MDM2 and p53: a Question of Balance

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Since the initial discovery of mutations in the p53 (also known as TP53) gene in colorectal carcinoma, notable progress has been made in elucidating the role of p53 in the regulation of gene expression, cycle progression, and apoptosis (7). It is now recognized that p53 is induced in response to some types of DNA damage and that it is a key component of the physiologic checkpoint pathway that governs the transition of cells from G1 to S phase in the cell cycle. In the absence of normal p53 function, cells may progress through the cell cycle despite genomic injury, producing daughter cells with ever increasing levels of genetic aberrations. In model systems, mutations in the p53 gene are, in fact, associated with an increased rate of aneuploidy and gene amplification (2). Biochemically, p53 exerts its regulatory effects as a transcription factor via binding to a specific recognition sequence in target genes. An unexpected connection between p53 and a previously described protein was made when immunoprecipitation studies (3,4) demonstrated that p53 interacts with the MDM2 oncoprotein. MDM2 (murine double minute-2) was originally identified as an amplified gene carried on double minutes in a transformed murine cell model (5). Overexpression of MDM2 was shown to produce a transformed phenotype, but its biochemical function remained a mystery until it was recognized that the 90-kd protein that coprecipitated with p53 was identical with MDM2. The p53/MDM2 interaction negatively regulates the transcriptional activating function of p53. Thus, overexpression of MDM2 results in an effect similar to mutational inactivation of p53. This finding has led to the proposition that MDM2 amplification may constitute an alternative pathway to mutational inactivation of p53 (3). This model would provide at least a partial solution to the puzzle of why p53 mutations only occur in a subset of otherwise apparently similar tumors and predicts that tumors with MDM2 amplification should lack p53 mutation and vice versa. The connection between these genes runs to another level, since p53 can induce expression of MDM2 via a p53 binding site in the MDM2 gene, suggesting the existence of an autoregulatory feedback loop in p53 function (6,7). The precise balance between the free and complexed forms of these proteins is probably critical in regulating p53 function.

Complementing these functional studies and strengthening the argument that MDM2 has a critical role in regulating p53, descriptive studies (3,8-10) have documented the amplification and overexpression of the MDM2 gene in human sarcomas and glial tumors. In addition, MDM2 amplification occurred only in the absence of p53 mutation, supporting the concept that amplification and p53 mutation are alternative mechanisms of p53 dysfunction (11). These observations are now extended by three studies reported in this issue of the Journal. Habuchi et al. (12) and Lianes et al. (13) add urothelial tumors to the list of cancers that carry MDM2 amplification and overexpression, while Floresen et al. (14) further associate MDM2 amplification and expression with p53 status in sarcomas. Amplification is, as expected, associated with high levels of MDM2 expression, and this association may be expected to result in altered p53 function in tumors bearing MDM2 amplification.

Nothing in cancer biology, including MDM2 amplification, is ever quite as simple as it first appears, and the genetic complexity of MDM2 amplification deserves comment. MDM2 maps to chromosome 12q14.3-15 (15), and the amplification unit is frequently quite large, including numerous flanking genes. Genes located centromeric to MDM2 that are variably amplified with it include SAS (16), CDK4 (17), CHOP (18), and GLI (19) as well as other less fully characterized transcripts (20). Each of these genes can be plausibly related to the regulation of cell growth. GLI has been demonstrated to have transforming activity. CHOP, normally a growth arrest-associated transcription factor, is the target of the tumor-specific t(12;16) in myxoid liposarcoma in which it forms a fusion protein with the FUS/TLS gene on chromosome 16 (21). SAS belongs to a large family of integral membrane proteins that may be related to signal transduction. CDK4 encodes a cyclin kinase with specificity for D cyclins. MDM2, SAS, and CDK4 are most frequently included in the amplification unit. A substantial proportion of sarcomas with amplification of chromosome 12 sequences have both CDK4 and MDM2 amplification, while only one of these genes is amplified in other tumors. This finding is of some importance in interpreting descriptive studies of tumors and is particularly interesting because of the role of CDK4 in cell cycle progression. CDK4 is known to be inhibited by WAF1/CIP1, a gene that was identified as a target of p53 induction (22). Recently, mutational inactivation of another CDK4 inhibitor, p16, has been observed in several cancer cell

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See "Note" section following "References."
CDK4 gene amplification may lead to a change in the balance between CDK4 inhibitors and CDK4, which favors cyclin kinase activity. Overexpression of either MDM2 or CDK4 can be conceptualized as opposing p53 function and therefore promoting cell cycle progression. Amplification of both genes may produce an additive or synergistic effect, perhaps explaining the retention of both genes in a large amplicon.

The proposal that p53 mutations predispose tumor cells to additional genetic injury in the form of gene amplification is examined by Habuchi et al. (12) in urothelial tumors. Using a panel of 14 oncogene probes, they could find no association between p53 status and the occurrence of gene amplification. Some tumors had amplification of two loci despite the absence of p53 mutations. Although limited by the particular panel of probes used, this conclusion is not entirely surprising in view of data from other cancers, notably neuroblastoma, a tumor in which gene amplification is common in the absence of p53 mutation (24). While p53 clearly has a role in maintaining genomic integrity, factors other than p53 mutation must be involved in rendering tumor cells permissive to gene amplification.

Both Lianes et al. (13) and Florenes et al. (14) identified tumor subsets with high levels of MDM2 expression in the absence of MDM2 gene amplification. In these instances, MDM2 expression is being driven by an effect other than increased gene copy number, with p53 activity a logical candidate for the cause of this phenomenon. The majority of such cases in both studies have intact p53 genes consistent with the proposition that MDM2 induction in these instances represents a response to p53. Interestingly, in the study of Lianes et al., these tumors were primarily of low grade and low stage. Lianes et al. also describe several cases with MDM2 overexpression and mutations in exon 8 of p53 and suggest that exon 8 mutations retain the ability to activate MDM2 expression. However, the single tumor with both MDM2 overexpression and p53 mutation identified by Florenes et al. had an alteration in exon 7. It is likely that additional, as yet undefined, factors also contribute to the regulation of MDM2 expression. It also is not clear that overexpression of MDM2 in the absence of gene amplification is biochemically equivalent to overexpression in the presence of gene amplification. Because of the relentless expression of MDM2 from dysregulated amplified genes and the influence of coamplified genes such as CDK4, this situation may be functionally different from the regulated overexpression of an intact MDM2 gene.

The mechanism that regulates cell cycle progression depends on a balance between multiple factors, and the ratio of p53 to MDM2 is tightly regulated, thereby protecting the cell from the propagation of DNA damage and preventing growth arrest once DNA repair is accomplished. Increasingly, as in these three studies (12-14), genotypic and phenotypic analysis of tumor specimens will need to examine multiple members of a given regulatory pathway as completely as possible to generate data that can be interpreted in the context of the underlying biochemical networks that control cell proliferation.

References

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Note

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