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Mechanisms Regulating Microtubule Binding, DNA Replication, and Apoptosis are Controlled by the Intestinal Tumor Suppressor APC

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Abstract

Colorectal cancer (CRC) results from the progressive accumulation of both genetic and epigenetic alterations that lead to the transformation of normal colorectal epithelium to benign (adenoma) and invasive (carcinoma) disease. Since its discovery in mutated form as the causative gene for familial adenomatous polyposis coli (FAP), as well as in many sporadic CRCs, the *APC* tumor suppressor has been shown to possess numerous functions within the cell including regulation of WNT signaling pathways and its transcriptional effects, cell migration, and chromosome separation. In recent years, other novel roles for APC have been investigated and suggest that APC can also repress DNA replication and enhance apoptosis. Further insights into the mechanisms by which APC contributes to tumor suppression will accelerate the diagnosis and treatment of CRC.

Keywords

APC; Apoptosis; Colorectal cancer; DNA replication; Tumor suppressor

Introduction

Colorectal cancer (CRC) represents nearly 10% of all new cases of cancer worldwide each year. According to current estimates from the National Cancer Institute, there will be 102,900 new colon and 39,670 rectal cancers newly diagnosed and 51,370 deaths in the United States this year, making it the third most common cancer and second most common cause of cancer-related deaths.

CRC results from the progressive accumulation of both genetic and epigenetic alterations that lead to the transformation of the normal colorectal epithelium to benign (adenoma) and invasive (carcinoma) disease, a process that can take up to 25 years [1]. Over the course of this transition from a normal cell to adenoma and to carcinoma, six or more molecular alterations occur, although the sequence may not be as important as the cumulative changes. The initial genetic events in the majority of CRCs are often loss of function mutations in both copies of the *APC* gene, a gene initially discovered from study of a rare but dominantly inherited colon cancer syndrome, known as familial adenomatous polyposis coli or FAP.

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Family studies, linkage analyses, and positional cloning of the *APC* gene, as well as subsequent mutational studies, identified germ-line and somatic mutations within *APC*, the majority of which are frameshift or nonsense mutations occurring between codons 1250 and 1464 (the mutation cluster region [MCR]) and result in a truncated protein [2–4]. Similar mutations in *APC* were identified in sporadic CRCs, suggesting that in this example, inherited or sporadic mutations of *APC* are associated with the occurrence of CRC. Although less than 1% of all CRC cases are the result of this familial syndrome, the pathways affected in those with inherited CRC provided important clues to the more common sporadic CRCs.

Given that loss of *APC* in both sporadic and familial CRC is often the initiating mutation in colorectal tumors, the APC protein is regarded as a gatekeeper. Genetic, biochemical, and mouse modeling studies have demonstrated the effects of APC on numerous cellular functions including the regulation of transcription, cell migration and adhesion, and chromosome separation. Most notably, APC is known for its role in the WNT signaling pathway, facilitating the degradation of the transcriptional effector β-catenin within a large complex of proteins; this is known as the canonical WNT signaling pathway. Loss of APC leads to the accumulation of β-catenin, which then is free to form complexes with members of the TCF family of transcription factors to enter the nucleus and alter the transcription of numerous genes, including *MYC* and *cyclin D1,* that are pro-growth, anti-apoptotic, or anti-differentiation [5–7].

Many models of CRC initiation and progression assume that the loss of APC is only synonymous with dysregulation of WNT signaling and aberrant nuclear accumulation of β-catenin. However, in the past few years, our research group and others have begun to define additional roles for APC within the cell, including control of DNA replication and apoptosis, that are independent of the transcriptional upregulation of WNT target genes and whose disruption could also contribute to the initiation and progression of CRC. The contribution of these two non-canonical pathways of APC function to tumor suppression will be discussed below.

Non-canonical Functions of APC

Although *APC* is mutated in the majority of CRCs, there are more unusual tumor subsets that harbor a stabilizing mutation of β-catenin that also leads to constitutive activation of the WNT pathway, and some tumors without evidence of activated WNT signaling, although the latter are often found in association with the inflammatory bowel diseases. Interestingly, tumors from the mutated β-catenin subset of CRCs are associated with a more favorable prognosis and therefore may be clinically distinct from those with mutated APC [8]. These data suggest that a different sequence of mutational events has occurred in these tumors. Additionally, although β-catenin and many of its transcriptional targets, including *MYC* and *cyclin D1*, are elevated in expression in adenomas from FAP patients compared to adjacent normal tissues, Blaker et al. [9] reported that in mildly dysplastic FAP adenomas, β-catenin is not nuclear. This observation was further supported by data from Anderson et al. [10] that identified detectable nuclear β-catenin accumulation in highly dysplastic adenomas but not in microadenomas from FAP patients. These results suggest that prior to activation of WNT signaling, mutant APC could affect other cellular pathways related to tumor suppression.

