ESSENTIAL CELL BIOLOGY, FOURTH EDITION
CHAPTER 18: THE CELL-DIVISION CYCLE

Overview of the Cell Cycle

18-1 For each of the following sentences, fill in the blanks with the best word or phrase selected from the list below. Not all words or phrases will be used; each word or phrase may be used more than once.

The four phases of the cell cycle, in order, are G_1, _____, _____, and _____. A cell contains the most DNA after _____ phase of the cell cycle. A cell is smallest in size after _____ phase of the cell cycle. Growth occurs in _____, _____, and _____ phases of the cell cycle. A cell does not enter mitosis until it has completed _____ synthesis.

DNA	M	protein
G_1	nucleotide	S
G_2	organelle	

18-2 What would be the most obvious outcome of repeated cell cycles consisting of S phase and M phase only?

(a) Cells would not be able to replicate their DNA.
(b) The mitotic spindle could not assemble.
(c) Cells would get larger and larger.
(d) The cells produced would get smaller and smaller.

18-3 A mutant yeast strain stops proliferating when shifted from 25°C to 37°C. When these cells are analyzed at the two different temperatures, using a machine that sorts cells according to the amount of DNA they contain, the graphs in Figure Q18-3 are obtained.

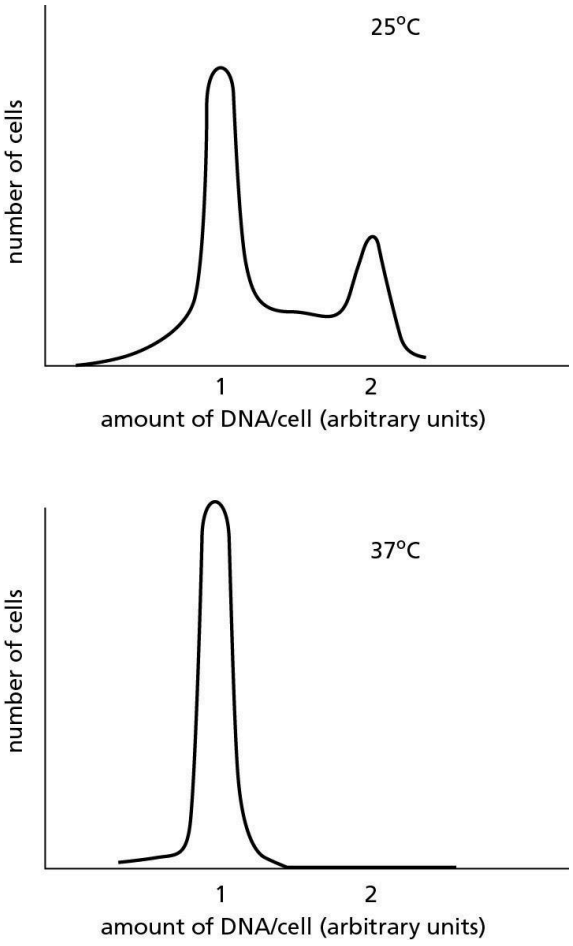

Figure 18-3

Which of the following would *not* explain the results with the mutant?

(a) inability to initiate DNA replication
(b) inability to begin M phase
(c) inability to activate proteins needed to enter S phase
(d) inappropriate production of a signal that causes the cells to remain in G_1

18-4 Which of the following events does *not* usually occur during interphase?

(a) Cells grow in size.
(b) The nuclear envelope breaks down.
(c) DNA is replicated.
(d) The centrosomes are duplicated.

18-5 In which phase of the cell cycle do cells check to determine whether the DNA is fully and correctly replicated?

(a) at the transition between G_1 and S
(b) when cells enter G_0
(c) during M
(d) at the end of G_2

18-6 What would happen to the progeny of a cell that proceeded to mitosis and cell division after entering S phase but had not completed S phase? Keep in mind that highly condensed chromatin, including the centromere region, is replicated late in S phase. Explain your answer.

18-7 Are the statements below *true* or *false*? Explain your answer.

A. Statement 1: Generally, in a given organism, the S, G_2, and M phases of the cell cycle take a defined and stereotyped amount of time in most cells.

B. Statement 2: Therefore, the cell-cycle control system operates primarily by a timing mechanism, in which the entry into one phase starts a timer set for sufficient time to complete the required tasks. After a given amount of time has elapsed, a molecular "alarm" triggers movement to the next phase.

18-8 Which of the following statements about the cell cycle is *false*?

(a) Once a cell decides to enter the cell cycle, the time from start to finish is the same in all eukaryotic cells.

(b) An unfavorable environment can cause cells to arrest in G_1.

(c) A cell has more DNA during G_2 than it did in G_1.

(d) The cleavage divisions that occur in an early embryo have short G_1 and G_2 phases.

18-9 Which of the following descriptions is consistent with the behavior of a cell that lacks a protein required for a checkpoint mechanism that operates in G_2?

(a) The cell would be unable to enter M phase.

(b) The cell would be unable to enter G_2.

(c) The cell would enter M phase under conditions when normal cells would not.

(d) The cell would pass through M phase more slowly than normal cells

The Cell-Cycle Control System

18-10 Match the following labels to the numbered label lines on Figure Q18-10.

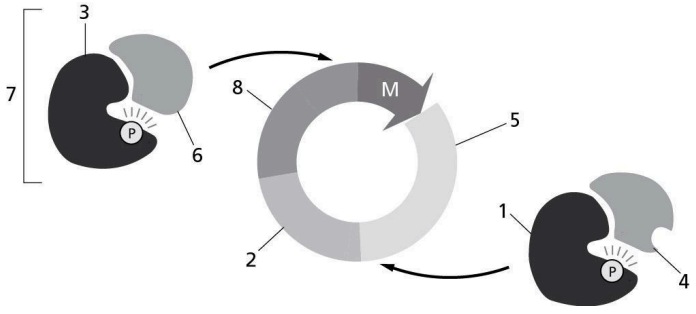

Figure Q18-10

A. G_1 phase

B. mitotic cyclin

C. mitotic Cdk

D. S phase

E. S-phase cyclin

F. S-phase Cdk
G. MPF
H. G_2 phase

18-11 Progression through the cell cycle requires a cyclin to bind to a Cdk because _____.
(a) the cyclins are the molecules with the enzymatic activity in the complex.
(b) the binding of a cyclin to Cdk is required for Cdk enzymatic activity.
(c) cyclin binding inhibits Cdk activity until the appropriate time in the cell cycle.
(d) without cyclin binding, a cell-cycle checkpoint will be activated.

18-12 Levels of Cdk activity change during the cell cycle, in part because _____.
(a) the Cdks phosphorylate each other.
(b) the Cdks activate the cyclins.
(c) Cdk degradation precedes entry into the next phase of the cell cycle.
(d) cyclin levels change during the cycle.

18-13 The concentration of mitotic cyclin (M cyclin) _____.
(a) rises markedly during M phase.
(b) is activated by phosphorylation.
(c) falls toward the end of M phase as a result of ubiquitylation and degradation.
(d) is highest in G_1 phase.

