

CULTURING BORRELIA

Explanation of Advanced Laboratory Services culture process for Borrelia, the bacteria that causes Lyme Disease

The culture for Borrelia was developed approximately two years ago and is based on the research of Dr. Eva Sapi [*Int. J. Med. Sci.* 2013, 10:362-376]. The test can be ordered only by an MD, DO, or Nurse Practitioner in the United States. Four red topped tubes of blood are drawn (approximately 35- 40 cc). One red topped tube (approximately 8 cc) is transferred at the time of the draw to the media used to culture Borrelia. All the tubes are transported to the laboratory within 24 hours of the time of the draw. Further processing takes place once the blood arrives in the laboratory.

All manipulations from this point are performed in a sterile environment. The tubes are centrifuged to remove red cells, the serum is removed, and the cultures are inoculated. Cultures are harvested and examined at 6 -10 days, at 8 weeks and at 16 weeks if an extension is requested.

Cultures are examined by preparing slides that are stained using polyclonal and/or monoclonal antibodies. The stains are accompanied by positive and negative controls included with each run. A stain is also used to illuminate any background collagen that may be floating in the bloodstream.

Once the slides are read, the final read is performed by the lab director before reports are prepared and sent out.

Pictured below are pictures of the positive control, the negative control and a patient sample. Although also grown in culture, the positive control, *B burgdorferi* B31, is visually distinguishable from strains that grow in culture from the patients' blood. This positive control has a growth rate of 2-3 days in culture but it takes patient strains at least 6 days and more often 8 to 16 weeks to grow in culture. The control strain appears long and stringy in culture and the patient strains appear as thick spirals or odd shaped.

