Miniprep

2 mL of LB medium (supplemented with the appropriate antiobiotic) with bacterial culture were inoculated and were left incubating overnight at 37 ºC with aggitation at 225 rpm. After the incubation period was finished, 1.5 mL of culture were deposited into a microcentrifuge compatible 2 ml tube and centrifugated for 30 seconds at 4 ºC. Once the centriguation was finished, the supernatant was discarded and resuspended in 100 µL of lisis solution 1, then it was vortexed for 5 min. Afterwards 200 µL of lisis solution 2 was added into the tube and mixed through inversion, then it was left to incubate for 15 min on ice. 150 µL of lisis solution 3 was then additioned into the tube and mixed softy through inversion to be left for a 15 min incubation on ice once again. Once the incubation period was completed, the tube was centrifugated for 5 min at maximum velocity. The supernatant was recovered in a new tube and an equal volume of phenol:chloroform:isoamyl alcohol 25:24:1 was added. The contents were then mixed by vortex and then centrifugated at max velocity in the microcentrifuge. The aqueos phase was transferred into a new tube, and mixedd with 2 volumes of 96% ethanol at room temperature and vortexed once more, then it was left incubating for 2 minutes at room temperature. The contents were centrifuged once more at maximum velocity for 5 min at 4 ºC and the supernatant was discarded. The tube was then left to dry open, inverted and over a towel until the remaining ethanol evaporated. 1 mL of 70% of ethanol was then added, mixed by inversion and then the contents were centrifugated for 5 min in maximum velocity once more. The contents were left to dry just as before open, inverted and over a towel until the ethanol had evaporated. The extracted nucleic acids were then resuspended on 50 µL of TE (pH 8.0) and gently vortxed for a few seconds. This solution was then stored at -20 ºC until it was used.