18-14 You have isolated a strain of mutant yeast cells that divides normally at 30°C but cannot enter M phase at 37°C. You have isolated its mitotic cyclin and mitotic Cdk and find that both proteins are produced and can form a normal M-Cdk complex at both temperatures. Which of the following temperature-sensitive mutations could *not* be responsible for the behavior of this strain of yeast?
(a) inactivation of a protein kinase that acts on the mitotic Cdk kinase
(b) inactivation of an enzyme that ubiquitylates M cyclin
(c) inactivation of a phosphatase that acts on the mitotic Cdk kinase
(d) a decrease in the levels of a transcriptional regulator required for producing sufficient amounts of M cyclin

18-15 You engineer yeast cells that express the M cyclin during S phase by replacing the promoter sequence of the M cyclin gene with that of S cyclin. Keeping in mind that yeast cells have one common Cdk that binds to all cyclins, which of the following outcomes is *least* likely during this experiment?
(a) There will be both M cyclin–Cdk and S cyclin–Cdk complexes in the cell during S phase.
(b) Some substrates that are normally phosphorylated in M phase will now be phosphorylated in S phase.
(c) G_1 cyclins will be expressed during S phase.
(d) S-Cdk targets will be phosphorylated during S phase.

18-16 You have isolated a mutant in which a fraction of the new cells die soon after cell division and a fraction of the living cells have an extra copy of one or more chromosomes. When you grow the cells under conditions in which they transit the cell cycle more slowly, the defect disappears, suggesting that the mitotic spindle and segregation machinery are normal. Propose a basis for the defect.

18-17 Which of the following statements is *false*?

(a) Mitotic Cdk must be phosphorylated by an activating kinase (Cak) before it is active.

(b) Phosphorylation of mitotic Cdk by the inhibitory kinase (Wee1) makes the Cdk inactive, even if it is phosphorylated by the activating kinase.

(c) Active M-Cdk phosphorylates the activating phosphatase (Cdc25) in a positive feedback loop.

(d) The activating phosphatase (Cdc25) removes all phosphates from mitotic Cdk so that M-Cdk will be active.

How We Know: Discovery of Cyclins and CDKs

18-18 For each of the following sentences, fill in the blanks with the best word or phrase selected from the list below. Not all words or phrases will be used; each word or phrase should be used only once.

Many features of _____ cells make them suitable for biochemical studies of the cell-cycle control system. For example, the cells are unusually large and are arrested in a _____-like phase. When the cells are triggered to resume cycling, the cell divisions have especially _____ G_1 and G_2 phases and occur _____. Studies with *Xenopus* eggs identified a partly purified activity called _____ that drives a resting *Xenopus* oocyte into M phase. MPF activity was found to _____ during the cell cycle, although the amount of its kinase component, called _____, remained constant. The regulatory component of MPF, called _____, has a _____ effect on MPF activity and plays a part in regulating interactions with its _____s. The components of MPF are evolutionarily _____ from yeast to humans, so that the corresponding human genes are _____ to function in yeast.

able	hexokinase	short
asynchronously	inhibitory	sperm
Cdk	long	steady
conserved	M	stimulatory
cyclin	maturation promoting factor	substrate
divergent	oscillate	synchronously
egg	PI 3-kinase	ubiquitin
fibroblast	regulin	unable
G_1	S	uniform
G_2		

18-19 MPF activity was discovered when cytoplasm from a *Xenopus* M-phase cell was injected into *Xenopus* oocytes, inducing the oocytes to form a mitotic spindle. In a control experiment, *Xenopus* interphase cytoplasm was injected into oocytes and shown not to induce the formation of a mitotic spindle. Which of the following statements is *not* a legitimate conclusion from the control experiment?

(a) The piercing of the oocyte membrane by a needle is insufficient to cause mitotic spindle formation.

(b) An increased volume of cytoplasm is insufficient to cause mitotic spindle formation.

(c) Injection of extra RNA molecules is insufficient to cause mitotic spindle formation.

(d) Components of an interphase nucleus are insufficient to cause mitotic spindle formation.

18-20 Which of the following is *not* good direct evidence that the cell-cycle control system is conserved through billions of years of divergent evolution?

(a) A yeast cell lacking a Cdk function can use the human Cdk to substitute for its missing Cdk during the cell cycle.

(b) The amino acid sequences of cyclins in plants are similar to the amino acid sequences of cyclins in humans.

(c) The Cdk proteins in humans share conserved phosphorylation sites with the Cdk proteins in yeast.

(d) Yeast cells have only one Cdk, whereas humans have many Cdks.

18-21 Your friend is intrigued by Cdks and purifies Cdk from the daisy plant. She is able to determine the sequence of 18 amino acids from the daisy protein. Using these data, she aligns the daisy sequence with one human Cdk protein and the Cdk proteins from two different kinds of yeast, as shown in Figure Q18-21. Sequences identical between the human and fungal proteins have been boxed.

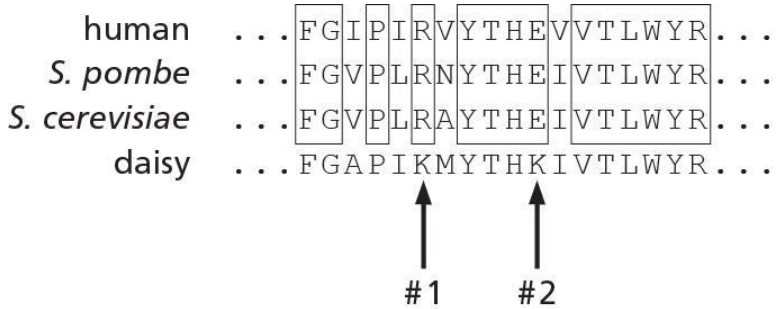

Figure Q18-21

Such conserved amino acid sequences are often involved in protein-protein interactions. Indeed, the threonine (T) in the central "YTHE/K" block is known to be phosphorylated by akinase that activates Cdks in human and yeast. The surrounding conserved sequences could thus be important for the interaction of this Cdk-activating kinase with Cdk. In the

daisy Cdk, however, not all of the amino acids in these conserved bloacks match the sequences from yeast and human, as indicated by the arrows marked #1 and #2.

A. When you replace the *S. cerevisiae* (budding yeast) Cdk with the daisy Cdk, you discover the daisy Cdk does not interact with the Cdk-activating kinase inside the yeast cell. Circle the amino acid in the daisy Cdk that is likely to be the most disruptive to its interaction with Cdk-activating kinase.

B. Based on the information in Figure Q18-21 and what you know about the Cdk1–Cdk-activating kinase interaction, would you predict that the human Cdk will interact with yeast Cdk-activating kinase? Explain.

G_1 Phase

18-22 Mitogens are _____.

(a) extracellular signals that stimulate cell division.
(b) transcription factors important for cyclin production.
(c) kinases that cause cells to grow in size.
(d) produced by mitotic cells to keep nearby neighboring cells from dividing.

18-23 The Retinoblastoma (Rb) protein blocks cells from entering the cell cycle by _____.

(a) phosphorylating Cdk.
(b) marking cyclins for destruction by proteolysis.
(c) inhibiting cyclin transcription.
(d) activating apoptosis.

18-24 Irradiated mammalian cells usually stop dividing and arrest at a G_1 checkpoint. Place the following events in the order in which they occur.

A. production of p21
B. DNA damage
C. inhibition of cyclin–Cdk complexes
D. accumulation and activation of p53

18-25 The G_1 DNA damage checkpoint _____.

(a) causes cells to proceed through S phase more quickly.
(b) involves the degradation of p53.
(c) is activated by errors caused during DNA replication.
(d) involves the inhibition of cyclin–Cdk complexes by p21.

18-26 Cells in the G_0 state _____.

(a) do not divide.
(b) cannot re-enter the cell cycle.
(c) have entered this arrest state from either G_1 or G_2.
(d) have duplicated their DNA.

18-27 What is the main molecular difference between cells in a G_0 state and cells that have simply paused in G_1?

