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Just when you thought it was safe to assume, in some small way, that you understood the regulation of the tumour suppressor p53 by the ubiquitin ligase MDM2, two papers published in *Cancer Cell* show just how wrong you can be.

Xiongbin Lu and colleagues investigated how wild-type p53-induced phosphatase 1 (WIP1, also known as PPM1D) functions to restore normal p53 levels following the resolution of DNA damage. They initially showed that overexpression of WIP1 in irradiated cells reduced levels of p53 and its transcriptional activity compared with controls, and that suppression of WIP1 increased p53 stabilization and activity after exposure to ionizing radiation. Moreover, p53 levels and transcriptional activity were increased in irradiated *Wip1*$^{-/-}$ mouse embryonic fibroblasts (MEFs). So, does WIP1 function simply to dephosphorylate p53 and reduce its stability by enabling binding with MDM2? Expression of a p53 protein in which most of the known phosphorylation sites were mutated did not affect the capacity of WIP1 to inhibit p53 after exposure to radiation, and subsequent experiments showed that p53 is ubiquitylated when WIP1 is overexpressed, so dephosphorylation of p53 is not required for the effect of WIP1. Interestingly, despite being a p53 target gene, levels of MDM2 were reduced in *Wip1*-null MEFs, whereas other p53 target genes showed increased expression, indicating that WIP1 might also stabilize MDM2. Further experiments showed that WIP1 binds MDM2 and dephosphorylates it at the ataxia telangiectasia mutated (ATM) phosphorylation site, reversing this destabilizing phosphorylation. The stability of MDM2 is thought to be determined through autoubiquitylation by the interaction of MDM2 monomers. Overexpression of WIP1 prevents this interaction and also increases the amount of MDM2 that is bound to the deubiquitylase HAUSP in response to radiation.

These authors conclude that the reduction of p53 expression induced by WIP1 is primarily through stabilizing MDM2 rather than by a direct effect on p53. Given that overexpression of WIP1 is seen in several types of human cancer, molecules that target WIP1 activity might prove to be useful therapeutics.

One could also conclude from these findings that the oncogenic effect of WIP1 is in part due to its promotion of MDM2 stability through preventing its autoubiquitylation, presumably through the E3 ligase domain. However, work by Koji Itahana and colleagues indicates otherwise.

These authors made a mutant MDM2 protein that lacks one of the cysteines (C462A) crucial for E3 ligase activity. However, this protein still binds to p53. *Mdm2*-null mice are embryonic lethal owing to the activation of p53 during embryogenesis, and knock-in mice that were homozygous for the *Mdm2*^{C462A} allele were also embryonic lethal. This indicates that MDM2 that is devoid of E3 ligase activity cannot regulate p53 activity, and this was confirmed by the finding that *Mdm2*$^{C462A/C462A}$;*Trp53*$^{-/-}$ mice are viable. The early lethality of the *Mdm2*$^{C462A/C462A}$ mice prevented further investigation so Itahana and colleagues crossed these mice with knock-in heterozygous mice expressing one copy of the tamoxifen-regulatable p53ER^{TAM} protein. This p53 fusion protein is non-functional in the absence of tamoxifen, enabling *Mdm2*$^{C462A/C462A}$ embryos to develop and p53 function to be restored after embryogenesis. MEFs isolated from *Mdm2*$^{C462A/C462A}$;*Trp53*$^{ER/-}$ and *Mdm2*$^{+/+}$;*Trp53*$^{ER/-}$ mice were compared. As expected, p53 expression levels were higher in *Mdm2*$^{C462A/C462A}$;*Trp53*$^{ER/-}$ MEFs owing to the inability of MDM2 to degrade p53. Previous findings had indicated that degradation of MDM2 should also be affected by the C462A mutation; however, analysis of MDM2 levels in wild-type, heterozygous and homozygous *Mdm2*^{C462A} MEFs showed no differences. Furthermore, MDM2 degradation after the induction of a DNA-damage response was still evident in MDM2C462A-expressing cells. Therefore the E3 ligase activity of MDM2 is required for p53 degradation, but not for the degradation of MDM2 itself. This finding is in disagreement with many published *in vitro* studies. If not through autoubiquitylation, how are MDM2 protein levels regulated? Although Itahana and colleagues did not identify the E3 ligase in question, they showed that MDM2C462A is still subject to polyubiquitylation and degradation by the proteasome.

So, both papers show that MDM2 is crucial for regulating p53 function, but that the simple feedback loop between p53 and MDM2 is more complex than previously anticipated.

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ORIGINAL RESEARCH PAPERS Lu, X. *et al.* The Wip1 phosphatase acts as a gatekeeper in the p53–MDM2 autoregulatory loop. *Cancer Cell* **12**, 342–354 (2007) | Itahana, K. *et al.* Targeted inactivation of MDM2 RING finger E3 ubiquitin ligase activity in the mouse reveals mechanistic insights into p53 regulation. *Cancer Cell* **12**, 355–366 (2007)