APC and DNA Replication

APC is a 310-kDa tumor suppressor protein normally expressed in non-proliferating colorectal epithelium and is essential for maintaining normal growth control and differentiation. As mentioned, APC contributes to a diverse set of cellular pathways and does so through several distinct functional domains, varied subcellular locations, and

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interactions with a diverse array of cellular proteins. Although APC is predominantly in the cytoplasm, normal, but not mutant, APC can be localized to the nucleus of some epithelial cell types [11]. Two nuclear localization signals (NLS) and four nuclear export sequences (NES) are found within APC [12, 13].

Accumulating evidence suggests a role for APC in DNA replication. Introduction of the *APC* gene into colon carcinoma cell lines prevents entry into and/or progression through S-phase [14–16]. APC controls entry into S-phase partially through its ability to regulate the cyclinD/RB pathway by disrupting β-catenin–mediated transcription. Other evidence points to a transcription-independent role for APC in cell cycle regulation. Cellular arrest induced by APC is only partially recovered by concomitant transfection of oncogenic β-catenin in the colon cancer cell line, SW480 [15]. In HCT116 colon cancer cells with wild-type *APC* and oncogenic β-catenin, the addition of exogenous APC also affects cell growth. Nuclear APC also counteracts the activation of WNT target genes by β-catenin [17].

Deka et al. [18] mapped three DNA-binding domains within APC: one in the β-catenin–binding domain and two in the carboxy-terminus. These domains have clusters of S (T)PXX sequence motifs that, similarly to other proteins containing these motifs, bind preferentially to A/T-rich DNA sequences rather than to specific DNA sequences [18]. APC also associates with microtubules (MTs) by direct binding and by indirect binding through EB1 and kinesin-2. This occurs within regions of the APC carboxy-terminus and overlaps with its DNA-binding regions. The interaction of APC with MTs is implicated in MT assembly and bundling in vitro [19], in MT stabilization in vitro [20], and spindle formation [21]. Additionally, the binding of APC and EB1 is required for EB1-induced MT polymerization in vitro. Formin mDia2 interacts in a complex with APC and EB1 and leads to Rho-induced MT stabilization [22]. Recent literature has defined new protein-binding partners of APC that also regulate its interactions with MTs. Murawala et al. [23•] demonstrated that the middle region of APC, its β-catenin–binding region, targets APC to the plus-ends of MTs and that this region of APC is associated with the growing end of the MT. Furthermore, they report that Nup358, implicated in nucleocytoplasmic transport, and kinesin-2 play a role in targeting APC to this specific set of MT-plus ends. In cells depleted of Nup358 by RNA interference, cytoplasmic APC fails to localize to the cell cortex. The interaction of APC with various components of the MT machinery may also affect its cellular localization to the nucleus when DNA replication is ongoing. More recently, Dikovskaya et al. [24•] demonstrated that APC directly interacts with importin-β, a transport factor involved in nuclear import of various cargo molecules during interphase that contribute to mitotic spindle formation by delivering spindle-promoting molecules to specific cellular regions during mitosis. They reported that importin-β inhibits the ability of APC to assemble and bundle MTs in vitro, and to promote the formation of MT structures in *Xenopus laevis* egg extracts. The MT-binding region of APC is deleted in most tumor-associated mutations of *APC*, suggesting subsequent effects on cell migration. Finally given the function of APC at mitotic spindles, *APC* mutation contributes to chromosomal instability [25, 26].