S Phase

18-28 Which of the following statements is false?
- (a) DNA synthesis begins at origins of replication.
- (b) The loading of the origin recognition complexes (ORCs) is triggered by S-Cdk.
- (c) The phosphorylation and degradation of Cdc6 help to ensure that DNA is replicated only once in each cell cycle.
- (d) DNA synthesis can only begin after prereplicative complexes assemble on the ORCs.

18-29 How does S–Cdk help guarantee that replication occurs only once during each cell cycle?
- (a) It blocks the rise of Cdc6 concentrations early in G_1.
- (b) It phosphorylates and inactivates DNA helicase.
- (c) It phosphorylates the Cdc6 protein, marking it for destruction.
- (d) It promotes the assembly of a prereplicative complex.

18-30 You create cells with a version of Cdc6 that cannot be phosphorylated and thus cannot be degraded. Which of the following statements describes the likely consequence of this change in Cdc6?
- (a) Cells will enter S phase prematurely.
- (b) Cells will be unable to complete DNA synthesis.
- (c) The origin recognition complex (ORC) will be unable to bind to DNA.
- (d) Cdc6 will be produced inappropriately during M phase.

18-31 Cells can pause in G_1 when DNA is damaged, and can pause in S when there are replication errors. Indicate whether the mechanism below applies to a G_1 arrest, an S-phase arrest, both types of arrest, or neither.
- A. p53 activates the transcription of a Cdk inhibitor.
- B. Cyclins are phosphorylated and destroyed.
- C. Cdk is unable to phosphorylate its substrates.
- D. The Cdc25 phosphatase is inhibited.

M Phase

18-32 For each of the following sentences, fill in the blanks with the best word or phrase selected from the list below. Not all words or phrases will be used; each word or phrase should be used only once.

The cell cycle consists of an alternation between _____, which appears as a period of dramatic activity under the microscope, and a preparative period called _____, which consists of three phases called _____, _____, and _____. During M phase, the nucleus divides in a process called _____, and the cytoplasm splits in two in a process called _____. The cell-cycle control system relies on sharp increases in

the activities of regulatory proteins called _____, or _____, to trigger S phase and M phase. Inactivation of _____ is required to exit from M phase after chromosome segregation.

APC	G_2 phase	metaphase
Cdks	interphase	microtubules
condensation	intraphase	mitosis
cyclin-dependent kinases	kinesins	myosins
cytokinesis	M phase	S phase
G_1 phase	M-Cdk	S-Cdk
G_1-Cdk	meiosis	

18-33 Which of the following does not occur during M phase in animal cells?
(a) growth of the cell
(b) condensation of chromosomes
(c) breakdown of nuclear envelope
(d) attachment of chromosomes to microtubules

18-34 Condensins _____.
(a) are degraded when cells enter M phase.
(b) assemble into complexes on the DNA when phosphorylated by M-Cdk.
(c) are involved in holding sister chromatids together.
(d) bind to DNA before DNA replication begins.

18-35 At the end of DNA replication, the sister chromatids are held together by the _____.
(a) kinetochores.
(b) securins.
(c) cohesins.
(d) histones.

18-36 Which of the following statements is *true*?
(a) The mitotic spindle is largely made of intermediate filaments.
(b) The contractile ring is made largely of microtubules and actin filaments.
(c) The contractile ring divides the nucleus in two.
(d) The mitotic spindle helps segregate the chromosomes to the two daughter cells.

Mitosis

18-37 Which stage of mitosis in an animal cell does each part of Figure Q18-37 represent?

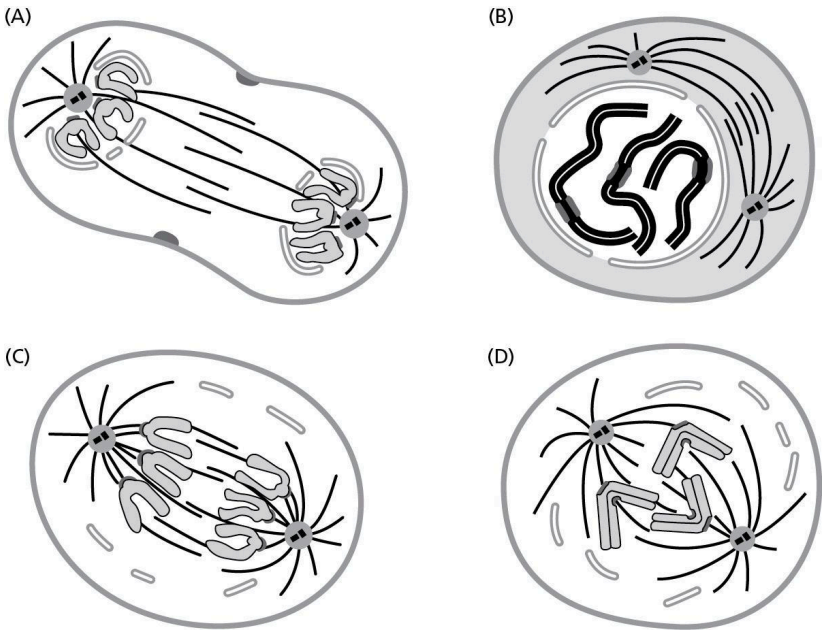

Figure Q18-37

18-38 Figure Q18-38 shows a living cell from the lung epithelium of a newt at different stages in M phase. Order these light micrographs into the correct sequence and identify the stage in M phase that each represents.

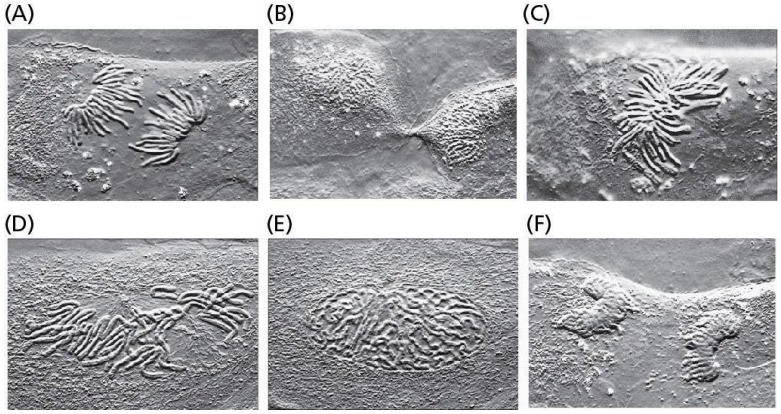

Figure Q18-38

18-39 Name the stage of M phase in which the following events occur. Place the numbers 1–8 next to the letter headings to indicate the normal order of events.

A. alignment of the chromosomes at the spindle equator
B. attachment of spindle microtubules to chromosomes
C. breakdown of nuclear envelope
D. pinching of cell in two
E. separation of two centrosomes and initiation of mitotic spindle assembly
F. re-formation of the nuclear envelope
G. condensation of the chromosomes
H. separation of sister chromatids

18-40 The principal microtubule-organizing center in animal cells is the _____.

(a) centrosome.
(b) centromere.
(c) kinetochore.
(d) cell cortex.

Questions 18-41 through 18-43 all utilize Figure Q18-41.

