Fertilized mouse eggs divide very slowly at first. They reach two cells after about 24 hours and eight cells by 48 hours. At the eight-cell stage they undergo a process known as compaction, as illustrated in Fig. 1. Although the mechanism is not clear, the cells appear to adhere to one another more strongly; consequently, they change from being a clump of loosely associated cells to a tightly sealed ball. You wish to know what kinds of intercellular junctions are present before and after this change in adhesion.

To study this question, you use very fine glass micropipettes, which allow you to measure electrical events and at the same time to microinject either the enzyme horseradish peroxidase (HRP), 40,000 daltons, or the fluorescent dye fluorescein, 330 daltons. Fluorescein glows bright yellow under UV illumination, while HRP can be detected by fixing the cells and incubating them with appropriate substrates. You inject embryos at various stages of development with the two marker substances. At both the two-cell and eight-cell stages, different results are obtained, depending on whether the injections are make immediately after cell division or later (Fig. 2). Some of this difference can be attributed to the cytoplasmic bridges that linger for a while before cytokinesis is truly completed.

A. Why do both HRP and fluorescein enter neighboring cells early, but not late, at the two-cell stage?

B. Why does fluorescein enter all cells in the compacted eight-cell embryo, whereas HRP is confined to the injected cell?

C. In which of the four stages of development diagrammed in Figure 2 would you detect electrical coupling if you injected current from the HRP injection electrode and recorded voltage changes in the fluorescein electrode?