MedicalXpress

Scientists discover remarkable editing system for protein production

January 3 2008

Even small mistakes made by cells during protein production can have profound disease effects, but the processes cells use to correct mistakes have been challenging to decipher. Recent work by scientists at The Scripps Research Institute, however, has uncovered two surprising new methods for such editing.

The work, published in the January 3, 2008 issue of the journal *Nature*, and led by Professor Paul Schimmel of the Skaggs Institute for Chemical Biology at Scripps Research, could help identify underlying causes of a range of diseases and, in time, even ways to correct the errors.

Producing proteins is an essential but complicated process involving a number of components. Messenger RNA within cells act as the instructions for protein synthesis. Ribosomes are cellular structures that read these instructions and follow their directions to bind with bits of transfer RNA carrying the amino acids needed for a given protein chain. In most cases, a single unique form of transfer RNA binds only to a single amino acid, and a specific enzyme called a synthetase is responsible for joining the two.

In very rare cases—typically a fraction of a percent—transfer RNA binds with the wrong amino acid. If this error, or mistranslation, is not corrected, that mistranslated amino acid is ultimately incorporated into a protein. Past research by the Schimmel team and others has shown that as little as one wrong amino acid can have profound consequences for

health.

"Even such a tiny defect can overwhelm a cell's ability to deal with misfolded proteins, ultimately causing specific neurological problems," says Scripps Research molecular biologist Kirk Beebe, first author of the new paper with Marissa Mock.

Quality Control

The prevailing thinking among researchers in the field has been that the portions of synthetases that recognize and bind transfer RNA and appropriate amino acids are so accurate that little editing is required. What little editing does occur was thought tied to the same checkpoints within the enzymes that perform recognition and binding.

But the current study suggests a new perspective is needed, at least for one widely studied enzyme, a synthetase found in everything from bacteria to humans that binds the amino acid alanine.

The Schimmel team's research revealed that a completely distinct segment of the enzyme acts as a second checkpoint responsible for identifying mistranslations and removing any amino acid besides alanine that might attach to the alanine transfer RNA. Remarkably, this second zone within the enzyme focuses its activity on the very same two nucleotides in the genetic code of the transfer RNA used by the first checkpoint, a guanine and uracil pair referred to as G3•U70.

"The part that is astonishing is that the information that each of these checkpoints is looking for is embedded in the same transfer RNA molecule," says Schimmel, "There's no precedent for this that we're aware of."

The researchers were able to show that this editor checkpoint, even when

separated from the rest of the enzyme, was able to efficiently cleave mistranslated amino acids from the alanine transfer RNA. Further experiments revealed that when the G3•U70 pair was transferred to a different type of transfer RNA, the editing unit still removed a non-alanine amino acid, showing clearly that the pair is the trigger for the activity.

Mystery Solved?

The research also led to a surprising find regarding segments of genome-encoded fragments common in cells and sometimes referred to as freestanding domains, whose functions have been debated for many years. The team found that certain freestanding domains with a genetic sequence very similar to the second checkpoint in the alanine synthetase can independently remove mistranslated amino acids in test tube experiments. This suggests that the fragments may act as yet another checkpoint to ensure that proteins are properly synthesized. The activity by the freestanding domain, called AlaXp, also targets the G3•U70 pair.

Whether AlaXp actually edits within cells is not certain, and how they could perform such a function is also not yet clear. "That's still a question to be resolved," says Beebe, "But our work presents a great possibility for how things are likely to occur." The group has already begun new experiments to study AlaXp activity in mouse cells.

"The results of the study were a complete surprise to us," says Schimmel.

Beebe and Schimmel agree that the apparent triple redundancy the team discovered for preventing mistranslation highlights the importance of accurate protein synthesis. It is unlikely such a robustly redundant system would have evolved, they say, if this were not the case.

The work's impact should go beyond adding to the limited understanding

of how cells avoid errors during protein synthesis. "We do think there are probably powerful connections to disease," says Schimmel.

Schimmel's and other groups have already shown clear connections between individual and cumulative errors in translation and a variety of problems tied to aging, such as neurodegenerative conditions. Now the Schimmel team has begun collaborative work with a number of other research groups to identify in synthetases telltale genetic mutations referred to as small nucleotide polymorphisms that can lead to mistranslation. Identifying such errors could reveal the underlying causes of diseases currently not understood and possibly identify potential paths for treatment.

Source: Scripps Research Institute