Cats Have Nine Lives, But Only One Liver: The Effects of Acetaminophen

by
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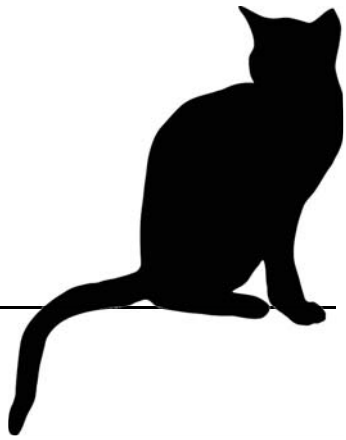


"Hi Mom, how are you?" Michelle asked her mother over the phone.

"I'm fine, is everything okay with you? You sound a bit down," said her mother.

"It's probably nothing, but Tabby doesn't seem quite right," Michelle replied.

"What's the matter with him?" Michelle's mother asked anxiously.

"He wasn't looking very well, and it felt as if he had a fever. Remember I told you he had a swollen leg? Well, it seemed as though it was causing him pain. He seemed so miserable, and so I gave him half of a regular-strength Tylenol®. He just doesn't seem right," Michelle blurted out.

"I saw a news item a month or so ago that said that Tylenol® wasn't very safe—something about it not being good for the liver. I remember they said it was bad for humans in high doses. I don't think they mentioned anything about animals. But don't worry," Michelle's mother reassured her. "Cats are very tough—they have nine lives, you know. And besides, you only gave him half a tablet."

"Thanks, Mom. I better go now. Talk to you later," Michelle hurried off.

The conversation with her mother had made Michelle uneasy. Tabby seemed very sad, tired, and out of breath. Michelle decided that she would search the Internet for information about acetaminophen, the active ingredient of Tylenol®. In particular, she wanted to know the safe dose for cats. She came across a 2003 article by someone named Steenbergen that alarmed her. It said that acetaminophen was particularly toxic for cats and that no dose was considered safe. Immediately, Michelle scooped Tabby up, loaded him in his carrier, and sprinted over to the vet. Luckily, the vet had a practice only a few blocks away.

Michelle was able to see the vet soon after she got there. She explained what had happened. The vet told her not to worry. He took the cat into his examination room and invited Michelle in. She noticed that he administered oxygen to the cat and took a blood sample. It was too much for Michelle! She stepped out of the examination room and sat anxiously in the waiting room. After a while, the vet came out. "Why don't you go home and have some rest," he said. "I've given Tabby an antidote. We'll call you tomorrow and let you know how Tabby responds to the antidote and supportive care."

"Is Tabby going to be okay?" Michelle asked.

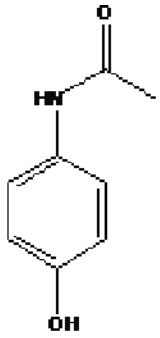
"With continued therapy and nursing care, Tabby has a good chance of overcoming the effects of the poisoning," the veterinarian told her.

Michelle could not bear to go home directly. She went to the campus library intending to study for her organic chemistry exam on reaction mechanisms the next morning, but she couldn't focus. She kept worrying about Tabby. She decided to learn some more about the liver toxicity of Tylenol® before she started studying.

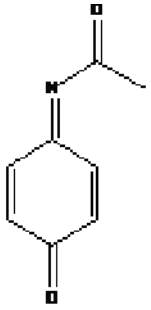
Michelle looked in several pharmacology textbooks and some of the articles they cited. She learned that the IUPAC name for acetaminophen is N-(4-hydroxyphenyl) acetamide and that it is metabolized primarily in the liver via the cytochrome P450 enzyme system. In most mammals the major portion of acetaminophen consumed is conjugated with glucuronic acid and, to a lesser degree, sulfate to form more water soluble but innocuous compounds that are eliminated by the kidneys. However, a very small portion of acetaminophen is metabolized to a compound, N-acetyl-p-benzo-quinone imine (NAPQI), which can lead to liver damage. She learned that the metaboloite NAPQI is toxic because it is electrophilic and reacts with nucleophilic groups of biomolecules to form adducts. It was also indicated that there is evidence to suggest that initially it is the sulfhydryl (-SH) group of the tripeptide, glutathione, that reacts with N-acetyl-p-benzo-quinone imine to form an adduct. This results in depletion of the glutatione levels; when the glutathione levels drop by 80% or greater, macromolecules in the liver react with the electrophilic metabolite, leading to liver damage.