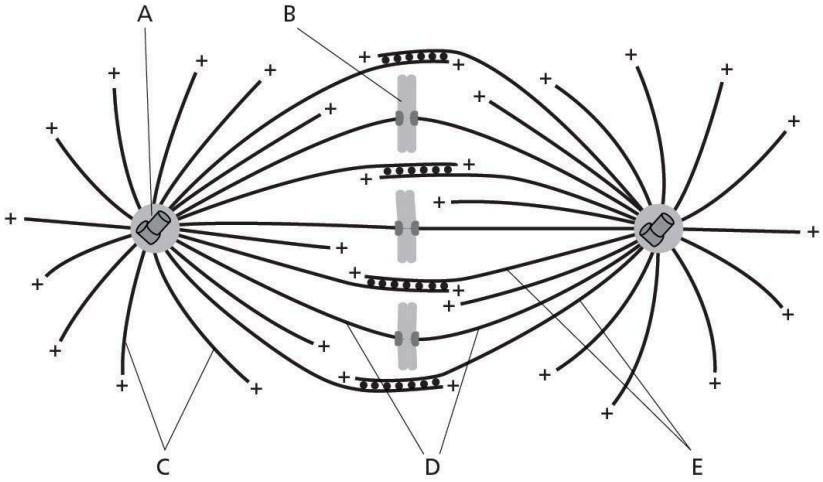

Figure Q18-41

18-41 Which word or phrase below best describes the phase in mitosis depicted in Figure Q18-41?

(a) anaphase
(b) prometaphase
(c) S-phase checkpoint
(d) metaphase

18-42 Which letter is associated with the line that is pointing to the interpolar microtubules in Figure Q18-41?

(a) A
(b) B
(c) C
(d) D
(e) E

18-43 Before a cell can enter M phase, two structures from Figure Q18-41 must be duplicated. Write the letters corresponding to the lines pointing to these structures, and write the names of the structures next to the letters.

18-44 Examine the schematic representation of centrosome duplication in Figure Q18-44. By analogy with DNA replication, would you classify centrosome duplication as conservative or semiconservative? Explain your answer.

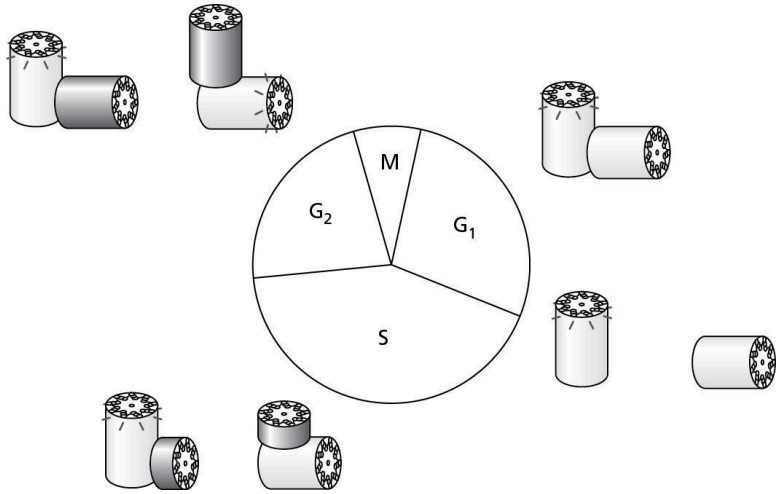

Figure Q18-44

18-45 Before chromosomes segregate in M phase, they and the segregation machinery must be appropriately prepared. Indicate whether the following statements are *true* or *false*. If false, change a single noun to make the statement true.

A. Sister chromatids are held together by condensins from the time they arise by DNA replication until the time they separate at anaphase.

B. Cohesins are required to make the chromosomes more compact and thus to prevent tangling between different chromosomes.

C. The mitotic spindle is composed of actin filaments and myosin filaments.

D. Microtubule-dependent motor proteins and microtubule polymerization and depolymerization are mainly responsible for the organized movements of chromosomes during mitosis.

E. The centromere nucleates a radial array of microtubules called an aster, and its duplication is triggered by S-Cdk.

F. Each centrosome contains a pair of centrioles and hundreds of γ-tubulin rings that nucleate the growth of microtubules.

18-46 The cytoskeleton of an animal cell changes markedly between G_1 and early M phase (prophase) of the cell cycle. For each of the following sentences, choose one of the options enclosed in square brackets that best describes the changes to the cytoskeleton and its components.

Before mitosis, the number of centrosomes must [**increase/decrease**]. At the beginning of [**anaphase/prophase**] in animal cells, the centrosomes separate in a process driven partly by interactions between the [**plus/minus**] ends of microtubules arising from the two centrosomes. Centrosome separation initiates the assembly of the bipolar mitotic spindle and is associated with a sudden [**increase/decrease**] in the dynamic instability of

microtubules. In comparison with an interphase microtubule array, a mitotic aster contains a [**smaller/larger**] number of [**longer/shorter**] microtubules. Extracts from M-phase cells exhibit [**increased/decreased/unchanged**] rates of microtubule polymerization and increased frequencies of microtubule [**shrinkage/growth**]. The changes in microtubule dynamics are largely due to [**enhanced/reduced**] activity of microtubule-associated proteins and [**increased/decreased**] activity of catastrophins. The new balance between polymerization and depolymerization of microtubules is necessary for the mitotic spindle to move the [**replicated chromosomes/daughter chromosomes**] to the metaphase plate.

18-47 Match each of the main classes of spindle microtubules from list 1 with their functions and features from list 2.

List 1	**List 2**
(A) Interpolar microtubules	1. Stabilized by interactions with each other via motor proteins
(B) Aster microtubules	2. Interact with the cell cortex
(C) Kinetochore microtubules	3. Link chromosomes to a spindle pole
	4. Depolymerize to promote anaphase A
	5. Depolymerize to promote anaphase B

18-48 Disassembly of the nuclear envelope _____.

(a) causes the inner nuclear membrane to separate from the outer nuclear membrane.
(b) results in the conversion of the nuclear envelope into protein-free membrane vesicles.
(c) is triggered by the phosphorylation of integrins.
(d) must occur for kinetochore microtubules to form in animal cells.

18-49 Consider an animal cell that has eight chromosomes (four pairs of homologous chromosomes) in G_1 phase. How many of each of the following structures will the cell have at mitotic prophase?

A. sister chromatids
B. centromeres
C. kinetochores
D. centrosomes
E. centrioles

18-50 Which of the following statements about kinetochores is *true*?

(a) Kinetochores assemble onto chromosomes during late prophase.
(b) Kinetochores contain DNA-binding proteins that recognize sequences at the telomere of the chromosome.
(c) Kinetochore proteins bind to the tubulin molecules at the minus end of microtubules.
(d) Kinetochores assemble on chromosomes that lack centromeres.

18-51 Is the following statement *true* or *false*?

After the nuclear envelope breaks down, microtubules gain access to the chromosomes and, every so often, a randomly probing microtubule captures a chromosome and ultimately connects to the kinetochore to become a kinetochore microtubule of the spindle.

18-52 A friend declares that chromosomes are held at the metaphase plate by microtubules that push on each chromosome from opposite sides. Which of the following observations does not support your belief that the microtubules are pulling on the chromosomes?

(a) the jiggling movement of chromosomes at the metaphase plate
(b) the way in which chromosomes behave when the attachment between sister chromatids is severed
(c) the way in which chromosomes behave when the attachment to one kinetochore is severed
(d) the shape of chromosomes as they move toward the spindle poles at anaphase

18-53 Why should it be that drugs such as colchicine, which inhibit microtubule polymerization, and drugs such as Taxol®, which stabilize microtubules, both inhibit mitosis?

18-54 Which of the following statements about the anaphase-promoting complex (APC) is *false*?

