Scientists discover new type of protein modification, may play role in cancer and diabetes

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Scientists at The Scripps Research Institute (TSRI) have discovered a new type of chemical modification that affects numerous proteins within mammalian cells. The modification appears to work as a regulator of important cellular processes including the metabolism of glucose. Further study of this modification could provide insights into the causes of diabetes, cancer and other disorders.

"It appears to be an intrinsic feedback mechanism in glucose metabolism , but I suspect that its other functions throughout the cell will prove at least as interesting when they are more fully elucidated," said Benjamin F. Cravatt, chair of the Department of Chemical Physiology and member of the Skaggs Institute for Chemical Physiology at TSRI.

Cravatt and his postdoctoral fellow Raymond E. Moellering reported the finding in the August 2, 2013 issue of the journal *Science*.

In Search of New Protein Modifiers

The Cravatt laboratory has long studied the natural chemical modifications that can change the functions of proteins "on the fly," switching their biological activities on or off or otherwise altering them. The better known of these modifications include phosphorylation, the addition of a small molecule known as a phosphate group, and acetylation, the addition of an acetyl group.

In search of new protein modifiers, Cravatt and Moellering, whose postdoctoral fellowship is sponsored in part by the Howard Hughes Medical Institute and the Damon Runyon Cancer Research Foundation, decided to investigate a small molecule known as 1,3-bisphosphoglycerate (1,3-BPG). The molecule's chemical makeup suggested that it might readily react with some proteins to form semipermanent, function-altering modifications. 1,3-BPG is one of the main "intermediate" molecules produced during glycolysis, which is a core metabolic pathway that converts glucose to cellular fuel.

"1,3-BPG's intrinsic reactivity seemed odd to us, considering that it is such a central metabolite," remembered Moellering.

Moellering's initial test-tube experiments showed that 1,3-BPG does indeed react with certain lysine amino acids to modify GAPDH, the enzyme that mediates the production of 1,3-BPG. "That gave us the first indication that this reaction does happen, and that we should therefore start looking for it in cells," he said.

A Role in Glucose Metabolism

After devising new methods to detect this unique lysine modification in human cell cultures, Moellering soon found it—on other glucose-metabolizing enzymes, as well as on proteins seemingly unrelated to glucose metabolism.

"With every step we took, the project became more interesting, because we were finding signs that this reaction occurs frequently in cells and in animal tissues, and in unexpected cellular locations, too," Moellering said.

He detected the signature of the new lysine modification not only on proteins in the main volume of the cell (the cytosol), but also in the DNA-

containing cell nucleus and even on the cell's membrane compartments.

"It appears that wherever GAPDH goes within cells, it is capable of catalyzing the localized production of 1,3-BPG, which in turn reacts with nearby proteins to modify their structure and function," said Cravatt.

Moellering found that when 1,3-BPG's lysine modification occurs on glucose-metabolizing enzymes, it tends to inhibit their activities, causing a slowdown of central glucose processing and a consequent buildup of certain glucose metabolites in the processing pathway. Moellering and Cravatt suspect that these overabundant metabolites may end up being shunted into other cellular processes besides basic fuel-making—processes that contribute to the synthesis of new molecules and even cell proliferation.

Moellering also discovered that 1,3-BPG and the modification it makes on proteins become more prevalent as glucose levels rise. Within the context of glucose metabolism, 1,3-BPG's modification thus seems to act as a "very old, maybe ancient feedback mechanism for regulating that central metabolic pathway," Moellering said.

Looking Ahead

The abnormal processing of glucose within cells features in a number of major diseases including cancer and diabetes. "Cancer cells, for example, bring in as much as 20 times more glucose than non-cancerous cells of the same type," Moellering noted. He now wants to find out whether 1,3-BPG is part of the problem in such cells. At abnormally high levels, it conceivably could help force glucose metabolism toward the runaway cell proliferation that is a hallmark of cancer.

Cravatt and Moellering also want to learn more about what 1,3-BPG's lysine modification does in the nuclei and membrane compartments of

cells, where they found evidence of it. "We suspect that it works to connect glucose metabolism to other pathways, perhaps as a kind of signaling mechanism," said Moellering.

Already Moellering has uncovered evidence that there are enzymes that work to reverse 1,3-BPG's modification of lysines—which underscores the likelihood that this modification represents a fundamental, dynamic mechanism in cells. "We'd like to discover which enzymes catalyze the removal of the modification," said Cravatt, "because then, in principle, we could use inhibitors of these enzymes to control the levels of the modification and get a better understanding of its biological functions as well as the conditions under which it occurs."

More information: "Functional Lysine Modification by an Intrinsically Reactive Primary Glycolytic Metabolite," *Science*, 2013.

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