To study the effect of APC on DNA replication directly our laboratory used an in vitro system that includes *Xenopus laevis* egg extract and exogenously added DNA template in order to separate the transcriptional effects of APC from its non-transcriptional effects. This cell-free system is transcriptionally and translationally quiescent as eggs are collected during maturation when these processes are inactive. These experiments demonstrated that full-length APC protein inhibited DNA replication and defined a specific carboxy-terminal segment, amino acids 2140-2421, that also conferred this inhibition. Again, this segment of APC is commonly deleted following *APC* mutation in CRCs. DNA binding by these S(T)PXX motifs was diminished by site-directed mutagenesis [27••]. Four cyclin-dependent kinase (CDK) consensus phosphorylation sites are also located within amino acids 2140–

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2421 of APC; phosphorylation of this region by CDKs weakens DNA binding and lessens replication inhibition, suggesting that APC-mediated DNA replication repression may be regulated by CDKs during the cell cycle. This region of APC that is required for repression of DNA replication also mediates the direct interaction of APC with microtubules [28, 29]. Amino acids 2244–2312 of APC are strikingly similar to the protein Tau, a microtubule-associated protein capable of repressing DNA replication by binding to DNA [19, 30]. Therefore, it is possible that APC may inhibit replication in a manner similar to Tau.

Many questions remain to be addressed. That the DNA-binding region and the MT-binding domains overlap suggests that these two functions may be interrelated. Are they competitive and are proteins required for either function competing for binding sites within APC? Is the effect of APC on replication tied to its interaction with MT proteins? Are there other protein-binding partners that contribute to or regulate the suppression of replication by APC? How does APC localize to the nucleus and under what conditions is it directed there? Inversely, when and how does it exit the nucleus? It is possible that APC interactions with MT may assist in its cellular transport and localization as demonstrated with Nup358 [23•]? Since the regions of APC involved in both cell migration and DNA replication are deleted by the majority of cancer-associated *APC* mutations, it is important to understand how these processes are regulated to better target therapeutics. We may also address how mutation of APC or other members of the WNT signaling pathway does not always result in the same cellular phenotypes.

APC and Apoptosis

Another tumor suppressor function of APC is its contribution to the control of apoptosis or programmed cell death. Our research group and others have focused on how APC promotes cell death mechanistically and how loss of this function contributes to tumor formation. Similarly to its effects on DNA replication, APC effects on apoptosis can occur through both WNT-dependent and WNT-independent mechanisms. Some of these will be explored below.

Apoptosis is a normal, physiological program necessary for proper development and cell turnover. It maintains a constant cell number in continuously renewing cell populations. Its improper regulation can facilitate tumor formation even if normal cell cycle control is maintained [31, 32]. There are generally two major pathways for apoptosis: the death receptor pathway and the mitochondrial pathway [33, 34]. Activation of cell surface death receptors of the Fas/tumor necrosis factor receptor family triggers activation of an initiator caspase, which in turn cleaves and activates the executioner caspase procaspase-3 [35, 36]. In the mitochondrial pathway, death stimuli induce mitochondrial outer membrane permeabilization, leading to the release of mitochondrial pro-apoptotic proteins that induce caspase activation, such as cytochrome c and Smac/Diablo, or trigger caspase-independent effectors such as the apoptosis-inducing factor (AIF) and HtrA2 [34, 37]. The death receptor pathway in some if not all types of cells requires the help of the mitochondrial pathway to amplify the downstream caspase cascade [38–40]. Given its function in energy metabolism, the mitochondria can be considered the central integrator and regulator of cell life and death.

The colorectal epithelium is a dynamic cell population. Replicating stem cells reside at the base of the colonic crypts; post-mitotic daughter cells differentiate and migrate along the crypt axis within the lumen. Limited apoptosis occurs along the crypt axis with the main population of apoptotic cells of the intestinal epithelium observed at the luminal aspect of the crypts [41–43]. Similarities in the expression of APC within the differentiated epithelium of the crypts first suggested that APC may function in processes of cell differentiation, migration, and apoptosis. Several other lines of experimental evidence also suggested a role for APC in apoptosis. One of the first of these was a study by Morin et al. [44] that induced

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an *APC* transgene in the colon carcinoma cell line HT-29, lacking endogenous APC, and consequently increased cell death.

