Edited out:

# Methods

### Pellet

In this experiment the goal was to scan bacteria as a pellet, with a low concentration of water and with as few other substances as possible. To achieve this, bacteria were washed as in the standard sample preparation but the final pellet was not re-suspended but scraped and loaded onto the aluminium cup. In order to get a large enough pellet, 2 starter cultures (15 ml each) were combined before the final centrifugation step.

### Saline

In this experiment the goal was to improve the bacterial Raman signal by suspending the bacteria in saline (0.9% NaCl). The rationale was that bacterial cells might be under osmotic stress in distilled water and are changing their optical properties. In this experiment all distilled water was replaced by autoclaved saline but the sample preparation was otherwise unchanged.

### Glass slides

In experiments done on glass slides the goal was to reduce scatter and reflectance from aluminium cup and (in some experiments) to reduce water content effect on the signal by drying the samples on the slides. After standard sample preparations, microscope glass slides (???) were loaded with 50 µL droplets of the samples. The slides were put into petri dishes and air-dried in an incubator at 37°C overnight.

### Aluminium slides

In this experiment the goal was to reduce the effect of water content on the signal by drying the samples. Roughly 10x3 cm cuttings of thick aluminum foil (???) were made and 50 µL droplets of samples were placed on top of the cuttings. The slides were put into petri dishes and air-dried in an incubator at 37°C overnight.

### “Dirty” experiment

In this experiment the goal was to keep the bacteria’s natural chemical environment in order to get a signal from both the bacteria and their environment. For this purpose, 15ml of overnight cultures were diluted in LB broth to OD of 1 and serial dilutions were made in LB broth to get a range of concentration between 106 – 10 CFU/ml. Samples were kept in ice to inhibit growth during transfer and scanning procedure, but brought to room temperature for actual Raman scanning.

### Milk

In this experiment the goal was to understand if the method will be applicable for use on milk samples. For this, bacterial cultures were washed as previously described and then diluted in Ultra-high-temperature treated (UHT) 3% fat milk (Tnuva, Israel). UHT milk was used because it is supposed to be sterile, and sterility was partially affirmed by inoculating 100µL of milk on agar plates and incubating for 72 hours in 37°C and 30°C.