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The E2F transcription factor as a regulator of cell proliferation

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The E2F family of transcription factors plays a crucial role in the control of cell cycle progression and regulates the expression of genes required for G1/S transition. These include genes encoding DNA replication proteins, enzymes involved in nucleotide synthesis and components of the origin recognition complex. E2F activity is modulated by multiple mechanisms including negative regulation by interaction with the product of the Rb tumor suppressor gene, RB. Binding of RB to E2F results in active transcriptional repression of E2F regulated genes and growth suppression. RB as well as molecules regulating the interaction between RB and E2F are often mutated in human cancer resulting in deregulated activity of E2F. The main goal of our laboratory is to elucidate the molecular mechanisms underlying both the regulation and the biological activities of E2F.

E2F up-regulates expression of genes involved in DNA replication, DNA repair and mitosis

We use DNA microarrays and cell lines containing either inducible E2F-1 or inducible E2F-3 to identify novel E2F target genes. Our data demonstrate that E2F up-regulates the expression of tens of genes not previously described as E2F target genes. Many of these novel E2F-regulated genes code for components of the DNA replication machinery. Among them are the genes coding for replication protein A (RPA). RPA is the most abundant ssDNA binding protein in eukaryotic cells and it plays an essential role in DNA replication. It is a heterotrimer composed of 70kDa (RPA1), 32kDa (RPA2) and 14kDa (RPA 3) subunits and our data show that expression of the genes, coding for these three subunits, is up-regulated by E2F. The regulation of the RPA2 gene was studied in greater detail and

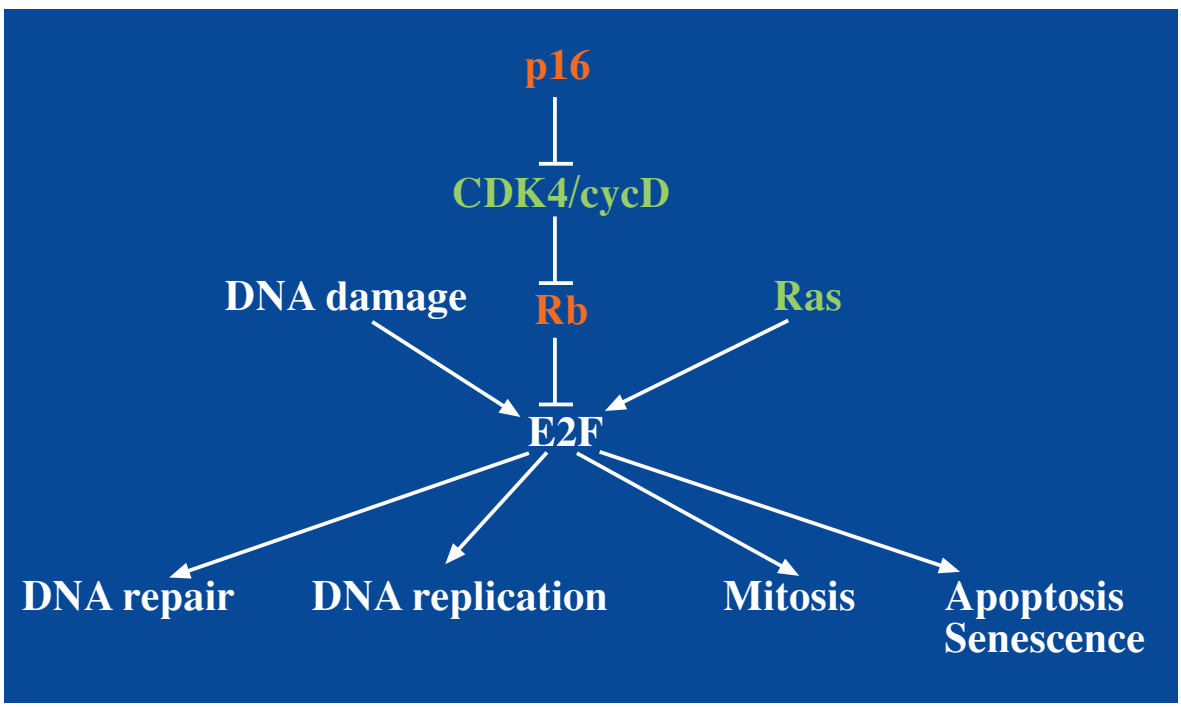

Fig. 1 *Regulations and activities of the transcription factor E2F. E2F is regulated by a number of mechanisms including negative regulation of its activity by RB and up-regulation of its levels by either DNA damage or activated Ras. E2F up-regulates the expression of genes involved in a number of biological processes including DNA replication, DNA repair, mitosis, apo ptosis and senescence.*

we show that it is directly regulated by E2F via E2F-binding sites in its promoter.

Other novel E2F-regulated genes, identified using the DNA microarray analysis, are involved in mitosis and DNA repair. The identification of mitotic genes, including the kinases Bub1b and cdc2, as E2F-regulated genes indicates that E2F affects cell cycle progression both at the S-phase and during mitosis. The molecular mechanism(s) of E2F-dependent regulation of mitotic genes is currently being studied. The identification of DNA repair genes, including MSH2, MSH6 and BRCA1, as E2F-regulated genes raises the possibility that E2F plays a role in the response to DNA damage. This notion is supported by recent reports demonstrating that various DNA damaging agents induce an increase in E2F-1 protein level.

Ras induces elevation of E2F-1 levels

E2F activity is often deregulated in human cancer, however, such deregulated activity is not sufficient to induce cellular transformation and additional molecular events must occur. One such additional event might be the activation of the proto-oncogene Ras that is frequently detected in human tumors. Co-expression of activated Ras and E2F-1 leads to the formation of morphologically transformed foci in primary fibroblasts. Furthermore, double transgenic animals overexpressing E2F-1 and activated Ras in their epidermis develop skin tumors. The molecular mechanisms underlying the cooperation between Ras and E2F-1 in cell transformation are currently not fully understood. Our recent data show that activated Ras induces an increase in E2F-1 mRNA and protein levels. This Ras-induced increase in E2F-1 levels is dependent on both MEK and PKB and it is RB-independent. The effect of Ras on up-regulation of E2F-1 mRNA is at the level of mRNA stability and the induced E2F-1 is transcriptional active. Our data describe a novel functional link between Ras and the RB/E2F pathway. Furthermore, we suggest that one of the molecular mechanisms underlying the collaboration between Ras and E2F-1 in cell transformation involves a Ras-induced elevation of transcriptionaly active E2F-1 levels.

Altogether, our studies provide new insights to the regulation and activities of E2F, thus increasing our understanding of the control of cell cycle progression and tumorigenesis at the molecular level.

Selected Publications

Lindeman, G.J., Gaubatz, S., Livingston, D.M. and Ginsberg, D. (1997) The subcellular localization of E2F-4 is cell-cycle dependent. Proc. Natl. Acad. Sci. USA, 94, 5095-5100.

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Berkovich E. and Ginsberg, D. (2001) Ras induces elevation of E2F-1 mRNA levels. J Biol Chem, In Press.

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