ID Lab Con 2025
Preconference Workshop

Optimizing Mycobacteriology Laboratory Operations:
Strategies for Supervisor Success

Quality Assurance & Quality Control for the Mycobacteriology Laboratory

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Objectives & Definitions

Objectives

- Understand the role and importance of regulatory bodies
- Understand requirements for validation and verification, including laboratory developed tests (LDT)
- Define and understand quality indicators/metrics
- Recognize common quality issues and understand steps in quality investigations

Definitions

- **Quality Assurance (QA)**
  - System of processes and requirements designed to prevent errors and ensure overall quality throughout laboratory operations
- **Quality Control (QC)**
  - Part of quality assurance
  - Active procedures and monitoring of testing to verify the accuracy, reliability, and validity of test results

Regulatory Bodies

Special focus on Clinical Laboratory Improvement Amendments (CLIA)

Clinical Laboratory Improvement Amendments (CLIA)

Federal regulatory program that sets quality standards for clinical laboratories

Applies to all laboratories that test human specimens for diagnosis or treatment

Important CLIA Standards

Proficiency Testing: §493.825 & §493.913

- Two testing events per year with five samples per testing event
- Must include samples for:
  - Acid-fast stain
  - Detection (presence and absence)
  - Identification
  - Susceptibility testing
- Test and report to the highest level of patient testing

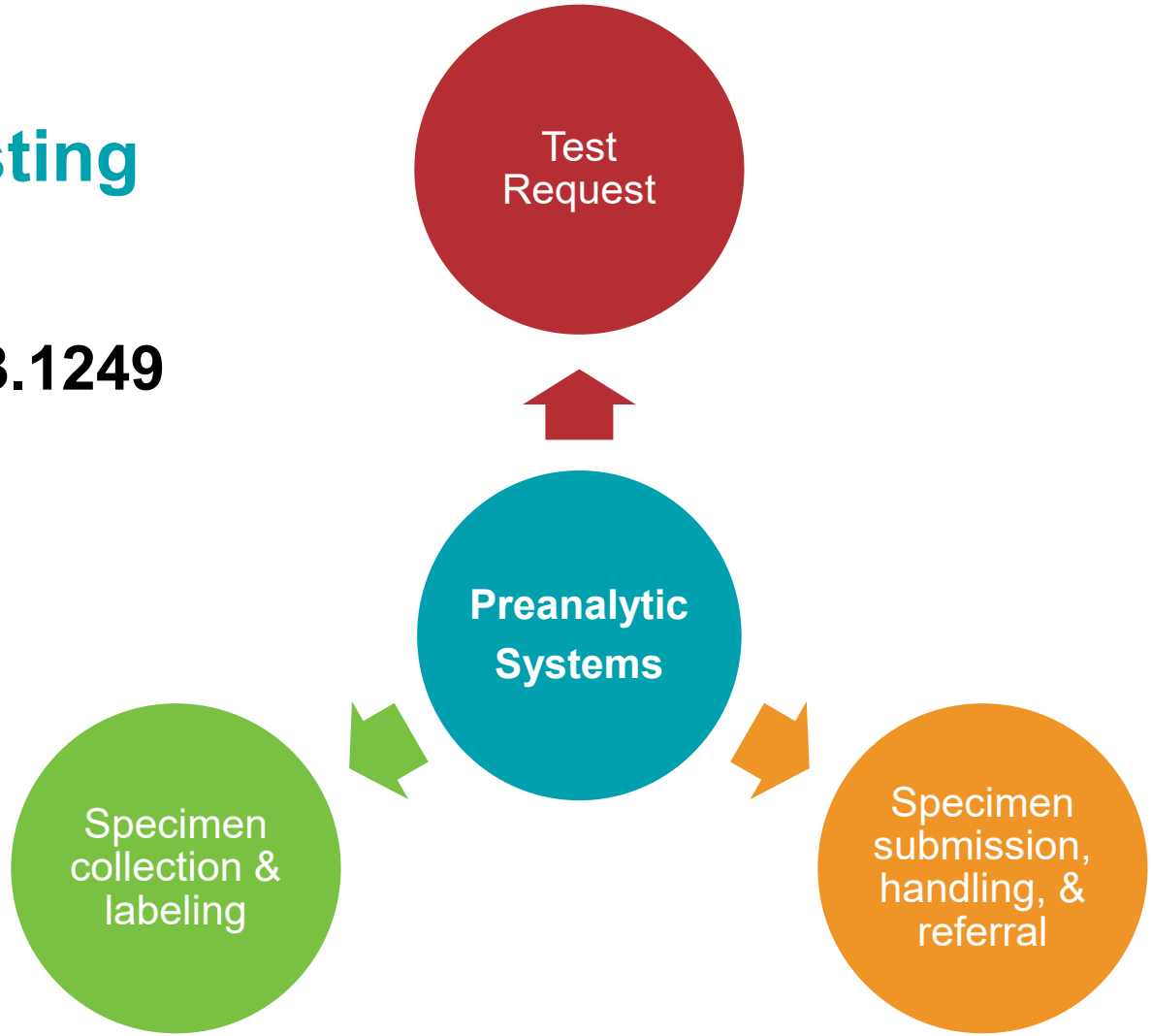
Personnel Requirements:

- High Complexity Testing Personnel
  - Qualifications: §493.1489
  - Responsibilities: §493.1495
- General Supervisor
  - Qualifications: §493.1461
  - Responsibilities: §493.1463
- Technical Supervisor
  - Qualifications: §493.1449
  - Responsibilities: §493.1451

Important CLIA Standards

Quality System for Nonwaived Testing

- **Preanalytic Systems** **§493.1240 – §493.1249**
  - Test request
  - Specimen submission, handling, and referral



Important CLIA Standards

Quality System for Nonwaived Testing

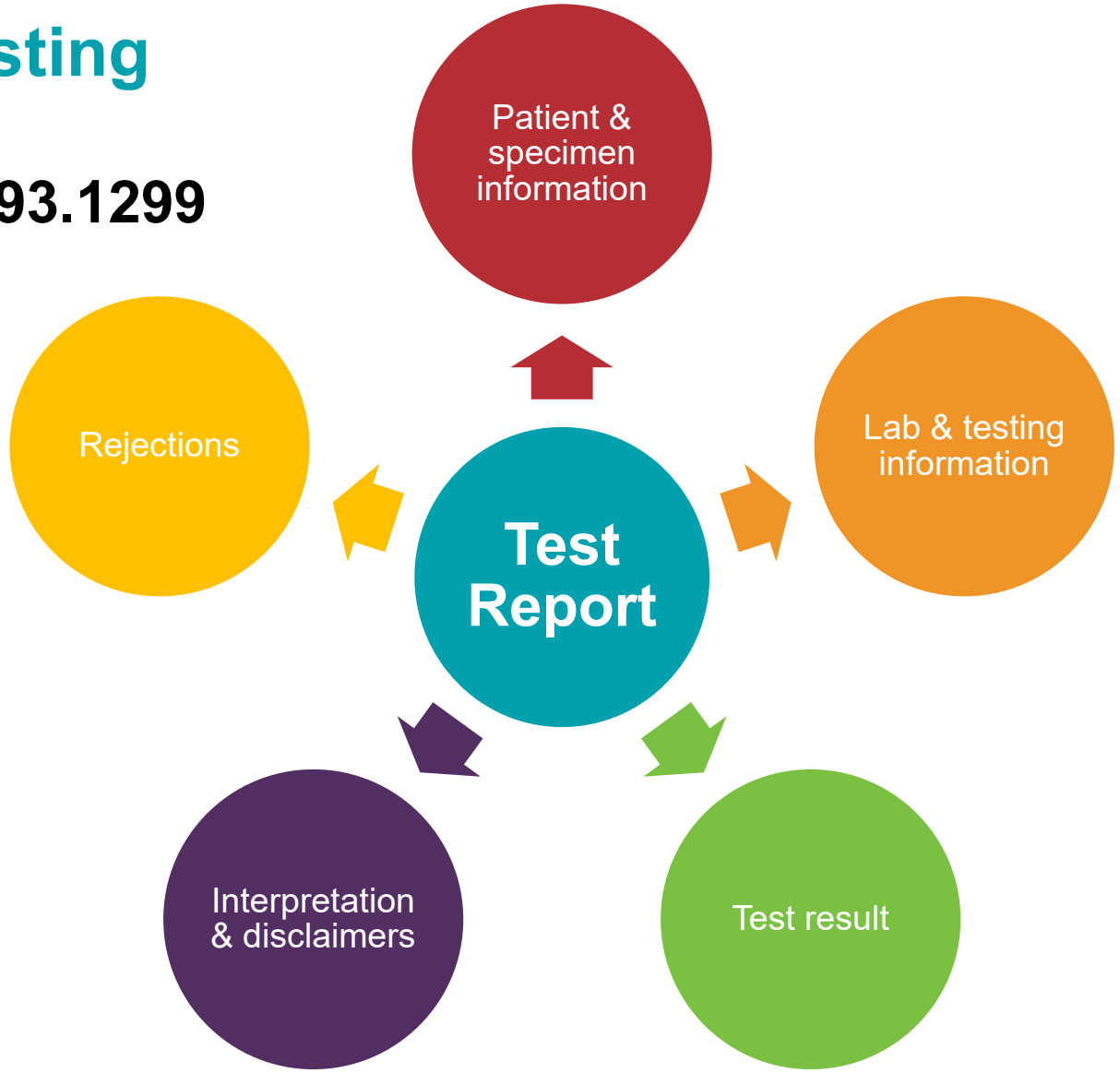
Analytic Systems §493.1250 – §493.1256, §493.1262

