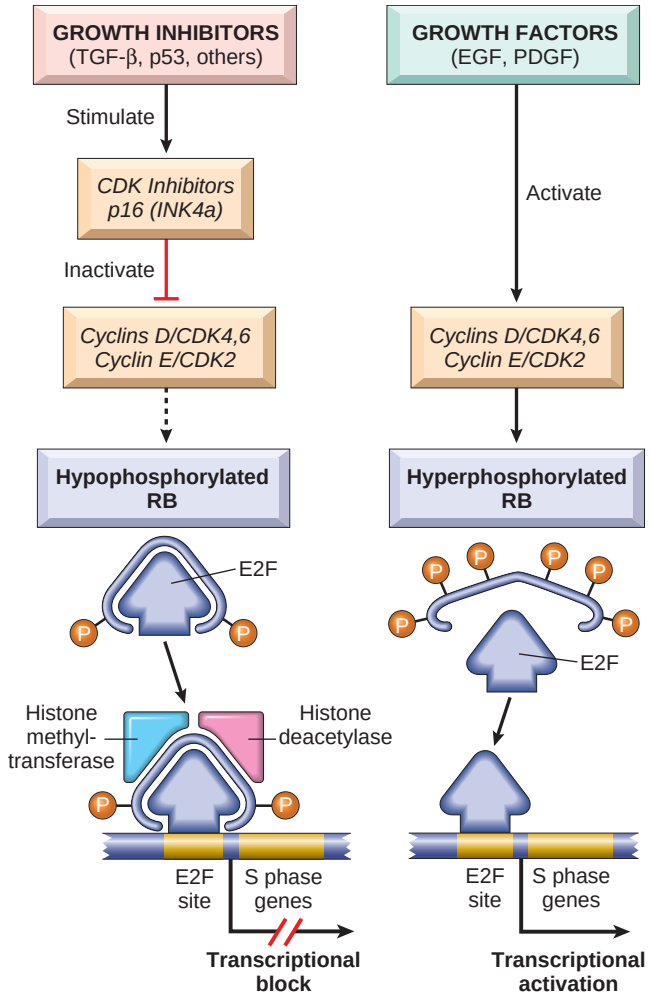

Figure 7-29 The role of RB in regulating the G_1-S checkpoint of the cell cycle. Hypophosphorylated RB in complex with the E2F transcription factors binds to DNA, recruits chromatin-remodeling factors (histone deacetylases and histone methyltransferases), and inhibits transcription of genes whose products are required for the S phase of the cell cycle. When RB is phosphorylated by the cyclin D-CDK4, cyclin D-CDK6, and cyclin E-CDK2 complexes, it releases E2F. The latter then activates transcription of S-phase genes. The phosphorylation of RB is inhibited by cyclin-dependent kinase inhibitors, because they inactivate cyclin-CDK complexes. Virtually all cancer cells show dysregulation of the G_1-S checkpoint as a result of mutation in one of four genes that regulate the phosphorylation of RB; these genes are *RB*, *CDK4*, the genes encoding cyclin D proteins, and *CDKN2A* (p16). TGF-β, transforming growth factor-β.

and drive cells through the G_1/S transition, whereas growth inhibitors tip the balance the other way by upregulating CDK inhibitors. RB is the point of integration of these opposing signals, making it a key player in regulation of cell cycle progression.

It was mentioned previously that germline and somatic loss-of-function mutations of the *RB* gene are associated with retinoblastoma and osteosarcoma, and analyses of cancer cell genomes have identified similar somatic *RB* mutations in a subset of glioblastomas, small-cell carcinomas of lung, breast cancers, and bladder carcinomas. However, given that RB is expressed in all cells, one may ask, why do patients with germline *RB* mutations preferentially develop only a few types of cancer? And, conversely, why aren't acquired mutations of *RB* found in all kinds of cancer? The reason why persons who inherit one defective allele of *RB* preferentially develop retinoblastoma is not understood, but a possible explanation is that other RB family members exist that may partially complement RB function in cell types other than retinoblasts.

With respect to the second question (i.e., why mutations of *RB* are not more widespread in human tumors), the answer is much simpler: mutations in other genes that control RB phosphorylation can mimic the effect of *RB* loss, and such genes are mutated in many cancers that have normal *RB* genes. Thus, for example, mutational activation of cyclin D or CDK4 and mutational inactivation of CDK inhibitors favors cell proliferation by facilitating the hyperphosphorylation and inactivation of RB. The current paradigm is that **loss of normal cell cycle control is central to malignant transformation and that at least one of four key regulators of the cell cycle (p16/INK4a, cyclin D, CDK4, RB) is dysregulated in the vast majority of human cancers**. In cells that harbor mutations in any one of these other genes, or in upstream factors that regulate their expression and function (e.g., receptor tyrosine kinases, RAS), RB may be functionally inactivated even if the *RB* gene itself is not mutated.

The transforming proteins of several oncogenic animal and human DNA viruses also act, in part, by neutralizing the growth-inhibitory activities of RB. In these cases, the RB protein is functionally inactivated by the binding of a viral protein and no longer acts as a cell cycle inhibitor. Simian virus 40 and polyomavirus large T antigens, adenovirus EIA protein, and HPV E7 protein all bind to the hypophosphorylated form of RB. The binding occurs in the same RB pocket that normally sequesters E2F transcription factors. Of note, in the case of HPV, viral types (such as HPV16) that confer a high risk for the development of cervical carcinoma express E7 protein variants with higher affinity for RB than do lower risk viral types. Thus, the RB protein, unable to bind the E2F transcription factors, is functionally inactivated by these viral oncoproteins, and the E2F factors are free to cause cell cycle progression.

KEY CONCEPTS

RB, Governor of the Cell Cycle

- When hypophosphorylated, RB exerts antiproliferative effects by binding and inhibiting E2F transcription factors that regulate genes required for cells to pass through the G1-S phase cell cycle checkpoint. Normal growth factor signaling leads to RB hyperphosphorylation and inactivation, thus promoting cell cycle progression.
- The antiproliferative effect of RB is abrogated in cancers through a variety of mechanisms, including:
  - Loss-of-function mutations affecting *RB*
  - Gene amplifications of CDK4 and cyclin D genes
  - Loss of cyclin-dependent kinase inhibitors (p16/INK4a)
  - Viral oncoproteins that bind and inhibit RB (E7 protein of HPV)

TP53: Guardian of the Genome. ***TP53*, a tumor suppressor gene that regulates cell cycle progression, DNA repair, cellular senescence, and apoptosis, is the most frequently mutated gene in human cancers.** Loss-of-function