**Trends in Cancer**

**Spotlight**

# Glucose regulated TET2 activity links cancer to diabetes

**Hari R. Singh[1] & Stefan H. Stricker[2-3]**

1. Physics Department E14, Technical University Munich, 85748 Garching, Germany.
2. MCN Junior Research Group, Munich Center for Neurosciences, Ludwig-Maximilian-Universität, Biomedical Center, Grosshaderner Strasse 9, Planegg-Martinsried 82152, Germany.
3. Epigenetic Engineering, Institute of Stem Cell Research, Helmholtz Zentrum, German Research Center for Environmental Health, Neuherberg, Germany.

**Email:** h.singh@tum.de, stricker@biologie.uni-muenchen.de

## Abstract

**Diabetes has long been associated with an increased risk of cancer. While likely many molecular connections exist between the diseases, a recent publication discovered a clear molecular link, demonstrating that a glucose dependent destabilization of the DNA de-methylase TET2 can promote malignant transformation via an AMPK dependent phospho-switch.**

Abnormal cellular metabolism and altered DNA methylation are hallmarks of cancer. In a recent report, Wu *and colleagues* [1] found a link between these two processes, and offered an explanation for why diabetic patients may have a significant higher risk of developing cancer [2].

Hyperglycemia, in particular chronically high glucose levels in blood, is a key indication in diabetes mellitus patients. But diabetes is not the only medical condition with an altered metabolic state; many tumors are characterized by an elevated glucose uptake, a phenomenon which is taken advantage of during positron emission tomography (PET) when cancer tissue is imaged using a glucose analogue. Evidence that this metabolic feature might be more than merely a consequence of proliferation comes from epidemiological studies indicating that the risk of several cancer types (e.g. colorectal, pancreas, breast and liver) is clearly increased in diabetic patients [2]. Moreover, it became recently clear that the anti-diabetic drug metformin can be used as an effective anti-cancer agent indicating that these two common diseases might also share therapeutic targets [3]. Metformin is known to act at

least partially via the AMP activated kinase pathway (AMPK). AMPK is the main energy sensor in the cell integrating both systemic and cellular energy states [4]. AMPK is a heterotrimeric protein complex that is activated upon starvation due to an increased ratio of AMP/ATP. The anti-diabetic drug metformin is a specific inhibitor of the mitochondrial Complex-I, thereby increasing AMP [4]. This indirect AMPK stimulating effect of metformin has been harnessed by clinicians for the treatment of type 2 diabetes for a long time [5]; only since recently it became clear that it also could be used as an anti-cancer agent. Whether this however, is also dependent on AMPK activation is unclear so far [5].

DNA is embedded in the nucleus of the cell, where its activity is regulated by a large number of epigenetic processes (including, but not limited to: interactions of the DNA with histone proteins, higher order packaging of DNA and last, but not least chemical modifications of histone proteins and DNA residues). DNA methylation, which in human cells consists of 5' methylation of cytosines that are part of CpG pairs, is a very common epigenomic mark. 5mC (5-methyl-cytosine) has been frequently associated to transcriptional repression and although it is thought to be rather stable it can be locally removed through the enzymatic activity of the TET (Ten eleven translocation) dioxygenases 1, 2 and 3 [6]. The product of this reaction is, however, not an unmodified cytosine, but a new epigenomic mark, 5-hydroxymethyl-cytosine (5hmC). 5hmC is much less frequently found in the human epigenome, and although it is often seen as merely a product of DNA de-methylation, evidence accumulates that it might have functional gene-regulatory roles itself [7].

The oxidation reaction catalysed by TET requires α-ketoglutarate, a TCA cycle intermediate that serves as one molecular link connecting the metabolic state of the cell with its epigenome [7]. Since in the Krebs cycle α-ketoglutarate is generated from glucose and a relationship between diabetes and α-ketoglutarate blood levels has already been investigated in the 50s [8] Wu *and colleagues* first aimed to investigate whether hyperglycemic conditions result in altered 5hmC levels in human cells. Because TET enzymes consume α-ketoglutarate in their enzymatic reaction they expected a global increase of this epigenomic mark. Instead, they found the opposite, both, in vitro (in cancer cells cultured in high glucose) and in vivo (in blood of diabetic patients) indicating that glucose regulated mechanisms in the cell can control TET2.

The single most important cellular mechanism for fast regulation of enzyme activity is the addition of chemical modifications to some residues of the mature protein. Such post-translational modifications can influence many aspects of the proteins behavior including its ability to interact, its stability, its cellular localization and its reaction kinetics. Interestingly

TET2 turns out to be a substrate of AMPK, which in low glucose condition phosphorylates the DNA-demethylase. This phosphorylation increases the stability of the protein by protection from the protease Calpain and as a consequence global 5hmC levels rise in the cell. Interestingly, phosphorylation is already the second post-translational mechanism described that has the capacity to regulate TET2 stability, as recently Zhang *and colleagues* presented similar findings for TET2 acetylation [9].

