Welcome to MIDL!

¡Bienvenido! Bienvenue! Herzlich Wilkommen!

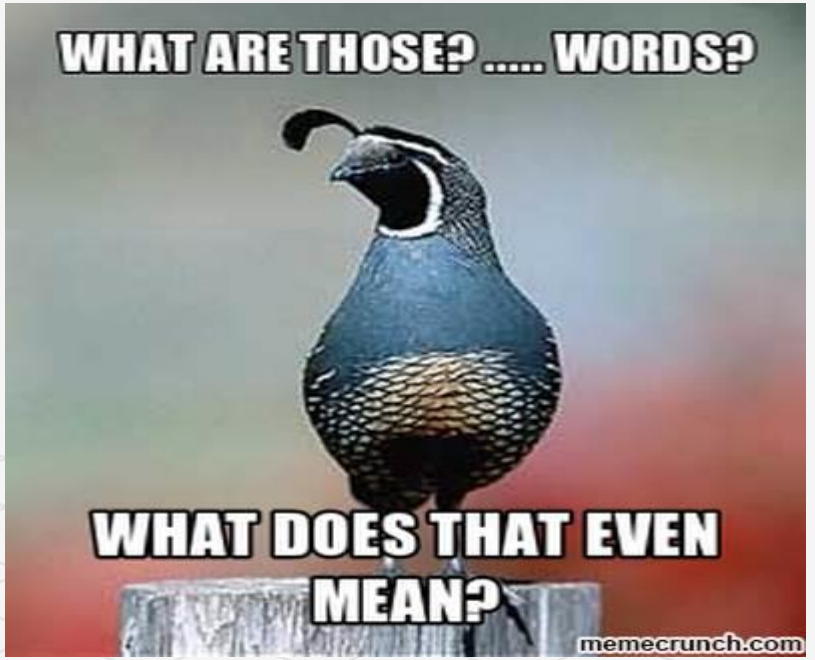
Molecular Infectious Disease Laboratory

What is the Molecular Infectious Laboratory?

- MIDL is one of the two clinical laboratories that offers molecular-based platforms for diagnostic testing.
- Diagnostic testing versus research laboratories
- MIDL's test directory and different PCR assay platforms
- Importance of tests (or assays) to VUMC's physicians

Tests Offered in the Molecular Infectious Disease Lab

- EHD
- VZD
- HSD
- CMD
- CMQ
- HNA
- HVG
- HCQ
- HC5
- EBD
- EBQ
- BKV
- PVB
- CGD
- GCD
- CTD
- TRICH
- HPP
- HBD
- GIP
- MEP
- RPP
- EVD



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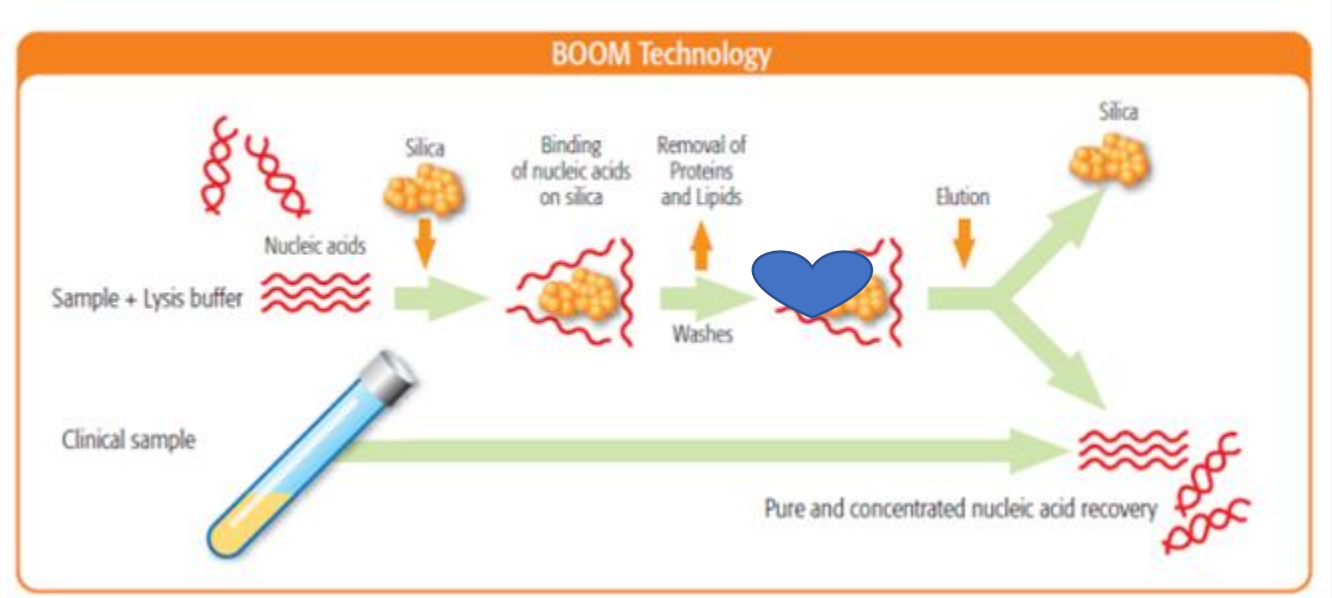
Basic Breakdown on Molecular Technology

