



news digest #003

In less than two hours from colony: *Legionella pneumophila* identification and subsequent differentiation of *L. pneumophila* serogroup 1

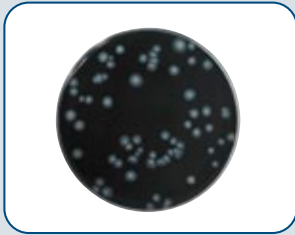
Legionellae are gram-negative waterborne bacteria. The genus *Legionella* was established in 1979 after a fast spread of pneumonia among members of the American Legion, hence called legionellosis or Legionnaires' disease. This outbreak was traced to a previously unknown bacterium, *Legionella pneumophila*. Most diagnostic tests are directed at the most severe species for human, *L. pneumophila* serogroup 1.

The detection of *Legionella* in water is one of the main tasks in water laboratories. After initial cultivation with the classical approach, the vast majority of laboratories require a subculture of suspect colonies on selective media and/or biochemical or molecular steps. Time-to-result for identification is often very long. The MALDI Biotyper® system allows a faster and reliable way of identifying species in minutes, starting from a single colony. This same-day identification result is achieved by matching the proteomic MALDI-TOF fingerprint mass spectrum to a library of reference mass spectral fingerprints. This elaborated library is the key-element of the MALDI Biotyper®, and Bruker have now reference spectra of more than 30 different *Legionella* species (listed on the right) for matching unknown colonies, in order to confirm *Legionella* spp. Even more species will be added during our annual library update 2021.

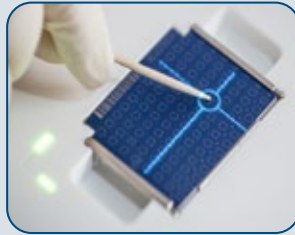
Legionella species in the MALDI Biotyper® reference library (status 2020, annually updated)

<i>L. anisa</i>	<i>L. jordanis</i>
<i>L. beliardensis</i>	<i>L. lansingensis</i>
<i>L. birminghamensis</i>	<i>L. longbeachae</i>
<i>L. brunensis</i>	<i>L. maceachernii</i>
<i>L. busanensis</i>	<i>L. moravica</i>
<i>L. cherrii</i>	<i>L. oakridgensis</i>
<i>L. cincinnatiensis</i>	<i>L. parisiensis</i>
<i>L. dresdenensis</i>	<i>L. pneumophila</i>
<i>L. erythra</i>	<i>L. rubrilucens</i>
<i>L. feeleii</i>	<i>L. sainthelensii</i>
<i>L. geestiana</i>	<i>L. santicrucis</i>
<i>L. gratiana</i>	<i>L. tucsonensis</i>
<i>L. hackeliae</i>	<i>L. wadsworthii</i>
<i>L. impletisoli</i>	<i>L. waltersii</i>

Selection of typical *Legionella* spp. colonies



Preparation of sample and control onto MALDI Biotarget 96 plate



***Legionella* species identification on the MALDI Biotyper®**



***Legionella* species identification in minutes**

Prepare a homogeneous suspension with the IR Biotyper® kit



Pipet samples and control onto the target plate, dry the samples



***Legionella pneumophila* serogroup 1 differentiation on the IR Biotyper®**



***L. pneumophila* serogroup 1 differentiation in less than 1 hour**

***Legionella* species identification and *L. pneumophila* serogroup 1 differentiation in less than 90 minutes**

Workflow: Starting from colony material and using both instruments in parallel or sequential, *Legionella* spp. confirmation and differentiation of serogroup 1 from all others can be achieved in less than 90 minutes.

Differentiation of *L. pneumophila* serogroup 1 from other serogroups is the next important question to answer, starting from the same colony material. The subspecies-level differentiation can be achieved by a second easy-to-use benchtop system, Bruker's IR Biotyper®, measuring non-protein-derived characteristic fingerprint signals, with a different technology than the MALDI Biotyper®.

The IR Biotyper® is based on Fourier Transform - Infra-Red (FT-IR) spectroscopy, offering an easy-to-use method for fast and effective microorganism strain typing, with a simple workflow and same-day result. The IR Biotyper® system analyses typical molecular vibrations caused by the absorption of infrared light. The absorption bands are assigned to certain chemical structures, with the wavelength range of the carbohydrates playing a major role for differentiation of the *L. pneumophila* serogroup 1 versus other serogroups. The new IR Biotyper® software version 3.0 extends the system's spectral range to also include lipids and peptides to enable further options for strain differentiation.

The IR Biotyper® workflow is easy and straightforward. From the colony a homogeneous suspension is prepared, using the IR Biotyper® kit. Aliquots of that suspension are pipetted onto a sample plate and a control is added as well. After drying, the sample plate is inserted in the IR Biotyper® for analysis, taking 30 minutes only for up to 30 microbial isolates. With the latest software version, *Legionella* strains can be classified as an immediate match to *Legionella* reference spectra, using the newly integrated *Legionella* classifier. Starting from colony material, *L. pneumophila* serogroup 1 can be discriminated from all other serogroups in a rapid workflow taking less than 1 hour. In addition, this classification model, based on Artificial Neural Networks (ANN), allows users to continually build up the model and train their system with common findings in water applications for improved classification power - not only for *Legionella*.

This powerful combination brings together the rapid microorganism identification capability (species level) of the MALDI Biotyper® (using MALDI-TOF mass spectrometry) and the strain typing/differentiation capability of the IR Biotyper® (using Fourier Transform - Infra-Red (FT-IR) spectroscopy).

Not for use in clinical diagnostic procedures. Please contact your local representative for availability in your country.

As of May 2021, Bruker Daltonik GmbH is now Bruker Daltonics GmbH & Co. KG.

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