Our laboratory used the cell-free system described earlier in this article, an in vitro system composed of transcriptionally and translationally inactive egg extracts from *Xenopus laevis,* to study the effect of APC protein on apoptosis [45]. APC directly accelerated apoptosis rate in vitro, as measured by a time-course of caspase-mediated lamin B cleavage. This acceleration was concentration dependent. Addition of β-catenin did not alter the apoptotic time course, suggesting that the effects of APC on apoptosis are independent of β-catenin, transcriptionally and non-transcriptionally The specific apoptosis-inducing domain of APC was mapped to the amino-terminal amino acids 1–777 [46], a segment that corresponds to a stable group II caspase cleavage product of APC normally released during apoptosis [45, 47, 48]. Mutation of the APC aspartic acid residue to an alanine at amino acid 777 completely abolished in vitro cleavage of APC by recombinant group II caspase and rendered the full-length protein unable to accelerate apoptosis in vitro. Truncated APC, corresponding to a mutation found commonly in both familial and sporadic CRC, was also unable to accelerate apoptosis in vitro or in vivo, and was resistant to group II caspases. More recently, some of these earlier findings were validated in vivo by pre-treating HCT116 cells with a caspase-3 inhibitor and an apoptosis-inducing drug, 5-fluorouracil (5-FU), to demonstrate that endogenous full-length APC was cleaved upon treatment with 5-FU to produce a 90-Da form of APC. This effect was partially blocked by pretreatment with a caspase-3 inhibitor. Additionally, truncated APC in the colon cancer cell lines DLD-1 and SW480 colon was not cleaved as efficiently as wild-type protein in vivo [49••]. These data suggest that caspase-cleaved APC assumes a modified protein confirmation that may result in differences in protein binding while truncated APC may be resistant to cleavage and protein binding.

Given that APC retains the armadillo repeat region (ARM) that mediates protein–protein interactions following caspase-cleavage, APC may accelerate apoptosis in part by interacting with other proapoptotic or antiapoptotic proteins. A yeast-two-hybrid study reported by Polakis [50] used this ARM-repeat domain of APC as bait to search for interacting proteins. The study identified a mitochondrial tumor suppressor protein, hTID-1, the human homologue of *dtid*, the lethal tumorous imaginal disc (l(2)tid) gene in *Drosophila*. Homozygous mutation of this gene in *Drosophila* leads to neoplastic growth of the larval imaginal discs. *hTID-1* encodes three splice-forms encoding three cytosolic (hTID-50, hTID-48, and hTID-46) and three mitochondrial (hTID-43, hTID-40, and hTID-38) proteins [51, 52]. Syken et al. [53] reported that *hTID-1* encoded two mitochondrial splice variants, 43-kDa and 40-kDa, that had opposing effects on responses to exogenous apoptotic stimulus. The 43-kDa hTID1 isoform facilitates apoptosis triggered by either TNF-α or DNA damage by mitomycin C treatment. Conversely, the 40-kDa hTID1 isoform suppresses apoptosis. The role of the 38-kDa hTID-1 isoform in apoptosis is unknown, although it is expressed at a low level in colon epithelium [54].

Experiments by Kurzik-Dumke et al. [52] demonstrated that full-length APC interacts with cytosolic hTID-1, both the 50-kDa and 48-kDa protein isoforms, in a complex with Hsp70, Hsc70, Actin, Dvl, and Axin. A subsequent study demonstrated that alteration of hTID-1 isoform expression is accompanied by APC redistribution within the cell, loss of polarity, and tumor progression [54]. Other recent data from our laboratory indicate that caspase-cleaved APC also partners with hTID-1, specifically the 43-kDa and 40-kDa mitochondrial isoforms, defining a novel role for this protein-protein interaction in apoptotic signaling. Consistent with the original finding by Syken et al. that the 40-kDa hTID-1 isoform suppresses apoptosis, our data also demonstrated that the 40-kDa isoform preferentially binds APC and negatively regulates the proapoptotic function of caspase-cleaved APC. These studies raise many interesting questions regarding the mechanism by which APC

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accelerates apoptosis. For instance, does the ratio of hTID-1 isoforms change in response to DNA damage and release APC from its negative suppression to bind other proapoptotic proteins? With which proteins does APC interact? Are hTID-1 and APC in a complex that responds to cellular stress and are they modified to initiate or repress an apoptotic cascade? One hypothesis is that hTID-1 acts to chaperone caspase-cleaved APC to the mitochondria.