Michelle was distressed to learn that in cats a much larger portion of acetaminophen is metabolized to NAPQI than in other mammals. Cats are deficient in their capacity to metabolize compounds via glucuronidation and sulfation. As such, toxicity occurs with much lower equivalent doses (mg/kg) of acetaminophen. The recommended dosage for humans is no more than 4,000 mg of acetaminophen per day, but there is no dose of acetaminophen that is considered safe for cats. Michelle knew that glutathione, which is widely distributed in mammalian tissues, is essential for cellular protection against oxidative injury by radicals. It was understandable why acetaminophen toxicity would lower glutathione levels and damage the liver and even injure cells in other parts of the body.

Michelle learned that cats are more likely to develop a condition called methemoglobinemia, in which there is an excess amount of methemoglobin (an abnormal form of hemoglobin) in the bloodstream, resulting in a decrease in the oxygen transport capacity of the blood. This occurs when the mechanisms that defend against oxidative stress within the red blood cell are overwhelmed and the oxygen carrying ferrous ion (Fe^{2+}) of the heme group of the hemoglobin molecule is oxidized to the non-oxygen carrying ferric state (Fe^{3+}). As methemoglobin levels start to rise, clinical signs develop, such as chocolate-brown mucous membranes, fast heart rate, labored breathing, depression, vomiting, edema (swelling) of the face, neck and limbs, hypothermia, ataxia (lack of coordination), and coma. Michelle understood why the vet took a blood sample and administered oxygen (O_2) to Tabby. As she read, she felt better when she realized that Tabby did not seem to have exhibited many of the clinical symptoms of methemoglobenia before she got him to the vet.



