

Supplementary material.

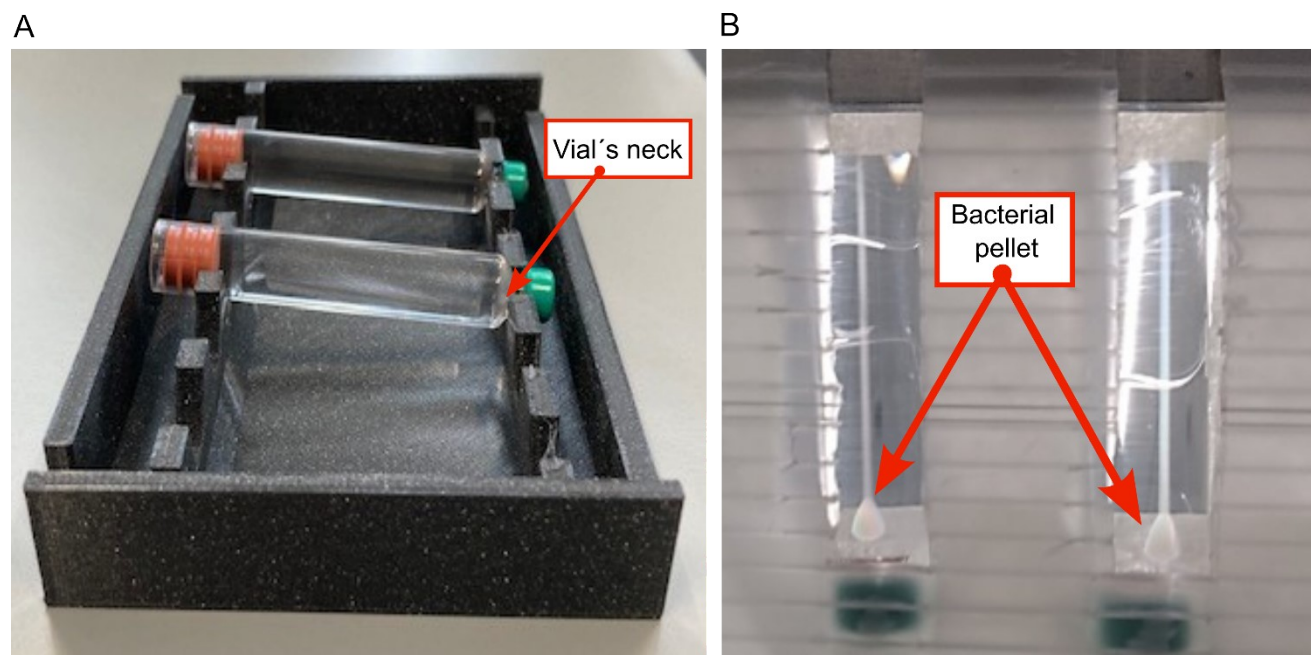


Figure 1. Representative pictures of the inclined centrifugation procedure. **(A):** Photo of custom-made plate shows the vial's position during centrifugation with a tilt of approx. 15 degrees. The lid is facing down (green metal) and the closing plunger (red rubber) facing up. Vial's neck is indicated (red arrow). **(B)** Photo of spiked vials with 10^{10} CFU of vegetative *Bacillus subtilis* and centrifuged in tilted position on prototype plates. The photo shown the tubes from below after centrifugation to visualize the location of the bacterial pellet. The pellet concentrates through the axial axis of the vial, with the highest concentration close to the neck of the vial.

Table 1: Results of incubated DP after Raman analysis. Incubation was performed up to 3 days at 35 °C following international standard procedures. The results for each sterile (negative controls) and spiked samples were considered by the absence of media turbidity as no growth (NG) or presence of turbidity as Growth (G). Results related to plate count of injected CFU amount (control of CFU spiked) are shown with their respective replicates.

Experiment	Growth result after Raman analysis																			Control of CFU spiked (by plate count)	
	Negative control samples										Spiked samples										
	1	2	3	4	5	6	7	8	9	10	1	2	3	4	5	6	7	8	9	10	Triplicates
<i>S. enterica</i> 50CFU	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	G	G	G	G	G	G	G	G	G	G	49 - 41 - 42 (av. 44 CFU)
<i>S. enterica</i> 10CFU	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	G	G	G	G	G	G	G	G	G	G	11 - 7 - 8 (av. 9 CFU)
<i>S. haemolyticus</i> 50CFU	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	G	G	G	G	G	G	G	G	G	G	76 - 61 (av. 68 CFU)
<i>S. haemolyticus</i> 10CFU	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	G	G	G	G	G	G	G	G	G	G	8 - 8 - 7 (av. 8 CFU)
<i>B. subtilis</i> 50CFU	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	G	G	G	G	G	G	G	G	G	G	41 - 43 - 50 (av. 45 CFU)
<i>B. subtilis</i> 10CFU	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	G	G	G	G	G	G	G	G	G	G	7 - 6 - 9 (av. 8 CFU)
<i>B. subtilis</i> 10CFU (validation)	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	G	G	G	G	G	G	G	G	G	G	9 - 8 - 8 (av. 8 CFU)