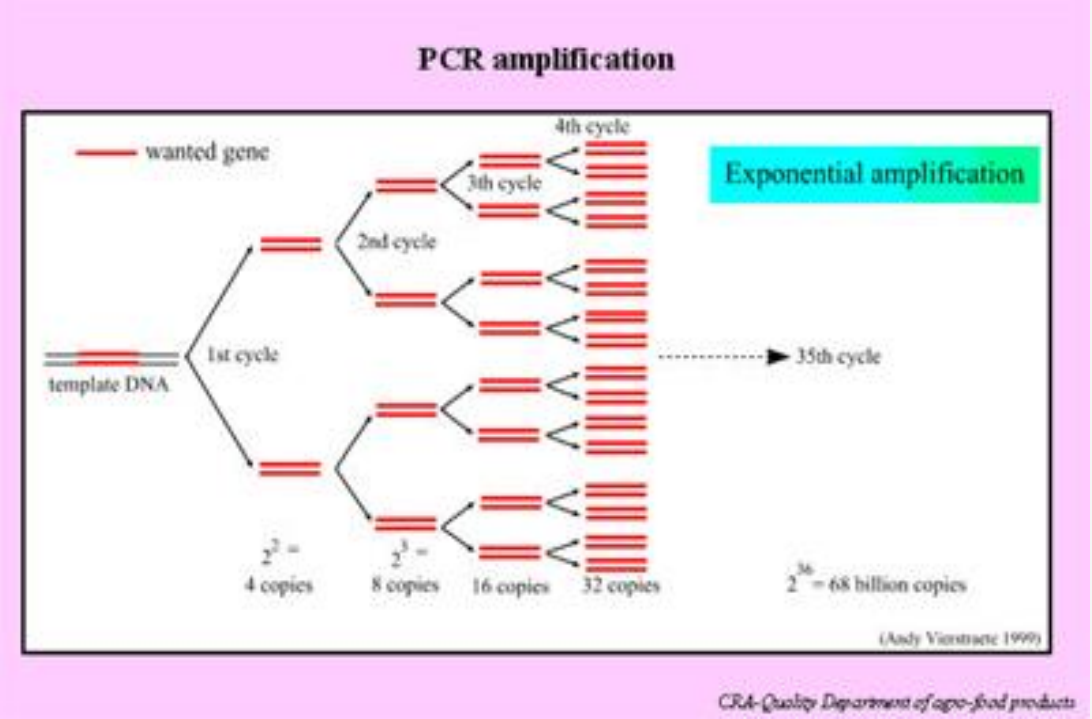
- MIDL incorporates many molecular platforms, but the first concept to understand is the breakdown on how we start the testing process from start to finish
- The first step is to extract DNA or RNA, depending on what organism is targeted, and the platform used to extract purified DNA or RNA.
- MIDL uses the EasyMAG system to lyse, bind DNA/RNA with magnetic silica (silica DNA/RNA), wash impurities from the silica-DNA/RNA hybrid, and finally elute the purified DNA or RNA from the silica.



