Fast and precise cultureindependent diagnosis by pathogen enrichment

Molzym's Micro-Dx is the first walk-away automation for selective pathogen DNA extraction, combined with broad-range species identification for daily clinical practice.

ulture diagnosis of infectious diseases is standard procedure in clinical microbiology, but its long identification time of two to four days is a major limitation. In clinical practice, therefore, administration of broadspectrum antibiotics is necessary until definitive information on the species identity is available for targeted treatment. Another limitation of culture diagnosis is its low positivity of 30–40%, as determined by Afshari et al's 2012 study.

German company Molzym has developed a platform that addresses these shortcomings by using a cultureindependent, 16S and 18S rDNA-based broad-range identification approach. Real-time PCR detection and sequencing identification of bacterial and fungal pathogens is possible on the day of sampling, enabling early guidance for the adaptation of antibiotic treatment of critically ill patients.

Micro-Dx is principally unlimited in the identification of bacterial and fungal pathogens. So far, clinical studies have identified species from more than 200 bacterial and 65 fungal genera. Difficult-to-culture organisms, including *Tropheryma whipplei, Finegoldia magna, Bacteroides fragilis* and *Ureaplasma urealyticum*, were found in synovial and peritoneal fluids, and in the wounds of sick patients. In addition, the diagnosis of cerebrospinal fluid from culturenegative patients treated with antibiotics before sampling (due to suspicion of meningitis) gave reliable identities of pathogens by Molzym's broad-range PCR system. This selection of examples shows the potential of Molzym's diagnostic system as a precise and rapid means of identifying pathogens – growing or static – directly in samples, without the need for cultivation.

Unique automated pathogen enrichment and DNA extraction technology

Generally, clinical samples like blood from infected patients contain just a few pathogens. Their DNA is largely outnumbered by human DNA, which is known to produce false-positive results and reduce the sensitivity of PCR assays by unspecific primer binding. Molzym's MolYsis technology is an approach to resolve this problem by selective lysis of human cells and degradation of released human DNA, followed by the isolation of purified microbial DNA. The Micro-Dx system is based on MolYsis and is fully operated by the SelectNA plus robot.



With the SelectNA plus robot, pathogens are concentrated by vacuum filtration from 0.2–1.0ml fluid samples and tissues on a membrane. Pathogens are enzymatically lysed in situ on the membrane, and the DNA is subsequently purified following a 'bind, wash, elute' process. The SelectNA plus robot enables the preparation of up to 12 samples in a single run. The DNA extraction is completed in 85 minutes.

Range of applications

Micro-Dx contains consumables and reagents for the operation of DNA extraction, and reagents for the diagnosis of bacterial and fungal pathogens. Sequencing primers are delivered with the kit to run a Sanger sequence analysis for strain identification. All consumables and reagents are guaranteed to be free of contaminating microbial DNA, ensuring the maximum sensitivity and accuracy of diagnosis. A single programme operates the automated extraction of a variety of specimens, including EDTA or citrate blood, BAL, aspirates from joints, CSF and other primary sterile body fluids, and biopsies for tissues and abscesses.

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