Recent experiments demonstrate that the subcellular localization of caspase-cleaved APC is mitochondrial [49]. These results complement data from Brocardo et al. [55••] that demonstrate mutant truncated APC proteins targeted to the mitochondria via the amino-terminus of APC. hTID-1 is a member of the DnaJ protein family and is a molecular co-chaperone of heat shock protein 70 (Hsp70) [53, 56]. hTID-1 interacts with HSP70 chaperones, which in turn mediate a variety of cellular activities in intercellular signaling linked to cell survival and growth regulation [57]. Emerging literature demonstrates antagonistic functions of HSP70 during apoptosis; the inhibitory effects of hTID-1 on APC functions in apoptosis may be mediated partially through HSP70 interactions. Unpublished data from our laboratory demonstrated that in the absence of hTID-1, caspase-cleaved APC continued to localize to the mitochondria, suggesting that the interaction of these proteins is not required to shuttle APC to the mitochondria. Again, these data point to unique protein partners of truncated and wild-type caspase-cleaved APC that modulate the ability of APC to enhance apoptosis. It is also unknown how APC is targeted to the mitochondria. Is the loss of the APC carboxy-terminus, either by mutation or by caspase-cleavage, responsible for alternatively tethering APC to the cytoplasmic network of MTs and localizing it within the cell? There may be an as yet unidentified mitochondrial-targeting sequence within the amino-terminus of APC.

Brocardo et al. [55••] reported that BCL2, another well-studied anti-apoptotic protein, interacts with mutant APC by recruiting and/or stabilizing BCL2 at the mitochondria. These data also suggest that truncated APC contributes to cell survival. *siRNA* knockdown of mutant APC in the SW480 colon carcinoma cell line increases apoptosis and correlates with loss of BCL2 at the mitochondria. It is well known that both mutant APC and BCL2 are elevated in many colon cancers. Does mutant APC have a higher affinity for BCL2 than wild-type APC? Are there proapop-totic BCL-2 family members, or other proapoptotic proteins, that more efficiently bind wild-type APC to enhance apoptosis? Suppression of mutant APC may relieve the cell of antiapoptotic protein interactions and allow apoptotic cascades to ensue.

APC transcriptionally represses expression of the gene encoding the antiapoptotic protein survivin, while APC mutation may also upregulate survivin protein to initiate colon cancer [58]. Survivin blocks apoptosis via caspase inhibition and antagonizing mitochondrial-dependent apoptosis. Huang et al. [59] observed that the status of APC in colon cells was related to sensitivity to histone-deacetylase (HDAC) inhibitor-induced apoptosis. Cells with wild-type APC were more sensitive to the HDAC inhibitors, VPA and SAHA, than those with mutant APC. Additionally, cells with mutant APC were sensitized to HDAC inhibitors when survivin levels were diminished using flavopiridol. These experiments suggest that survivin repression by APC is required for tumor sensitivity to this class of drugs. Other proteins with effects on apoptosis may also be transcriptionally regulated by APC and may be related to drug sensitivity. Understanding transcriptional and non-transcriptional effects of APC in the regulation of apoptosis will eventually enable tailoring of drug regimens to treat colon cancers.

Conclusions

APC has many functions as a tumor suppressor, including inhibition of cell motility, transcriptional regulation of other genes affecting growth, differentiation, and cell death. APC effects on WNT signaling are facilitated by the degradation of β-catenin but often overshadow other pathways through which APC contributes to the inhibition of tumor growth. We know from the literature that not all colon cancers are the same and that not all patients respond similarly to treatment. We also know that other mutations accumulate in the evolution of an adenoma to an adenocarcinoma, but that often this begins with an *APC* gene mutation. Understanding how APC truncation affects cellular proliferation or cell viability may influence how we think about treating patients with tumors. New targets could be used as biomarkers, with therapeutics then personalized by a tumor's molecular profile. Combinatorial therapeutics could be designed in light of these data. Many questions about APC functions in transcriptional regulation, microtubule dynamics, DNA replication, and apoptosis remain unanswered. Although APC has been studied for many years, it remains an intriguing protein with many undiscovered avenues for investigation.

Acknowledgments

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References

Papers of particular interest, published recently, have been highlighted as:

• Of importance

•• Of major importance.