Wu *and colleagues* find that the glucose induced alterations (on the epigenome and transcriptome) are fairly reminiscent to those occurring during tumorigenesis. Although this first came as a surprise, it is plausible when one takes into account that epigenomic perturbations are hallmarks of many types of cancer and the global loss of 5hmC is an especially frequent event [10]. How these epigenomic changes are established and whether they have causal roles in the development (or progression) of the disease is still vividly discussed [6, 10, 11]. Evidence that loss of the TET2-5hmC axis in cancer is more than merely a consequence (for example of metabolic changes) is provided by the finding that TET proteins are mutated in some cancer types (most prominently in those of myeloid origin [12]). To investigate whether glucose controlled regulation of TET2 stability is of relevance for cancer cells the authors turn towards a xenograft model using human melanoma cells and show that the tumor suppressing effect of TET2 expression is significantly reduced when mice were kept under diabetic conditions. This suggests that blood sugar levels might influence tumorigenicity of some cancer types. Moreover, in this model the anti-tumorigenic effects of (the anti-diabetic drug) metformin act downstream of glucose and depend on a working AMPK-TET2 axis.

What remains unclear, is to which degree glucose regulated hydroxymethylation levels are indeed causal in the two human diseases.Ten eleven translocation proteins have pleiotropic enzymatic (and non-enzymatic) entanglements in gene regulation and metabolic circuits alike [7, 13]. In this context it should be noted that only few of the many glucose induced transcriptomic changes *Wu* and colleagues find depend on a catalytically active TET2 and correlate a differentially 5-hydroxymethylated region (DhMRs). This might challenge to which degree diabetic and/or tumor suppressing roles of TET2 really depend on the detected epigenomic alterations. In that regard it is worth noticing that in the few cases in which cancer specific chromatin features have been globally rectified, no reduction of tumorigenicity has been detected [14, 15].

In summary, Wu et al. present a molecular link between two of the largest burdens of humanity, diabetes and cancer, by showing that the tumor suppressor TET2 is adamantly

controlled by glucose levels. This finding might not only offer an explanation why diabetic patients develop more often certain forms of cancer, but also why the diabetic drug metformin has tumor suppressing potential.

**Figure1 (Legend): A phospho-switch that connects metabolic state with cancer epigenome.** Figure 1 Shows the pathway that employs a phospho-switch on the TET2 demethylase. Low glucose upon starvation is sensed by AMPK which gets activated upon AMP binding, upon auto-phosphorylation AMPK gets activated and phosphorylates TET2 on serine 99 that prevents it from degradation by calpain mediated cleavage and therefore allows it to convert the Methyl-Cytosine(mC) to 5-hydroxymethy-Cytosine (5hmC). Chronic high glucose levels in diabetic patients will ultimately destabilize the TET2 by preventing its phosphorylation by pAMPK therefore leading to a loss of 5hmC, a hallmark of cancer epigenomes. Further, authors identify that metformin mediated activation of the AMPK is one of the mechanisms whereby metformin impart its anti-cancer properties.

## References


1. Wu, D. et al. (2018) Glucose-regulated phosphorylation of TET2 by AMPK reveals a pathway linking diabetes to cancer. Nature 559 (7715), 637-641.
2. Vigneri, P. et al. (2009) Diabetes and cancer. Endocr Relat Cancer 16 (4), 1103-23.
3. Evans, J.M. et al. (2005) Metformin and reduced risk of cancer in diabetic patients. BMJ 330 (7503), 1304-5.
4. Lin, S.C. and Hardie, D.G. (2018) AMPK: Sensing Glucose as well as Cellular Energy Status. Cell Metab 27 (2), 299-313.
5. Kasznicki, J. et al. (2014) Metformin in cancer prevention and therapy. Ann Transl Med 2 (6), 57.
6. Stricker, S.H. et al. (2017) From profiles to function in epigenomics. Nat Rev Genet 18 (1), 51-66.
7. Wu, X. and Zhang, Y. (2017) TET-mediated active DNA demethylation: mechanism, function and beyond. Nat Rev Genet 18 (9), 517-534.
8. Smith, M.J. and Taylor, K.W. (1956) Blood pyruvate and alpha-ketoglutarate in normal and diabetic subjects. Br Med J 2 (5000), 1035-8.
9. Zhang, Y.W. et al. (2017) Acetylation Enhances TET2 Function in Protecting against Abnormal DNA Methylation during Oxidative Stress. Mol Cell 65 (2), 323-335.
10. Ficz, G. and Gribben, J.G. (2014) Loss of 5-hydroxymethylcytosine in cancer: cause or consequence? Genomics 104 (5), 352-7.
11. Baylin, S. and Bestor, T.H. (2002) Altered methylation patterns in cancer cell genomes: cause or consequence? Cancer Cell 1 (4), 299-305.
12. Delhommeau, F. et al. (2009) Mutation in TET2 in myeloid cancers. N Engl J Med 360 (22), 2289-301.

13. Lian, H. et al. (2016) The emerging insights into catalytic or non-catalytic roles of TET proteins in tumors and neural development. Oncotarget 7 (39), 64512-64525.
14. Stricker, S.H. et al. (2013) Widespread resetting of DNA methylation in glioblastoma-initiating cells suppresses malignant cellular behavior in a lineage-dependent manner. Genes Dev 27 (6), 654-69.
15. Chao, M.P. et al. (2017) Human AML-iPSCs Reacquire Leukemic Properties after Differentiation and Model Clonal Variation of Disease. Cell Stem Cell 20 (3), 329-344 e7.