THE DOSE MAKES THE POISON

Which would you prefer to drink—a cup of caffeine or a cup of trichloroethylene? Chances are good that your response was "caffeine." Caffeine occurs naturally in coffee, tea, and chocolate, and it is added to sodas and other types of drinks and foods. Trichloroethylene, on the other hand, is a solvent used to dissolve grease, and it is also a common ingredient in glues, paint removers, and cleaning fluids. Trichloroethylene does not occur naturally in the environment, but it is sometimes found as a pollutant in groundwater and surface water.

So, which would be better to drink? Believe it or not, caffeine is more poisonous than trichloroethylene. At low concentrations, caffeine is used as a food additive because of its effects as a stimulant—it helps people to stay awake and to feel lively. However, at concentrations higher than those found in food products, caffeine can cause insomnia, dizziness, headaches, vomiting, and heart problems. In studies of laboratory animals, high doses of caffeine have caused birth defects and cancer.

Does this mean you should think twice about reaching for that cup of cocoa or tea? No, there's more to the story than that. What it does mean is that many common substances found in food and drinks are *toxic*, or poisonous, if you eat or drink large enough quantities. The amount of caffeine in a normal human diet does not cause illness, but just 50 times this amount is enough to be fatal.

Toxicity indicates how poisonous a substance is to biological organisms.

Trichloroethylene is less toxic over the short term than caffeine, but it is not harmless. In fact, long-term exposure may cause a variety of health problems, including cancer as well as damage to liver and kidneys.

ANY CHEMICAL CAN BE TOXIC

Any chemical can be toxic if you eat, drink, or absorb too much of it. Even water can kill you if you drink too much too quickly! Back in the early 1500s, a Swiss doctor named Philippus Aureolus Theophrastus Bombastus von Hohenheim-Paracelsus wrote:

> All substances are poisons; there is none which is not a poison. The right dose differentiates a poison from a remedy.

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Topic: trichloroethylene
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Any chemical can be toxic if you eat, drink, or absorb too much of it.

Paracelsus was one of the first people to recognize that a chemical can be harmless or even beneficial at low concentrations but poisonous at higher ones. That is why it is so important to take medicine in the correct dosage. Even vitamin pills can kill you if you swallow too many in too short a period of time. For example, vitamin D is an important nutrient, but it also is a highly toxic chemical. In tiny amounts it is good for you, but taking more than the recommended dose can cause serious health problems, including kidney stones, high blood pressure, deafness, and even death.

ARE NATURAL CHEMICALS SAFER?

Toxins are toxic chemicals created by plants and animals, usually for their own defense.

Synthetic chemicals are made by people rather than nature. They are composed of natural elements such as carbon, hydrogen, nitrogen, and chlorine. We manufacture synthetic compounds to use in a wide variety of products such as cleaners, deodorants, food additives, and pesticides.

Many people believe that chemicals produced by nature are safe and synthetic ones are harmful. They fear that synthetic chemicals will cause cancer and that any exposure to them must be dangerous. It is true that some synthetic chemicals cause cancer, and others are highly toxic. But it also is true that many synthetic chemicals are harmless at doses normally encountered in food, water, air, and other sources.

The same is true for natural chemicals—they range from relatively harmless to highly toxic. Some plants and animals create toxic chemicals called *toxins,* either for self-defense or for assistance in catching their prey. Think about rattlesnakes, scorpions, and poison ivy—each produces a natural toxin that is hazardous to humans as well as to other organisms in the environment.

The _dose_ is the total amount of a chemical that an individual eats, drinks, breathes, or absorbs through the skin.

The distinction between synthetic and natural is not always clear-cut because people can manufacture many chemicals that occur in nature. For example, the vitamin C in an orange is identical to ascorbic acid created in a laboratory. There are additional benefits to eating an orange that you do not get from taking a vitamin C tablet, but the vitamin itself is identical from either source.

HOW MUCH IS TOO MUCH?

To measure a chemical's short-term toxicity, scientists carry out something called a "dose/response" study. The word *dose* refers to the total amount of a substance to which an individual is exposed through the mouth, lungs, or skin. Your total dose of a chemical includes the amount of the chemical that you eat, either by itself or contained in food or drinks, and the amount that you inhale with the air you breathe. It also includes absorption through your skin, which could happen if the chemical were dissolved in your bath water or included in your shampoo or skin care products. All of these sources together make up your *exposure* to the chemical.

Exposure means coming in contact with a chemical—through food, water, air, or other sources.