(a) It promotes the degradation of proteins that regulate M phase.
(b) It inhibits M-Cdk activity.
(c) It is continuously active throughout the cell cycle.
(d) M-Cdk stimulates its activity.

18-55 Which of the following statements is *true*?

(a) Anaphase A must be completed before anaphase B can take place.
(b) In cells in which anaphase B predominates, the spindle will elongate much less than in cells in which anaphase A dominates.
(c) In anaphase A, both kinetochore and interpolar microtubules shorten.
(d) In anaphase B, microtubules associated with the cell cortex shorten.

18-56 When introduced into mitotic cells, which of the following is expected to impair anaphase B but not anaphase A?

(a) an antibody against myosin
(b) ATPγS, a nonhydrolyzable ATP analog that binds to and inhibits ATPases
(c) an antibody against the motor proteins that move from the plus end of microtubules to the minus end
(d) an antibody against the motor proteins that move from the minus end of microtubules toward the plus end

18-57 Which of the following precede the re-formation of the nuclear envelope during M phase in animal cells?

(a) assembly of the contractile ring
(b) decondensation of chromosomes
(c) reassembly of the nuclear lamina
(d) transcription of nuclear genes

18-58 A cell with nuclear lamins that cannot be phosphorylated in M phase will be unable to _____.

(a) reassemble its nuclear envelope at telophase.
(b) disassemble its nuclear lamina at prometaphase.
(c) begin to assemble a mitotic spindle.
(d) condense its chromosomes at prophase.

18-59 The lengths of microtubules in various stages of mitosis depend on the balance between the activities of catastrophins, which destabilize microtubules, and microtubule-associated proteins (MAPs), which stabilize them. If you created cells with an increased number of catastrophin molecules, do you predict the length of the mitotic spindle will be longer, shorter, or unchanged, relative to the corresponding stage of mitosis in wild-type cells? What do you predict for a cell with increased numbers of MAPs? Explain your reasoning.

18-60 You have discovered a new protein that regulates microtubule dynamics. First, you isolated proteins from a cellular extract that bound to a tubulin affinity column. You then separated the proteins from each other by loading the mixture of proteins on an ion-exchange column, eluting the column with increasing salt concentration, and collecting small "fractions" of protein as they dripped from the column. To test whether each fraction contained microtubule regulators, you mixed it with fluorescent tubulin and purified centrosomes, and then analyzed the reaction microscopically to measure the size of the aster microtubules formed. You found that fractions 8, 9, and 10 promoted the formation of unusually long aster microtubules. Because electrophoretic separation of the fractions on a gel revealed a plentiful protein with an apparent molecular mass of 98 kD, you named the protein p98.

A. Does p98 behave like a MAP or a catastrophin?
B. Propose two ways in which p98 might change the dynamic behavior of microtubules to account for the observed change in microtubule length. (Hint: There are four simple possible mechanisms.)
C. Video microscopy of fluorescent tubulin in reactions with purified centrosomes allowed you to follow the behavior of individual microtubules over time. You graphed the changes in microtubule length in the absence (Figure Q18-60A) and presence (Figure Q18-60B) of p98. Five representative microtubules are shown for each condition. Does p98 alter the rate of microtubule growth or shrinkage? Does p98 alter the frequency of catastrophes (a sudden and rapid decline in microtubule length) or rescues (when a microtubule switches from shrinking to growing)? Explain your answers.
D. After demonstrating the consequences of p98 addition on microtubule dynamics *in vitro* with the use of purified components, you want to determine whether the

protein has the same effects in a complex cellular extract that naturally contains p98. Briefly describe an experiment that will allow you to determine what p98 normally does in mitotic extracts.

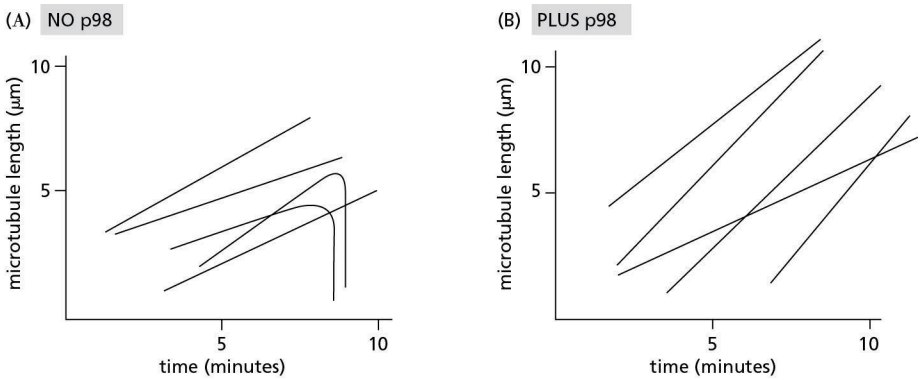

Figure Q18-60

Cytokinesis

18-61 Cytokinesis in animal cells _____.

(a) requires ATP.
(b) leaves a small circular "scar" of actin filaments on the inner surface of the plasma membrane.
(c) is often followed by phosphorylation of integrins in the plasma membrane.
(d) is assisted by motor proteins that pull on microtubules attached to the cell cortex.

18-62 Which of the following statements is *false*?

(a) The cleavage furrow is a puckering of the plasma membrane caused by the constriction of a ring of filaments attached to the plasma membrane.
(b) The cleavage furrow will not begin to form in the absence of a mitotic spindle.
(c) The cleavage furrow always forms perpendicular to the interpolar microtubules.
(d) The cleavage furrow always forms in the middle of the cell.

18-63 Which of the following statements is *false*?

(a) Cytokinesis in plant cells is mediated by the microtubule cytoskeleton.
(b) Small membrane vesicles derived from the Golgi apparatus deliver new cell-wall material for the new wall of the dividing cell.
(c) The phragmoplast forms from the remains of interpolar microtubules of the mitotic spindle.
(d) Motor proteins walking along the cytoskeleton are important for the contractile ring that guides formation of the new cell wall.

18-64 Which organelle fragments during mitosis?

(a) endoplasmic reticulum
(b) Golgi apparatus
(c) mitochondrion

(d) chloroplast

Control of Cell Numbers and Cell Size

18-65 Programmed cell death occurs _____.
(a) by means of an intracellular suicide program.
(b) rarely and selectively only during animal development.
(c) only in unhealthy or abnormal cells.
(d) only during embryonic development.

18-66 Apoptosis differs from necrosis in that necrosis _____.
(a) requires the reception of an extracellular signal.
(b) causes DNA to fragment.
(c) causes cells to swell and burst, whereas apoptotic cells shrink and condense.
(d) involves a caspase cascade.

18-67 Which of the following statements about apoptosis is *true*?
(a) Cells that constitutively express Bcl2 will be more prone to undergo apoptosis.
(b) The prodomain of procaspases contains the catalytic activity necessary for procaspase activation.
(c) Bax and Bak promote apoptosis by binding to procaspases in the apoptosome.
(d) Apoptosis is promoted by the release of cytochrome *c* into the cytosol from mitochondria.

18-68 Imagine that you could microinject cytochrome *c* into the cytosol of both wild-type cells and cells that were lacking both Bax and Bak, which are apoptosis-promoting members of the Bcl2 family of proteins. Would you expect one, both, or neither of the cell lines to undergo apoptosis? Explain your reasoning.

18-69 The number of cells in an adult tissue or animal depends on cell proliferation. What else does it depend on?

18-70 What is the cause of the massive amount of programmed cell death of nerve cells (neurons) that occurs in the developing vertebrate nervous system, and what purpose does it serve?