1. Fearon ER, Vogelstein B. A genetic model for colorectal tumorigenesis. Cell. 1990; 61:759–67. [PubMed: 2188735]
2. Nishisho I, Nakamura Y, Miyoshi Y, et al. Mutations of chromosome 5q21 genes in FAP and colorectal cancer patients. Science. 1991; 253:665–9. [PubMed: 1651563]
3. Groden J, Thliveris A, Samowitz W, et al. Identification and characterization of the familial adenomatous polyposis coli gene. Cell. 1991; 66:589–600. [PubMed: 1651174]
4. Leppert M, Dobbs M, Scambler P, et al. The gene for familial polyposis coli maps to the long arm of chromosome 5. Science. 1987; 238:1411–3. [PubMed: 3479843]
5. Nathke I. Cytoskeleton out of the cupboard: colon cancer and cytoskeletal changes induced by loss of APC. Nat Rev Cancer. 2006; 6:967–74. [PubMed: 17093505]
6. Nathke IS. The adenomatous polyposis coli protein: the Achilles heel of the gut epithelium. Annu Rev Cell Dev Biol. 2004; 20:337–66. [PubMed: 15473844]
7. Watson SA. Oncogenic targets of beta-catenin-mediated transcription in molecular pathogenesis of intestinal polyposis. Lancet. 2001; 357:572–3. [PubMed: 11558478]
8. Samowitz WS, Powers MD, Spirio LN, et al. Beta-catenin mutations are more frequent in small colorectal adenomas than in larger adenomas and invasive carcinomas. Cancer Res. 1999; 59:1442–4. [PubMed: 10197610]
9. Blaker H, Scholten M, Sutter C, et al. Somatic mutations in familial adenomatous polyps. Nuclear translocation of beta-catenin requires more than biallelic APC inactivation. Am J Clin Pathol. 2003; 120:418–23. [PubMed: 14502807]
10. Anderson CB, Neufeld KL, White RL. Subcellular distribution of Wnt pathway proteins in normal and neoplastic colon. Proc Natl Acad Sci USA. 2002; 99:8683–8. [PubMed: 12072559]
11. Neufeld KL, White RL. Nuclear and cytoplasmic localizations of the adenomatous polyposis coli protein. Proc Natl Acad Sci USA. 1997; 94:3034–9. [PubMed: 9096341]

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12. Zhang F, White RL, Neufeld KL. Phosphorylation near nuclear localization signal regulates nuclear import of adenomatous polyposis coli protein. Proc Natl Acad Sci USA. 2000; 97:12577–82. [PubMed: 11050185]

13. Rosin-Arbesfeld R, Townsley F, Bienz M. The APC tumour suppressor has a nuclear export function. Nature. 2000; 406:1009–12. [PubMed: 10984057]

14. Baeg GH, Matsumine A, Kuroda T, et al. The tumour suppressor gene product APC blocks cell cycle progression from G0/G1 to S phase. EMBO J. 1995; 14:5618–25. [PubMed: 8521819]

15. Heinen CD, Goss KH, Cornelius JR, et al. The APC tumor suppressor controls entry into S-phase through its ability to regulate the cyclin D/RB pathway. Gastroenterology. 2002; 123:751–63. [PubMed: 12198702]

16. Carson DJ, Santoro IM, Groden J. Isoforms of the APC tumor suppressor and their ability to inhibit cell growth and tumorigenicity. Oncogene. 2004; 23:7144–8. [PubMed: 15273719]

17. Sierra J, Yoshida T, Joazeiro CA, Jones KA. The APC tumor suppressor counteracts beta-catenin activation and H3K4 methylation at Wnt target genes. Genes Dev. 2006; 20:586–600. [PubMed: 16510874]

18. Deka J, Herter P, Sprenger-Haussels M, et al. The APC protein binds to A/T rich DNA sequences. Oncogene. 1999; 18:5654–61. [PubMed: 10523845]

19. Deka J, Kuhlmann J, Muller O. A domain within the tumor suppressor protein APC shows very similar biochemical properties as the microtubule-associated protein tau. Eur J Biochem. 1998; 253:591–7. [PubMed: 9654054]

20. Zumbrunn J, Kinoshita K, Hyman AA, Nathke IS. Binding of the adenomatous polyposis coli protein to microtubules increases microtubule stability and is regulated by GSK3 beta phosphorylation. Curr Biol. 2001; 11:44–9. [PubMed: 11166179]

21. Dikovskaya D, Newton IP, Nathke IS. The adenomatous polyposis coli protein is required for the formation of robust spindles formed in CSF Xenopus extracts. Mol Biol Cell. 2004; 15:2978–91. [PubMed: 15075372]

22. Wen Y, Eng CH, Schmoranzer J, et al. EB1 and APC bind to mDia to stabilize microtubules downstream of Rho and promote cell migration. Nat Cell Biol. 2004; 6:820–30. [PubMed: 15311282]

23•. Murawala P, Tripathi MM, Vyas P, et al. Nup358 interacts with APC and plays a role in cell polarization. J Cell Sci. 2009; 122:3113–22. This article presents and discusses experiments to demonstrate that APC is targeted to the plus-ends ofMTs by kinesin-2 and the nuclear-cytoplasmic transport protein Nup358. [PubMed: 19654215]

