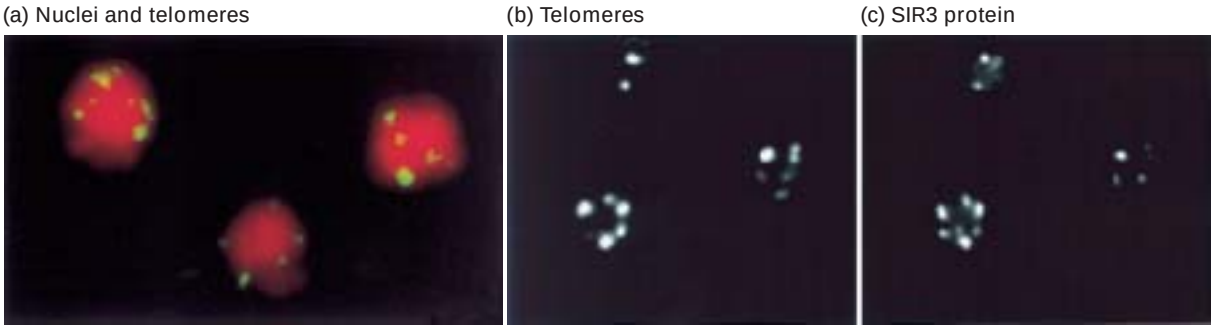
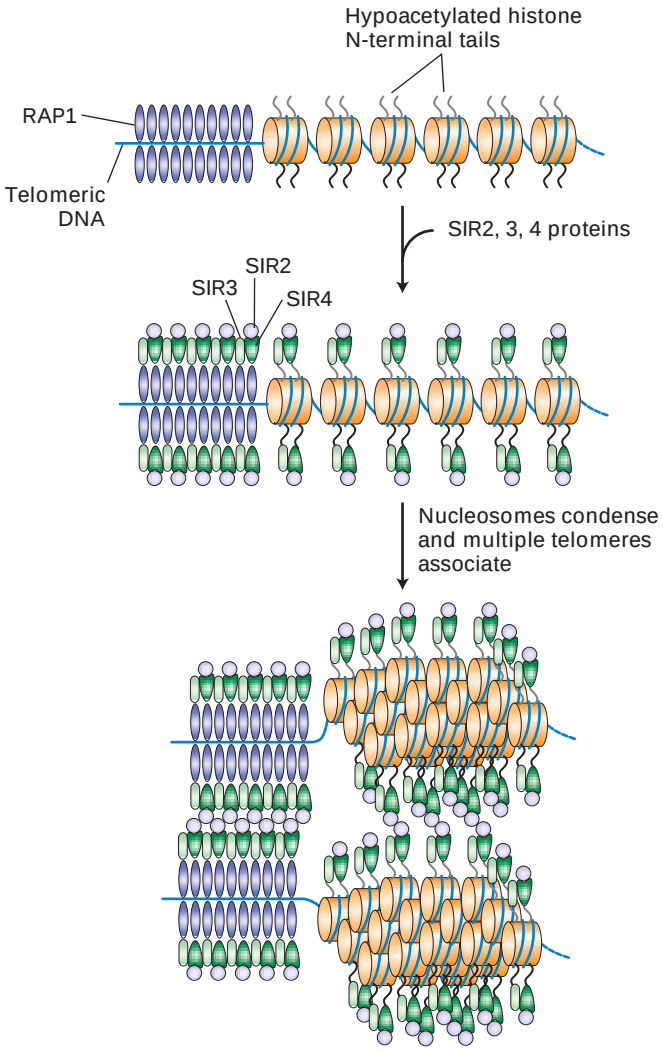


▲ EXPERIMENTAL FIGURE 11-29 **Antibody and DNA probes colocalize SIR3 protein with telomeric heterochromatin in yeast nuclei.** (a) Confocal micrograph 0.3 μm thick through three diploid yeast cells, each containing 68 telomeres. Telomeres were labeled by hybridization to a fluorescent telomere-specific probe (yellow). DNA was stained red to reveal the nuclei. The 68 telomeres coalesce into a much smaller number of regions near the nuclear periphery. (b, c) Confocal micrographs of yeast cells labeled with a telomere-specific hybridization probe (b) and a fluorescent-labeled antibody specific for SIR3 (c). Note that SIR3 is localized in the repressed telomeric heterochromatin. Similar experiments with RAP1, SIR2, and SIR4 have shown that these proteins also colocalize with the repressed telomeric heterochromatin. [From M. Gotta et al., 1996, *J. Cell Biol.* **134**:1349; courtesy of M. Gotta, T. Laroche, and S. M. Gasser.]



to associate with the silencer region, where it is thought to cause further assembly of the multiprotein telomeric silencing complex so that it spreads farther from the end of the chromosome, encompassing *HML* and *HMR*.

An important feature of this model is the dependence of silencing on hypoacetylation of the histone tails. This was shown in experiments with yeast mutants expressing histones in which lysines in histone N-termini were substituted with arginines or glutamines. Arginine is positively charged like lysine, but cannot be acetylated. It is thought to function in histone N-terminal tails like an unacetylated lysine. Glutamine on the other hand simulates an acetylated lysine. Repression at telomeres and at the silent mating-type loci was defective in the mutants with glutamine substitutions, but not in mutants with arginine substitutions. Hyperacetylation of the H3 and H4 tails subsequently was found to interfere with binding by SIR3 and SIR4.

Although chromatin-mediated repression of transcription is also important in multicellular eukaryotes, the mechanism of this repression is still being worked out. Genetic and

◀ FIGURE 11-30 **Schematic model of silencing mechanism at yeast telomeres.** Multiple copies of RAP1 bind to a simple repeated sequence at each telomere region, which lacks nucleosomes (*top*). This nucleates the assembly of a multiprotein complex (*bottom*) through protein-protein interactions between RAP1, SIR2, SIR3, SIR4, and the hypoacetylated N-terminal tails of histones H3 and H4 of nearby nucleosomes. SIR2 deacetylates the histone tails. The heterochromatin structure at each telomere encompasses ≈4 kb of DNA neighboring the RAP1-binding sites, irrespective of its sequence. Association of several condensed telomeres forms higher-order heterochromatin complexes, such as those shown in Figure 11-29, that sterically block other proteins from interacting with the DNA. See the text for more details. [Adapted from M. Grunstein, 1997, *Curr. Opin. Cell Biol.* **9**:383.]