



Objectives

After reading this chapter, you will understand:

- How to apply deductive reasoning to a series of analytical data.
- The limitations of presumptive (screening) tests.
- The relationship between the electromagnetic spectrum and spectroscopic analysis.
- The difference between qualitative and quantitative analysis.
- The dangers of using prescription drugs, controlled substances, over-the-counter medications, and alcohol.

You will be able to:

- Chemically identify illicit drug types.
- Classify the types of illicit drugs and their negative effects.
- Discuss the federal penalties for possession and use of controlled substances.
- Explain the need for confirmatory tests.
- Describe IR, UV-VIS spectroscopy, and GC-MS, and explain how they are used in forensic science.
- Present and interpret data with graphs.
- Use the *Physicians' Desk Reference (PDR)* to identify pills.



"Every form of addiction is bad, no matter whether the narcotic be alcohol or morphine or idealism."

—*Carl Jung (1875–1961), Swiss psychiatrist*



Teacher Note

The TRCD for this chapter includes a PowerPoint presentation, which is an overview of the chapter; it can be used as introductory material or at the end as a review.

The TRCD also contains a crossword puzzle that can be used after students have learned the vocabulary from Chapter 7.

The estimated time chart is included as a suggested timeline for this chapter.

Drugs and Crime

A drug is a natural or synthetic substance designed to affect humans (or other animals) psychologically or physiologically. A drug can affect the function or structure of living tissue through various chemical reactions. When any type of drug is taken in excessive amounts and causes illness or death, it shows toxic effects and is classified as a poison. Most drugs are legitimately manufactured by drug companies and are prescribed for particular medical problems.



Sometimes, however, they can have effects that people find pleasurable. When these substances, produced legally or illegally, are taken strictly for pleasure, they are considered "drugs of abuse" or "illicit drugs."

In the United States, all drugs covered by law that are somehow restricted are called "controlled substances." They are listed in a part of the Federal



Code called the Controlled Substances Act (Act 21 U.S.C. 812).

Recreational use of drugs may cause dependency, where one's energy and time are spent getting the drugs needed to feel good. This can lead to theft to support such a habit. It also can lead to violence committed during robberies as well as for control of drug distribution and profits. Drug abuse can lead to extreme mental and physical health problems, creating a burden on the public health system.

The Office of National Drug Control Policy estimated that illegal drug use cost American society \$181 billion in 2002, and it appears the trend increases by about 5 percent a year. About 30 percent of prisoners committed their current offense while under the influence of drugs. In 2002, 4.7 percent of the 14,263 homicides in the United States were drug-related. One-quarter to one-half of all incidents of domestic violence are drug-related. Scary, isn't it?



And, of course, there is a social, moral, and religious stigma associated with drug abuse, as well as loss of productivity and pride among users.

In the United States, as much as 75 percent of the evidence being examined in forensic laboratories is considered drug-related, either the drugs themselves or evidence from drug-related crimes. Forensic chemists have many tests for identifying drugs and poisons. As soon as a pharmaceutical company produces a new drug, it sends a sample to the FBI Crime Lab so they can develop procedures to identify both large and minute quantities. These procedures are then filed for reference when unknown samples are collected at a crime scene.

Salem Witch Trials and LSD

Ergot is a fungus that infects the grain of rye plants in cold, wet weather. Ingested in large doses, it restricts blood flow, causing gangrene in the extremities. It was called St. Anthony's Fire in the Middle Ages and killed tens of thousands in Europe. It has been suggested that the visions of the girls of Salem, Massachusetts, and the frenzied response of others in the famous witch hunts of 1692 were caused by ergot-tainted rye! Twenty innocent townsfolk were executed.

-www.crimelibrary.com/notorious_ murders/not_guilty/salem_witches/1.html

There are five categories of **controlled drugs**, as defined by the Controlled Substances Act; see Appendix A at the end of the book. The legal penalties are listed in Appendix B for marijuana and in Appendix C for other controlled substances.

The following list is a widely used classification scheme for illicit drugs, based on their pharmacological effects:

• Hallucinogens are mostly naturally occurring substances that can change normal thought processes, perceptions, and moods. The most widely used hallucinogen in the United States is marijuana, a plant that has been used as medicine, to make rope (hemp), and to produce euphoria for thousands of years. The active ingredient in marijuana can range from 1 percent in low-grade material to 10 percent in sinsemilla, a cultivated female **cannabis**. A resinous oil ("hash oil") extracted from the plant can contain as much as 50 percent of the active ingredient; more often, this oil is only partially extracted from the ground leaves and sold as hashish, or "hash." Marijuana decreases the user's ability to concentrate, slows reactions, and impairs coordination.

LSD (*d*-lysergic acid diethylamide, or "acid") is chemically derived from ergot alkaloids found in a fungus that grows on grain. As little as 25 µg can cause visual and auditory hallucinations. LSD is often sold soaked into blotter paper ("blotter acid"), as tiny

Teacher Note

Appendices A–E can also be found on the TRCD (Blackline Masters 7.1–7.9).

controlled drugs: material whose possession is controlled by law

cannabis: a tall, annual hemp plant native to central Asia from which marijuana is harvested; also refers to the harvested product



Blotter tabs of LSD

teratogen: an agent that can cause birth defects in an embryo or fetus. Two well-known examples are alcohol and thalidomide.



as tiny colored tablets ("microdots"), or in small pieces of dried gelatin ("window panes"). It is an extremely dangerous drug that can cause psychosis, flashbacks (hallucinogenic episodes), and impaired

memory and attention span. It is also a possible **teratogen**.

PCP (phencyclidine) was sold as an intravenous anesthetic, but it was dropped because of its bizarre side effects, including insensitivity to pain, feelings of super strength, rage, memory loss, and paranoia. It is often mixed with other drugs, for example, with marijuana ("wobble-weed" or "Jim Jones") or with LSD or amphetamines ("angel dust"). MDMA (methylenedioxymethamphetamine, or "ecstasy") is a synthetic drug that both stimulates the user and causes hallucinations. Many of its less desirable effects are similar to those of cocaine and amphetamines: psychological problems such as confusion, depression, addiction, severe anxiety, and paranoia-sometimes even weeks after use-and increased blood pressure and heart rate, which may cause death. Recent research links MDMA use to long-term damage to the parts of the brain that are important in thought and memory.

Ketamine ("Special K"), used legitimately as an animal tranquilizer, is a powerful hallucinogen. It is sometimes called a "club drug," along with MDMA, GHB, GBL, rohypnol, LSD, PCP, and methamphetamine, because it is most commonly found at nightclubs and "rave" parties. Ketamine can cause delirium, amnesia, depression, and long-term memory difficulties.

• **Stimulants** act on the central nervous system to make the user feel better and increase his or her energy alertness while suppressing

alkaloid: one of a class of bitter-tasting, basic organic compounds with nitrogen-containing rings, which are normally obtained from plants. Alkaloids often have powerful effects on living things. Examples are cocaine, nicotine, strychnine, caffeine, and morphine. appetite and fatigue. The downside of these drugs is the accompanying restlessness and anxiety, and, after the drug wears off, depression. Stimulants vary from mild, such as caffeine, to strong, such as amphetamines ("uppers" or "bennies") and methamphetamines ("speed" or "crank"). Cocaine is considered a stimulant. It is an **alkaloid** that comes from the leaf of the coca plant, grown almost

In Yemen, drug dependency grows

Chewing qat leaves is a national pastime, with few opposing its usage

The primary active stimulant in qat is cathinone, which is structurally and chemically similar to



The structure of cathinone

(top) and d-amphetamine

chewed to release the stimulant, which produces euphoria. It is estimated that 8 are stoned every day diverts resources and and culture

d-amphetamine. Fresh leaves are wadded

in the cheek and



Qat (khat)

It is estimated that 80 percent of the inhabitants of Yemen are stoned every day! This decreases productivity and diverts resources and income, deeply influencing Yemeni life and culture.

-from news article, 2001

exclusively on the Amazon slopes of the Andes. It takes about 500 pounds of coca leaves to produce one pound of cocaine powder. Most cocaine used to be snorted as powder, but now smoking the recrystallized freebase ("crack") is more popular. Crack is highly addictive. Regular use can cause anxiety, insomnia, and weight loss and leads to paranoia and mental deterioration.

Nicotine is also a habit-forming stimulant, but it is not a controlled substance.

• **Narcotics** are **analgesics**, that is, substances affecting the central nervous system to relieve pain. Mild analgesics are found in many over-the-



analgesics: drugs that relieve pain

counter (OTC) drugs such as aspirin, Tylenol, and Motrin. Illicit narcotics come from opium, harvested from a particular type of poppy flower

grown mostly in Asia. Opium contains from 4 to 21 percent morphine, often used medicinally as a powerful painkiller. Heroin ("horse" or "smack") is easily made from morphine and is highly addictive. Overdoses can cause death; serum hepatitis is common in users. Codeine is commercially prepared from morphine and is often found in OTC drugs in Canada.



