**CDC Polio Lab Video 2019**

0:00 - 1:07

A CDC scientist is performing an environmental surveillance technique, concentrating poliovirus from sewage so that the virus can be grown in cultured cells and then tested using molecular methods. It is a standard WHO method for processing and concentrating sewage and environmental waters. There is no routine polio environmental surveillance in the U.S., but surveillance is done in many countries. A few other countries, like Haiti, send their sewage to CDC for analysis.

1:07 - 2:46

A CDC scientist adds stool to chemicals and puts the chemical mix into a centrifuge. To mix the stool and the chemicals, she puts the stool in with the cell culture media and glass beads, suspending the stool to get it into solution. Then she shakes it to get the stool to come off onto the liquid using a vortex mixer. She puts it in a centrifuge that spins the sealed buckets to separate the solid material from the liquid supernatant.

2:47 - 3:07

CDC scientists are observing cultured cells that have been infected with stool material containing poliovirus. The scientists are using a microscope to observe cells on the video monitor, scanning various areas of cells on the dish.

3:08 - 3:47

A CDC scientist is doing a virus plaque assay to determine the amount of virus in the cell culture isolates. She adds liquid, and then removes it to see which cells are stained. Those that are stained blue or purple have not been killed by virus. The dead cells are the unstained plaques – the clear open circles – and she can count the number of plaques to determine the amount of virus. A CDC scientist is also shown taking a photo of a plate with stained virus plaques. The CDC uses computer software to count the number of plaques and measure the sizes to compare one virus to another.

3:48 - 4:41

A CDC scientist adds chemical reagents to tubes to prepare them for molecular testing using polymerase chain reaction (PCR). He is using molecular testing to test for different types of polio; the 6-assay screen can determine which samples are polio, which serotype of polio, and whether they are vaccine or wild strains.

4:42 - 5:25

A CDC scientist prepares samples for next generation sequencing.

5:26 - 5:59

CDC scientists carry out a molecular test to sequence a virus. This is one of the last steps in testing process. The outer coat of the virus (the capsid) is sequenced.

The machine is a sequencing instrument – a Sanger sequencer. Sanger sequencing is the traditional type of sequencing. The plates go in and the machine analyzes the reactions. Dyes are used to determine

which of the nucleotides (bases of genetic material) is present in a sequence. Each base has a different color. The machine processes the information, producing a chromatogram of four colors. The peaks on the graph correspond to the intensity of color, and translates each color into a letter. By reading left to right, we can see the sequence of the material.

A computer compares one sequence to another to see how closely related different viruses are, and identify where a virus lives and spreads, within a country in order to focus vaccination programs. These sequences can be used to create a family tree of viruses that allows us to visualize how the viruses are moving around a country (spreading). We use this to identify reservoirs where the virus survives and spreads. Sequence gives us a window to where the virus has been and how long it has been circulating, allowing the immunization program to focus vaccination programs on certain parts of countries.

6:00 - 7:16

A CDC scientist uses 96-well plates to measure the amount of antibodies in a sample of blood. Each plate tests four serum samples for antibodies against one poliovirus serotype. The antibodies are able to block poliovirus from infecting cells; the amount of antibody can be determined using this assay. He is using a robot plate stacker to collect data from the stained serology plates. Purple wells mean the antibodies in the serum sample prevented poliovirus from infecting and killing the cells. If the well is clear, the antibodies did not block the virus from infecting and killing the cells. The presence of antibodies in serum capable of blocking poliovirus from infecting cells means that the person is protected from developing paralysis after exposure to poliovirus.