18-71 For each of the following sentences, fill in the blanks with the best word or phrase selected from the list below. Not all words or phrases will be used; each word or phrase should be used only once.

The survival, _____, and size of each cell in an animal are controlled by extracellular signal molecules secreted by neighboring and distant cells. Many of these signal molecules bind to a cell-surface _____ and trigger various intracellular signaling pathways. One class of signal molecules, called _____, stimulates cell division by releasing the molecular brakes that

keep cells in the _____ or _____ phase of the cell cycle. Members of a second class of signal molecules are called _____, because they stimulate cell growth and an increase in cell mass. The third class of signal molecules, called _____, inhibits _____ by regulating members of the _____ family of proteins. In addition to such stimulatory factors, some signal proteins such as _____ act negatively on other cells, inhibiting their survival, growth, or proliferation.

anaphase	differentiation	myostatin
annihilation	G_0	nourishment
apoptosis	G_1	nutrition
arrestase	G_2	phosphatases
Bcl2	growth factors	proliferation
biosynthetic	interphase	receptor
cascades	ligand	S
caspase	M	survival factors
Cdk	mitogens	transcription
cyclin		

18-72 Of the following mutations, which are likely to cause cell-cycle arrest? If you predict a cell-cycle arrest, indicate whether the cell will arrest in early G_1, late G_1, or G_2. Explain your answers.

A. a mutation in a gene encoding a cell-surface mitogen receptor that makes the receptor active even in the absence of the mitogen
B. a mutation that destroyed the kinase activity of S-Cdk
C. a mutation that allowed G_1-Cdk to be active independently of its phosphorylation status
D. a mutation that removed the phosphorylation sites on the Rb protein
E. a mutation that inhibited the activity of Rb

ANSWERS

18-1 The four phases of the cell cycle, in order, are G_1, **S**, $\mathbf{G_2}$, and **M**. A cell contains the most DNA after **S** phase of the cell cycle. A cell is smallest in size after **M** phase of the cell cycle. Growth occurs in $\mathbf{G_1}$, **S**, and $\mathbf{G_2}$ phases of the cell cycle. A cell does not enter mitosis until it has completed **DNA** synthesis.

18-2 (d) The cells produced would get smaller and smaller, as they would not have sufficient time to double their mass before dividing.

18-3 (b) At 37°C, the cells all have one genome-worth of DNA, meaning that they have not replicated their DNA and therefore have not entered S phase. Cells that are unable to begin M phase should have two genomes-worth of DNA, as they would have completed DNA replication and arrested in G_2.

18-4 (b)

18-5 Choice (d) is correct. It does not make sense to monitor DNA replication before S phase because DNA replication has not yet occurred [choice (a)]. When cells enter G_0, they do not replicate their DNA [choice (b)]. During M phase, chromosomes are condensed for chromosome segregation, so it would be difficult for the cell to examine the replicated DNA for errors at that point [choice (c)].

18-6 The daughter cells would probably die. Those chromosomes that had not completed replication in S phase would have only one centromere, because the centromere is the last part of the chromosome to be replicated; the chromosome would therefore be segregated to only one of the two daughter cells at random. At least one, and probably both, of the daughter cells would thus receive an incomplete set of chromosomes and would be unlikely to be viable. Even if one daughter cell, by chance, received a full set of chromosomes, some of these chromosomes would be incompletely replicated and the cell would probably still not be viable.

18-7 A. True. In nearly all cells in an organism, the S, G_2, and M phases of the cell cycle take the same amount of time. The different timing of cell division in different cell types is due to variable lengths of the G_1 phase or to withdrawal into the G_0 state.

B. False. The cell-cycle control system does use a timing mechanism of sorts, but it also employs various surveillance and feedback mechanisms (checkpoints). Cells will not embark on later events or phases until the earlier events or phases have been completed successfully. In response to a defect or a delay in a cell-cycle event, cells engage molecular brakes to arrest the progression of the cell cycle at various checkpoints, to allow time for completion or repair.

18-8 (a)

18-9 (c) Normal cells arrest at the G_2 checkpoint if DNA replication is incomplete or DNA is damaged. Cells without this mechanism may enter M phase with unreplicated or damaged DNA, whereas normal cells would not.

18-10 A—5; B—6; C—3; D—2; E—4; F—1; G—7; H—8

18-11 Choice (b) is correct. Cyclins have no enzymatic activities themselves [choice (a)], and cyclin binding to Cdk activates the Cdk [choice (c)]. As far as we know, cyclin–Cdk binding is not directly monitored by checkpoints [choice (d)].

18-12 Choice (d) is correct. Cdks do not phosphorylate each other [choice (a)]. The Cdks do not activate the cyclins [choice (b)], and Cdks are not degraded during specific phases of the cycle [choice (c)]. The cyclins, however, are degraded in a cell-cycle-dependent fashion, and they are required for Cdk activity.

18-13 (c) The concentration of mitotic cyclin rises gradually during G_2 and it is ubiquitylated and degraded during late M phase.

18-14 (b) A cell with a mutation that prevents ubiquitylation of M cyclin could enter mitosis but could not exit mitosis properly.

18-15 (c) The transcription of the G_1 cyclins should not be affected by S-Cdk (which normally occurs in S phase) or M-Cdk (which likely now exists within the cell due to the inappropriate expression of M cyclin from the S cyclin promoter).

18-16 This mutant may be lacking the checkpoint mechanism that delays the onset of anaphase and chromosome segregation until all chromosomes have attached properly to the mitotic spindle. If cells attempt chromosome segregation before all chromosomes have attached properly, some of the daughter cells will receive too few chromosomes (and thus will probably die) and other cells will receive additional chromosomes. Normally, cells use such a surveillance system to monitor the spindle attachment of each chromosome and engage molecular brakes until all chromosomes have attached properly. The molecular brakes will be dispensable in some dividing cells if all of the chromosomes rapidly become properly attached to the spindle. In other dividing cells, it will take longer for some of the chromosomes to find their appropriate attachments, and thus the molecular brakes will be essential to ensure faithful segregation of the duplicated copies of each chromosome to the two daughter cells. Slowing the cycle will allow more cells to segregate their chromosomes properly even in the absence of this “spindle attachment” checkpoint.

18-17 (d)

18-18 Many features of **egg** cells make them suitable for biochemical studies of the cell-cycle control system. For example, the cells are unusually large and are arrested in a **G_2**-like phase. When the cells are triggered to resume cycling, the cell divisions have especially

short G_1 and G_2 phases and occur **synchronously**. Studies with *Xenopus* eggs identified a partly purified activity called **maturation promoting factor** that drives a resting *Xenopus* oocyte into M phase. MPF activity was found to be **oscillate** during the cell cycle, although the amount of its kinase component, called **Cdk**, remained constant. The regulatory component of MPF, called **cyclin**, has a **stimulatory** effect on MPF activity and plays a part in regulating interactions with its **substrate**s. The components of MPF are evolutionarily **conserved** from yeast to humans, so that the corresponding human genes are **able** to function in yeast.

18-19 (d) The interphase cytoplasmic extract used as the control would not have contained nuclear components (because the nuclear membrane was intact) and so one cannot conclude that components of an interphase nucleus are insufficient to cause mitotic spindle formation. The other statements are all true.

18-20 (d) Although it is true that yeast cells have one Cdk and human cells have many, this statement does not provide any evidence for the conservation of function across evolutionary time. All the other statements do provide such evidence.

