Finding that 1-in-a-billion that could lead to disease

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Errors in the genetic code can give rise to cancer and a host of other diseases, but finding these errors can be more difficult than looking for the proverbial needle in the haystack. Now, scientists at Johns Hopkins have uncovered how the tiny protein-machines in cells tasked to search for such potentially life-threatening genetic damage actually recognize DNA errors.

Appearing online next week in *Nature*, the Hopkins team describes how the UDG enzyme (for uracil DNA glycosylase) scrutinizes the shape of DNA building blocks by holding onto them and testing their fit into a specially sized pocket. The UDG pocket holds onto mistakes only — the enzyme loses its grip on the right building blocks, which fall back in line with the rest of the DNA.

“Locating damage in DNA is critical for a cell’s survival: So much can go wrong if damage goes unrepaired; cells can’t tolerate any of this going on,” says study author James Stivers, Ph.D., professor of pharmacology and molecular sciences at Hopkins. “But the question is how these enzymes find the few mistakes among the billions of correct building blocks in DNA.”

One typical error that occurs is to the DNA building block cytosine, being chemically converted to a similar-looking building block not normally found in DNA: uracil. “Even water can cause DNA damage,” says Stivers. “It’s not a fast reaction, but water does convert the occasional cytosine into an unwanted uracil.”

To figure out how the enzyme responsible for cutting unwanted uracils out of DNA works, Stivers and colleagues studied a tiny segment of DNA. The research team then asked whether the “breathing” properties of DNA played a role in the search process of UDG. “Although the bases in the DNA double helix resemble the rungs of a ladder, the rungs are not that sturdy,” says Stivers. “They actually pop in and out of the helix a bit, randomly.”

Each time a base pops out of the helix, it exposes itself to water. Thus, using a special chemical trick, the team magnetically labeled water, which allowed them to follow the interaction of water with bases that had randomly popped out of the DNA helix. The researchers could then follow which bases pop out, and for how long, using a strong magnet.

After studying DNA breathing by itself, the researchers then added UDG into the mix. They saw that UDG holds onto the normal DNA building block thymine (T) after it pops out of the DNA on its own. However, because T is not identical to U, UDG then lets it fall back into DNA helix.

When the DNA contains an unwanted U, the UDG enzyme actually grabs on and pulls it all the way out and holds it in the enzyme’s pocket. Once sitting in this pocket, the enzyme clips out the U, leaving a gap in the DNA for other repair machinery to fill in with the correct building block.

“This is the first time we’ve been able to actually see how an enzyme discriminates between right and wrong bases in DNA,” says Stivers. “Our discovery helps us appreciate what properties of DNA itself might lead to errors that are not repaired. “The finding may help address how and where diseases like cancer arise in the genome.”

Source: Johns Hopkins Medical Institutions