Molecular Biology

DNA Polymerases and the Repair of Double-Strand Breaks in DNA

All living cells suffer constant damage, but even the lowliest bacteria have a complex array of repair mechanisms.

DNA polymerases—special enzymes that exist in all living cells—not only can "read" DNA and make copies of it, but they also play a vital role in repairing DNA damage. Fifteen or so different DNA polymerases have been identified, but little is known about them. A Santa Clara University professor and his student researchers are doing their best to shed more light on this marvelous repair mechanism.

Life is a miracle. It is also lethal. Our cells, the cells of all living things, are being damaged constantly just by normal metabolism. The very oxygen we breathe, which helps us sustain life, also damages our DNA. If the damage is not repaired, it can be immortalized as a mutation, a change in information to the DNA. Eventually, that can result in a tumor.

There are a variety of ways of damaging DNA, and a variety of damages that can ensue. Repairs, too, happen in different ways, but what all of them have in common is the help of enzymes called DNA polymerases, which are responsible for duplicating the entire DNA genome of a cell when it divides.

"The question is how they are being used in the repair process," says Ángel Islas, Santa Clara University assistant professor of biology. In his Molecular Biology Lab, he leads a small team of student researchers who are helping him study this question and other areas related to the "three R's" of DNA: recombination, replication, and repair.

DNA's Mr. Fix-it

DNA has a two-strand structure—the famous spiraling helix—with nucleotide pairs connecting the strands like steps on a ladder. Of the types of DNA damage that occur, the worst is a double-strand break. That is because, unlike when only one strand is broken or damaged, there is no undamaged nearby strand to act as a template for repair. With double-strand breaks, the repair mechanism has to bridge a complete gap in DNA. Exactly how DNA polymerases help do that is still unknown.

Just as there are different types of DNA damage, there are different methods of DNA repair. Some are long and involved, and produce the highest quality results. Others are quick and dirty, but they get the job done. The sloppier methods are likely involved in repairing double-strand breaks in humans.

"It's estimated that eight to ten double-strand breaks occur every day in each and every one of our cells," says Islas. "That's a lot of repairs being done all the time, and we don't even think about it, at least not until something goes wrong."

What can go wrong is the DNA repair mechanism itself. In fact, whenever mutation occurs, it is because the DNA repair failed or made a mistake—which happens on average once in every 100,000 repairs.

"We have to understand that these DNA repair mechanisms, whatever they are, are imperfect at times," Islas points out. "That's actually a good thing, from an evolutionary point of view. But from a human individual's point of view, too much mutation can lead to cancer."

A number of different kinds of cancers and diseases are directly caused by defects in DNA repair. And behind the research of Islas and his undergraduates, the hope of eliminating some human suffering lends a poignancy to their efforts.

For example, victims of one rare disease completely lack the DNA polymerase enzymes necessary to repair damage caused by ultraviolet radiation. The genetic defect usually becomes apparent when a newborn is taken home from the hospital and develops an immediate rash or burn when first exposed to sunlight.

An Exciting Experience

Islas delegates work on individual aspects of the overall research to his student assistants. He usually has two or three undergraduates working with him in the lab, engaged in a variety of tasks.

