to determine the concentration of a solution is dependent on the precision of the volumes injected into the GC for analysis. Small deviations in injection volumes will affect the accuracy and precision of the analysis.

To compensate for any deviations in injection volumes that may occur, a known concentration of internal standard is placed into the standard and unknown solutions prior to analysis. The concept of a given concentration producing a given peak area is just as true for the internal standard as the samples in which they are placed. This being the case, the ratio of peak area of sample to peak area's internal standard (IS) will not change for a solution, no matter what volume is injected into the GC.

When quantitatively analyzing organic liquids, the chemist must dilute the sample until the unknown sample produces approximately the same compound-to-internal-standard ratio that exists in the standard solution. With the known dilution factor, the chemist can calculate the original concentration. If the sample in the previous example had a 20:1 dilution factor, the concentration of the original sample would have been 25.2 mg/ml.

By converting the concentration term into its basic units of weight (W) and volume (V), the concentration equation can be manipulated into an equation that describes the percentage of the unknown that contains the target compound.

5.4 Summary

The complete analysis of clandestine lab samples is an essential portion of the forensic investigation. The information derived from the testing can only be obtained through the scientific examination of the evidence. This information is used to meet the burden of proof required to establish the presence of a controlled substance, beyond a reasonable doubt. The cumulative effect of the examinations that can only establish a preponderance of evidence can be used to formulate expert opinions concerning the operation.

The tools the clandestine lab chemist uses to analyze these samples are the same tools that the forensic chemist uses to analyze controlled substance samples. The only difference is the clandestine lab chemist may apply certain techniques in a method to produce more detailed information concerning the sample. The scope of the analysis goes beyond the forensic chemist's desire to determine whether or not a controlled substance exists. The clandestine lab chemist needs to know what else is in the sample so that he can develop opinions concerning the operation.