24•. Dikovskaya D, Li Z, Newton IP, et al. Microtubule assembly by the Apc protein is regulated by importin-beta-RanGTP. J Cell Sci. 2009; 123:736–46. This article presents and discusses experiments to demonstrate that APC directly interacts with importin-β, a transport factor involved in nuclear import, to inhibit the ability of APC to assemble and bundle MTs. [PubMed: 20144988]

25. Fodde R, Kuipers J, Rosenberg C, et al. Mutations in the APC tumour suppressor gene cause chromosomal instability. Nat Cell Biol. 2001; 3:433–8. [PubMed: 11283620]

26. Dikovskaya D, Schiffmann D, Newton IP, et al. Loss of APC induces polyploidy as a result of a combination of defects in mitosis and apoptosis. J Cell Biol. 2007; 176:183–95. [PubMed: 17227893]

27••. Qian J, Sarnaik AA, Bonney TM, et al. The APC tumor suppressor inhibits DNA replication by directly binding to DNA via its carboxyl terminus. Gastroenterology. 2008; 135:152–62. This article presents and discusses experiments to demonstrate that the carboxy-terminal of APC contains DNA-binding sequences to inhibit DNA replication directly. [PubMed: 18474248]

28. Munemitsu S, Souza B, Muller O, et al. The APC gene product associates with microtubules in vivo and promotes their assembly in vitro. Cancer Res. 1994; 54:3676–81. [PubMed: 8033083]

29. Smith KJ, Levy DB, Maupin P, et al. Wild-type but not mutant APC associates with the microtubule cytoskeleton. Cancer Res. 1994; 54:3672–5. [PubMed: 8033082]

30. Li W, Wang XS, Qu MH, et al. Human protein tau represses DNA replication in vitro. Biochim Biophys Acta. 2005; 1726:280–6. [PubMed: 16226838]

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31. Jacobson MD, Weil M, Raff MC. Programmed cell death in animal development. Cell. 1997; 88:347–54. [PubMed: 9039261]
32. Hengartner MO. The biochemistry of apoptosis. Nature. 2000; 407:770–6. [PubMed: 11048727]
33. Kaufmann SH, Hengartner MO. Programmed cell death: alive and well in the new millennium. Trends Cell Biol. 2001; 11:526–34. [PubMed: 11719060]
34. Green DR, Reed JC. Mitochondria and apoptosis. Science. 1998; 281:1309–12. [PubMed: 9721092]
35. Salvesen GS, Dixit VM. Caspase activation: the induced-proximity model. Proc Natl Acad Sci USA. 1999; 96:10964–7. [PubMed: 10500109]
36. Budihardjo I, Oliver H, Lutter M, et al. Biochemical pathways of caspase activation during apoptosis. Annu Rev Cell Dev Biol. 1999; 15:269–90. [PubMed: 10611963]
37. Green DR, Evan GI. A matter of life and death. Cancer Cell. 2002; 1:19–30. [PubMed: 12086884]
38. Youle RJ, Strasser A. The BCL-2 protein family: opposing activities that mediate cell death. Nat Rev Mol Cell Biol. 2008; 9:47–59. [PubMed: 18097445]
39. Green DR, Chipuk JE. Apoptosis: stabbed in the BAX. Nature. 2008; 455:1047–9. [PubMed: 18948940]
40. Chipuk JE, Bouchier-Hayes L, Green DR. Mitochondrial outer membrane permeabilization during apoptosis: the innocent by-stander scenario. Cell Death Differ. 2006; 13:1396–402. [PubMed: 16710362]
41. Bedi A, Pasricha PJ, Akhtar AJ, et al. Inhibition of apoptosis during development of colorectal cancer. Cancer Res. 1995; 55:1811–6. [PubMed: 7728743]
42. Hall PA, Coates PJ, Ansari B, Hopwood D. Regulation of cell number in the mammalian gastrointestinal tract: the importance of apoptosis. J Cell Sci. 1994; 107(Pt 12):3569–77. [PubMed: 7706406]
43. Merritt AJ, Potten CS, Watson AJ, et al. Differential expression of bcl-2 in intestinal epithelia. Correlation with attenuation of apoptosis in colonic crypts and the incidence of colonic neoplasia. J Cell Sci. 1995; 108(Pt 6):2261–71. [PubMed: 7673346]
44. Morin PJ, Vogelstein B, Kinzler KW. Apoptosis and APC in colorectal tumorigenesis. Proc Natl Acad Sci USA. 1996; 93:7950–4. [PubMed: 8755583]
45. Steigerwald K, Behbehani GK, Combs KA, et al. The APC tumor suppressor promotes transcription-independent apoptosis in vitro. Mol Cancer Res. 2005; 3:78–89. [PubMed: 15755874]
46. Qian J, Steigerwald K, Combs KA, et al. Caspase cleavage of the APC tumor suppressor and release of an amino-terminal domain is required for the transcription-independent function of APC in apoptosis. Oncogene. 2007; 26:4872–6. [PubMed: 17297457]
47. Webb SJ, Nicholson D, Bubb VJ, Wyllie AH. Caspase-mediated cleavage of APC results in an amino-terminal fragment with an intact armadillo repeat domain. FASEB J. 1999; 13:339–46. [PubMed: 9973322]
48. Browne SJ, MacFarlane M, Cohen GM, Paraskeva C. The adenomatous polyposis coli protein and retinoblastoma protein are cleaved early in apoptosis and are potential substrates for caspases. Cell Death Differ. 1998; 5:206–13. [PubMed: 10200466]
49••. Qian J, Perchiniak EM, Sun K, Groden J. The mitochondrial protein hTID-1 partners with the caspase-cleaved adenomatous polyposis cell tumor suppressor to facilitate apoptosis. Gastroenterology. 2010; 138:1418–28. This article presents and discusses experiments to demonstrate that caspase cleavage of APC releases an amino-terminal segment that relocates to the mitochondria and interacts with an hTID-1 isoform to promote apoptosis. [PubMed: 19900451]
50. Polakis P. The adenomatous polyposis coli (APC) tumor suppressor. Biochim Biophys Acta. 1997; 1332:F127–47. [PubMed: 9196022]
51. Yin X, Rozakis-Adcock M. Genomic organization and expression of the human tumorous imaginal disc (TID1) gene. Gene. 2001; 278:201–10. [PubMed: 11707338]
52. Kurzik-Dumke U, Czaja J. Htid-1, the human homolog of the Drosophila melanogaster l(2)tid tumor suppressor, defines a novel physiological role of APC. Cell Signal. 2007; 19:1973–85. [PubMed: 17588722]