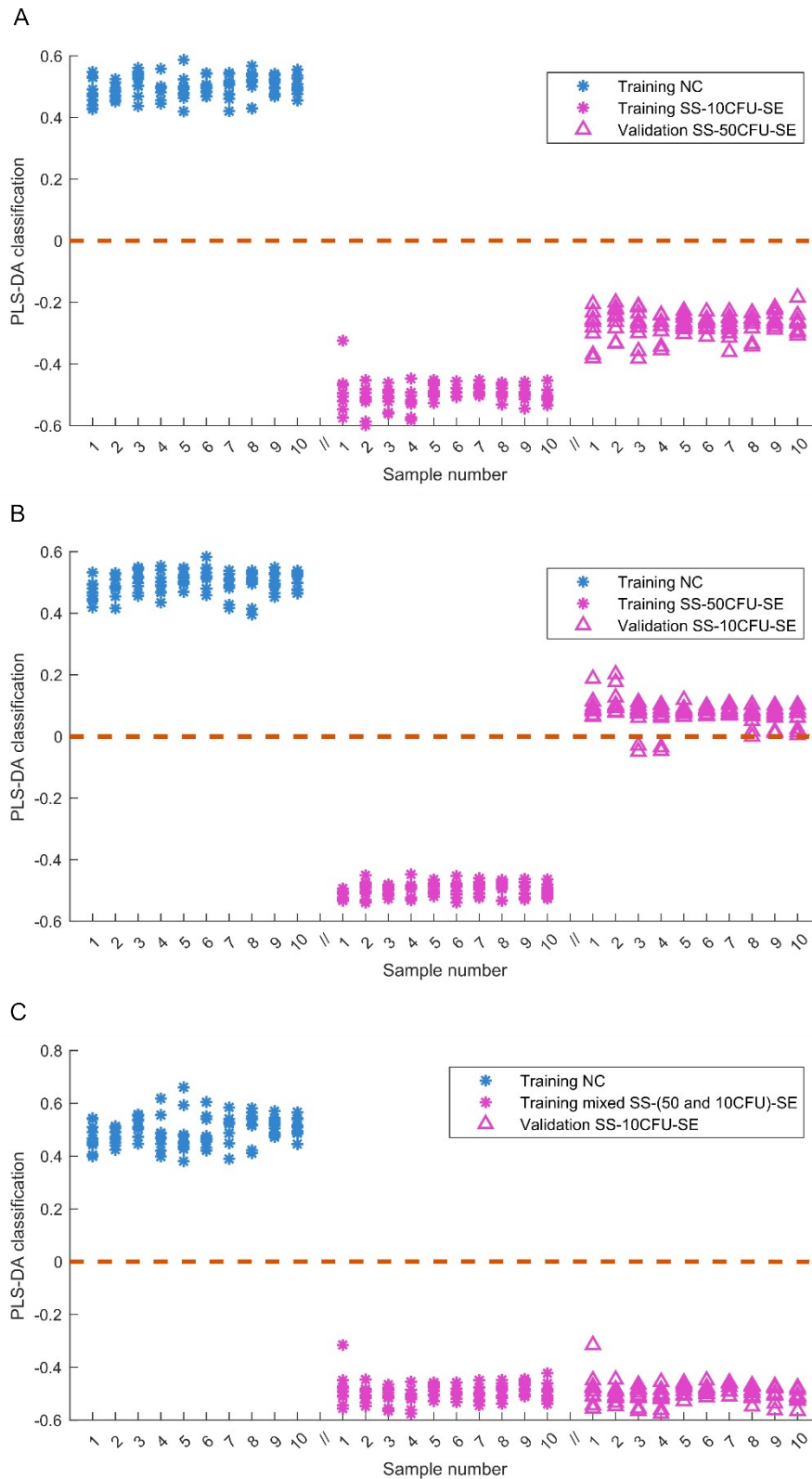


Figure 2: Model validation of spiked samples with different concentrations of *S. enterica* using PLS-DA and modelling with 10-fold cross-validation. The classification is based on a four-component model. Orange dotted lines represents the classification decision line from the model calibration. **(A)** Predicted classification of 10 spiked samples with 50 CFU/ml of *S. enterica* (SS-50CFU-SE) (magenta triangles). Spiked samples with 10 CFU/ml of *S. enterica* and respective NC were used as training dataset (magenta and blue stars respectively). **(B)** Predicted classification of 10 spiked samples with 10 CFU/ml of *S. enterica* (SS-10CFU-SE) (magenta triangles). In this model 10 samples with 50 CFU/ml of *S. enterica* and respective NC were used as training dataset (magenta and blue stars respectively). **(C)** A mixed sample training model was built to test the model performance to predict spiked samples with 10 CFU/ml. Here, for the training dataset corresponding to spiked sample were constructed with half SS-10CFU-SE and half 50CFU-SE. For NC training dataset comprised 10 sterile samples.