Opium comes from the milky fluid in the ripe seed bulb of the opium poppy. It is estimated that in 2007 Afghanistan supplied 92 percent of the world's illicit production.



Opium poppy seed bulbs.

Synthetic narcotics, also classified as opiates, are often prescribed for pain but are also abused. Methadone is used in this country as a heroin substitute to wean addicts from heroin. Propoxyphene (Darvon) and oxycodon (Percocet) are two prescription drugs commonly abused. Fentanyl is an anesthetic and is 100 times stronger

designer drugs: drugs synthesized with particular pharmacological characteristics, designed for abuse and evasion of specific drug laws than morphine. It spawned a series of **designer drugs** known as "China White."

• Depressants: Ethyl alcohol is a common depressant—alcohol is a \$40 billion industry in America. Alcohol, however, is not a controlled substance. The effects of overuse of alcohol will be covered in the next chapter.



Barbiturates ("downers") are highly addictive; withdrawal is difficult and dangerous. Most (such as Phenobarbital, Nembutal, and Seconal) are prescribed to reduce anxiety and help the user sleep; they pose no problems in small amounts. Other synthetic depressants include meprobamate (Miltown), methaqualone (Quaaludes), and benzodiazepines (tranquilizers such as Librium and Valium).

Many abused substances are controlled by prescription. Legitimately manufactured pills, tablets, and capsules can often be identified using the *Physicians' Desk Reference (PDR)*, which is an illustrated dictionary of medicines and drugs. A typical illustration and description—in this case, of oxycodon—is shown in Figure 7.1.

There are many different editions of this reference, which is updated each year; usually one can be found in a town or city library or a bookstore. The school nurse may have previous years' copies available.

Investigators test drugs and poisons with gas, paper, and thin-layer chromatography (TLC); UV and infrared spectrophotometry; mass spectrometry; spot tests; and qualitative analysis.

Some over-the-counter (OTC) analgesics can be harmful when taken in excess or combined with other medications or alcohol. For example, some people are allergic to aspirin, which can cause their throat to swell, even suffocating them. People with asthma and chronic sinusitis are more likely to have this extreme reaction. The same symptoms can occur with





Barbituric acid from which commercial barbiturates are derived. Sodium pentothal, the "truth serum," is also a derivative.

Teacher Note

A common "cause of death" in classroom final exam crime scenes is an allergic reaction to an NSAID. Traces of the white powder residue or unmarked pills can be tested by the methods developed in this chapter.



Percocet

Pronounced: PERK-o-set

Generic ingredients: acetaminophen, oxycodone hydrochloride Other brand names: Roxicet, Tylox NSAIDS (nonsteroidal anti-inflammatory drugs) such as Advil or Motrin. See Table 7.1 below for a list of some OTC analgesics.

Table 7.1: Some OTC Analgesics and Cold Remedies			
Trade Name	Active Ingredients		
Actifed	pseudoephedrine hydrochloride, triprolidine hydrochloride		
Advil	ibuprofen		
Aleve	naproxen sodium		
Anacin	acetylsalicylic acid, caffeine		
aspirin	acetylsalicylic acid		
Benadryl	diphenhydramine hydrochloride		
Contac	acetaminophen, pseudoephidrine hydrochloride, dextromethorphan hydrobromide, chlorpheniramine maleate		
Excedrin	acetaminophen, acetylsalicylic acid, caffeine		
Motrin	ibuprofen		
No-Doz	caffeine		
Sudafed	pseudoephidrine hydrochloride		
Tylenol	acetaminophen		
Tylenol PM	acetaminophen, diphenhydramine hydrochloride		

See www.nlm.nih.gov/medlineplus/druginformation.html for brand names, chemical names, proper use, precautions, side effects, and the like.

Anabolic steroid abuse has been associated with a wide range of adverse side effects ranging from some that are physically unattractive, such as acne and breast development in men, to others that are life-threatening, such as heart attacks and strokes, even in athletes younger than 30. Use of steroids can also cause increased irritability and aggression. Anabolic steroids are schedule III drugs (see Appendix C) and are illegal. Simple possession of illicitly obtained anabolic steroids carries a maximum penalty of one year in prison and a minimum \$1,000 fine for the first offense.



Spot Tests

A spot test is a chemical reaction that occurs when a particular substance is added to an unknown. Color reactions for spot tests on drugs are a

relatively easy and quick method used to detect the presence of certain substances. You can take this procedure into the field for preliminary testing. These tests are not conclusive, however, because a few substances may give *false-positive* results, indicating drug existence even though no drugs are present. If you get a positive result, you can use further tests, such as chromatography or infrared **spectrophotometry**, to confirm that the drug is found in the sample. A negative test result indicates that the drug in question is not present.

Reminder

false positive: a test result that comes out positive when the material being tested for is not present

spectrophotometry:

measurement of the absorption of electromagnetic radiation of a substance at different wavelengths of the spectrum

Spot Test Lab

This activity will give you practice in identifying some over-the-counter drugs with spot tests.



Color spot tests

Laboratory Activity 7.1

Advance Preparation

Use a mortar and pestle to grind the pills into powder, or have the students do it, but this will increase the risk of contamination.

Twelve-well white ceramic spot plates are best for this activity because they don't stain or dissolve by inadvertent contact with solvents such as acetone; however, they are more expensive than the white plastic type. Check with your Kendall/Hunt representative or your preferred science supplier. You will need two per group.

For 0.6 M HCl, dilute the 6 M HCl used in Chapter 6 ("Fibers") 1:10; that is, 1 ml 6 M HCl to 9 ml water. Add 3 g of FeCl₃ \cdot 6H₂O to 100 ml water (3 g of the hydrated iron chloride gives about 2 g of the iron chloride itself). You will also use this reagent later in the chapter.

Advance Preparation, continued

For each group, you will need a spatula, four labeled dropping bottles or reagent bottles with Beral pipettes for the reagents (include the water), seven labeled containers for the drugs and unknowns, toothpicks for mixing, and a grain of rice.



Procedure

Do not write in your textbook. Take notes in your science notebook.

 In the first horizontal row of the spot plate, place a small amount of aspirin (no bigger than a grain of rice) into each of the three depressions. Label the row.

Laboratory Activity 7.1, *continued*

- 2. Repeat step 1 for each of the other five powders. Label each row.
- 3. Examine each of the powders, noting color and texture. Make a data table and record your observations on it.
- 4. Examine each of the powders under the microscope. Record your observations.



Spot plate

- 5. Add five drops of distilled water to each powder in column #1 of the spot plate. Record your observations.
- 6. Add one drop of universal indicator to each of the depressions in column #1. Use a different toothpick to stir each one. Record the color and pH of each powder. Note whether the substances are acidic or basic.

- 7. Add two drops of HCl to each of the powders in column #2. Record your observations.
- 8. Add two drops of the ferric chloride solution to each of the powders in column #3. Use a different toothpick to stir each one. Record your observations.
- 9. Carefully discard all solutions into the sink and rinse the plate with water.

Analysis Questions

- Aspirin (acetylsalicylic acid), the most widely used drug in the world, is usually taken as a pain reliever. An acidified solution of ferric chloride can be used to detect the presence of aspirin in an unknown powder. The aspirin hydrolyzes to form acetic acid, and the ferric ion reacts with the salicylic acid to form a compound of what color?
- 2. Acetaminophen, a widely used pain reliever, is not acidic; therefore, it is often taken by people who cannot tolerate aspirin. How can you tell that a sample contains acetaminophen?
- 3. Antacids are slightly basic compounds used to treat hyperacidity, too much hydrochloric acid in the stomach. Many of these products contain carbonates that react with or neutralize the acid in the stomach to produce a salt, water, and carbon dioxide gas. How can you tell that a sample contains an antacid (sodium bicarbonate)?
- 4. Alka-Seltzer contains sodium bicarbonate, citric acid, and aspirin; it reacts with water to produce carbon dioxide gas. How can you tell that Alka-Seltzer is present in a sample?
- 5. Excedrin is a mixture of aspirin, acetaminophen, and caffeine. What would be a good test for Excedrin? Would you need more than one test?
- 6. Use what you know about color spot tests to identify unknown powders #1 and #2.

Drug possession defendant Christopher Jansen, on trial in Pontiac, Michigan, said he had been searched without a warrant. The prosecutor responded that the officer didn't need a warrant because a bulge in Jansen's jacket could have been a gun. Nonsense, said Jansen, who happened to be wearing the same jacket that day in court. He handed it over so the judge could see it. The judge discovered a packet of cocaine in the pocket and laughed so hard he required a five-minute recess to compose himself.

Answers to Analysis Questions

- 1. The universal indicator turns purple; ferric chloride turns violet.
- 2. The universal indicator turns green; ferric chloride turns violet.
- There will be no reaction with ferric chloride, the universal indicator turns greenish blue, and an addition of HCl fizzes.
- **4.** The addition of water causes fizzing.
- **5.** Excedrin reacts the same as aspirin and acetaminophen.
- **6.** You can use any of the powders to make unknowns for student analysis.