- Procedure manual
- Test systems, equipment, instruments, reagents, materials, and supplies
- Establishment and verification of performance specifications
- Maintenance and function checks
- Calibration and calibration verification procedures
- General control procedures
  - Test in the same manner as patient specimens & document all QC
  - Reagent, media, and supply checks
  - Follow manufacturer specifications
- Mycobacteriology specific control procedures (§493.1262)
  - Susceptibility testing

Important CLIA Standards

Quality System for Nonwaived Testing

- **Postanalytic Systems** **§493.1290 – §493.1299**
  - Test report
  - Turnaround times
  - Reporting errors
  - Patient access to results



Regulatory Inspections

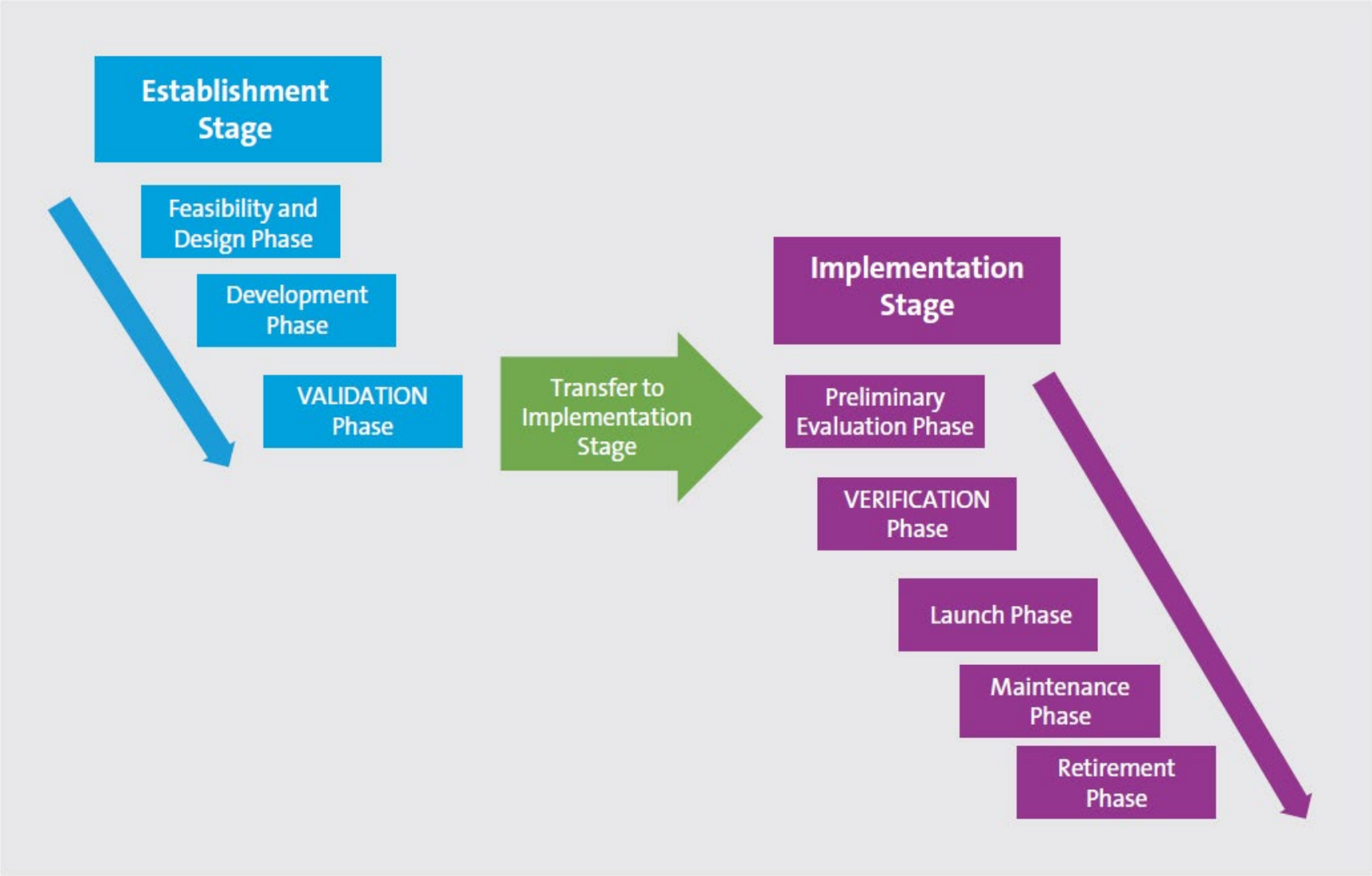
- CLIA conducts standard inspections of certified laboratories every two years
- CLIA Interpretative Guidelines
- APHL CLIA Resources
- Inspection preparation:
  - Internal audits (horizontal & vertical)
  - Routine review and ongoing quality assessments within the laboratory
  - APHL CLIA Audit Checklist
- Addressing deficiencies
  - Conditional deficiency from 2024 DCLS inspection – pre-analytic specimen transport

College of American Pathologists (CAP)

- CAP Laboratory Accreditation Program
  - Centers for Medicare and Medicaid Services (CMS) granted deeming authority
    - Allows CAP inspection in lieu of CMS (CLIA) inspection
  - CLIA is a federal regulatory body that sets minimum quality standards, while CAP is an accreditation program that goes beyond those minimums
  - Inspections performed every two years
    - Internal inspection on “off” year
  - Additional fees
  - More comprehensive approach to laboratory quality systems
  - CAP offers accreditation checklists that can be customized based on laboratory test menu

Validations & Verifications

Test Life Phases Model



CLSI EP19

Test Method Type

- FDA-approved Test Method
  - Requires extensive design and production controls and verification and validation studies by the manufacturer
  - Requires additional verification in the laboratory
- Laboratory Developed Test (LDT)
  - Test developed by a laboratory
  - Includes modification to an established test method (including an FDA-approved method)
  - Requires validation in the laboratory

