Spiking diabetes

By Lauren Martz, Staff Writer

U.S. and Australian researchers have unraveled a mechanism that could explain the correlation between transient spikes in glucose levels and an increased risk for cardiovascular complications in diabetes patients.¹ The findings could change the way that diabetes is treated and monitored because standard glucose monitoring often fails to detect these fluctuations.

The current standard measure of blood glucose control involves determining levels of hemoglobin A1c (HbA1c) over time. HbA1c is the amount of hemoglobin in the blood that has bound to glucose and is typically established by calculating a time-averaged area under the time curve of HbA1c concentrations.² According to the **American Diabetes Association**, doctors should measure a patient's HbA1c at least twice a year.

About 30–40% of diabetes patients have glucose levels outside of the normal range despite monitoring and insulin treatment, said Jaime Flores-Riveros, SVP of endocrinology and metabolism at **Hollis-Eden Pharmaceuticals Inc.** This leads to macrovascular complications such as heart attacks and coronary artery disease (CAD), and microvascular complications including diabetic retinopathy and neuropathy.³

Previous research has shown that increased risk of complications is correlated not only with high average blood glucose,⁴ but also with high glycemic variability—fluctuations in glucose levels.⁵ Now, Assam El-Osta and colleagues took those findings a step further to identify the actual mechanisms linking glycemic variability to vascular complications.

El-Osta, an associate professor in the epigenetics in human health and disease laboratory of the **Baker Medical Research Institute**, was joined by researchers from the **Juvenile Diabetes Research Foundation International**.

In a paper published in the *Journal of Experimental Medicine*, the team showed that transient hyperglycemia caused long-lasting epigenetic changes in the promoter of NF- κ B's p65 subunit. Those changes induced transcription of proinflammatory genes in vascular cells.

El-Osta and colleagues incubated bovine and human aortic endothelial cells in high-glucose medium for 16 hours and then returned the cells to normal medium for 6 days to simulate diabetic hyperglycemia spikes.

Both cell types had higher transcription of p65, more activation of NF- κ B and higher levels of the p65-associated SET domain containing (lysine methyltransferase) 7 (SETD7) than cells grown at normal glucose levels, and this persisted over the six-day incubation period.

Knockdown of SETD7 prevented upregulation of p65 and the NF-

κ B-dependent proinflammatory genes *chemokine (C-C motif) ligand 2 (CCL2; MCP-1)* and *vascular cell adhesion molecule-1 (VCAM-1)*. According to the authors, the finding suggests that methylation of lysine 4 of histone 3 (H3K4) by SETD7, which enhances transcription of p65, results from hyperglycemia and is necessary for hyperglycemia-induced upregulation of genes associated with vascular inflammation.¹

Furthermore, upregulation of uncoupling protein 1 (UCP1) or manganese superoxide dismutase (MnSOD) prevented both hyperglycemia-induced increases in superoxide production and changes in p65 transcription associated with the glucose spikes. The effects of UCP1 and MnSOD indicate that the epigenetic changes resulting from hyperglycemia are also dependent on mitochondrial overproduction of superoxide.

When the team exposed mice to six hours of hyperglycemia, this led to the same persistent epigenetic changes observed *in vitro*.

"These findings explain long-term deleterious effects of previous episodes, albeit transient, of elevated glucose levels," El-Osta told *SciBX*. "These studies suggest that it is critical to achieve the best control of glucose to try to avoid glucose spikes, because even after you correct glucose levels, there may still be repercussions on the vasculature in the future."

Glycemic control

Companies contacted by *SciBX* agreed that the most important aspect of the *JEM* paper is the evidence that a very tight control over glycemic fluctuations is essential to prevent cardiovascular complications. The question is how to achieve such level of control. Some researchers suggested that altering existing monitoring techniques will be sufficient, whereas others expect that new therapeutic interventions and increased patient compliance will be required.

"If you can aggressively control sugar, you can mitigate the risk" of complications, said David Simmons, VP and CMO of the Diabetes Care division of the Bayer Healthcare unit of **Bayer AG**. "This article opens a potential new explanation for how glycemic variability might be involved in causing complications."

Diabetes Care's therapeutics include Precose acarbose, an oral α -glucosidase inhibitor, and its marketed diagnostics include the Contour Blood Glucose meter.

"HbA1c is used to measure the average levels of glycemia. It is a marker of glycemic control over time," Flores-Riveros said. The *JEM* paper "stresses the need to closely monitor blood sugar levels on a continuous day-to-day basis in order to avoid the epigenetic changes that are proven to occur here."

Jay Lichter, chairman and CEO of **Akesis Pharmaceuticals Inc.**, agreed that "this suggests you need to measure patient blood sugar levels more often. The question now to ask is: With the technology moving in the direction of self-monitoring at home, will it be possible for patients to take measurements more often?"

Akesis is developing AKP-020, a vanadium insulin-enhancing compound that is in Phase III testing to treat diabetes.

Epigenetic corrections

In addition to suggesting the need to avoid glucose spikes, the *JEM* paper could open new avenues to help prevent cardiovascular complications of diabetes. Indeed, many companies were optimistic about targeting the epigenetic changes at the points of p65 transcription, histone methylation or superoxide production, which link hyperglycemia to vascular damage.

El-Osta said his lab is now focused on showing that post-transcriptional changes to p65 expression is the primary mechanism underlying the vascular complications of diabetes.

“At this stage we have started to identify the potential regulatory factors and pathways that could be involved and hopefully we will be able to determine the critical step that will become a therapeutic target,” he told *SciBX*.

Flores-Riveros noted that preventing epigenetic changes could help avoid inflammation and therefore limit complications.

Hollis-Eden is developing Triolex, an NF- κ B inhibitor that is in Phase II testing to treat diabetes. Flores-Riveros said the product mediates cytokines and chemokines that control inflammation, specifically MCP-1.

“The researchers have looked at the effects of transient hyperglycemia on the two proinflammatory genes—*MCP-1* and *VCAM-1*—which is especially relevant because these have been linked in the long-term to development of cardiovascular disease,” Flores-Riveros said.

He added that increased expression of those proinflammatory genes has been linked not only to development of cardiovascular disease, but also to progression from insulin intolerance to full-blown type 2 diabetes.

Flores-Riveros thinks preventing epigenetic changes could be a good add-on therapy to insulin. Insulin can help control fluctuations in glucose levels, whereas targeting the epigenetic alterations could prevent damage when fluctuations do occur.

However, Bayer's Simmons noted that “there have been lots of candidate pathways to prevent these downstream perturbations, but they have all fallen by the wayside.”

He added: “One problem is that they tend to be common, shared

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**—Assam El-Osta,
Baker Medical Research Institute**

pathways. It would be difficult to design a drug without many unwanted side effects.”

Vincent Mooser, director of disease genetics at **GlaxoSmithKline plc**, agreed. “For therapeutic purposes, drugs designed to modify mitochondrial functions and reactive oxygen species generation have been tested but have been disappointing,” he said.

Mooser noted that compounds such as histone deacetylase (HDAC) inhibitors could prevent activation of p65 transcription. But these tend to be very toxic in their oncology settings and might not be useful for a chronic disease.

GSK markets several diabetes therapeutics, including the peroxisome proliferation activated receptor (PPAR) agonist Avandia rosiglitazone.

El-Osta told *SciBX* that the findings have not been patented and are not available for licensing.

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