

# Salmonella Confirmation & Strain Typing with a Two-Step Detection Solution Using Mass Spectrometry & Infrared Spectroscopy in Combination



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## Introduction & Purpose

The MALDI Biotyper (MBT) is a benchtop mass spectrometry instrument and can be applied to identify microorganism in an undirected approach and/or to confirm foodborne pathogens starting from colony material. We launched our new MBT sirius platform (see fig. 1A) applicable for food & agriculture laboratories; and two instrument versions are available MBT sirius (GP) with positive/negative ion mode and MBT sirius one (GP) with positive ion mode only. Part01 of our study should show the application and performance of the new MBT sirius instrument platform for rapid *Salmonella* spp. confirmation compared to classic MBT models.



Fig.1B: IR Biotyper

The IR Biotyper is an infrared spectroscopy solution for source tracking and rapid serogroup differentiation (see fig 1B). In part02 we evaluated a new two-step approach: Confirmation with the MBT in combination with serogroup differentiation of *Salmonella* starting from colonies.

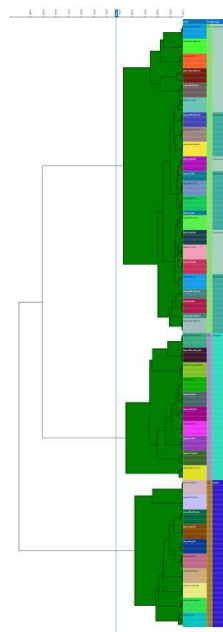
## Methods

For mass spectrometry analysis MALDI target plates were used for applying of colony material. Isolates are identified by comparing the generated spectra to the reference library, which covers currently 3,239 species.

For infrared spectroscopy analysis starts with a short sample preparation of colony material which was performed using Brukers IR Biotyper Kit. Samples and the IR test standard were applied on silicon microtiter plates dedicated to the IR Biotyper. The new IR Biotyper software calculates distance values of sample sets, and dataset can be shown as dendrograms and 2D scatter plots.



Fig.1A: MBT sirius



## Results

In the first study part we evaluated a high number of mass spectra on the MBT sirius and on MBT smart instruments. Results of cultures from non-selective and selective media were compared to each other plus different sample preparation procedures (DT, eDT and Ext) were applied in parallel.  $\text{Log}(\text{scores}) \geq 2.0$  showed reliable identification on genus level. Results of 5 different instruments were successfully compared for rapid confirmation of *Salmonella* spp. However, the protein-based MALDI technology does not allow subspecies differentiation e.g. from different *Salmonella enterica* subspecies which are frequent in the food and livestock industry.

In part02, we analyzed different *Salmonella enterica* subspecies with importance in the meat & poultry sector: In our set we included 10 Enteritidis, 10 Infantis, 20 Typhimurium strains (10 out of 20 monophasic variants) and successfully started with colony material. The IR Biotyper software 3.1 allows clustering of *Salmonella* samples with a new O-antigen-classifier. Here we present a 2D scatter plot and a dendrogram of all samples (see fig.2A & 2B). The IR Biotyper allowed differentiation of *Salmonella* O-groups on the same day within 3 hours. We could successfully show different well distinguishable groups in our test set with a mixture of different *Salmonella enterica* subspecies. Further research is necessary to determine whether subtypes within an O-group can also be distinguished in the future.

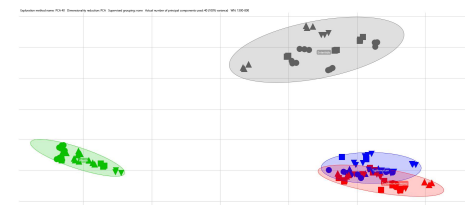


Fig.2A: 2D scatter plot, incl Enteritidis (grey group), Infantis (left/ green group) and 20xTyphimurium (right groups).

Fig.2B: In the dendrogram Infantis strains (blue bar) are close together in the lower part of the tree and are clearly separated from Enteritidis and Typhimurium ones.

## Conclusions

The MALDI Biotyper sirius system is a mass spectrometry platform useful for confirmation of *Salmonella* spp. Reliable and fast differentiation of foodborne *Salmonella* subspecies based on O-groups is now possible with the IR Biotyper. With the parallel use of two devices the user has a significant lower time-to-result compared to traditional confirmation and subtyping methods.

MALDI & IR Biotyper