N-(4-hydroxyphenyl)acetamide



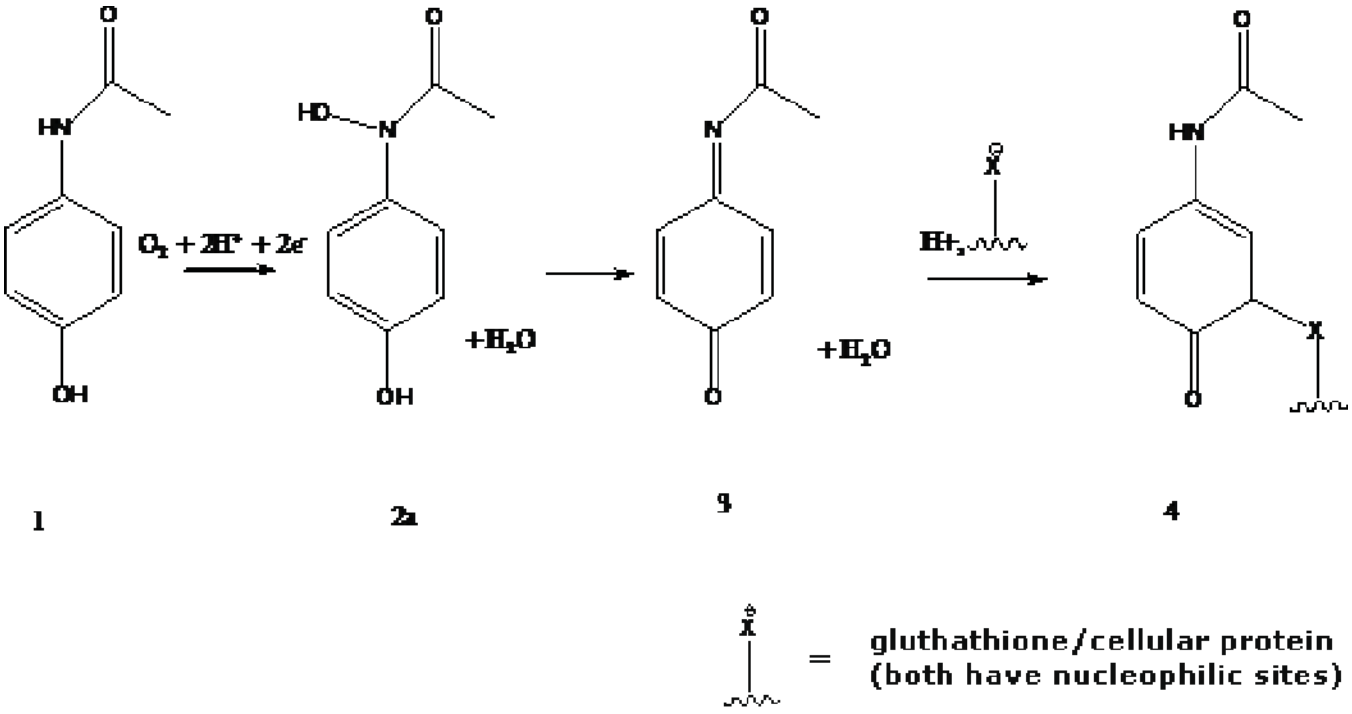
N-acetyl-p-benzo-quinone imine

Michelle read that it has been postulated that the metabolism of acetaminophen to N-acetyl-p-benzo-quinone imine is via one or more pathways. The most common reaction catalyzed by cytochrome P450 is one catalyzed by a family of enzymes known as monooxygenases. These catalyze insertion of one atom of oxygen into an organic substrate (RH) while the other oxygen atom is reduced to water.

$$RH + O_2 + 2H^+ + 2e^- \rightarrow ROH + H_2O$$

The net result is that an organic compound is converted to a hydroxyl derivative, which is more water soluble and easily excreted. A proposal for the formation of the electrophilic metabolite, NAPQI, is outlined in Scheme 1 below. Structure 3 corresponds to NAPQI.

