**Question:** Tom Zahrt isolated a strain of *Salmonella typhimurium* that could grow as well as the wild-type parent on rich medium, but when transferred from rich medium to minimal medium the strain a very long lag phase compared to the wild-type parent. In the figure below, growth of the wild-type is shown by black squares and the other strain is shown by blue triangles.

How could you determine if the growth that occurred after the long lag phase was (i) due to a second mutation that permitted growth in minimal medium or (ii) simply due to a requirement for longer time for adaptation to the minimal medium.

**Answer:** A simple approach is to take an aliquot from the culture that grew in minimal medium after the long lag phase and to subculture the bacteria in rich medium. After the culture grew in rich medium, subculture into minimal medium. If the original growth in minimal medium was due to a mutation, the strain should begin growing soon after subculture. In contrast, if the original growth in minimal medium was due to adaptation, it should require the strain just as long to adapt upon subculture from rich to minimal medium as it took following the first subculture.