E1. The expected result would be that the embryo would develop with two anterior ends. It is difficult to predict what would happen at later stages of development. At that point, the genetic hierarchy has already been established so its effects would be diminished. Also, at later stages of development, the embryo is divided into many cells so the injection would probably affect a smaller area.

E2. *Drosophila* is more advanced from the perspective that many more mutant alleles have been identified that alter development in specific ways. The hierarchy of gene regulation is particularly well understood in the fruit fly. *C. elegans* has the advantage of simplicity and a complete knowledge of cell fate. This enables researchers to explore how the timing of gene expression is critical to the developmental process.

E3. The term *cell fate* refers to the final cell type that a cell will become. For example, the fate of a cell may be a muscle cell. A lineage diagram depicts the cell lineages and final cell fates for a group of cells. In *C. elegans*, an entire lineage diagram has been established. A cell lineage is a description of the sequential division patterns that particular cells progress through during the developmental stages of an organism.

E4. To determine that a mutation is affecting the timing of developmental decisions, a researcher needs to know the normal time or stage of development when cells are supposed to divide, and what types of cells will be produced. With this information (i.e., a lineage diagram), one can then determine if particular mutations alter the timing when cell division occurs.

E5. A bag of worms phenotype can be due to an aberrancy in the development of several cell types. It is also an easy phenotype to observe and study. In some cases, the inability to lay eggs may be due to the abnormal timing of developmental steps in particular cell lineages (although this is not always the case). A researcher can watch the division patterns of cells that may be affected in the bag of worms phenotype to see if the timing of cell division is out of sync with the rest of the animal.

E6. Mutant 1 is a gain-of-function allele; it keeps reiterating the L1 pattern of division. Mutant 2 is a loss-of-function allele; it skips the L1 pattern and immediately follows an L2 pattern.

E7. These results indicate that the gene product is needed from 1 to 3 hours after fertilization for the embryo to develop properly and survive. The gene product is not needed at the other developmental stages that were examined in this experiment (0–1 hours, or 3–6 hours after fertilization).

E8. As discussed in Chapter 15, most eukaryotic genes have a core promoter that is adjacent to the coding sequence; regulatory elements that control the transcription rate at the promoter are typically upstream from the core promoter. Therefore, to get the *Antp* gene product expressed where the *abd-A* gene product is normally expressed, you would link the upstream genetic regulatory region of the *abd-A* gene to the coding sequence of the *Antp* gene. This construct would be inserted into the middle of a P element (see next). The construct shown here would then be introduced into an embryo by P element transformation.

P element	***abd-A* regulatory region**	**/**	***Antp* coding sequence**
P element			

The *Antp* gene product is normally expressed in the thoracic region and produces segments with legs, as illustrated in Figure 23.11. Therefore, because the *abd-A* gene product is normally expressed in the anterior abdominal segments, one might predict that the genetic construct shown above would produce a fly with legs attached to the segments that are supposed to be the anterior abdominal segments. In other words, the anterior abdominal segments would probably resemble thoracic segments with legs.

E9. A. Yes, because Krüppel protein acts as a transcriptional repressor, and its concentration is low in this region anyway.

B. Probably not, because Bicoid protein acts as a transcriptional activator.

C. Probably not, because Hunchback protein acts as a transcriptional activator.

D. Yes, because giant protein acts as a repressor, and its concentration is low in this region anyway.

E10. A. The female flies must have had mothers that were heterozygous for a (dominant) normal allele and the mutant allele. Their fathers were either homozygous for the mutant allele or heterozygous. The female flies inherited a mutant allele from both their father and mother. Nevertheless, because their mother was heterozygous for the normal (dominant) allele and mutant allele, and because this is a maternal effect gene, their phenotype is based on the genotype of their mother. The normal allele is dominant, so they have a normal phenotype.

B. *Bicoid-A* appears to have a deletion that removes part of the sequence of the gene and thereby results in a shorter mRNA. *Bicoid-B* could also be a deletion that removes all of the sequence of the *bicoid* gene or it could be a promoter mutation that prevents the expression of the *bicoid* gene. *Bicoid-C* seems to be a point mutation that does not affect the amount of the *bicoid* mRNA.

With regard to function, all three mutations are known to be loss-of-function mutations. *Bicoid-A* probably eliminates function by truncating the Bicoid protein. The Bicoid protein is a transcription factor. The *bicoid-A* mutation probably shortens this protein and thereby inhibits its function. The *bicoid-B* mutation prevents expression of the *bicoid* mRNA. Therefore, none of the Bicoid protein would be made, and this would explain the loss of function. The *bicoid-C* mutation seems to prevent the proper localization of the *bicoid* mRNA in the oocyte. There must be proteins within the oocyte that recognize specific sequences in the *bicoid* mRNA and trap it in the anterior end of the oocyte. This mutation must change these sequences and prevent these proteins from recognizing the *bicoid* mRNA.

C. If we only used the technique of Northern blotting, we would not have understood how *bicoid-C* was abnormal. Likewise, if we had only used the technique of *in situ* hybridization, we would not have understood how *bicoid-A* was abnormal.

E11. You could follow the strategy of reverse genetics. Basically, you would create a *HoxD-3* gene knockout. This inactivated *HoxD-3* gene would be introduced into a mouse by the technique of gene replacement described in Chapter 19. By making the appropriate crosses, homozygous mice would be obtained that carry the loss-of-function allele in place of the wild-type *HoxD-3* gene. The phenotypic characteristics of normal mice would then be compared to mice that were homozygous for a defective *HoxD-3* gene. This would involve an examination of the skeletal anatomies of mice at various stages of development. If the *HoxD-3* plays a role in development, you might see changes in morphology suggesting anterior transformations. In other words, a certain region of the mouse may have characteristics that are appropriate for more anterior segments.

E12. An egg-laying defect is somehow related to an abnormal anatomy. The *n540* strain has fewer neurons compared to a normal worm. Perhaps the *n540* strain is unable to lay eggs because it is missing neurons that are needed for egg laying. The *n536* and *n355* strains have an abnormal abundance of neurons. Perhaps this overabundance also interferes with the proper neural signals that are needed for egg laying.

E13. A. The larva would develop with two posterior ends. The larva would not survive to the adult stage.

B. The posterior end of the larva and adult fly would develop structures that were appropriate for thoracic segments. The adult fly may not survive.

C. The dorsal side of the larva and adult fly would develop structures that were appropriate for the ventral side. The adult fly may not survive.

E14. Geneticists who are interested in mammalian development have used reverse genetics because it has been difficult for them to identify mutations in developmental genes based on phenotypic effects in the embryo. This is because it is difficult to screen a large number of mammalian embryos in search of abnormal ones that carry mutant genes. It is easy to have thousands of flies in a laboratory, but it is not easy to have thousands of mice. Instead, it is easier to clone the normal gene based on its homology to invertebrate genes and then make mutations in vitro. These mutations can be introduced into a mouse to create a gene knockout. This strategy is opposite to that of Mendel, who characterized genes by first identifying phenotypic variants (e.g., tall vs. dwarf, green seeds vs. yellow seeds, etc.).