Exposure to a toxic chemical can be either intentional or unintentional. For example, a person who chooses to swallow too many pills is taking an intentional overdose. Someone who accidentally becomes poisoned by eating contaminated food receives an unintentional overdose. Similarly, a smoker intentionally inhales whatever substances are contained in cigarette smoke, whereas nearby people get exposed unintentionally when they inhale second-hand smoke.

The word *response* refers to the changes in living things caused by exposure to a specified chemical or mixture. Typically, the higher the concentration of a toxic compound, the more powerful its effect. Scientists study this relationship by carrying out dose/response experiments to determine the response of laboratory organisms to various doses of a test chemical.

Scientists measure *response*, the biological changes in living things caused by exposure to a toxic substance.

Dose/Response Bioassays

Dose/response experiments are called *bioassays* (the word *assay* means test, and *bio* is short for biological). For any given chemical, the question is, "How much is too much?" At low enough doses, the test organisms are not harmed and may even benefit. At high enough doses, they all die. For each chemical, there is an intermediate range in which some individuals will be affected and others will not.

A *bioassay* uses living things to determine chemical toxicity.

In a typical dose/response bioassay, laboratory rats are each fed a single dose of the chemical being tested. Some rats get an extremely high dose, and others receive doses ranging from moderate to very low. Exposure to the chemical occurs only on the first day, but the experiment continues for 14 days in order to give the organisms time to react. At the end of this period, scientists count the number of dead rats and note any health-related responses in those that are still alive. At the highest dose, it is likely that all of the rats will have died. At the lowest dose, most of the rats probably will have survived. If the experiment has been properly designed, there should be several doses that have killed some but not all of the exposed rats.

A *dose/response bioassay* measures "How much is too much?"

The end result is a number called the LD_{50}, which stands for the lethal dose for 50% of the treated organisms. In other words, half of the rats that received the LD_{50} dose have died by the end of the 14-day test period. LD_{50}s are expressed in terms of milligrams of the chemical per kilogram of body weight (mg/kg).

The *LD50* is the dose causing death of half of the organisms exposed at this level.

The experiment should also include a control group. The rats in the control group are treated exactly the same as the other rats except that they are not exposed to the chemical being tested—their dose of this chemical is zero.

Within any species, some individuals will die at lower doses than others. When rats are fed caffeine, some may die after eating only 100 mg, while others may tolerate 20 or 30 times this amount. Humans show these same kinds of differences. A cup of coffee at bedtime may have no effect on one person, yet may keep someone else awake through the whole night. Therefore, rather than relying on individuals, toxicity tests are based on group responses. The more individuals tested, the better the chance of accurately estimating the LD_{50} and of identifying low doses to which only the most sensitive individuals respond.

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Topic: bioassays
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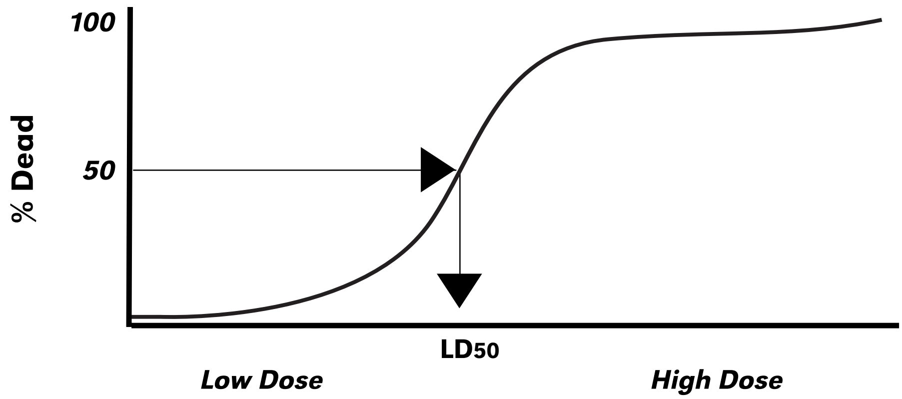
FIGURE 1.1
A Typical Dose/Response Curve



Follow the arrows to see how the percentage of deaths is used to figure out the LD_{50} (the dose representing death of 50% of the treated organisms).

Some individuals are more sensitive than others to any particular chemical.

LD_{50} experiments measure lethal dose—the amount of a chemical that will kill 50% of the test organisms. But of course chemical exposures can affect organisms in ways other than death, such as causing nausea, dizziness, skin rashes, or paralysis. When scientists carry out LD_{50} experiments, they also look for health effects such as these and record the doses at which such effects occur.