18-21 A. The lysine (K) at the position of arrow #2 in the daisy Cdk is likely the culprit for the disrupted interaction between Cdk and Cdk-activating kinase (see Figure A18-21). This changes a conserved glutamic acid (E) to a lysine, which results in a change from a negatively charged amino acid to a positively charged amino acid. Although there is also a difference in the daisy sequence at the position of arrow #1, this change from an arginine (R) to a lysine is less dramatic, as both are positively charged amino acids.

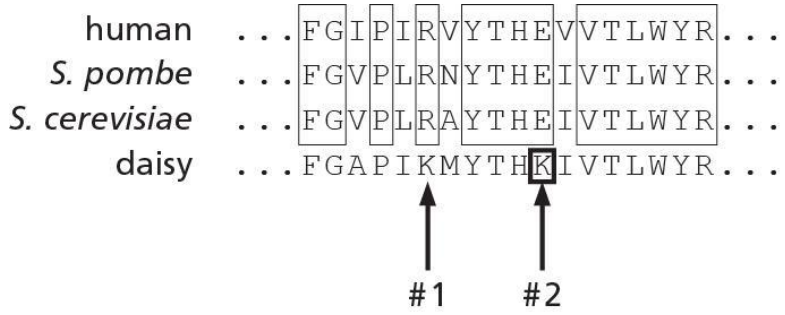

Figure A18-21

B. You would predict that the human Cdk will interact with the yeast Cdk-activating kinase, because the amino acids at the positions of arrows #1 and #2 are identical between human and yeast.

18-22 (a)

18-23 (c)

18-24 B, D, A, C

18-25 (d)

18-26 (a)

18-27 In G_0, the cell-cycle control system is partly dismantled, so that some of the Cdks and cyclins are not present. Cells paused in G_1, by contrast, still contain all the components of the cell-cycle control system. Whereas the latter cells can rapidly progress through the cycle when conditions are right, G_0 cells need to synthesize the missing cell-cycle control proteins so as to re-enter the cycle, which usually takes 8 hours or more.

18-28 (b)

18-29 (c) The concentration of Cdc6 rises early in G_1, independent of S–Cdk [choice (a)]. Cdc6 guides the assembly of the prereplicative complex at an origin [choice (d)]. By phosphorylating Cdc6, S-Cdk targets the protein for destruction, so that once an origin fires and replicates, Cdc6 will not be around to re-initiate replication. S-Cdk phosphorylates and activates DNA helicases [choice (b)].

18-30 (b)

18-31 A. G_1 arrest
B. neither
C. both
D. S-phase arrest

18-32 The cell cycle consists of an alternation between **M phase**, which appears as a period of dramatic activity under the microscope, and a preparative period called **interphase**, which consists of three phases called **G_1 phase**, **S phase**, and **G_2 phase**. During M phase, the nucleus divides in a process called **mitosis**, and the cytoplasm splits in two in a process called **cytokinesis**. The cell-cycle control system relies on sharp increases in the activities of regulatory proteins called **cyclin-dependent kinases**, or **Cdks**, to trigger S phase and M phase. Inactivation of **M-Cdk** is required to exit from M phase after chromosome segregation.

18-33 (a) Cell size increases throughout interphase and not during M phase. All of the other phenomena are observed in M phase.

18-34 (b)

18-35 (c)

18-36 (d)

18-37 A—telophase; B—prophase; C—anaphase; D—prometaphase

18-38 The correct order is listed as follows, with stages in parentheses: E (prophase), D (prometaphase), C (metaphase), A (anaphase), F (telophase), and B (cytokinesis).

18-39
A. 5, metaphase
B. 4, prometaphase
C. 3, prometaphase
D. 8, cytokinesis
E. 2, prophase
F. 7, telophase
G. 1, prophase
H. 6, anaphase

18-40 (a)

18-41 (d)

18-42 (e)

18-43 A. centrosome; B. chromosome

18-44 Centrosome duplication is semiconservative. The paired centrioles in the centrosome separate, and each serves to nucleate the assembly of a new centriole. As a consequence, each new centrosome consists of one old and one new centriole. Thus, centrosome duplication is analogous to DNA replication, in which the new double helix consists of one old DNA strand and one newly replicated DNA strand.

18-45
A. False. Sister chromatids are held together by **cohesins** from the time they arise by DNA replication until the time they separate at anaphase.
B. False. **Condensins** are required to make the chromosomes more compact and thus to prevent tangling between different chromosomes.
C. False. The **contractile ring** is composed of actin filaments and myosin filaments.
D. True.
E. False. The **centrosome** nucleates a radial array of microtubules called an aster, and its duplication is triggered by S-Cdk.
F. True.

18-46 Before mitosis, the number of centrosomes must **increase**. At the beginning of **prophase** in animal cells, the centrosomes separate in a process driven partly by interactions between the **plus** ends of microtubules arising from the two centrosomes. Centrosome separation initiates the assembly of the bipolar mitotic spindle and is associated with a sudden **increase** in the dynamic instability of microtubules. In comparison with an interphase microtubule array, a mitotic aster contains a **larger** number of **shorter** microtubules. Extracts from M-phase cells exhibit **unchanged** rates of microtubule polymerization and increased frequencies of microtubule **shrinkage**. The changes in

microtubule dynamics are largely due to **reduced** activity of microtubule-associated proteins and **increased** activity of catastrophins. The new balance between polymerization and depolymerization of microtubules is necessary for the mitotic spindle to move the **replicated chromosomes** to the metaphase plate.

18-47 A—1; B—2, 5; C—3, 4

18-48 (d) In animal cells, kinetochore microtubules cannot form if the chromosomes in the nucleus are separated from the microtubules in the cytoplasm because of the presence of the nuclear envelope. (But in some other cells, such as the yeast *S. cerevisiae*, the nuclear envelope never breaks down and yet chromosomes can attach to microtubules emanating from the spindle poles within the nucleus.) The nuclear envelope disassembles by breaking up into vesicles containing lipids from both the outer and inner envelopes [choice (a)]. Integral membrane proteins of the nuclear envelope and some of the nuclear lamins remain associated with the vesicles [choice (b)]. Phosphorylation of lamins (not integrins) triggers the breakdown of the nuclear lamina [choice (c)].

18-49 A—16; B—16; C—16; D—2; E—4

18-50 (a)

18-51 The statement is true.

18-52 (a) The jiggling movement [choice (a)] is simply a sign that the chromosomes are subject to forces from both sides, which could be the microtubules pulling, pushing, or both. All the other observations suggest pulling. When the attachment between sister chromatids is severed [choice (b)], both daughter chromosomes move toward their respective poles, which suggests that they are being pulled. When the attachment to one kinetochore is severed [choice (c)], the whole chromosome moves to the opposite pole, showing that the kinetochore microtubules are pulling on their attached chromatid, not pushing it. Similarly, the shape of the chromosomes as they move toward the pole [choice (d)] suggests that the chromosomes are being pulled.

18-53 Mitosis requires that the spindle microtubules behave dynamically—continuously polymerizing and depolymerizing—to probe the cell cortex, to seek attachments to kinetochores, and to segregate the chromosomes. Static microtubules are unable to do any of these things.

18-54 (c) The APC becomes activated in mid to late M phase.

18-55 Choice (d) is correct. Anaphase A and anaphase B generally occur at the same time [choice (a)]. In cells in which anaphase B predominates, the spindle will elongate more than in cells in which anaphase A predominates [choice (b)]. In anaphase A, only the kinetochore microtubules shorten [choice (c)].