Laboratory Is It Ibuprofen? Activity 7.2

Procedure Note

Have students set up a test protocol. They don't know if the sample is one of the drugs they have tested or if it is ibuprofen. They haven't tested a known ibuprofen yet, so that must be done first. Then their results can be compared to those from the previous color tests.

It turns out that ibuprofen shows no reaction with any of the three reagents. Here is a case in which no result (that is, no reaction) is a result; it defines ibuprofen in this particular field of tests.

Different investigative groups of students can be assigned different unknowns; just add more canisters to the passenger's suitcase.

You may want the students to weigh the sample and determine the penalty for possession. The students should realize that a confirmatory test must be performed subsequently.

At the international arrivals area of the Detroit airport, a random inspection of a passenger's suitcase reveals a film canister containing a white powder. The passenger indignantly says it is ibuprofen, powdered for quick dissolving as a headache remedy, and that he never travels without it. Customs agents need to have a quick spot test done because they don't know whether to call the DEA or let the man go. You run a quick test. What do you advise them to do?



may occur in patients allergin

WARNING: ASPIRIN SENSI TIVE PATIENTS. Do not take this product if you have had a severe allergic reaction to aspirin, e.g asthma, swelling, shock o hives, because even though this product contains no aspirin o salicylates, cross-reaction:





Discuss with your teacher how you would solve this problem. Write a procedure and run the tests. Remember, you must support your conclusions.

Thin-Layer Chromatography

Forensic scientists use several types of confirmatory tests based on chromatography.

Chromatography is a way to separate the components of a mixture. There are many types of chromatography: gas chromatography (GC), liquid chromatography (LC) and highperformance liquid chromatography (HPLC), paper chromatography, and thin-layer chromatography (TLC).

In all types of chromatography, there is contact between a stationary phase and

a mobile phase, which cannot be mixed. The sample is carried through the chromatographic system by the mobile phase and continuously interacts with the stationary phase. The interactions of each component in the sample are based on their physical and chemical properties.

Separation takes place as each component is repeatedly adsorbed on or desorbed from the stationary phase at different rates. Think about an oily mixture of spheres and cubes poured onto a slanted plane. Which will land at the bottom first, the spheres or the cubes? Why? The components of this mixture were separated because of their shape. What is the mobile phase? What is the stationary phase?

In the stationary phase of TLC, a thin layer of adsorbent particles is attached to an inert substrate. In the following activity, you will use silica (SiO_2) on plastic, aluminum, or glass. A small amount of a sample is applied ("spotted") near the bottom of the plate, and the plate is placed in a beaker

so that the mobile phase can be drawn up the plate by capillary action. Each part of the sample that is soluble in the mobile phase will interact differently with the stationary phase, creating individual areas or spots on the plate (see Figure 7.2). Often the spots are colorless, and you must develop them to make them visible, as with latent fingerprints.

The retention factor, R_f , of each component is an individual characteristic used to



Spotting a TLC plate

Chromatography comes from the Greek *chroma*, color, and *graphe*, writing. A chromatogram is the record of the separation.





compare the components in the various samples. You can find the R_f values for various substances in reference manuals. In your lab, you simply want to compare the R_f values of the suspected powders with the known standards:

 $R_{f} = \frac{\text{distance from origin to spot}}{\text{distance from origin to solvent front}}$

7.1: "The Drugs Made Me Do It"

What are the legal aspects of violent behavior associated with the ingestion of prescribed drugs? Let's use Prozac as an example.

Prozac was introduced in 1987 by Eli Lilly & Company as an antidepressant. It is one of a type of antidepressants known as SSRIs (selective serotonin reuptake inhibitors), which includes Paxil, Zoloft, Luvox, Celexa, and Effexor. Prozac became so popular that 4.5 million Americans had taken it by 1992.

In a small percentage of the population, side effects included suicide, violence, and other criminal acts. Sometimes the behavior can be quite bizarre. A man taking Prozac as well as the tranquilizer Xanax robbed the bank where his wife worked, disguised with only a fake mustache. He drove off from the front of the bank in his easily identifiable vintage automobile in a hail of bullets. At trial, the judge attributed this irrational behavior to the drugs and found him not guilty.

Indeed, a claim of involuntary intoxication can be admitted under the law in many states as a mitigating factor in a criminal case when there is no history of drug abuse or prior violence. This does not excuse the offense, but it allows the judge to reduce the sentence, advise psychiatric help, or even entertain a plea of temporary insanity. 'PROZAC MADE OUR FATHER KILL OUR MOTHER THEN HIMSELF'

COURT FINDS PROZAC AND XANAX CAUSE CRIMINAL CONDUCT PROZAC'S * ROLE IN * MAUI DEATHS GOING TO COURT

PROZAC BLAMED FOR YOUTH'S MURDER SPREE

CASE STUDY



Teacher Note

This case study can be used to open a discussion on responsibility and culpability of criminal acts performed under the influence of drugs, including alcohol. You can refer back to the section on the insanity defense (pages

23–24 in Chapter 1) to lead an openended discussion.

Qualitative Analysis by Thin-Layer Chromatography (TLC)

While working quietly at his desk, N. Ron Leigh, an executive of a multinational energy company, was stricken with what appeared to be a severe allergic reaction. His throat swelled to such an extent that he could hardly breathe. Rapid and effective response to a 911 call saved his life. Both the paramedics and the police wondered what might have brought on such a sudden attack. A bottle of Tylenol was found on Mr. Leigh's desk. It was well known that he suffered from asthma, bouts of sinusitis, and, lately, tension headaches.

Several caplets from the Tylenol bottle have been submitted to your lab for analysis with the thought that perhaps they were not Tylenol but that someone had substituted an aspirin or ibuprofen product, hoping to cause a severe allergic reaction.



- 1. Pulverize (pound into powder) a tablet from each of the following known standards:
 - aspirin No-Doz Tylenol Excedrin Motrin

Laboratory Activity 7.3

Advance Preparation

Aspirin is a good choice for the questioned material ("Tylenol"). Students can then discuss how the crime could have been committed and how to pursue the investigation. Use Table 7.1 on page 174 to pick out drugs with only one active component; for example, Benadryl could be used as the diphenhydramine standard.

You can make capillaries by drawing out a Pasteur pipette over the flame from a Bunsen burner. It takes a little practice, but it is fun. It is important to have a clean, even cut on the end of the capillary (scratch with the edge of a file, grasp each side of the scratch, and snap), or the stationary phase will get scratched off the supporting plate. *Be sure to protect your hands with thick gloves or a towel when snapping the glass.*

Use only a few crystals of iodine in an 800-cc or 1-L beaker with a cover; 2 percent FeCl_3 solution is used as in the previous activity.

Use a mortar and pestle to pulverize the pills; a metric ruler to measure the chromatograms; filter paper to line the beaker; six 10×75 -mm test tubes per group; openended capillary tubes (both ends open) such as ones sold by Flinn; and an 800-cc or 1-L beaker for developing the chromatogram, with a watch glass or equivalent cover.

You will need a black light (ultraviolet light) with both short and long wavelengths. You will use the short wavelength (254 nm) for this lab. Lamps with both short and long wavelengths are available from a rock shop or places such as Central Scientific or Cole Parmer.

Caution: Ultraviolet light can damage your eyes. Do not look directly into any UV light source!

Reminder

The known sample originates from a verifiable or known source; it can also be called the exemplar in forensic science.

The questioned, or unknown, sample has been collected from a known location but is of unknown origin.

Reminder

Elute means to separate one material from another with a solvent.

Reminder

A fluorescent material absorbs light of a shorter wavelength (often in the ultraviolet) and gives off light of a longer wavelength (often in the visible part of the spectrum).

Procedure Notes

For an eluting solvent, use ethyl acetate (25 parts by volume), ethanol (1 part), and glacial acetic acid (1 part). You will probably need about 30 to 50 cc per developing tank (beaker).

Wear lab apron, chemical safety goggles, and
 disposable lab gloves. Avoid inhalation, ingestion, and skin contact with chemicals.