Comparing Chemical Toxicities

The more toxic the compound, the lower its LD_{50} . That makes sense if you think of poisons—the more poisonous a chemical is, the less it takes to kill you. For caffeine, the LD_{50} is roughly 200 mg in laboratory rats. For trichloroethylene, it is over 7,000 mg. This means that on average, rats can survive eating over 35 times as much pure trichloroethylene as caffeine. (This is a 14-day test and does not consider possible long-term impacts on health and survival.)

Even "Just a Taste" Can Be Too Much

It only took a little! This is the message behind the story of Loretta Boberg, a 62-year-old woman from Wisconsin who always tastes food before serving it to company. In this case, the company can be very thankful she did.

When Mrs. Boberg opened a jar of home-canned carrots last January, she dipped in a finger to taste the juice. Not liking the taste, she served home-canned beans to her guests instead. Within two days, Mrs. Boberg became dizzy and had difficulty walking. At first, hospital staff thought she had suffered a stroke because of her slurred speech and muscle weakness. The doctor did ask her if she had eaten any spoiled food lately, however. Too weak to speak, Mrs. Boberg wrote "carrots" on a piece of paper.

> If this physician had not suspected botulism, even though he had seen only a few cases, Mrs. Boberg would probably have died. The toxin moved through the respiratory system, paralyzing her muscles. A sample from the jar was fed to a laboratory mouse and it died instantly. The road to recovery for this lady was very slow.
>
> Mrs. Boberg used a boiling water canner for the carrots that gave her botulism. Yes, this was the same method she had used—and only by luck had gotten away with—for the past 44 years. This year she was not so lucky. If, like Mrs. Boberg, you are canning low-acid foods such as vegetables (except tomatoes), red meats, seafood, and poultry in a boiling water canner or by the open kettle method, you may wish to think twice before taking another chance...
>
> (*Andress 1991*)

Botulin, the compound that came close to killing Mrs. Boberg in the story above, is one of the most highly toxic chemicals known. It is created by bacteria in improperly canned foods. People eating these foods suffer a severe form of food poisoning called botulism. As you can tell by comparing LD_{50} values in Table 1.1, the compound that causes botulism is a million times more toxic than cyanide, and twenty million times more toxic than caffeine. In its pure form, less than one drop of botulin toxin is enough to kill 500 adult humans.

To get an idea what LD_{50} numbers mean, you can compare them to the amounts it would take to kill a typical human adult (see Table 1.2).

The LD_{50} values in Table 1.1 are based on experiments in which the compounds were fed to rats. LD_{50} values should always include information about the type of animal and how it was exposed to the chemical being tested. Otherwise, it is impossible to interpret what the values mean or to compare them to values reported by other scientists.

Species may respond in different ways to toxic chemicals.

For some compounds, there is a big difference in LD_{50} values from one species to another. Dioxin is a good example. The LD_{50} for dioxin is 5,000 times higher for hamsters than for guinea pigs. How could this be? Many factors affect how sensitive each species will be to a particular compound. One of these factors is how the chemical gets metabolized. How much gets absorbed into the animal's blood, or stored in its liver, kidneys, or other tissues? How much passes right through and is excreted? How much gets converted into other chemical forms? The answers to these questions may vary from one species to another.

For example, a human being would have a hard time dying from eating too much chocolate. This is not true for dogs—eating just a few chocolate bars can be fatal to dogs because they cannot digest and break down the chemicals in chocolate in the same way that humans do.

TABLE 1.1
Lethal Doses of Some Common Compounds

Substance	Comments	LD50* (mg/kg)
Botulin	An extremely toxic compound formed by bacteria in improperly canned foods; causes botulism, a sometimes fatal form of food poisoning	0.00001
Aflatoxin	A cancer-causing chemical created by mold on grains and nuts; can be found in some peanut butter and other nut and grain products	0.003
Cyanide	A highly poisonous substance found in apricot and cherry pits and used in industrial processes such as making plastics, electroplating, and producing chemicals	10
Vitamin D	An essential part of the human diet but toxic in doses higher than those found in normal human diets	10
Nicotine	The addictive agent that occurs naturally in tobacco and is added to some cigarettes to make them more addictive	50
Caffeine	A compound that occurs naturally in cocoa and coffee beans and is a common food additive	200
Acetylsalicylic acid	The active ingredient in aspirin	1,000
Sodium chloride	Table salt	3,000
Ethanol	Alcohol in beer, wine, and other intoxicating beverages	7,000
Trichloroethylene	A solvent and a common contaminant in groundwater and surface water supplies	7,200
Citric acid	An ingredient in citrus fruits such as oranges, grapefruits, and lemons	12,000
Sucrose	Sugar, refined from sugar cane or sugar beets	30,000