CLIA §493.1253 Establishment and verification of performance specifications

Accuracy

- Closeness of agreement between a measured quantity value and a true quantity value
- Testing reference materials
- Compare results to reference method
- Compare split sample results to previously validated method

Precision

- Closeness of agreement between replicate results
- Repeat testing over time
- Patient, QC or calibration materials

Reportable Range

- Span of values for the test system
- Test low and high calibration/control materials
- Test known samples of high and low values

Reference Interval

- Interval between the lower reference limit to the upper reference limit of the population (95% of persons presumed to the healthy)
- Use manufacturer's reference range or published reference range

CLIA §493.1253 Establishment and verification of performance specifications

Analytical Sensitivity

- The lowest concentration or amount of the analyte that can be measured
- Limit of detection (LOD)

Analytical Specificity

- The extent to which the method can detect only the intended target
- Test materials containing interfering substances

Any Other Performance Characteristics

- Specific to the test method

Calibration and Controls

- Based upon the performance specifications verified
- Frequency, type, number and concentration

Verification/Validation Requirements

- Sample numbers may vary depending on:
  - FDA approved versus LDT
  - Qualitative versus quantitative
  - Difficulty obtaining samples
  - Cost of reagents
- Laboratory director has final discretion to determine requirements are met
  - Number of samples needed
  - Acceptable performance criteria

Common microbiology V/V suggestions

- Verification:
  - 20 - 40 samples
  - ≥ 20 positive samples
- Validation
  - ≥ 50-100 samples
- Acceptable performance
  - >90% accuracy and precision

Validation vs. Verification

- **Validation = Establishment**
  - Provide evidence that the accuracy, precision, analytical sensitivity, and analytical specificity is adequate
  - Performed before results can be used for decisions regarding patient care
- **Verification = Confirmation**
  - Verify the system is performing within manufacturer's specifications
  - Verify the ability of the laboratory staff to produce accurate and reproducible results
  - Fulfill regulatory requirements

Validation

- Reportable range
- Precision
- Accuracy
- Reference interval
- Analytical sensitivity
- Analytical specificity
- Interfering substances

Verification

- Reportable range
- Precision
- Accuracy
- Reference interval

Verification/Validation Steps

STEP 1

Develop validation plan and submit proposal

STEP 2

Validation Testing

STEP 3

Validation Report: data for each requirement, discuss any discrepancies, acceptance criteria met