53. Syken J, De-Medina T, Munger K. TID1, a human homolog of the Drosophila tumor suppressor l(2)tid, encodes two mitochondrial modulators of apoptosis with opposing functions. Proc Natl Acad Sci USA. 1999; 96:8499–504. [PubMed: 10411904]

54. Kurzik-Dumke U, Horner M, Czaja J, et al. Progression of colorectal cancers correlates with overexpression and loss of polarization of expression of the htid-1 tumor suppressor. Int J Mol Med. 2008; 21:19–31. [PubMed: 18097612]

55••. Brocardo M, Lei Y, Tighe A, et al. Mitochondrial targeting of adenomatous polyposis coli protein is stimulated by truncating cancer mutations: regulation of Bcl-2 and implications for cell survival. J Biol Chem. 2008; 283:5950–9. This article presents and discusses experiments to demonstrate that truncated APC interacts with Bcl2 to stabilize Bcl2 at the mitochondria and to promote cell survival. [PubMed: 18160396]

56. Schilling B, De-Medina T, Syken J, et al. A novel human DnaJ protein, hTid-1, a homolog of the Drosophila tumor suppressor protein Tid56, can interact with the human papillomavirus type 16E7 oncoprotein. Virology. 1998; 247:74–85. [PubMed: 9683573]

57. Trentin GA, Yin X, Tahir S, et al. A mouse homologue of the Drosophila tumor suppressor l(2)tid gene defines a novel Ras GTPase-activating protein (RasGAP)-binding protein. J Biol Chem. 2001; 276:13087–95. [PubMed: 11116152]

58. Zhang T, Otevrel T, Gao Z, et al. Evidence that APC regulates survivin expression: a possible mechanism contributing to the stem cell origin of colon cancer. Cancer Res. 2001; 61:8664–7. [PubMed: 11751382]

59. Huang X, Guo B. Adenomatous polyposis coli determines sensitivity to histone deacetylase inhibitor-induced apoptosis in colon cancer cells. Cancer Res. 2006; 66:9245–51. [PubMed: 16982769]

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