18-56 (d) The motor proteins used in anaphase B to push the interpolar microtubules apart move toward the plus end of microtubules, whereas the motor proteins used in anaphase A move toward the minus end. Myosin [choice (a)] is not involved in either anaphase A or anaphase B. Motor proteins require ATP hydrolysis (they are ATPases) to function and are used in both anaphase A and anaphase B, so both types of anaphase will be affected by ATPγS [choice (b)]. The motor proteins used in anaphase A move toward the minus end of microtubules, as do the motor proteins used in anaphase B that are attached to the cell cortex [choice (c)].

18-57 (a) The contractile ring in an animal cell begins to assemble in anaphase. The chromosomes do not decondense [choice (b)], the lamina does not re-form [choice (c)], and transcription does not begin [choice (d)] until the formation of the nuclear envelope is complete and nuclear proteins have been imported through the nuclear pores.

18-58 (b) If the lamins cannot be phosphorylated during mitosis, the cells will be unable to disassemble their nuclear lamina, preventing the breakdown of the nuclear envelope at prometaphase. The mitotic spindle begins to form before the nuclear envelope breaks down, forming a sort of cage around the nucleus [choice (c)]. Lamins are not involved in chromosome condensation [choice (d)].

18-59 In a cell with excessive catastrophin molecules, the balance between catastrophins and MAPs will be disrupted. Increased catastrophin activity relative to MAP activity will lead to an increased frequency of microtubule catastrophes, the sudden shift from growth to shrinkage. Thus, microtubules will be shorter on average because they spend less time growing slowly and more time shrinking rapidly. Shorter microtubules would probably result in a shorter mitotic spindle. Conversely, increased MAP activity relative to catastrophin activity will stabilize microtubules by enhancing polymerization or inhibiting depolymerization, thereby promoting the formation of longer microtubules and probably a longer mitotic spindle. However, in some cases, the normal balance between catastrophins and MAPs has been shown to be necessary for the formation of a bipolar spindle. It is possible that increasing the amount of MAP or catastrophin proteins will result in microtubules that are so long or so short that they cannot form a spindle at all.

18-60 A. p98 behaves like a MAP, because it promotes microtubule growth.

B. The four possible mechanisms by which a protein can promote microtubule growth are (1) increasing the rate of polymerization, (2) decreasing the rate of depolymerization, (3) inhibiting the occurrence of catastrophes (when a microtubule shifts abruptly from growing to shrinking), and (4) stimulating the occurrence of rescues (when a microtubule switches from shrinking to growing).

C. p98 increased the rate of microtubule growth, as seen by the steeper slopes of the lines when p98 was added. The data do not allow a conclusion to be made about the rate of microtubule shrinkage. The protein decreased the rate of catastrophes: two of the five microtubules in the absence of p98 underwent catastrophe, whereas none did so in the presence of p98. No rescues were observed in either

the presence or the absence of p98, so it is uncertain whether the protein alters the rescue rate.

D. The p98 protein can be removed from an extract of *Xenopus* eggs in mitosis by using antibodies that specifically recognize p98. The depleted extract with little to no p98 protein can then be mixed with sperm nuclei, centrosomes, and fluorescent tubulin. On the basis of the previous experiments, the p98-depleted extract would be expected to have shorter and more dynamic microtubules. Ideally, this kind of experiment would be supplemented with the production and examination of intact cells in which p98 function has been abolished by mutation or another means, such as RNA-mediated interference.

18-61 (a) All cell movement requires ATP, and in cytokinesis actin and myosin molecules are moving relative to one another to cause contraction of the contractile ring. The myosin is an ATPase that hydrolyzes ATP to power this movement. The assembly of the contractile ring requires a general rearrangement of the filaments in the cell cortex. The contractile ring in animal cells disassembles completely after mitosis, leaving no trace [choice (b)]. Phosphorylation of integrins (which weakens the hold of these transmembrane proteins on the extracellular matrix, thereby allowing cells to round up) generally *precedes* cytokinesis and is part of the general rearrangement of cell structure that accompanies cell division [choice (c)]. Microtubules do not have an important role in animal cytokinesis [choice (d)].

18-62 Choice (d) is false. Although the furrow always forms perpendicular to the interpolar microtubules about midway between the spindle poles [choice (c)], if the spindle were in an asymmetrical position (which can occur normally during development), cell division would not always occur in the middle of the cell.

18-63 (d) No contractile ring is formed during plant cytokinesis.

18-64 (b)

18-65 (a) Programmed cell death results from an intracellular suicide program that eliminates unneeded, unwanted, or damaged cells. It occurs frequently and happens even in healthy cells throughout the lifetime of an individual.

18-66 (c)

18-67 Choice (d) is correct. Bcl2 tends to inhibit rather than promote apoptosis [choice (a)]. The prodomain of procaspases does not contain the catalytic activity needed for procaspase activation and is usually discarded from the active caspase [choice (b)]. When activated, Bax and Bak promote apoptosis by stimulating the release of cytochrome *c* from mitochondria into the cytosol, not by binding to procaspases in the apoptosome [choice (c)].

18-68 The presence or absence of Bak and Bax would not affect whether a microinjection of cytochrome *c* would promote apoptosis, because Bax and Bak act upstream of cytochrome *c* by promoting its release from mitochondria. By promoting the formation of the apoptosome and the activation of procaspases, microinjection of cytochrome *c* bypasses the need for Bax or Bak in promoting apoptosis.

18-69 Programmed cell death also influences cell numbers. Most animal cells require survival signals from other cells to avoid programmed cell death, so that the levels of such signals can help determine how many cells live and how many die.

18-70 Immature neurons are produced in excess of the number that will eventually be required. They compete for the limited amount of survival factors secreted by the target cells they contact. Those cells that fail to get enough survival factor undergo programmed cell death. Up to half or more of the original nerve cells die in this way. This competitive mechanism helps match the number of developing nerve cells to the number of target cells they contact.

18-71 The survival, **proliferation**, and size of each cell in an animal are controlled by extracellular signal molecules secreted by neighboring and distant cells. Many of these signal molecules bind to a cell-surface **receptor** and trigger various intracellular signaling pathways. One class of signal molecules, called **mitogens**, stimulates cell division by releasing the molecular brakes that keep cells in the $\mathbf{G_0}$ or $\mathbf{G_1}$ phase of the cell cycle. Members of a second class of signal molecules are called **growth factors**, because they stimulate cell growth and an increase in cell mass. The third class of signal molecules, called **survival factors**, inhibits **apoptosis** by regulating members of the **Bcl2** family of proteins. In addition to such stimulatory factors, some signal proteins such as **myostatin** act negatively on other cells, inhibiting their survival, growth, or proliferation.

18-72 B and D are likely to cause cell-cycle arrest.

A. Because ligand-independent activation of a mitogen receptor will probably make the cell divide when it otherwise might not, this mutation is unlikely to cause a cell-cycle arrest.

B. This mutation is likely to cause cell-cycle arrest in late G_1. Without S-Cdk activity, the cells will probably be unable to enter S phase.

C. Phosphorylation-independent activity of G_1-Cdk is unlikely to lead to cell-cycle arrest; the cells should progress through the cycle, although the kinetics and fine regulation of the cycle may be altered.

D. This mutation is likely to cause cell-cycle arrest in early G_1. Unphosphorylated Rb will inhibit the transcription of genes required for progression through G_1 and into S phase. This inhibition is normally released on phosphorylation of Rb. If Rb cannot be phosphorylated, it will always inhibit transcription of these genes, leading to arrest in early G_1.

E. If Rb is inactivated by a mutation, cells will be more likely to divide in the absence of extracellular mitogens, which is the opposite of a cell-cycle arrest.