Special Instructions

Cultivation of *Borrelia*

Borreliae have a characteristic spiral shape like all spirochetes, are about 8 – 30 µm long and about 0.2 – 05 µm wide and they are neither Gram-positive nor Gram-negative. They are highly motile organisms, with corkscrew and oscillating motility. Borreliae are fastidious organisms and require complex media for their cultivation. Most of them do not survive lyophilization and hence are delivered as **actively growing cultures** from the DSMZ. Cultures may still continue growing or may suffer from adverse conditions during the transportation process, thus, it is possible that they reach their destination in a less than optimal state. **Therefore, it is extremely important to transfer the obtained cultures into freshly prepared media immediately upon receipt.**

Borrelia

All *Borrelia* strains available from the DSMZ grow in **BSK- Medium**. This can either be prepared according to the instructions given on the DSMZ web site http://www.dsmz.de/microorganisms/medium/pdf/DSMZ_Medium403.pdf or bought as BSK-H medium (complete or incomplete (SIGMA, BIO&SELL (www.bio-sell.de)); in the latter case supplements (serum, gelatin, glutamine) have to be added according to the instructions given in the DSMZ catalogue of strains). Turbidity of the medium does not occur during the growth of *Borrelia*. Fermentation is indicated by a color change of the medium to yellow, however, at this point most of the cells are already dead.

You will receive from the DSMZ 5 ml of an actively growing culture in BSK-H medium. The cultures are checked directly prior to transport microscopically for cell number (at least two bacteria per field of vision at magnification x1000) and viability. When subculturing, combine fresh medium and culture in equal amounts and additionally prepare a dilution of 1:3 or 1:5 of the original culture. Cultivation is carried out at 37°C in a **candle jar** or under **microaerophilic conditions** (gas-pack; OXOID). After one or maximally two days the culture should be ready for further cultivation or preservation. To cultivate large quantities of cells it is recommended to double the volume of a well grown culture; **do not dilute more than 1:5**.

When strains are frozen at -80°C add glycerol (20%, sterile) to a final concentration of 10%, mix well and freeze. After thawing, add 0.5 ml of frozen stock to 3 ml of medium for activation. Check for growth microscopically, it may take 5 days or more until you see larger quantities of living cells.

Notes:

1. The information contained herein is offered for informational purposes only and is based on the present state of our knowledge. Recipients of our microorganisms must take responsibility for observing existing laws and regulations. DSMZ accepts no responsibility for the accuracy, sufficiency, reliability or for any loss or injury resulting from the use of the information.

2. If you have any questions or comments to this page, please send an e-mail to the following address: sgr06(at)dsmz.de