The TLC sheets (silica gel on plastic or aluminum backing) can be obtained from many scientific supply houses such as Carolina Biological Supply Company (www. carolina.com) in 2.5×7.5 -cm sheets. Flinn sells single 20×20 -cm sheets, but they are not fluorescent. Each sheet can be cut with scissors

Laboratory Activity 7.3, *continued*

- 2. Pulverize a tablet from the Tylenol bottle on Ron's desk, the questioned sample.
- 3. Label six 10 imes 75-mm test tubes.
- 4. Add a small amount, equivalent to a grain of rice, of each sample to a test tube.
- 5. Add five drops of methanol to each sample.
- 6. Add five more drops of methanol to the known aspirin and Tylenol samples.
- Agitate each test tube for a minute or so. *Hint:* To agitate a small test tube without putting a stopper in it, simply hold its neck between your thumb and forefinger and flick it repeatedly with your middle finger. Be careful not to splash out the contents. The methanol will dissolve the active ingredients and leave behind the starch, cellulose, and other products used as "fillers" to form the pills.
- 8. Prepare a 10×10 -cm TLC sheet by lightly drawing a pencil line across it 1 cm from the bottom. Lightly mark six tick marks equally spaced along the line for each sample, and label.
- 9. Dip an open-ended capillary tube into each of the sample test tubes.
- 10. Gently spot each sample on the appropriately labeled mark. Be careful not to scratch the silica coating.
- 11. After each spot dries, repeat the spotting two or three times. You can make the spots dry faster by placing the TLC sheet on a hot plate set on "low."
- 12. The eluting solvent has already been prepared. Pour up to ½ cm into an 800- to 1,000-ml beaker. Do not put enough solvent in the beaker to cover any of the spots. You can line the beaker with

filter paper, as in Figure 7.3. Separation is quicker in the solvent atmosphere provided by the soaked filter paper.

Two TLC plates, hinged together at the top with tape, form a stable tent to place in the beaker. Cover the beaker and allow the solvent to rise to within 1 cm of the top. This should take about 15 to 20 minutes. Then remove the sheets, mark the solvent front, and let the chromatograms dry.



Figure 7.3 Lining a beaker with filter paper

- 13. Look at the chromatogram under short-wave ultraviolet light. Outline each spot that you see. The TLC plate being used in this experiment has a fluorescent substance added to the silica. The spots on the chromatogram block the fluorescence. *Caution: Do not look directly into the light; it can damage your eyes*.
- 14. To develop the spots so they are visible, put each plate in a beaker with a few crystals of iodine. Cover the beaker and watch to see what happens.

Laboratory Activity 7.3, *continued*

Note which spots become totally cloudy. *Caution: lodine vapor is poisonous; be careful.* After using iodine to make the spots visible, blot each spot with a paper towel wet with FeCl, solution. Write down all your observations in your lab book and draw the chromatograms. Calculate the R, values and present your data in a table.

Analysis Questions

- 1. What can you say about the identity of the questioned material? Support your conclusions.
- 2. What is the mobile phase in this experiment?
- 3. What is the stationary phase in this experiment?
- 4. What is meant by an eluting solvent?

ADVANCED TOPICS in the Analysis of Drugs

A **plasma** sample taken from N. Ron Leigh has been submitted to the lab for analysis. Did Mr. Leigh actually take aspirin, and if so, how much?

The salicylate ion that comes from aspirin forms a violet or slightly red complex with the Fe(III) ion. The intensity of the color is directly related to the concentration of salicylate, and thus aspirin; it can be measured using a UV-VIS spectrophotometer (Spec 20 or equivalent).

Thus, with the proper information, you can determine the number of aspirin tablets taken by Mr. Leigh.



Acetylsalicylic acid (aspirin)

plasma: the fluid portion of blood, obtained by centrifuging a whole blood sample

The charge on an ion can be expressed in two ways: Fe(III) or Fe⁺³



electromagnetic spectrum.

Answers to Analysis Questions

- **1.** Answers will vary depending on what the unidentified substance was.
- **2.** the eluting solvent system
- 3. the TLC plate
- 4. the solvent that will separate the components in the analgesics

Fe⁺³ salicylate ion

tetraaquosalicylatroiron (III) complex

from a science lab in zipper-type plastic bags or empty plastic containers so that the custodial staff doesn't get upset when emptying the trash. If the questioned material in

the Tylenol bottle is identified as containing aspirin, then this activity can be continued to

Procedure Notes, continued to make four 10 imes 10-cm

plates. Unfortunately, paper chromatography will not work.

sheet per group.

You will need one 10×10 -cm

It is important to use fluorescent

TLC plates in this lab to visualize the separated spots. lodine

gives a violet color only to aspirin.

Aspirin and acetaminophen will

of methanol are used to extract

these particular standards. Everything can be disposed of down the sink and in the wastebasket. It is always prudent to place powdery substances

spread and tail if too much is present; that is why 10 drops

visualization delineates all the spots but stains only the acetaminophen. The FeCl, solution



Beer's law describes the linear relationship between absorbance and concentration, discovered by August Beer in 1852:

 $A = abc = \log \left[1/T \right]$

where A = absorbance, T = transmittance, a = a constant dependent on the dissolved substance (solute), b = the path length of light (width of cuvette), and c = concentration.

A linear calibration curve is also called a Beer's law plot.



In the following activity, you will prepare a set of standards, that is, solutions of different known salicylate ion concentrations. Each of these will be complexed with an Fe(III) solution, and the amount of light absorbed at a fixed wavelength will be measured. You can then draw a calibration curve (which is more often than not a straight line) relating the intensity of color, that is, how much light the solution absorbs (absorbance = A) to the concentration of salicylate (see Figure 7.4).

You can complex the salicylate in a sample of blood plasma taken from Mr. Leigh at some time after the incident and measure the absorbance. You can then read the concentration from the calibration curve. Acetylsalicylic acid (aspirin) is metabolized in the body to salicylate ion at a known rate:



Thus, with the proper information, you can determine the number of aspirin tablets taken by Mr. Leigh.

The Quantitative Analysis of Aspirin by Spectrophotometry

Laboratory Activity 7.4



Procedure

Do not write in your textbook. Take notes in your science notebook.

Standard solutions have been prepared from sodium salicylate that will yield the following concentrations of salicylate ion when mixed with exactly 5 ml of Fe(III) stock solution:

100 ppm

- 80
- 60
- 40

20

```
0 "Blank"
```

unknown (Mr. Leigh's plasma sample)

- 1. Pipette 5.00 ml of Fe(III) stock solution into each labeled container of salicylate solution. Swirl to mix and develop the color.
- 2. Read the directions on use of the UV-VIS spectrophotometer.

Try different wavelengths to determine the best absorption wavelength for your sample. The salicylate-iron complex looks violet-red because it absorbs all wavelengths of the visible spectrum except those in the violet and red. These colors are transmitted and reflected, and that is what you

Advance Preparation

Preparation of Standard Stock Solutions

- Wear lab apron, chemical safety goggles, and
- disposable lab gloves. Avoid inhalation, ingestion, and skin contact with chemicals. The iron chloride solution will stain clothing.
- Dissolve 0.232 g sodium salicylate (available from Flinn) in 1 L water to make a 200-ppm solution of salicylate ion.
- Dissolve 5.0 g FeCl₃ · 6H₂0 in 1 L water. This is equivalent to 1 g/L of Fe⁺³ ion. The final concentration of Fe⁺³ in all the prepared samples will be 0.05 percent.

The idea is to prepare exactly 5 ml of different salicylate concentrations and have each investigative group add exactly 5 ml of the FeCl_3 stock solution to each in order to develop the color.

100 parts per million (ppm) = 0.01\% = 1 part in 10,000

Only about 5 ml is needed for each absorbance measurement.

- Add 5.00 ml of the 200-ppm salicylate stock solution to a 25- or 30-ml beaker or Erlenmeyer flask. This, diluted with exactly 5 ml Fe(III) stock solution by the students, will make the 100-ppm standard.
- 2. Add 4.00 ml salicylate stock solution to another suitable container, along with 1.00 ml water. This will be the 80ppm standard when diluted with exactly 5 ml of the Fe(III) solution.

Advance Preparation, continued

- 3. Repeat with 3.00, 2.00, and 1.00 ml of the stock solution, adding the required amount of water to make 5.00 ml of solution. These will become the 60-, 40-, and 20-ppm standards.
- 4. The "blank" is prepared by adding 5 ml of water only to the labeled container.

Laboratory Activity 7.4, *continued*

see. To choose the best wavelength for absorption by the complex, you will have to scan one sample at different wavelengths, looking for the particular wavelength that gives the maximum absorbance.

Remember that the spectrophotometer responds differently when the wavelength changes, so you must reset the display to 0.00A with the blank solution every time you change the wavelength.

- 3. Choose a wavelength of 450 nm. Fill a cuvette (test tube specifically for use with the spectrophotometer) half full with the blank solution. Wipe the test tube with a tissue to remove liquid, dust, and fingerprints.
- 4. Put the test tube in the sample compartment, lining up the guide mark on the tube with the guide mark at the front of the sample compartment.
- 5. Close the lid of the sample compartment.
- 6. Adjust the display to 0.00A by turning the %T/A selector knob.

Look at the color wheel; if the solution appears violetred, then the light of blue, green, yellow, and orange wavelengths is absorbed. The absorption maximum will be in the complementary green-yellow part of the visible spectrum, barring other absorbers.

See http://science.csustan.edu/tutorial/color/index. htm about colors, absorption, and chromatography.



