The isolation of novel bacteriophages for phage therapy is necessary to find phages against an increasing number of pathogenic bacteria and to provide phages against bacteria that become resistant to previously used phages. The basic procedure for isolating bacteriophages remains relatively unchanged from the original procedures pioneered by Felix D’Herelle. An environmental sample appropriate for the target bacteria is obtained, processed to remove bacteria and larger material, and the resulting viral suspension is mixed with an isolation host bacterial culture. After incubation, any remaining bacteria are removed and the putative phage culture is screened for the presence of bacteriophages.

A number of researchers have developed alternative versions of this basic procedure. These include omitting the sample clearing before adding the isolation host; use of multiple isolation hosts that are closely related; use of isolation hosts that are distantly related; multiple rounds of isolation with varying hosts for each round. Each of these variations represents a way to deal with particularities of some phages and hosts such as phages that infect bacteria found in soil, phages which are often difficult to remove from soil particles. Sometimes the variation is meant to increase the numbers of phages isolated while other times the change is intended to increase the chances of isolating phages with a specific property, commonly increased host range.

Here we review these variations and discuss the strengths and weaknesses of each compared to the standard protocol. Finally, we make recommendations of when particular variations might be most useful in isolating novel bacteriophages for use in phage therapy.