https://www.biomerieux-diagnostics.com/sites/clinic/files/boom-technology.jpg

Basic Breakdown on Molecular Technology

- The second step is to amplify the requested target(s). This step uses mastermix, which contains primers, probes, and enzymes needed for the PCR process. The mastermix is added to the purified DNA or RNA extract., usually in a cuvette or 96-well plate, for amplification



http://stratfeed.cra.wallonie.be/img/page/PCR_web_page5.jpg

Basic Breakdown on Molecular Technology

- The third step in molecular testing is the detection of a target or signal.
- Many assays offer different detection steps, depending on the platform's manufacturer, which includes:
  - Electrochemical
  - Chemiluminescence
  - Hybrid capture or ELISA
  - XMAP (or bead technology)

Real-Time PCR

Tests that use RT-PCR technology:

- CMV, EBV, HBV, HCV, HIV-1, BKV  These are tests where a viral load is measured.
- CMV, EBV, and BKV are lab orders generated by treating physicians to monitor viral loads in immunocompromised and/or transplant recipients. The morbidity and mortality of these viruses to this subset of patients are well known.
- HIV-1, HCV, and HBD genomic copy levels are monitored by doctors who have prescribed these patients specific medications to help bring these viruses to undetectable loads. If a viral load changes drastically, then a physician will order a genotype to determine if the genetic structure of the virus mutated.
- HPV and CT/NG are sexually transmitted viruses that are measured by this testing platform.

Importance of Genotyping HCV and HIV

HIV-1 is an RNA virus that mutates. When a physician requests a genotype, he or she is trying to monitor current medication efficacy.

Current types of Antiretroviral therapy (ART):

- Nucleoside/Nucleotide Reverse Transciptase inhibitors
- Non-nucleoside Reverse Transcriptase Inhibitors
- Protease Inhibitors

Goals of these drug are to:

- Control the growth of the virus
- Improve how well the immune system works
- Slow or stop symptoms
- Prevent transmission of HIV to others.

MIDL extracts, amplifies, sequences, and then analyzes the patient's consensus sequence for accuracy against a reference sequence. The software has a built-in algorithm that determines if the patient is susceptible or resistant to different ART drugs currently available.

Importance of Hepatitis C Genotyping

- Hepatitis C has different genotypes, which are Types 1a, 1b, 2b, 2a, 2c, 3, 4, 5, 6
- New drugs developed as a cure for Hep. C
- What does this mean?
- People on transplant waiting lists can receive organs that are Hepatitis C positive and then receive the drug Harvoni.

Biofire

- This is a new testing platform that has a simple specimen set-up and rapid turn-around-time for multiple targets…infectious organisms. The FILMARRAY is an FDA-cleared multiplex PCR system that integrates sample preparation, amplification, detection and analysis.

Respiratory Pathogen Panel:

- One specimen (300 uL) is all that is needed to use this technology, which can detect viruses and bacteria that are known culprits for lower and upper respiratory infection.

Meningitis and Encephalitis Panel:

- Only 200 uL of cerebrospinal fluid (CSF) is needed to detect multiple targets responsible for meningitis and encephalitis.

Gastrointestinal Pathogen Panel:

- A small amount of stool specimen can detect multiple viruses, parasites, and bacteria in as little as one hour.

Importance of the Molecular Infectious Disease Lab

- Laboratory testing is a key part of the medical field. The Medical Laboratory Scientist provides data to the physician, which aids in the diagnosis and treatment for a patient
- The laboratorian maintains lab-wide safety standards because if such work is not done properly, it could mean life or death for a patient. In MIDL, contamination is a risk that could have serious implications.
- When health risks are diagnosed early, they usually cost less to treat. Medical testing plays an important role in managing these financial factors.
- As fully trained, educated medical professionals, we understand the mechanisms and purpose of the many tests that physicians order.