- 7. Take the blank out of the sample compartment and save it.
- 8. Fill another test tube with the 60-ppm solution, wipe it off, and put it in the sample compartment, lining up the guide marks.
- 9. Close the lid of the sample compartment.
- 10. Record the absorbance, *A*, from the display.
- 11. Take the test tube out of the compartment and save it for subsequent measurements.
- 12. Choose 470 nm as the next wavelength, and repeat steps 3 through 7.
- Put the 60-ppm solution in the sample compartment again and line up the guide marks. Repeat step 9 and so on every 20 nm to 550 nm.
- 14. Plot A versus wavelength (λ) for an absorption spectrum. Estimate the optimum wavelength for measuring the absorption of the salicylate-iron complex. Now you are ready to prepare a calibration curve.

- 15. Set the wavelength on the spectrophotometer to the best one found in the previous exercise.
- 16. Put the blank you used previously into the sample compartment, lining up the guide mark on the tube with the guide mark at the front of the sample compartment.
- 17. Close the lid of the sample compartment.
- 18. Adjust the display by turning the %T/A selector knob.
- 19. Take the blank out of the sample compartment and save it.
- 20. Fill a test tube with the 100-ppm salicylate standard, wipe it, and put it in the sample compartment, lining up the guide marks.
- 21. Close the lid of the sample compartment.
- 22. Record the absorbance, *A*, from the display.
- 23. Take the test tube out of the compartment and put in the one containing the 80-ppm salicylate standard. (Because the wavelength is fixed, you don't need to run a blank each time.)
- 24. Repeat the above steps until all the standards have been measured.
- 25. Plot absorbance (A) of salicylate versus concentration of salicylate, in ppm (see Figure 7.4). This is the calibration curve you will use to determine the concentration of salicylate in the plasma sample taken from Mr. Leigh three hours after he allegedly took the aspirin in the Tylenol bottle.
- 26. Measure the absorbance of Mr. Leigh's plasma sample.
- 27. Using your calibration curve, determine the concentration of salicylate in the plasma sample. Remember that this sample was diluted by a factor of two when the Fe(III) solution was added.



Teacher Note

The optimum wavelength for absorption by the complex is around 530 nm.

The Metabolism of Aspirin

The **metabolism** of acetylsalicylic acid takes place rather rapidly in the body; you can follow this process by sampling urine or blood plasma at different times after ingestion. Figure 7.5 shows how the relative

metabolism: the sum of all chemical processes occurring in an organism. Metabolites are organic molecules involved in the process of metabolism; they can form more complex molecules, or they may result from the degradation of compounds. concentration of salicylate ion changes over time in an average adult male.

You must adjust the concentration of salicylate you found in Mr. Leigh's plasma to correct for the time because some of it has been metabolized to products other than salicylate. Use the graph in Figure 7.5 to determine the maximum amount of salicylate that was in Mr. Leigh's plasma. Record your result.

The number of aspirin tablets that would have produced the maximum amount of salicylate found in the plasma sample can be estimated from Figure 7.6. Calculate how many aspirin tablets Mr. Leigh took. Does this number make sense?

Write a report analyzing the incident with N. Ron Leigh. Your report should include a summary of what happened as well as a brief description of the laboratory tests and the results and your opinion about those results. This report may be used as evidence if this case comes to trial, so it must be defendable in court.



Figure 7.5 Changes in plasma salicylate concentration with time



Figure 7.6 Relationship between plasma salicylate and ingested acetylsalicylic acid

Testing for Marijuana

A **presumptive test**, or screening test, for illicit drugs is based on spot tests that produce specific colors for specific drugs. The most commonly used test for marijuana is the Duquenois-Levine test, developed in 1941, which creates a purple color when the active ingredients of marijuana are present in a sample.

Detecting Marijuana

A police officer pulls a car over for a minor traffic violation. The officer thinks she smells marijuana in the car. A search uncovers a plastic bag containing plantlike material stuffed under the front seat. The occupants of the car insist it is "stuff used to make incense, like oregano and cloves" and is strictly innocent.

A sample has been submitted to your laboratory for preliminary analysis. Does it contain marijuana?



Laboratory Activity 7.5, Advance Preparation

Wear lab apron, chemical safety goggles, and

 disposable lab gloves. Avoid inhalation, ingestion, and skin contact with chemicals.

Obviously, a real sample of marijuana cannot be used for this exercise unless someone with a DEA Class I drug license and local authorization supervises. Some brands of green tea come pretty close to duplicating a positive Duquenois-Levine test. However, it is easier and simpler to simulate the test reagent on simulated marijuana. So make up your samples by adding

> oregano, cloves, and anything else that comes to mind to the simulated marijuana.

You will need four or more 10 \times 75 test tubes per group, hexane, potassium iodide (KI), ferric chloride (FeCl₃ · 6H₂0), tea, oregano, and cloves.

Laboratory Activity 7.5

presumptive test: test

that presumes the presence of the

questioned substance; also called a

screening test. A blue-purple color

in the test for marijuana can also

be caused by other substances. In

such a case, it is a false positive.

confirms that there is no marijuana

A negative test result, however,

in the sample.

Advance Preparation, continued

Soak the tea in about 10 percent FeCl₂ solution (add about 15 g to 100 ml water). Filter and dry. This is the "marijuana." The "Duquenois reagent" is merely a 2 percent solution of KI: 2 g KI in 100 ml H₂O. (The real reagent is made up of acetaldehyde and vanillin in ethanol.) The Fe⁺³ oxidizes the I⁻¹ to I₂, imparting a purplish color to the solution. The I_o is more soluble in hexane than in water, and so gives the characteristic red-violet color to the top hexane layer. Sometimes it takes a few minutes to develop the color.

 $Fe^{+3} + I^{-1} \rightarrow Fe^{+2} + I_{2}$

Advance Preparation, continued

If the "marijuana"/KI mixture is light brown, the tea did not absorb enough $FeCI_3 \cdot 6H_2O$. Too dark a color indicates that too much Fe^{+3} was absorbed in the tea; dilute it with fresh material.

After the class has completed the experiment, combine all the material from the test tubes, filter out the solids, and allow the liquid portion to separate. Pour the bottom, aqueous layer down the sink. The hexane can be poured into a shallow vessel and allowed to evaporate in a well-ventilated spot, preferably a fume hood.

immiscible:

describes materials that do not mix. When shaken, two immiscible liquids will separate to their original volumes.

Dumb Crook

Forty-five-year-old Amy Brasher was arrested in San Antonio, Texas, after a mechanic reported to police that 18 packages of marijuana were packed in the engine compartment of the car that she had brought in for an oil change. According to police, Brasher later said that she didn't realize that the mechanic would have to raise the hood to change the oil.

Materials • "Duquenois reagent" test tubes (simulated) hexane • FeCl_a \cdot 6H_aO oregano potassium iodide (KI) cloves alleged marijuana • tea marijuana standard ° **SAFETY ALERT!** CHEMICALS USED Always wear goggles and an apron when working in the labaratory SAFETY NOTE Also wear disposable lab gloves. Avoid inhalation, ingestion,

Laboratory Activity 7.5, *continued*

Procedure

Do not write in your textbook. Take notes in your science notebook.

and skin contact with chemicals.

- 1. Place about 2 to 3 mm of the suspect material in a 10×75 -mm test tube. Add just enough "Duquenois reagent" to cover the material, then add about 2 to 3 mm more. Note the color.
- 2. Add a volume of hexane equal to one-half the volume of the Duquenois reagent in the test tube, and shake it for a minute. A red-purple color in the aqueous phase as well as in the **immiscible** (hexane) layer at the top of the test tube indicates the presence of THC, the active ingredient of marijuana.

You should also test a control sample or standard of known marijuana (simulated in this case) exactly the same way, as well as each of the alleged ingredients of the "incense," oregano and cloves.

3. Prepare a brief report of your results and justify your conclusion.

Spot tests like this are used just for screening. Negative results mean that the drug you are testing for is *not* present. There are, however, substances that can cause a false positive, so you should do another test to confirm so it will be conclusive as evidence. The presence of marijuana can be confirmed with a microscopic examination, if possible, or thin-layer chromatography (TLC).

Drug Tests That Use Color

As with the Duquenois-Levine test for marijuana, you can use a number of spot tests to determine the presence or absence of a particular drug or type of drug. A positive result implies the presence of the drug, but these tests are not conclusive, and they must be confirmed by a more specific test to rule out false positives.

Table 7.2 below summarizes the more common spot tests used by law enforcement agencies.

Table 7.2: Color Tests for Drugs			
Drug	Reagent	Color	
marijuana	Duquenois-Levine (D-L)	blue-violet	
LSD	Erlich/Van Urk (ERL)	blue-violet	
amphetamines	Marquis (MARQ)	red-orange $ ightarrow$ brown	
cocaine	cobalt thiocyanate (CO)	blue flaky precipitate	
heroin	Marquis (MARQ)	purple	
barbiturates	Dille-Kopanyi (D-K)	violet	

Presumptive Color Tests for Drugs

This activity will give you some practice with testing samples for the presence of drugs.



