CP6 culturing

- 1. Mix 5ul stock of CP6 + 95ul Thioglycolate broth
- 2. Inoculate into Blood agar base (Sigma-Aldrich®, MO) supplemented with 5% of sheep blood (LAMPIRE biological laboratories, PA).
- 3. Incubate anaerobically with CO2 anaerobic pack (AnaeroPack® System, Mitsubishi Gas Chemical Company, INC., NY) in sterile bags at 37°C for 24 hours.
- 4. Pick 5-10 CP colonies and sub-culture it in 50 mL tubes containing thioglycolate broth with resazurine (Sigma-Aldrich®, MO) supplemented with 2% beef extract (Bioworld, bioPLUS Inc.) and incubate at 37 °C for 24 hours.
- 5. A second subculture mix this 50 ml broth with CP into, a total of 200 mL of thioglycolate broth with resazurine (Sigma-Aldrich®, MO) supplemented with 2% beef extract for 24 h at 37 °C under anaerobic conditions.
- 6. At 24h of incubation, the optical density of the culture, will be measured at 600nm with a spectrophotometer (BioTek Epoch spectrophotometer). The expected optimal density should be 0.8-1.2.