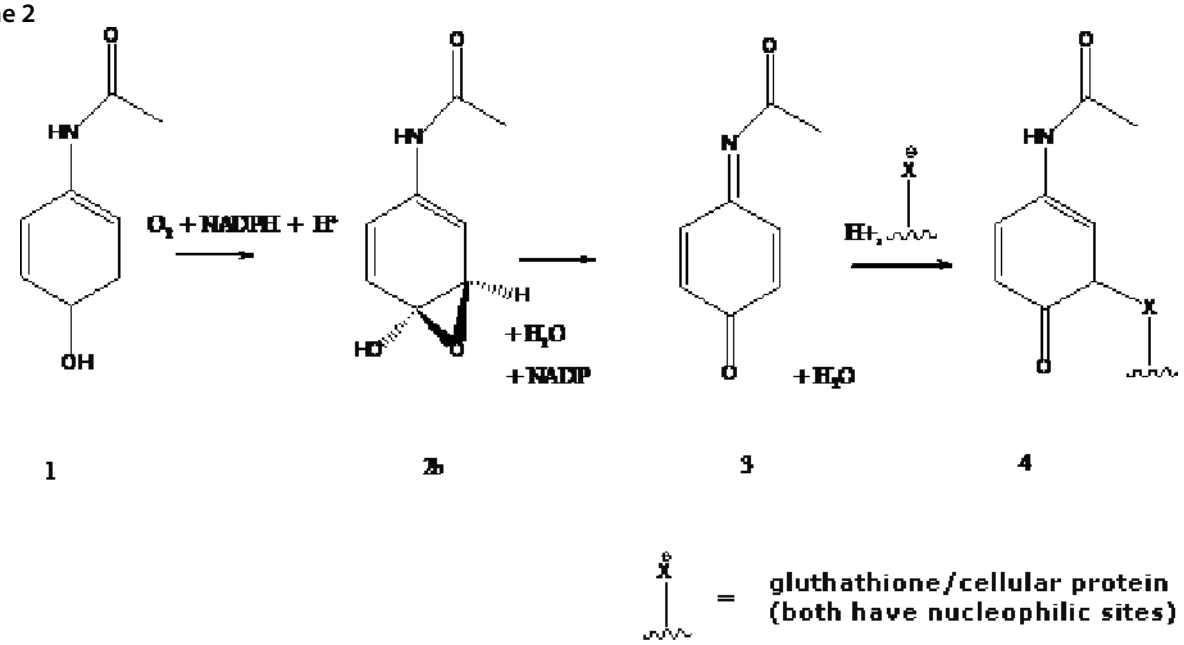
Scheme 1



1 2a 3 4

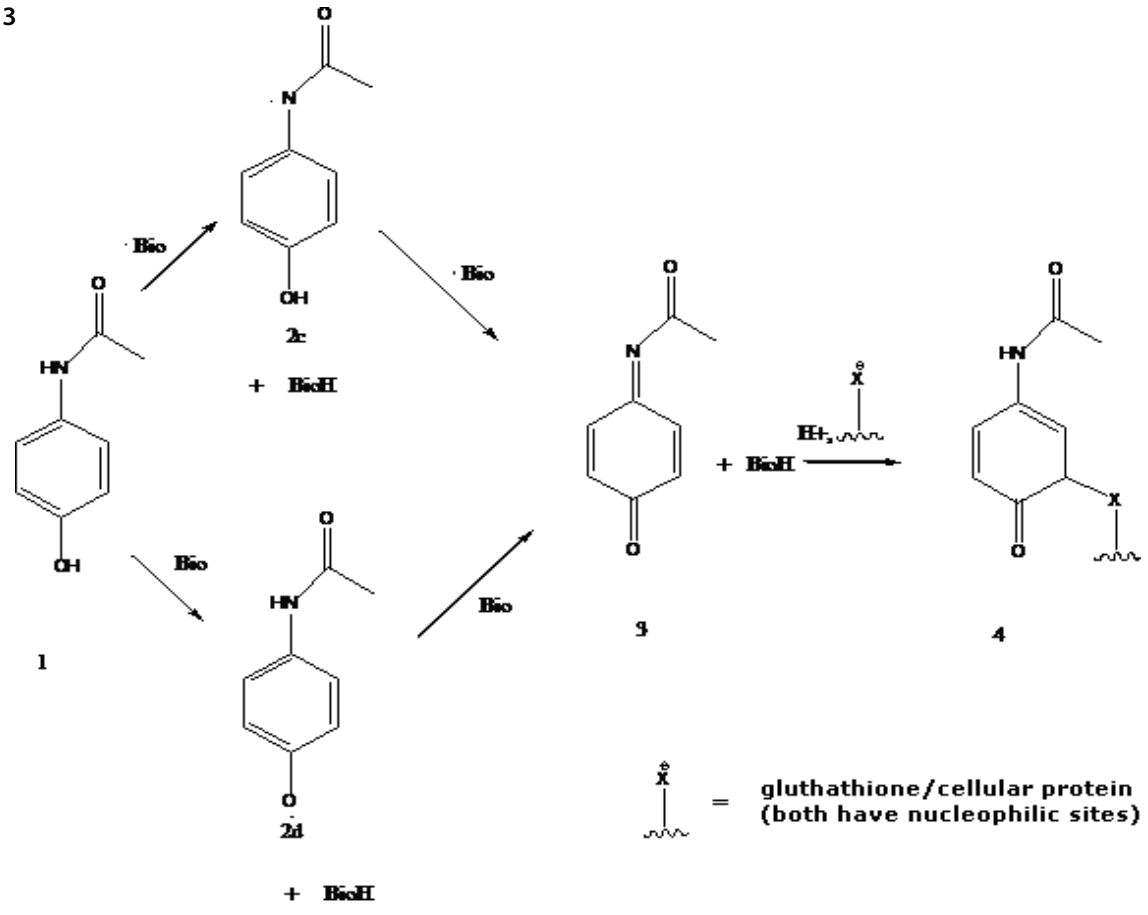
Scheme 2 outlines another proposed reaction scheme for the formation of the electrophilic metabolite 3 and involves acetaminophen epoxidation. Although expoxidation of aromatic rings does not occur readily under laboratory conditions, many aromatic compounds (including polycyclic aromatic hydrocarbons) are known to be metabolized to more water-soluble metabolites via an epoxide intermediate *in vivo*.

Scheme 2



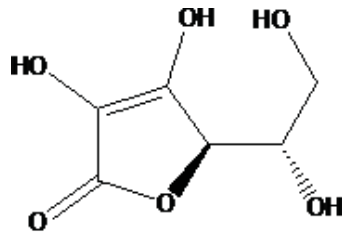
A third possibility, shown in Scheme 3, is hydrogen abstraction via an acetamininophen radical. The enzyme system (cytochrome P450) has a Fe moiety and is associated with coenzymes that allow it to participate in redox reactions.

