

Explaining the DHR test for diagnosing CGD

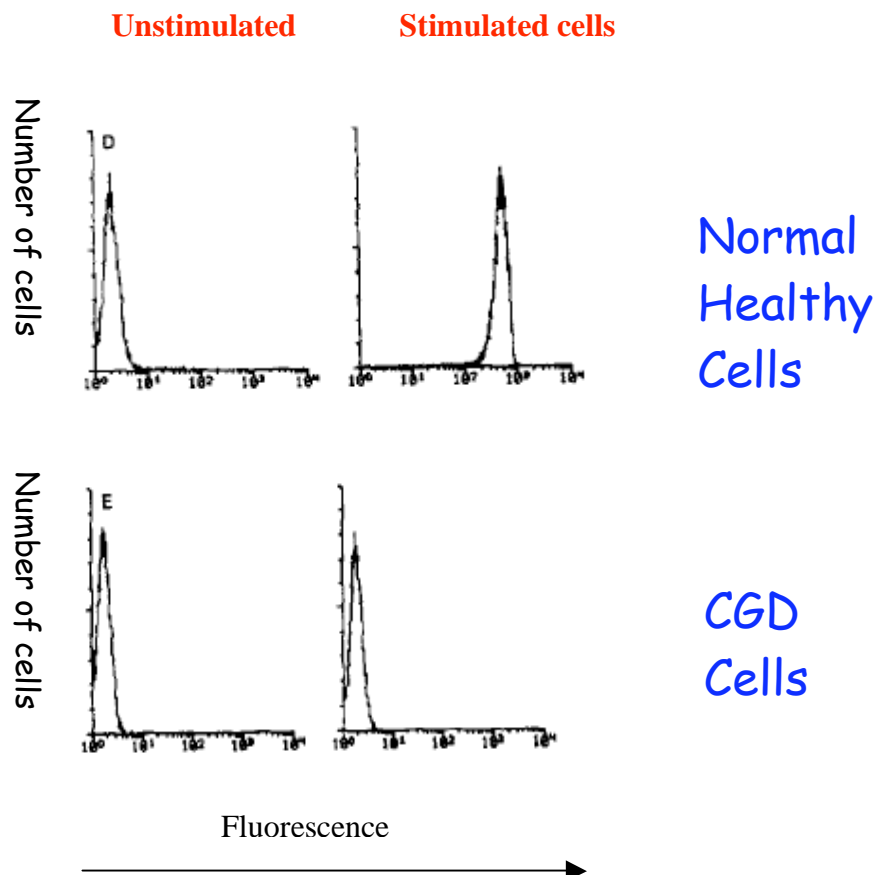
A different method called the dihydrorhodamine (DHR) test is now being used to help diagnose CGD. This test replaces the nitroblue tetrazolium (NBT) test but uses the same principle of testing for the production of reactive oxygen species (ROS) by white blood cells.

The DHR method works by testing whether a person's cells can oxidise dihydrorhodamine to the strongly fluorescent compound rhodamine when they are artificially stimulated to produce ROS. The production of rhodamine is then measured by passing the cells through the path of a laser that detects and measures the fluorescence, and the machine works out how much fluorescence is produced per cell. The laser system forms part of a sophisticated piece of machinery called a flow cytometer, equipment that is now commonly used in hospital laboratories.

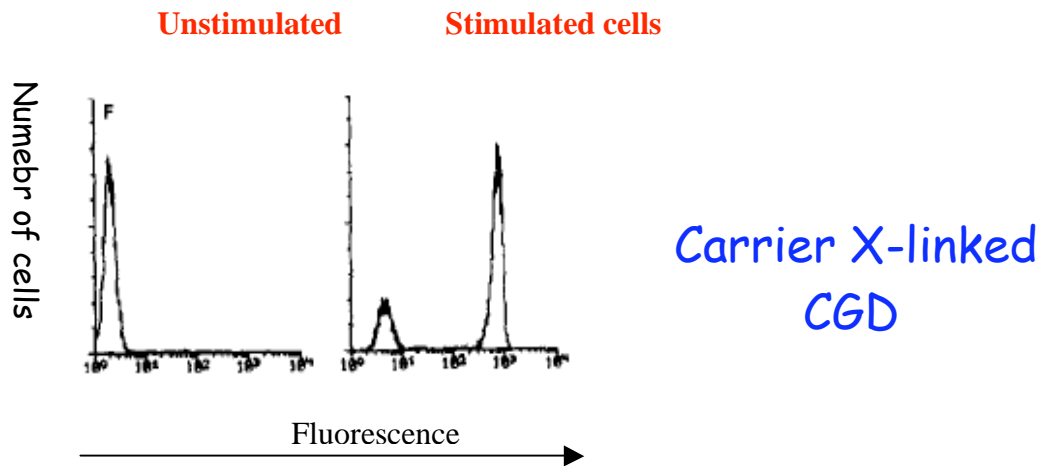
How does it work?

In healthy cells there will be lots of fluorescent signal produced, indicating the cells are producing the ROS necessary to kill bacteria. An abnormal result indicative of CGD or a carrier is found if the cells do not produce the fluorescent signal or if there is a change in the pattern of the fluorescence compared to a healthy sample (see below for examples).

Clinicians look at the pattern of the fluorescence and can determine the diagnosis of CGD. The DHR method can be used to test for all types of CGD.



This test can also be used to detect carriers of X-linked CGD, where two distinct peaks are seen, one from a population of cells that can produce ROS, and thus produce rhodamine in the test, and one from those that cannot, as shown below. If such a pattern is seen, this is indicative for X-linked CGD in the family. If not, the patient in the family can still have X-CGD. In all cases, genetic analysis is necessary for deciding the patient's CGD subtype.



'In contrast to the NBT test that relies on a trained eye to interpret the results, the DHR test can be easily interpreted by laboratory and clinicians and is much more sensitive', commented Professor Roos, of the Blood Cell Research Sanquin Research Centre in Amsterdam. We are now developing a method to combine the DHR with other reagents that will further pinpoint which part of the enzyme affected in CGD is not working properly. This will allow us to cut down on the amount of time it takes us to map exactly where the genetic mutation causing CGD in a person occurs.'