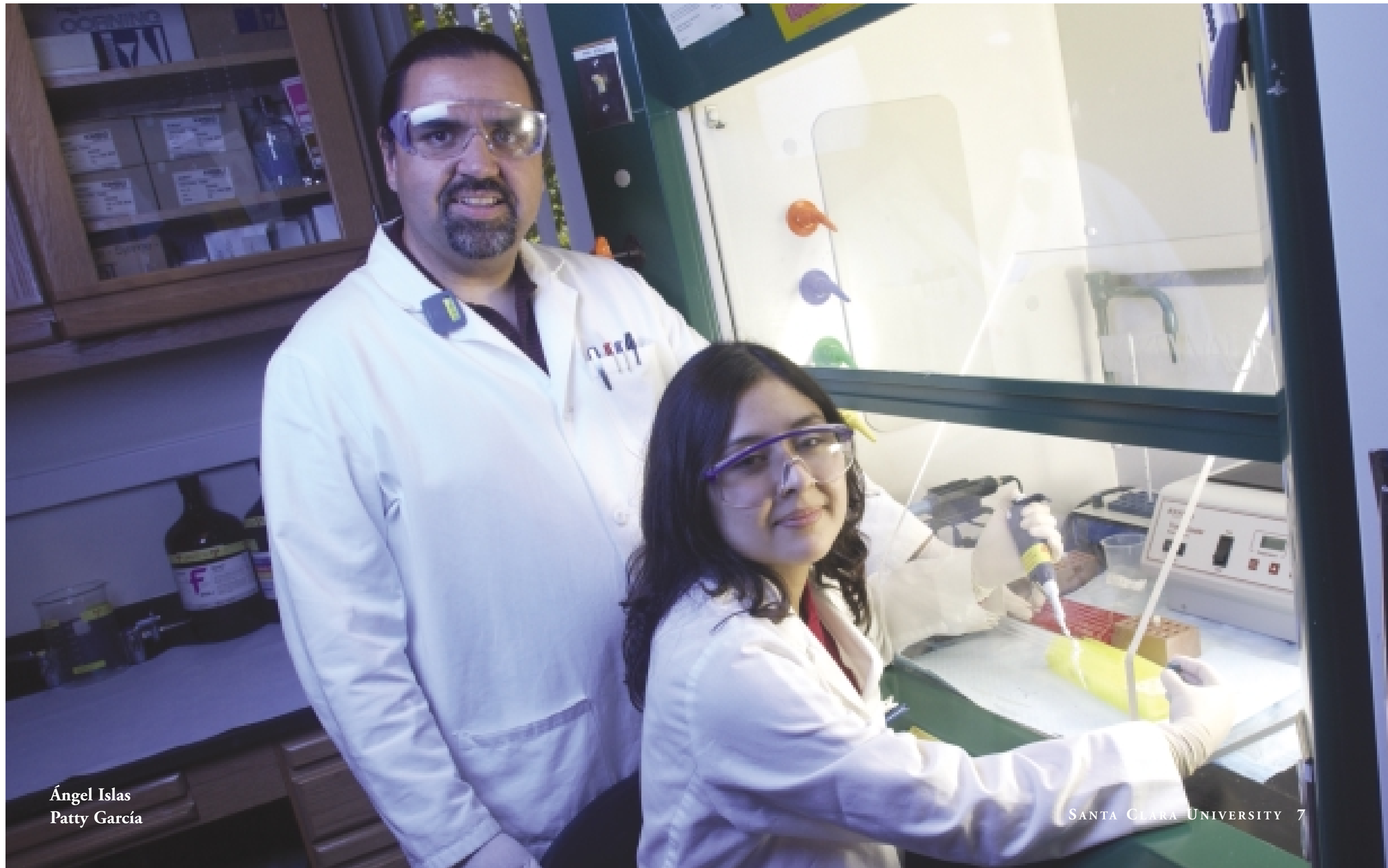
Students learn how to use advanced equipment in the lab and how to perform a number of sophisticated techniques. For example, one system employs chromatography to help them separate and purify proteins and DNA. Other equipment allows them to separate pieces of DNA according to length, through a process called gel electrophoresis.

"I try to take on new students during the summer between their sophomore and junior years," Islas says. "Becoming productive takes a little time for them because there's so much training involved, both in technique and equipment."

He sees how they find the work exciting, perhaps partly because of its implications for cancer research. "I know that I really enjoy and benefit from my collaboration with them," Islas says.

"Fortunately for all of us, every organism has at least one and usually several different kinds of mechanisms or pathways for repairing the different kinds of DNA damage."

Ángel Islas
assistant professor of biology



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