Laboratory Activity 7.6, Advance Preparation

Wear lab apron, chemical
safety goggles, and
disposable lab gloves. Avoid
inhalation, ingestion, and
skin contact with chemicals.

Simulated Test Reagents

Erlich (ERL) contains Fe^{+3} . Make up a 2 percent solution of $FeCl_3 \cdot 6H_2$ O by adding 3 g to 100 ml, or use what you made up for the spot test lab. The real Erlich solution for the detection of LSD contains paradimethylaminobenzaldehyde in ethanol and HCI.

Marquis (MARQ) is the same as Erlich, above. The real Marquis reagent is nasty stuff—it is formaldehyde in concentrated $H_2SO_{a^*}$

Cobalt thiocyanate (CO) is actually used in drug testing for cocaine and PCP. It is a 2 percent aqueous solution. The compound can be purchased from Aldrich

Laboratory Activity 7.6

Advance Preparation, continued

(www.sigmaaldrich.com/Brands/ Aldrich.html), but it is quite expensive. Make your own: Add 2.7 g cobalt chloride hexahydrate, $CoCl_2 \cdot 6H_2O$, and 2.2 g potassium thiocyanate (KSCN) to 100 cc water. This gives approximately a 2 percent solution of cobalt thiocyanate, $Co(SCN)_2$. If you happen to have on hand only other cobalt and thiocyanate salts, work out the quantities stoichiometrically.

Dille-Kopanyi (DK) is 1 percent 1-naphthol dissolved in ethanol. The 1-naphthol is available from suppliers such as Flinn. The real Dille-Kopanyi reagent is made from cobalt acetate in methanol and a little acetic acid. It is a two-step reaction as simulated, but the second step uses isopropylamine in

Advance Preparation, continued

methanol rather than concentrated sulfuric acid (H_2SO_4). *Caution: Concentrated sulfuric acid is a strong acid and is severely corrosive to eyes, skin, and other tissue.* Put a little more than what you need in a dropper bottle, suitably labeled, and place it in a plastic tray.

Simulated Illicit Drugs

"LSD": The basis for this reaction is the formation of Prussian blue by combining Fe^{+3} (ERL) with the ferrocyanide ion:

 $\begin{array}{c} 4 \; \mathrm{Fe^{+3}} + \; 3 \; \mathrm{Fe(CN)}_6 \rightarrow \\ \mathrm{Fe_4[Fe(CN)_6]_3} \end{array}$

Prepare about a 5 percent solution of potassium ferrocyanide, K,Fe(CN),; put 5 g in 100 ml water

Reminder

A *metabolite* is a by-product of a chemical reaction in the body. For example, over 31 metabolites of marijuana can be detected in urine.

enzyme: a protein that acts as a catalyst in a living organism

antibody: a protein produced by a body's immune system that tags a molecule in order to destroy it

EMIT: an enzyme immunoassay test to detect particular types of metabolites in body fluids, commonly used for screening drug use

Reminder

A false positive is a test result that shows a substance is present, when it is not. That is why confirmatory tests are required.

Laboratory Activity 7.6, *continued*

Procedure

Do not write in your textbook. Take notes in your science notebook.

- 1. Put a small amount (the size of a grain of rice—no more!) of each simulated drug in the cavity of a white ceramic or plastic spot plate, according to the diagram shown in Figure 7.7. The LSD sample is the only one impregnated on paper, so some of the tests are not required, as indicated by the X in the circle.
- Add one drop from the labeled bottle of reagent to the edge of each sample. Mix if necessary. Note and record any changes, initially and over a period of 15 minutes. The D-K test for barbiturates is a two-step process: Add the D-K reagent; then add one drop of concentrated sulfuric acid.

Caution: Concentrated H_2SO_4 is a strong acid; it is severely corrosive to skin, eyes, and other tissue. Wear safety goggles!



There are a number of new preliminary or screening tests now being used to detect the use of major classes of illicit drugs. Most of these use urine samples. They are based on competition between a drug metabolite, an **enzyme**-labeled metabolite, and a specific **antibody**. These tests result in a color reaction that is proportional to the amount of drug present. The Enzyme Multiple Immunoassay Test (**EMIT**) is simple to administer and inexpensive. It is even sold for home use; however, it is subject to many false positives.

Confirmatory Tests

In court, forensic scientists are often required to explain the testing method to the jury, so now you will learn about the more sophisticated analytical techniques for testing drugs.

Most **confirmatory tests** for drugs are based on one of two analytical techniques, infrared spectroscopy and mass spectrometry.

Spectroscopy is the interaction of electromagnetic radiation with matter. In infrared (IR) spectroscopy, material absorbs energy in the near-IR region of the electromagnetic spectrum (Figure 7.8).

This range or band of energies is just enough to make the bonds that hold the atoms of a molecule together bend and stretch. Each bond, or group of bonded atoms, has a characteristic excitation energy. So, when a band of near-IR light passes through a substance, certain energies, frequencies, or wavelengths are absorbed by the many different molecular bonds. Comparing the IR light beam before and after it passes through a transparent sample results in a transmission or an absorption spectrum. The IR spectrum of each substance is a combination of the many different bonds that make up that substance, so it's not surprising to find that its IR spectrum gives a unique view of the substance, much as a fingerprint is unique to a single person.





The equations E = hv and $v = hc/\lambda$ describe the relationships between energy (E), frequency (v), and wavelength (λ). h is a proportionality constant and c = speed of light. IR spectroscopists measure the energy absorbed by molecular vibrations as $1/\lambda$, called wave numbers and measured in cm⁻¹.

Advance Preparattion, continued

and wipe it on a poor-quality paper that will absorb some of it (photocopier paper will do). Dry it and cut it up into small squares, simulating a "hit" on each one. It looks more authentic if you can print out on your computer a page

of symbols or designs.

For "amphetamines," use potassium iodide, KI. It generally is somewhat crystalline, so grind it with a mortar and pestle. With the FeCl₃ solution (MARQ), it forms a deep orange that darkens with time, just as in the real

test for amphetamines.

confirmatory tests: tests

spectroscopy: the branch of

science that involves the study of

electromagnetic radiation and its

interaction with matter

that specifically identify one

substance

Your simulated "cocaine" is Benadryl. Dump the white powder from a capsule into your labeled container. This particular screening test for real cocaine is replete with false positives. It is the reaction of the antihistamine that causes a beautiful blue precipitate with the $Co(SCN)_2(CO)$, which does not easily wash off the spot plate without an abrasive soap.

The "heroin" can be aspirin, or use the sodium salicylate required for the quantitative analysis on page 185 (Laboratory Activity 7.4). The Fe^{+3} (MARQ) forms a violet complex as described.

"Barbiturates" are powdered sugar. A nice violet color develops upon the addition of the concentrated H_2SO_4 in the second step (DK).

The spot plates can be washed in the sink. Emphasize the problem of contamination, which can lead to false results. Spatulas should be wiped clean between uses; the tip of the eyedropper or pipette should never touch the powder; spills should be reported and the possibility of contamination assessed. *It is recommended that safety glasses be worn for this lab.*

The spot test results are shown below and represent the positive test colors for the real drugs: NR indicates no change in color (that is, no reaction). The shaded circles are the positives for the drugs indicated. See Table 7.2 on page 191.

Drugs 193



Figure 7.9 shows IR transmission spectra of ethyl alcohol and several other two-carbon molecules. Note the difference in their spectra.

The abscissa (*x*-axis) in the IR diagrams is a function of the energy of the IR radiation, measured in wave numbers. The ordinate (*y*-axis) measures the amount of IR energy

transmitted through the sample, that is, energy that is not absorbed. The molecular structure determines the position of the peak. For example, the "peak" at 1,050 cm⁻¹ in ethanol (ethyl alcohol) is caused by absorption of energy from stretching of the carbon–oxygen–hydrogen bond in the molecule. You can confirm identification of a substance by comparing the IR spectrum of an unknown sample with that of a known compound.



Source: SDBSWeb: http://riodb01.ibase.aist.go.jp/sdbs/ (National Institute of Advanced Industrial Science and Technology, 2005) Figure 7.9 IR spectra of C, compounds

The IR spectrum of unknown liquid 1 is shown below. Is it ethanol, acetaldehyde, or acetic acid?



Teacher Note

peak at 3,000.

An acid: There is a large peak at

about 1,750 cm^{-1} and a broad

Ethanol



Source: SDBSWeb: http://riodb01.ibase.aist.go.jp/sdbs/ (National Institute of Advanced Industrial Science and Technology, 2005)

What is the nature of unknown liquid 2, whose IR spectrum is shown below? Is it an alcohol, an aldehyde, or an acid? Justify your answer.



Source: SDBSWeb: http://riodb01.ibase.aist.go.jp/sdbs/ (National Institute of Advanced Industrial Science and Technology, 2005)

The basic instrument now in use for taking IR spectra is the Fourier Transform Infrared (FTIR) spectrophotometer. A good one costs about \$25,000. A simple schematic of an IR spectrophotometer is shown in Figure 7.10.