** These LD50s are based on oral ingestion by rats. They represent single doses that cause death of 50% of the treated animals within 14 days of exposure. LD50s are expressed in terms of milligrams of the substance per kilogram of body weight (mg/kg).*

TABLE 1.2
Toxicity Categories Used for Human Poisons

Toxicity Category	LD50 (mg/kg)	Probable Lethal Dose for 70 kg Human Adult	Example Compounds
Super toxic	<5	<0.35 g	Botulin Aflatoxin
Extremely toxic	5–50	0.35–3.5 g	Cyanide Vitamin D (calciferol)
Very toxic	50–500	3.5–35 g	Nicotine Caffeine
Moderately toxic	500–5,000	35–350 g	Aspirin (acetylsalicyclic acid) Salt (sodium chloride)
Slightly toxic	5,000–15,000	350–1,050 g	Ethanol Trichloroethylene
Practically nontoxic	>15,000	>1,050 g	Sugar (sucrose)

Many animals, including dogs and humans, will vomit if they eat something disagreeable. Rats and other rodents cannot vomit. Does this mean we should not use rodents in laboratory tests of chemical toxicity? Obviously they do not represent an exact model of how a person might respond to the same chemical. However, they do provide information that we can use to make limited conclusions about possible health effects on people.

Another concern related to use of laboratory animals for toxicity testing is the issue of animal rights. This is a complicated issue. Some people find it unethical to carry out experiments that may cause suffering or death of the test animals. However, we all want to be confident that we will not become sick, blind, or otherwise injured by the medicines, cleaning products, cosmetics, and huge range of other chemicals that we use on a daily basis.

Over the past few decades, scientists have developed a variety of new techniques to reduce the number of laboratory animals used in toxicology experiments. For example, some tests are carried out on single cells or on blood samples rather than on whole organisms. However, it has not been possible to eliminate the need for animal experiments. This is because there is no guarantee that the response of molecules, cells, or tissues will provide a reasonable model of the response of whole animals or humans.

The toxicity of a chemical depends on many factors, including whether it gets broken down, is stored in the body, or is excreted.

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Topic: animal experiments
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LONG-TERM VERSUS SHORT-TERM TOXICITY

For most of human history, concern about the toxic effects of chemicals has focused on poisons that cause a rapid death. The earliest descriptions of human life include stories about use of toxic plant and animal extracts—to coat arrows and spears used in hunting or fighting battles, or to create poisonous drinks used to kill prisoners. These are examples of *acute* toxicity, the effects of a single exposure to a toxic compound. LD_{50} experiments are designed to assess acute toxicity by measuring the short-term response of test organisms to a single dose of a chemical. Acute toxicity experiments provide useful information but give a limited view of overall toxicity because they address only short-term responses to single doses.

***Acute* effects are caused by exposure to a single dose, such as death caused by walking into a room filled with toxic fumes.**

For some chemicals, the same total dose can be either deadly or harmless, depending on the rate of exposure. For other chemicals, this is not true, and even tiny doses can add up to toxic concentrations over time. This is because our liver and kidneys work to break down and get rid of toxic chemicals, but these systems work better for some types of chemicals than others.

Lead is an example of a chemical that builds up in our bodies over time rather than getting broken down or excreted. Lead poisoning has been linked with stunted growth and mental retardation in children. These are not sudden effects, but ones that develop gradually with long-term, low-level exposures to lead in air, food, and drinking water. Children living in homes with lead paint receive additional doses when they eat chips of paint or breathe dust-filled air. Even though the daily doses may be quite low, lead accumulates in bones. When the concentrations become too high, lead poisoning damages the nervous system and kidneys, causing problems such as hearing loss and mental retardation.

For many other types of chemicals, low daily doses do not cause problems such as these, and toxic effects occur only with short-term exposure to relatively large doses. For example, the oxalic acid found in rhubarb and spinach is harmless at the low concentrations found in these foods, but it would lead to kidney damage or death if you managed to eat 10 to 20 pounds of these foods at one meal.

Alcoholic drinks work the same way. A person who drinks too many drinks in a short period of time may die from acute alcohol poisoning. At the rate of only one drink per day, that same total amount of alcohol might do little or no harm. At this slower rate, most people's livers would have time to break down the alcohol rather than allowing it to build up to harmful levels in the body. However, "most people" does not include everyone, and there are some individuals with extra sensitivity to the toxic effects of any particular chemical. In the case of alcohol, pregnant women are cautioned not to drink because of the heightened sensitivity of their unborn children to alcohol toxicity.

Within limits, our bodies can break down or get rid of many types of toxic compounds before they harm our health. However, it is possible to expect too much of our bodies. With continued exposure to a toxic chemical, the liver can become damaged. Alcoholics frequently suffer from this problem, as do people who have had long-term exposure to toxic compounds through their work or through living in a contaminated environment.

