BIOCHEMISTRY

Closing In on a Stomach-Sparing Aspirin Substitute

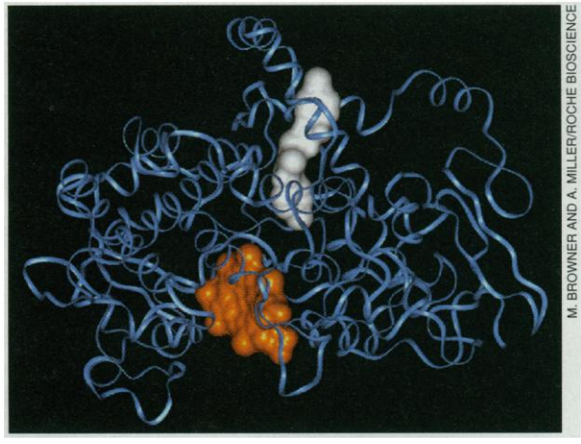
Few people think of aspirin as a dangerous drug. Yet chronic use of aspirin and other so-called nonsteroidal anti-inflammatory drugs (NSAIDs) can cause stomach bleeding, kidney failure, or worse. In the United States alone, these and other side effects kill more than 7000 people every year and send roughly 10 times that number to the hospital. Behind this Jekyll-and-Hyde behavior is a lack of discrimination: While the compounds go about their job of subduing a key enzyme involved in triggering inflammation and pain, they also stifle a nearly identical enzyme that helps maintain tissues such as the stomach lining. Now researchers at a number of drug companies are making progress toward "superaspirins"—compounds capable of discriminating between the twin enzymes, which would enable them to fight pain without causing dangerous side effects.

One superaspirin candidate that is moderately selective for the pain-causing enzyme has already reached the market, while more selective counterparts remain in clinical trials. But researchers are also beginning to unravel atomic differences between the two enzymes that could lead to even more discriminating compounds. At a pair of recent meetings, for example, groups from Roche Bioscience in Palo Alto, California, Searle in St. Louis, and the Merck Frosst Research Centre in Pointe Claire, Quebec, announced that they had independently solved the x-ray crystal structure of the pain-inducing enzyme bound to different inhibitors. The new structures "give clues as to how you can make an agent selective for one compound and not the other," says Louise Johnson, a molecular biophysicist at Oxford University. "I think that's exciting."

The excitement is proving contagious. "If [superaspirins] live up to the promise of the early data, there's the potential to really revolutionize therapy and replace NSAIDs," says Peter Isakson, who heads anti-inflammation research at Searle. With a projected market of about $5 billion, it is not surprising that, as Ravi Kurumbail, an x-ray crystallographer at Searle, puts it, "It's a very competitive field." He adds, "It will be exciting to see who will come out first."

What is already clear is what it will take to win. Traditional aspirin and other NSAIDs block the activity of so-called cyclooxygenase (COX) enzymes. These enzymes create prostaglandins, hormones that have local or temporary effects—pain, inflammation, and fever—that are triggered by injury or infection, as well as long-term effects, such as protecting the stomach lining against acidity and maintaining the proper balance of electrolytes in the kidneys. The first of these enzymes, COX-1, has been familiar to researchers for decades.

COX-1 produces the "good" prostaglandins that act in the stomach and other tissues. But researchers suspected that another COX enzyme, responsible for the trouble-some prostaglandins made in response to injury, might still be at large. That suspicion was confirmed in 1991 when a trio of labs independently cloned the gene for COX-2. The race was on for compounds that block COX-2 without interfering with the vital role of COX-1.



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Inhibited. Structure of the inflammation-causing enzyme COX-2, with a bound inhibitor molecule *(white)*.

The race has been run on two parallel tracks. Screening large numbers of compounds against the two enzymes in a trial-and-error search for ones that inhibit COX-2 preferentially is one strategy. The other is trying to determine the three-dimensional structures of the two enzymes. The hope is that comparing the structures will yield clues to guide the rational design of drugs that would stop COX-2 while leaving COX-1 untouched.

The rational-design strategy got a boost in 1994, when Michael Garavito and his colleagues, then at the University of Chicago, solved the structure of COX-1. Matching data for COX-2 came last April from Michelle Browner and her colleagues at Roche, who presented a detailed view of the structure at a small conference on inflammation in New Orleans, and from Kurumbail and his Searle colleagues at an International Union of Crystallography meeting in Seattle last month. A group from the Merck Frosst Research Centre offered glimpses of similar data at the April meeting and plans a full report this week at the Tenth International Conference on Prostaglandins and Related Compounds in Vienna, Austria.

Placed side by side, the COX-1 and -2 structures show that the sites where NSAIDs bind the enzymes are very similar. "But there is at least one important amino acid difference," says Browner: A bulky isoleucine in the COX-1 binding site is replaced by a slimmer valine in COX-2. The isoleucine, says Kurumbail, blocks a side pocket that branches off the main binding site in both enzymes. The blocked pocket in COX-1 apparently doesn't hamper the binding of existing NSAIDs. But an inhibitor molecule that needed the extra toehold provided by the side pocket would presumably bind more readily to COX-2 than COX-1. Researchers at Roche, Searle, and Merck say they are all working on designing next-generation inhibitors with a strong preference for the open side pocket of COX-2.

Already, though, trial-and-error screening has yielded some superaspirin candidates that seem to be sensitive to the structural difference between the enzymes. Kurumbail, Browner, and Zhaoyin Wang, a medicinal chemist at Merck Frosst, all say their teams have developed candidate compounds that have at least 200 times greater affinity for COX-2 than for COX-1. Both Searle's Isakson and Merck's Wang say that their companies' inhibitors have passed initial toxicity tests in humans and are now undergoing tests to help researchers gauge proper doses. The German pharmaceutical company Boehringer Ingelheim in Ingelheim, Germany, has a less discriminating COX inhibitor—one with at least a twofold preference for COX-2—that went on the market earlier this year in Europe, Latin America, and South Africa. Early indications are that even this level of selectivity is enough to reduce side effects, says Ralf Brassat, a product manager at Boehringer Ingelheim.

How much more selective a superaspirin needs to be is an open question, says Chicago's Garavito. "Do you need 100% selectivity to prevent any side effects?" he asks. The answer is likely to determine whether a superaspirin worthy of the name is already on the market, approaching it, or still waiting to be designed.

–Robert F. Service