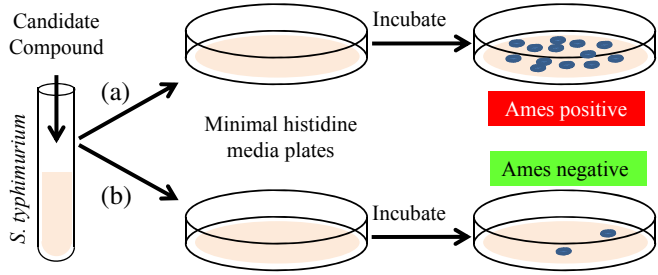

FIGURE 8.10 In the Ames assay, cells genetically engineered to be incapable of producing histidine (such as *Salmonella typhimurium*) are cultured in the presence of candidate compounds and a limited supply of histidine. Once the histidine is depleted, only cells with mutations that restore histidine synthesis pathways will survive. (a) Candidate compounds that promote mutations (mutagenic compounds) will lead to an increase in cell survival in the absence of a histidine-rich media. This is referred to as an Ames positive result. (b) Compounds that do not increase cell survival in this assay are referred to as being Ames negative.

is exhausted, only bacteria that have mutated to reactivate the histidine synthesis pathway will survive. If the test compound is mutagenic, it will produce mutations that restore the histidine synthesis pathway, allowing more bacteria to survive after the histidine supply is exhausted (as compared to the same bacteria grown in the absence of the test compound). This can be quantified by counting the number of bacterial colonies present after a set time interval. The assay can also be run in the presence of rat liver extract (specifically the S9 fraction) in order to determine if a metabolite of the test compound has mutagenic properties. Like all assays, the Ames assay is not perfect, as both false positives and false negatives have been reported. However, the speed at which this assay can be run and the overall cost/time saving as compared to *in vivo* screening in rats or mice are substantial advantages. Also, regulatory bodies have provided specific guidance regarding acquiring Ames data on candidate compounds.

Another important method used to identify potentially risky compounds is the Micronucleus Assay (Figure 8.11).[27] Unlike the Ames assay, this

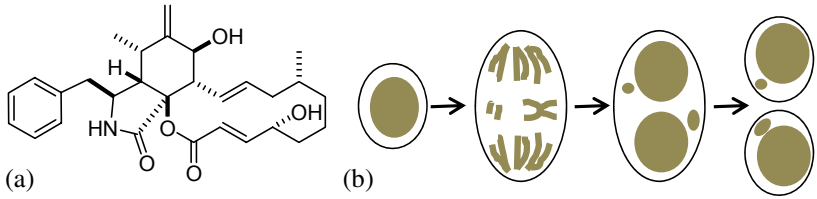

FIGURE 8.11 (a) Cytochalasin B. (b) In the Micronucleus Assay, Chinese hamster ovarian cells (CHO cells) are grown in the presence of a candidate compound for a set period of time and then exposed to cytochalasin B. This blocks cytokinesis, leading to a buildup of binucleated cells. The presence of micronuclei indicates that a candidate compound can cause chromosomal damage.