E1. A. False, this would suggest an infectious disease, because people living in the same area would be exposed to the same kinds of infectious agents. Relatives living together and apart would exhibit the same frequency for a genetic disease.

B. This could be true, because the individuals living in one area may be more genetically related. On the other hand, a particular infectious agent may be found only in southern Spain, and this might explain the high frequency in this region.

C. A specific age of onset is consistent with a genetic disease.

D. A higher likelihood of developing a disease among monozygotic twins compared to dizygotic twins is consistent with a genetic basis for a disease because monozygotic twins are more genetically similar (in fact, identical) compared to dizygotic twins.

E2. Perhaps the least convincing is the higher incidence of the disease in particular populations. Because populations living in specific geographic locations are exposed to their own unique environment, it is difficult to distinguish genetic versus environmental causes for a particular disease. The most convincing evidence might be the higher incidence of a disease in related individuals and/or the ability to correlate a disease with the presence of a mutant gene. Overall, however, the reliability that a disease has a genetic component should be based on as many observations as possible.

E3. The term *genetic testing* refers to the use of laboratory tests to determine if an individual is a carrier or affected by a genetic disease. Testing at the protein level means that the amount or activity of the protein is assayed. Testing at the DNA level means that the researcher tries to detect the mutant allele at the molecular or chromosomal level. Examples of approaches are described in Table 22.4.

E4. You would probably conclude that it is less likely to have a genetic component. If it were rooted primarily in genetics, it would be likely to be found in the Central American population. Of course, there is a chance that very few or none of the people who migrated to Central America were carriers of the mutant gene. This is somewhat unlikely for a large migrating population. By comparison, one might suspect that an environmental agent that is present in South America but not present in Central America may underlie the disease. Researchers could try to search for this environmental agent (e.g., pathogenic organism, etc.).

E5. A. As seen in lane 3, the α–galactosidase A polypeptide is shorter in cells obtained from Pete. This indicates that Pete's disease is caused by a mutation that either is a deletion in the gene or introduces an early stop codon. In Jerry's case (lane 6), there does not appear to be any of the α–galactosidase A polypeptide in his cells. This could be due to a deletion that removes the entire gene, a promoter mutation that prevents the expression of the gene, a mutation that prevents translation (e.g., a mutation in the start codon), or a mutation that results in a polypeptide that is very unstable and rapidly degraded.

B. Amy appears to have two normal copies of the α–galactosidase A gene. She will not pass a mutant allele to her offspring. Nan is a heterozygote. She has a 50% chance of passing the mutant allele. Half of her sons would be affected with the disease. Likewise, Aileen also appears to be a heterozygote because the amount of α–galactosidase A polypeptide seems to be about 50% of normal. She also would have a 50% chance of passing the mutant allele to her offspring; half of her sons would be affected.

E6. Males I-1, II-1, II-4, II-6, III-3, III-8, and IV-5 have a normal copy of the gene. Males II-3, III-2, and IV-4 are hemizygous for an inactive mutant allele. Females III-4, III-6, IV-1, IV-2, and IV-3 have two normal copies of the gene, whereas females I-2, II-2, II-5, III-1, III-5, and III-7 are heterozygous carriers of a mutant allele.

E7. You would not expect a high number of malignant foci. Mutations in tumor-suppressor genes that cause malignancy are due to an inactivation of the tumor-suppressor gene. The NIH3T3 cells must have normal (nondefective) tumor-suppressor genes, otherwise they would be malignant. If a defective tumor-suppressor gene was transformed into NIH3T3 cells, it would have no effect because the NIH3T3 cells already have normal tumor-suppressor genes that prevent malignant growth.

Note: In the experiment of Figure 22.9, the sources of DNA that led to a large number of malignant foci (e.g., MC5-5-0) must have contained oncogenes. Oncogenes have mutations that lead to the overexpression of genes that control cell division. When an oncogene is taken up by the NIH3T3 cells, it causes malignant growth due to gene overexpression. The NIH3T3 cells are not able to prevent this overexpression that leads to uncontrolled cell growth. In other words, the normal tumor-suppressor genes in NIH3T3 cells are not strong enough to overcome the effects of oncogenes.

E8. A transformed cell is one that has become malignant. In a laboratory, this can be done in three ways. First, the cells could be treated with a mutagen that would convert a proto-oncogene into an oncogene. Second, cells could be exposed to the DNA from a malignant cell line. Under the appropriate conditions, this DNA can be taken up by the cells and integrated into their genome so that they become malignant. A third way to transform cells is by exposure to an oncogenic virus.

E9. If the DNA sample had been treated with RNase or protease, the results would have been the same. If they had been treated with DNase, no transformation would have occurred.

E10. By comparing oncogenic viruses with strains that have lost their oncogenicity, researchers have been able to identify particular genes that cause cancer. This has led to the identification of many oncogenes. From this work, researchers have also learned that normal cells contain proto-oncogenes that usually play a role in cell division. This suggests that oncogenes exert their effects by upsetting the cell division process. In particular, it appears that oncogenes are abnormally active and keep the cell division cycle in a permanent "on" position.

E11. Most inherited forms of cancer are inherited in a dominant manner. This can oftentimes be revealed by a pedigree analysis, since affected individuals are much more likely to have affected offspring. Since cancer may be caused by the sequential accumulation of mutations, the correlation between affected parents and affected offspring may be relatively low because the offspring may not accumulate the other postzygotic mutations that are necessary for cancer to occur.

E12. One possible category of drugs would be GDP analogues (i.e., compounds that resemble the structure of GDP). Perhaps one could find a GDP analogue that binds to the Ras protein and locks it in the inactive conformation.

One way to test the efficacy of such a drug would be to incubate the drug with a type of cancer cell that is known to have an overactive Ras protein, and then plate the cells on solid media. If the drug locked the Ras protein in the inactive conformation, it should inhibit the formation of malignant growth or malignant foci.

There are possible side effects of such drugs. First, they might block the growth of normal cells, since Ras protein plays a role in normal cell proliferation. Second (if you have taken a cell biology course), there are many GTP/GDP-binding proteins in cells, and the drugs could somehow inhibit cell growth and function by interacting with these proteins.

E13. Mammalian cells grow as a monolayer on solid growth media, whereas malignant cells tend to pile on top of each other and form malignant foci. A malignant focus can be formed from a single cancer cell that has divided many times. (It is also possible that multiple independent cancer cells could form a malignant focus, but this would not be necessary.)