Molecules can be identified through mass spectrometry by breaking them apart into pieces and then measuring the quantity of the different pieces. Each molecular compound has its own unique fragmentation pattern or spectrum. You can identify this pattern by comparing it to a catalog of known spectra. The mass spectra of ethanol and similar two-carbon molecules are shown in Figure 7.11. Note the differences in their spectra.



Teacher Note

In the incident at the Detroit airport already described, if the contents of the canister test positive for amphetamines, cocaine, or barbiturates, have the students confirm preliminary identification by comparing the IR spectra. Appendix D contains a spectral library of 12 known

> substances including two different amphetamines, cocaine, and two barbiturates. Use any of these as the unknown by photocopying them with the identification covered. Note that



Figure 7.11 Mass spectra of C₂ compounds

Teacher Note, continued

appendices are at the back of the book and can also be found on the Teacher Resource CD. The mass spectrometer works by bombarding the vaporized sample with electrons, which have enough energy to break a molecule apart. Those

fragments that have become positive **ions**



ions: atoms or molecules that have lost or gained one or more electrons and, thus, have a net positive or negative charge

FTIR spectrophotometer

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(that is, the fragments that lose an electron and, therefore, have a net positive charge) are accelerated by a negative electric field through either a magnetic or oscillating electric field that sorts them according to their

mass-to-charge ratio (m/e or m/z; z or e is usually 1). The resulting signal is a mass spectrum. Each molecular species has its own unique mass spectrum; therefore, a catalog can be created, as with IR spectra.

Three equations describing fragmentation of ethanol are:

$$CH_{3}CH_{2}OH + e^{-} \rightarrow CH_{3}CH_{2}O^{+} + H + 2e^{-}$$
$$CH_{3}CH_{2}OH + e^{-} \rightarrow CH_{3}CH_{2}^{+} + OH + 2e^{-}$$
$$CH_{3}CH_{2}OH + e^{-} \rightarrow CH_{3}^{+} + CH_{2}OH + 2e^{-}$$

```
Atomic Mass Units:

H = 1; C = 12; 0 = 16

To find the molecular mass, or

molecular weight, of CH_3CH_2OH:

2 C \times 12 = 24

6 H \times 1 = 6

1 0 \times 16 = \underline{16}

46 units
```

Note which peak is related to which ion in the spectrum. What other equations can be written to account for the peaks seen on the mass spectrum of ethanol? Account for the major peaks in the other two spectra.

What is the substance whose mass spectrum appears below?



Acetic acid



Source: SDBSWeb: http://riodb01.ibase.aist.go.jp/sdbs/ (National Institute of Advanced Industrial Science and Technology, 2005)

Mass spectrometry works fine in identifying pure substances, just like IR spectrophotometry. However, mixtures are difficult to identify in both techniques because their spectra become superimposed.

Chromatography can separate a mixture into its component parts, but the technique does not specifically identify what the component is. In a gas chromatograph-mass spectrometer (GC-MS), a sample is vaporized and swept through a column containing some sort of adsorbent by using a "carrier gas" (usually helium), and each separated component Illicit drugs are often diluted with other products to cut the purity, making them safer to take as well as increasing the profit for the dealer; examples of materials used to dilute these drugs include quinine, starch, lactose, and lidocaine. Dilution of an illicit drug complicates penalties based on weight of the substance (see Appendix C, for example).



analyte: a substance being analyzed

(or **analyte**) is detected as an electronic signal when it emerges from the chromatographic column. An example of output from a gas chromatograph is shown in Figure 7.12.

Each peak represents a component of the mixture, but the analyte cannot be identified directly. You can identify an analyte by introducing each component coming from the chromatograph into the mass spectrometer. A schematic for a GC-MS is represented in Figure 7.13. A good GC-MS costs about \$60,000.



GAS CHROMATOGRAPH Figure 7.13 Schematic of a GC-MS



Cocaine



Crack cocaine

Checkpoint Questions

Answer the following questions. Keep the answers in your notebook, to be turned in to your teacher at the end of the unit.

- 1. What are the differences among a controlled substance, an illicit drug, a prescription drug, and an OTC drug?
- 2. What is a presumptive test? When can it be useful? What are its limitations?
- 3. How are illicit drugs classified?
- 4. A sample of light brown powder found in the kitchen of an alleged drug house gives a blue precipitate with cobalt thiocyanate. What is it? Is there enough evidence to prosecute?
- 5. What is the electromagnetic spectrum (EMS)? Which end of the EMS is the high-energy part? How is the EMS used in analytical analyses?
- 6. What is the difference between absorbance and transmittance in spectroscopy? What is their relationship to analyte concentration?
- 7. What are the types of chromatography?
- 8. A bag of pills was confiscated from a student. They were small, white, and heart-shaped, possibly with a numeral on one side. Should the student be arrested?
- 9. What are Xanax, Zithromax, and Zocor?
- 10. Why is Ritalin prescribed? Can it be addictive? What common side effects may occur with its prescribed use? What are some symptoms of Ritalin overdose?

Teacher Note

Some answers may come from the presentations on the TRCD. Otherwise, students could use outside sources, such as PDR or Internet.

Answers

- A controlled substance is identified under Federal Schedules of the Controlled Substances Act. An illicit drug is an illegal substance used for pleasure but not necessarily defined in the Federal Schedules. A doctor prescribes a prescription drug for a specific reason; misuse can make it an illicit drug. Over-the-counter (OTC) drugs have no restrictions on purchase, although the use of pseudoephedrine in making meth may curtail access to cold remedies such as Sudafed.
- 2. Presumptive tests determine if a substance is present. If the result is negative, the substance is absent. A positive result could be a false positive and requires a confirmatory test.
- **3.** one such scheme: hallucinogens, stimulants, narcotics, and depressants
- **4.** This could be crack cocaine, but without a confirmatory test there is not enough evidence to prosecute.
- 5. The EMS includes the full range of energies (wavelengths, frequencies) of light, from gamma rays to radio waves. The high part is the longwavelength portion (low energy). Various EMS energies excite molecules and atoms, and the resulting effects are related to what the material whose spectrum is being shown is.

METH BILL RESTRICTING COLD PILLS ADVANCES IN LEGISLATURE

Sales of cold medicines used to make methamphetamine would be restricted under proposed legislation.

The new law would require stores to sell Sudafed, Nyquil, and other medicines containing pseudoephedrine only from behind the pharmacy counter.

—abstracted from the Boston Globe, July 29, 2005

Answers, continued

- **6.** Absorbance is the measurement of the amount of light going into a substance; transmittance is a measure of the amount of light coming out of a substance. Mathematically, they are related by A = abc = log [1/T], where A = absorbance, T = transmittance, c = concentration of the substance (analyte), and a and b are constants.
- 7. The common ones are paper, thin-layer, gas, and liquid; others include high-performance, size-exclusion, and gel.
- **8.** According to the PDR, the pills are Visken, a high-blood-pressure medicine, but even if they were not, they would be confiscated and analyzed before any arrest would be made.
- **9.** Xanax is a tranquilizer, Zithromax is an antibiotic, and Zocor is used to treat high cholesterol.
- Ritalin is prescribed for attention deficit disorder (ADD). The effects of the drug can be addictive. The main side effects can be insomnia and nervousness. Overdose can cause many symptoms, such as delirium, confusion, and an exaggerated feeling of elation (which accounts for illicit use).
- PDR, *Physicians' Desk Reference*; IR, infrared; OTC, over-the-counter; DEA, Drug Enforcement Administration; THC, tetrahydrocannabinol; MARQ, Marquis reagent, presumptive test for amphetamines; GC-MS, gas chromatograph(y)-mass spectrometer(-try); LSD, lysergic acid diethylamide; ppm, parts per million; AMU, atomic mass unit; PCP, phencyclidine; TLC, thin-layer chromatography; NSAID, non-steroidal anti-inflammatory drug; R, retention factor; Co(II) or Co⁺⁺, cobaltous ion.
- **12.** a. pertaining to the amount or quantity, as in quantitative analysis; b. a screening test only, as in a color spot test; c. a range of values, as in the electromagnetic spectrum of light; d. chemical changes in a living body, as in the change of aspirin to other chemicals; e. the amount of light a substance absorbs, as measured in IR spectrophotometry; f. an instrument for measuring absorbance, used to identify drugs; g. a known sample that can be prepared by a chemist for an analysis or evidence of a known origin; h. a standard that can be diluted to prepare more standards; i. a charged particle such as a cobaltous ion; j. a measure of energy of light, such as red light having a wavelength of 0.65 µm; k. an OTC medicine, type NSAID; I. the relative configuration of atoms in a molecule; m. a method of separation of mixtures, such as paper chromatography; n. a test that positively identifies a substance, such as GC-MS for

- **11.** Define each of the following abbreviations:
 - a. PDRf. MARQk. PCPb. IRg. GC-MSI. TLCc. OTCh. LSDm. NSAIDd. DEAi. ppmn. R_r e. THCj. AMUo. Co(II)

12. Define the following terms, with examples:

- a. quantitative
- b. presumptive
- c. spectrum
- d. metabolism
- e. absorbance
- f. spectrophotometer
- g. a standard
- h. a stock solution
- i. an ion
- j. wavelength

- k. Motrin I. molecular structure
- m. chromatography
- n. confirmatory test
- o. analyte
- p. questioned sample
- q. narcotic
- r. allergic reaction
- s. barbiturate
- t. false positive
- 13. You have tested a sample of a white powder with cobalt thiocyanate reagent and obtained a blue precipitate. You think you have ______, but you ask the spectroscopy lab for a confirmation. They provide the following spectrum:



Source: SDBSWeb: http://riodb01.ibase.aist.go.jp/sdbs/ (National Institute of Advanced Industrial Science and Technology, 2005)

What is your sample? (Compare with the spectra in Appendix D at the end of this book.)