In recent years, people have become increasingly concerned about the effects of long-term exposure to relatively low doses of contaminants. These are called *chronic* effects. If you lived in a house with a leaky furnace, you might be exposed to either acute or chronic carbon monoxide poisoning. Acute poisoning would occur if your house were tightly sealed, with so little ventilation that carbon monoxide fumes could build up to lethal levels. If your house were better ventilated, you would be more likely to suffer chronic effects such as headaches and fatigue from exposure to lower concentrations of the toxic fumes.

Chronic **effects develop slowly due to long-term exposure to contaminants in water, food, or the environment.**

TESTING CHRONIC TOXICITY

The easiest way to test chemical toxicity is to count how many test organisms suffer serious health effects or die when exposed to large doses. However, for most types of environmental pollution, these acute toxicity measurements do not provide answers to the questions we are interested in asking. For example, we might wonder whether it is harmful to drink water that contains low concentrations of a chemical such as trichloroethylene. The concentrations are not high enough to cause acute poisoning, but we would also want to know whether it might be dangerous to drink the water every day for many years. Would this cause a disease such as cancer or asthma? Would it result in birth defects, reduced growth rates, or lowered intelligence in children? These questions concern chronic toxicity.

Tests for chronic toxicity measure health problems rather than death rates.

To measure acute toxicity, you count how many test animals die within a couple of weeks after a single exposure to a chemical. For chronic toxicity, we want to know how the animals' health is affected by continuing exposure over a much longer time period. Rats, mice, or other lab animals are fed relatively low doses of the test chemical each day for months or years. During this time, the experimenters look for various effects such as lowered growth rates, changes in behavior, increased susceptibility to disease, or reduced ability to produce healthy young. Since lab animals lead much shorter lives than humans, it is possible to study effects on life span and reproduction without having to wait decades for the results.

In the case of trichloroethylene, chronic exposure has caused cancer as well as damage to the liver, kidneys, and central nervous system of laboratory animals. Whether trichloroethylene causes cancer in humans is still uncertain. Limited data are available on humans who have used trichloroethylene in poorly ventilated areas. These people have suffered from dizziness, headaches, slowed reaction time, sleepiness, and facial numbness. Data on the concentrations causing health effects such as these are used by the government in setting standards for acceptable chronic exposure to trichloroethylene through water, air, and other sources.

CONCLUSION

This chapter describes the process of measuring how a chemical affects laboratory animals such as rats. You may be wondering how data from these dose/response experiments can be used in the real world. For example, suppose that scientists have determined that rats tend to develop liver disease when exposed to a certain concentration of trichloroethylene in their daily diets. How can the government use this information in deciding the maximum concentration to allow in human drinking water?

Toxicity experiments provide the basis for government regulations that specify what concentrations of certain chemicals are allowed in human food, drinking water, drugs, and cosmetics. The next chapter explains how this process occurs, starting with laboratory data and ending with regulations about chemical use.

FOR DISCUSSION

- What do you think that Paracelsus meant when he wrote that the right dose differentiates a poison from a remedy? Can you think of a substance that is good for you at one dose and poisonous at another?
- Why might it be useful to know the LD_{50} for a chemical? How might you use this information?
- If a compound is shown to be practically nontoxic in a dose/response bioassay, can you conclude that this compound will have no toxic effects on living things? What other sorts of tests might be useful in helping you to make this decision?

TOXICITY CALCULATIONS

Name________________________________ Date___________________

Based on the LD_{50} for caffeine (see Table 1.1), how many cups of coffee would you estimate that it would take to kill an average human of your size (assuming that humans respond in the same way as rats to this compound)? You can calculate this using the steps below:

1. Convert your weight to kilograms:

   _____ lbs × 0.45 kg/lb = _____ kg

2. Calculate the average lethal dose for a human your size:

   _____ mg/kg × _____ kg = _____ mg caffeine
   LD_{50} your weight

3. Assuming that each cup of coffee contains 90 mg caffeine,
   calculate how many cups it would take to kill an average person about your size:

   _____ mg caffeine ÷ 90 mg/cup = _____ cups of coffee

What Does This Number Mean?

A. Take a look at the number you calculated in Step 3. If you were to drink one cup of coffee per day for this number of days, would you be likely to die from an overdose of caffeine? Why or why not?

B. If you could drink exactly the number of cups of coffee you calculated in Step 3 all at one sitting, would you be guaranteed to die? Why or why not?

C. What is the most important assumption that we make when we use LD_{50}s to estimate lethal doses for humans?