Scheme 3



Bio = a biological molecule with a radical moiety such as heme, an Fe^{3+} containing conenzyme

Michelle wondered which mechanism was the correct one. She continued to research the literature for evidence that supported or refuted each of these mechanisms. She learned that when acetaminophen was metabolized in the presence of $^{18}O_2$ *in vitro,* the metabolites produced were not labeled with ^{18}O. In addition, she learned that a specific enzyme (of the cytochrome P450 system) that is induced by alcohol (ethanol) was shown to be responsible for the metabolism of acetaminophen to the electrophilic metabolite, 3. The hepatoxicity (liver toxicity) of acetaminophen in ethanol-fed animals was compared with normal animals. It was found that alcoholic animals had increased hepatoxicity, and that Vitamin C (ascorbic acid) protected the animals. Furthermore, it was found there is an increase in acetaminophen hepatoxicity in alcoholics.



Ascorbic Acid

Michelle realized in order to figure out which was the correct pathway, she needed to understand the mechanism for each of the schemes. She hadn't reviewed her lecture notes or the textbook chapter on mechanisms yet. But she found a few websites that provided her with a good review of writing mechanisms. She studied these and then tried writing reaction mechanisms for each of the schemes. She found that she was able to do so. She felt better. "Aha! Now I know which pathway is the correct one."

Michelle was about to start studying her notes on mechanisms when she heard an announcement that the library was closing. She packed her books and went home. She went to bed promptly, promising herself that she would wake up early and study for a few hours before heading to class to take her exam on mechanisms.

Michelle woke up the next morning with a start. It was already 8:10 a.m. and her exam was at 9:15 a.m., leaving her no time to study. She dressed hurriedly, grabbed her backpack and a bagel, and rushed out of her apartment and towards the subway station. She made it to class with only a few minutes to spare. She sat near to her friend Silvia and related her experience of the previous day. Silvia advised her not to take the exam and ask the professor if she could take a make-up exam instead at a later time. But Michelle decided to take the exam anyway. She was exhausted and just wanted to get it over with.

A few days later she picked Tabby up from the vet. He seemed to be recovering remarkably well. He appeared to have lost some weight, but his eyes shone. The vet assured her that Tabby would be his old self soon. In parting, the vet advised her: "Remember: Cats have nine lives, but only one liver."

That evening, as Michele lay cuddled up on her sofa with Tabby, the phone rang. "How did you do on the exam?" her friend Silvia asked.

Michelle replied, "I got an A."

"You told me that you didn't have a chance to study," her friend reminded her.

"You know, I've found out that you can learn a lot of organic chemistry by trying to make sense of things that happen in your daily life. Organic chemistry is really about understanding life at a molecular level," Michelle observed.

Your Task

Use curve arrows to show the mechanism for the proposed conversion of 2a, 2b, 2c, and 2d to 4. Based on the evidence presented, which is the most likely pathway? Explain the rationale for your choice.

Resources

You can download a free drawing program from: **http://www.acdlabs.com/download/chemsk.html**

The following sites provide a review of writing organic reaction mechanisms:

- **http://www.chemguide.co.uk/mechmenu.html**
- **http://www.abdn.ac.uk/curly-arrows/**
- **http://www.chem.ox.ac.uk/vrchemistry/nor/notes/conj.htm**

References

ABC News. 2006. FDA increases acetaminophen labeling. 12/20/2006. **http://abcnews.go.com/Video/playerIndex?id=2740985**. Last accessed: April 8, 2008.

Sondheimer, N. 2007. Methemoglobinemia. MedinePlus. National Library of Medicine/ National Institutes of Health. **http://www.nlm.nih.gov/medlineplus/ency/article/000562.htm**. Last accessed: April 8, 2008.

Steenbergen, V. 2003. Acetaminophen and cats: a dangerous combination. *Veterinary Technician*, p. 43–45. Available online at **http://www.aspca.org/site/DocServer/vettech_0103.pdf?docID=642&AddInterest=1101** Last accessed: April 8, 2008.

Wikipedia. 2008a. Liver. **http://en.wikipedia.org/wiki/Liver**. Last accessed: April 8, 2008.

Wikipedia. 2008b. Cytochrome P450. **http://en.wikipedia.org/wiki/Cytochrome_P450** Last accessed: April 8, 2008.