14. You get a violet-blue color with Marquis reagent on a sample submitted to your lab. You ask the mass spectroscopy lab to check it for you, and they



16. Infrared spectra of butanol (butyl alcohol) and butanal (butyraldehyde) are given below. Which do you think is the aldehyde? Why?



Source: SDBSWeb: http://riodb01.ibase.aist.go.jp/sdbs/ (National Institute of Advanced Industrial Science and Technology, 2005)

Answers, continued

- cocaine; o. a substance being analyzed, such as a salicylate in plasma; p. a sample of unknown origin, such as a powder on a victim; q. an analgesic, such as heroin; r. abnormal immune response of the body to a substance, such as poison ivy; s. a depressant, such as phenobarbital; t. a positive test result when there is no product present; needs confirmatory test
- **13.** The IR spectrum shows ephedrine, which can cause a false positive. The color spot test is a presumptive test only; infrared analysis is a confirmatory test, and it shows no cocaine.
- **14.** You think you have cocaine, but the spectra confirm quinine (a dilutant or cutting agent) and morphine.
- **15.** Answers to the first part may vary widely. The answer to the second part is mass spectrometry.
- **16.** Comparing the spectra of ethyl alcohol and acetaldehyde on page 194 of this chapter, note that the biggest difference is the absorption of an aldehyde at about 1,500 wave numbers; so the second spectrum is butyraldehyde.

Teacher Note

Appendices can also be found on the Teacher Resource CD (Blackline Masters 7.1–7.9).

 $CH_3 - (CH_2)_3 - OH$

butanol

Answers, continued

17. five years

18. With this amount of cocaine, the man is assumed to be selling, and he could serve 5 to 40 years in prison. If there were just one canister of cocaine, with no other incriminating circumstances, he would probably get a misdemeanor conviction for simple possession with a sentence of no more than one year.

19. yellow

20. You would say that you analyzed different standards—preferably one made up by another lab, or even better, one that has been certified as accurate—and compared these results to the unknown. If applicable, you would have investigated the effect of other materials in the unknown upon your results. You would have also run replicate analyses to check the precision of your method.

Optional Website Activity

Have students conduct the virtual autopsy case that can be found in the Chapter 7 student resources area (SCSI tab) on the *Forensics* website. See the teacher resource section on the site for more information.

- 17. In the "incense" incident described on page 189, about two pounds of the material was retrieved from the car. What would be the maximum sentence if the driver were convicted as a first offense? (See Appendix B.)
- 18. If, in the incident at the Detroit airport, the film canister contained 50 grams of cocaine and 10 more grams were found in the man's camera bag, what could be the sentence if he were convicted as a first offense? (See Appendix C.)
- **19.** What color is a solution of a compound that has an absorption spectrum like the one below?



20. If you were asked in court to assess the accuracy of your analytical results, how would you respond?

Project: Both Sides of the Issue; Legalization of Drugs

Write a paper analyzing the arguments about legalizing drugs in the United States. Billions of dollars have been spent on the "war on drugs." Seventyfive percent of people in prison are there on drug or drug-related crime charges. Do we need more severe drug laws? Would legalization benefit society? Which drugs should be legal? How would the cost of drugs be affected? How would the cost of drugs affect violent crimes? Should the state protect people from harm to themselves? What has history taught us about government control? Can the success or failure of drug laws in other countries help us decide?

So you can gain an understanding of both sides of the issue and get experience in identifying and defending the side of the issue that you disagree with, structure your paper as follows:

TITLE: Would the Legalization of Drugs Benefit American Society?

- AUTHOR: Your name
- INTRODUCTION: Write one or two paragraphs briefly explaining what specific laws would be changed and the controversy surrounding the issue.
- PRO SIDE: Write one sentence saying that legalization of drugs in the United States would benefit society.
- SUPPORT: Write a short statement explaining why society would benefit. Write at least three paragraphs supporting the statement, using at least three different sources.
- CON SIDE: Write one sentence saying that legalization of drugs in the United States would not benefit society.
- SUPPORT: Write a short statement explaining why legalization of drugs in the United States would not benefit society. Write at least three paragraphs supporting the statement, using three different sources.
- PERSONAL OPINION: Write your views and conclusions based on the arguments you have already presented. You must support one side or the other.
- WORKS CITED: List references for all sources that you have used.

Schools add tobacco to list for student drug screening

Critics say tests are an invasion of kids' privacy

Some schools are administering urine tests to teenagers to determine if they have been using tobacco. Random drug testing of students involved in extracurricular activities has been affirmed by the U.S. Supreme Court, but opponents say such tests violate the students' rights.

This article appeared in a local newspaper in October 2002. Do you think it is an invasion of individual privacy to be subjected to random drug testing in school? How about for participating in after-school events? What about for attending after-school events? Do you think that tobacco products should be included in the drug-testing programs? Is tobacco a drug? What constitutional amendment pertains to individual privacy?

Answers

- Certain molecules in the atmosphere allow solar radiation to reach the Earth. As this heat energy "bounces" back into the atmosphere at longer wavelengths (infrared), it is absorbed by greenhouse gases such as water vapor, carbon dioxide, methane, and nitric oxide. Not only do these gases prevent this heat from escaping, but they radiate heat from the absorption. This effect keeps the temperature of our planet livable, but now greenhouse gases are building up to the point where global warming is initiated.
- **2.** Open-ended. The teacher might wish to assign different areas of the EMS to different investigative groups.
- **3.** Open-ended. The teacher might wish to assign different drugs to different investigative groups.
- 4. See news insert, page 171.
- **5.** open-ended; related to the forensics project on the previous page
- 6. This is a nice experiment to do as a time filler.
- 7. Doping is drugging racehorses to excel. It is related to the use of drugs to enhance athletes' performance. Good topic for discussion.

Additional Projects

- 1. How is IR absorption related to the greenhouse effect?
- 2. Research where each area of the EMS originates and how that frequency of radiation can be used by humankind. For example: Ultraviolet light originates from the sun. It causes sunburn and skin cancer. A black light can cause fluorescence in many substances and is useful in forensic investigation.
- **3.** Research the history of opium—its use and the regulations governing it. The history of other naturally occurring drugs such as marijuana, mescaline (from peyote cactus), and psilocybin mushrooms is also interesting.
- **4.** What incident brought the designer drug fentanyl into the news in 2002? What went wrong?
- **5.** Is the illicit drug trade the fault of the supplier or the user? Support your opinion.
- Analyze different brands of aspirin tablets to determine

 (a) the amount of acetylsalicylic acid in each and (b) the
 type of filler. (See "The Chemistry of Over-the-Counter
 Drugs," Flinn Scientific, www.flinnsci.com.)
- **7.** What is meant by the "doping" of racehorses? Is it a problem?

References

Books and Articles

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Websites

- www.drugabuse.gov/NIDAHome.html; source of information on drugs
- www.usdoj.gov/dea/concern/concern.htm; information on many drugs
- www.ojp.usdoj.gov/bjs/dcf/contents.htm; drugs and crime statistics
- www.drugabuse.gov/DrugPages; source of information on drugs
- www.whitehousedrugpolicy.gov/streetterms/Default. asp; index of street names of drugs
- http://science.csustan.edu/tutorial/ir/index.htm; good college-level tutorial with problems on IR spectroscopy
- www.chem.arizona.edu/massspec/intro_html/intro. html; a good tutorial on mass spectrometry
- www.chemistry.nmsu.edu/Instrumentation/GC.html; explanation of gas chromatography
- www.chem.vt.edu/chem-ed/sep/gc/gc.html; good tutorial on gas chromatography
- webbook.nist.gov/chemistry/name-ser.html; database for structures, spectra
- http://riodb01.ibase.aist.go.jp/sdbs/cgi-bin/cre_index. cgi?lang=eng; spectral database for organic compounds
- www.aist.go.jp/RIODB/SDBS/cgi-bin/cre_index.cgi; database for structures, spectra
- www.rxlist.com/drugs/alpha_a.htm; index of drugs
- www.rxlist.com/cgi/rxlist.cgi; prescription drug identification