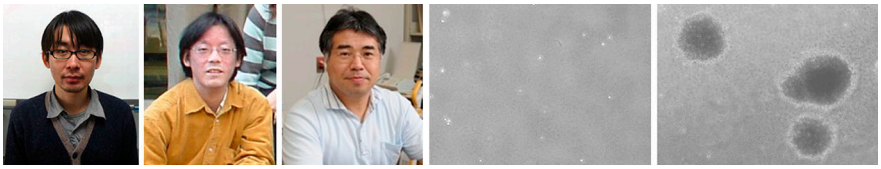
Text by Ben Short
bshort@rockefeller.edu



# Mdm2 pulls the plug on glycolysis

The ubiquitin ligase induces senescence by promoting degradation of the glycolytic enzyme phosphoglycerate mutase.

## Focal Point



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**(Left to right) Takumi Mikawa, Takeshi Maruyama, Hiroshi Kondoh, and colleagues (not pictured) reveal that, in response to DNA damage or oncogene expression, the ubiquitin ligase Mdm2 induces cell senescence by down-regulating the glycolytic enzyme phosphoglycerate mutase (PGAM). In collaboration with the kinase Pak1, Mdm2 targets PGAM for degradation, thereby inhibiting glycolysis and suppressing tumorigenesis. Mouse embryonic fibroblasts expressing PGAM and constitutively active Ras become senescent in the presence of wild-type Mdm2 (second from right), but expression of a catalytically inactive Mdm2 mutant enhances glycolysis and promotes cell transformation (far right).**

Cancer cells have long been known to have higher rates of glycolysis than normal cells, a phenomenon named the Warburg effect after its discoverer Otto Warburg (1). Enhanced glycolysis is thought to allow cancer cells to survive the hypoxic conditions they experience in the center of solid tumors. But Mikawa et al. reveal how damaged cells switch off glycolysis as they enter senescence and show that defects in this down-regulation may contribute to the early stages of tumorigenesis (2).

Various stresses, such as DNA damage or oncogene expression, can cause cells to cease proliferating and become senescent, thereby preventing the cells from transforming into tumor cells. In 2005, Hiroshi Kondoh and co-workers found that cells down-regulate glycolysis as they enter senescence and that overexpressing the glycolytic enzyme phosphoglycerate mutase (PGAM) prevented fibroblasts from exiting the cell cycle (3). PGAM is up-regulated in many tumors, stimulating not just glycolysis (4) but also the biosynthetic pentose phosphate pathway (5), but how cells regulate the enzyme's expression is unknown.

> "This suggests that enhanced glycolysis is important at a very early stage of tumorigenesis."

Working at Kyoto University in Japan, Kondoh and colleagues, led by Takumi Mikawa and Takeshi Maruyama, found that PGAM was ubiquitinated and degraded in primary fibroblasts following DNA damage or expression of oncogenic Ras, thereby inhibiting glycolysis as the cells entered senescence (2). "Many proteins must be phosphorylated before they can be ubiquitinated," Kondoh explains, "so we looked for kinases that affected PGAM stability."

Pak1 has previously been shown to phosphorylate PGAM (6), and Mikawa et al. found that overexpressing this kinase stimulated PGAM ubiquitination and degradation, inhibiting glycolysis and inducing cell senescence. Pak1's activity was enhanced by senescence-inducing stresses such as DNA damage. On the other hand, PGAM was stabilized by mutations that prevented the enzyme's phosphorylation.

Mikawa et al. then looked for the E3 ligase responsible for ubiquitinating PGAM. The researchers noticed that DNA damage could induce PGAM ubiquitination in cancer cells expressing wild-type p53 but not in cells lacking this tumor suppressor. p53 induces the E3 ubiquitin ligase Mdm2, and Mikawa et al. found that this enzyme bound to PGAM in vivo, an association enhanced by DNA damage and Pak1 phosphorylation. Mdm2 could ubiquitinate PGAM in vitro and was required in cells for the glycolytic enzyme's down-regulation in response to senescence-inducing stresses.

"Mdm2's role in tumorigenesis is controversial," Kondoh explains. "Many people consider it to be an oncogene because it blocks the transcriptional activity of p53 and promotes its degradation. But Mdm2 is also suggested to behave as a tumor suppressor that can inhibit cell proliferation." Mdm2's ability to down-regulate PGAM and induce cell senescence could fit with a tumor suppressor function for the E3 ligase, so Mikawa et al. tested the activity of several Mdm2 mutants found in human cancers. Each of these mutants could still inhibit p53-dependent transcription, but their ability to induce PGAM degradation was impaired.

One mutant in particular—Mdm2-M459I—was completely incapable of ubiquitinating and down-regulating PGAM and therefore helped to transform cells expressing oncogenic Ras. Primary fibroblasts expressing constitutively active Ras normally shut down glycolysis and enter senescence. But in the presence of Mdm2-M459I and constitutively expressed PGAM, cells increased glycolysis and continued to proliferate, allowing them to form tumors when injected into mice. "So PGAM can up-regulate glycolysis and transform cells," Kondoh says. "And Mdm2 can clearly, in some cases, act as a tumor suppressor by destabilizing PGAM. This suggests that enhanced glycolysis is important at a very early stage of tumorigenesis." Kondoh and colleagues now plan to investigate the in vivo effects of PGAM by generating transgenic and knockout mice.

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