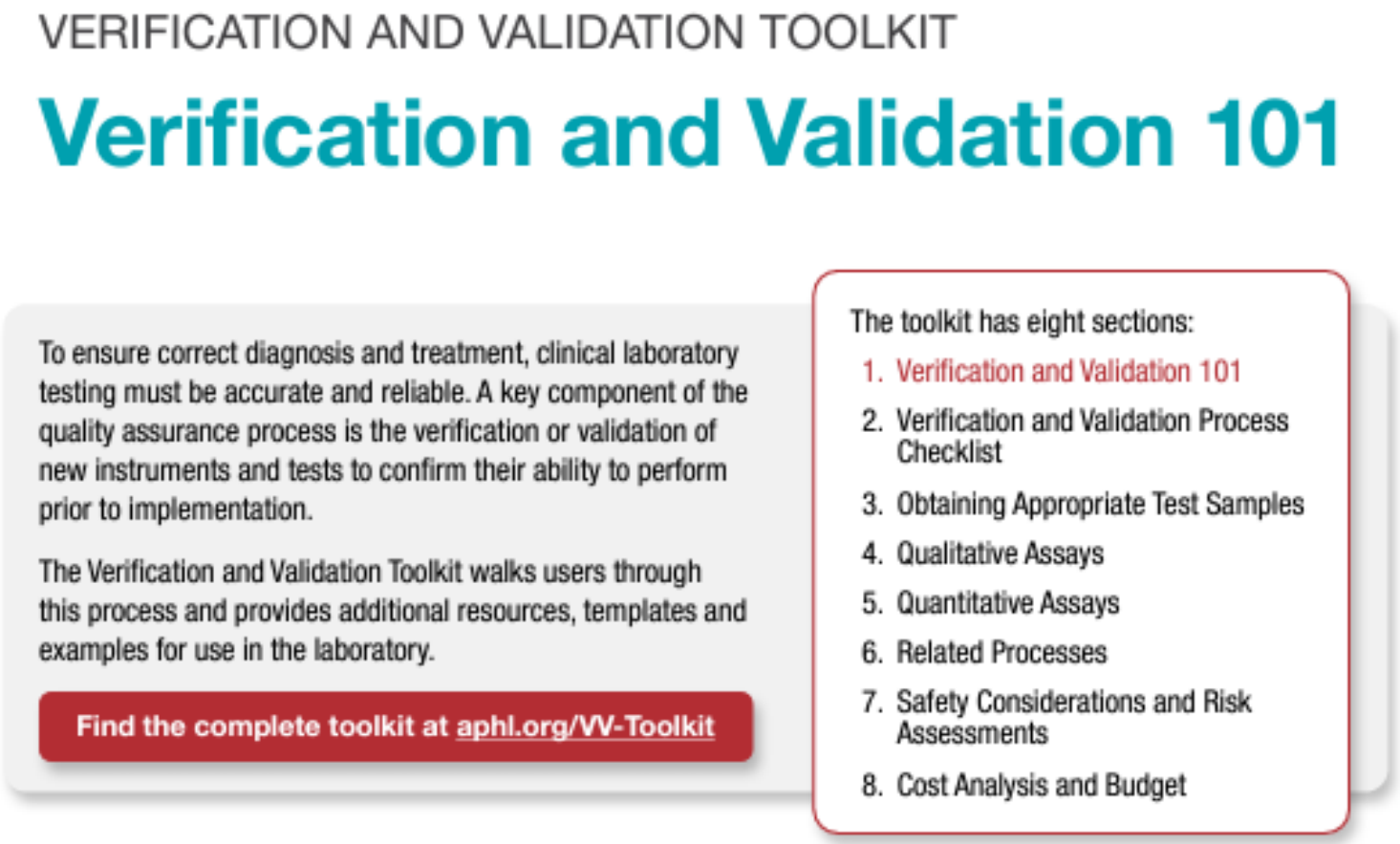
STEP 4

Write SOP for validated method, including sample types, media validated, limitations

STEP 5

Implementation including training and competency

APHL Verification and Validation Toolkit



APHL VV Toolkit

APHL Toolkit Verification/Validation Process Checklist

- Provided to assist in determining whether a verification or validation needs to be performed and the events or steps that should occur for implementation of a new testing method
  - Should a verification or validation be performed?
  - Develop a plan or proposal (reason for study, safety considerations, methodology, acceptance criteria, data analysis and evaluation)
  - Approval of plan by laboratory director
  - Initiate verification or validation plan
  - Completion of testing and summary report creation
  - Completion of associated documents
  - Approval by laboratory director
  - Test implementation

APHL Toolkit Reference Documents

Templates

- Biological Risk Assessment for all New Assays Template
- Breakpoint Implementation Toolkit, CRO 2021 (APHL)
- Breakpoint Implementation Toolkit, MIC 2023 (CLSI)
- Bridging, Addendum and Extension Studies Template
- CLIA-compliant Analytical Method Validation Plan and Template FOR LRN-C Laboratories (APHL)
- Method Verification Validation Plan Approval Checklist
- NGS Method Validation Plan Template
- NGS Method Validation Summary Report Template (NGS Quality Initiative)
- Printable Checklist for the Verification or Validation Process
- Reagent Comparison QC Studies Template
- Software Update SOP and Template
- Verification Plan Template
- Validation Summary Report Template
- Validation Plan Template
- Verification Summary Report Template

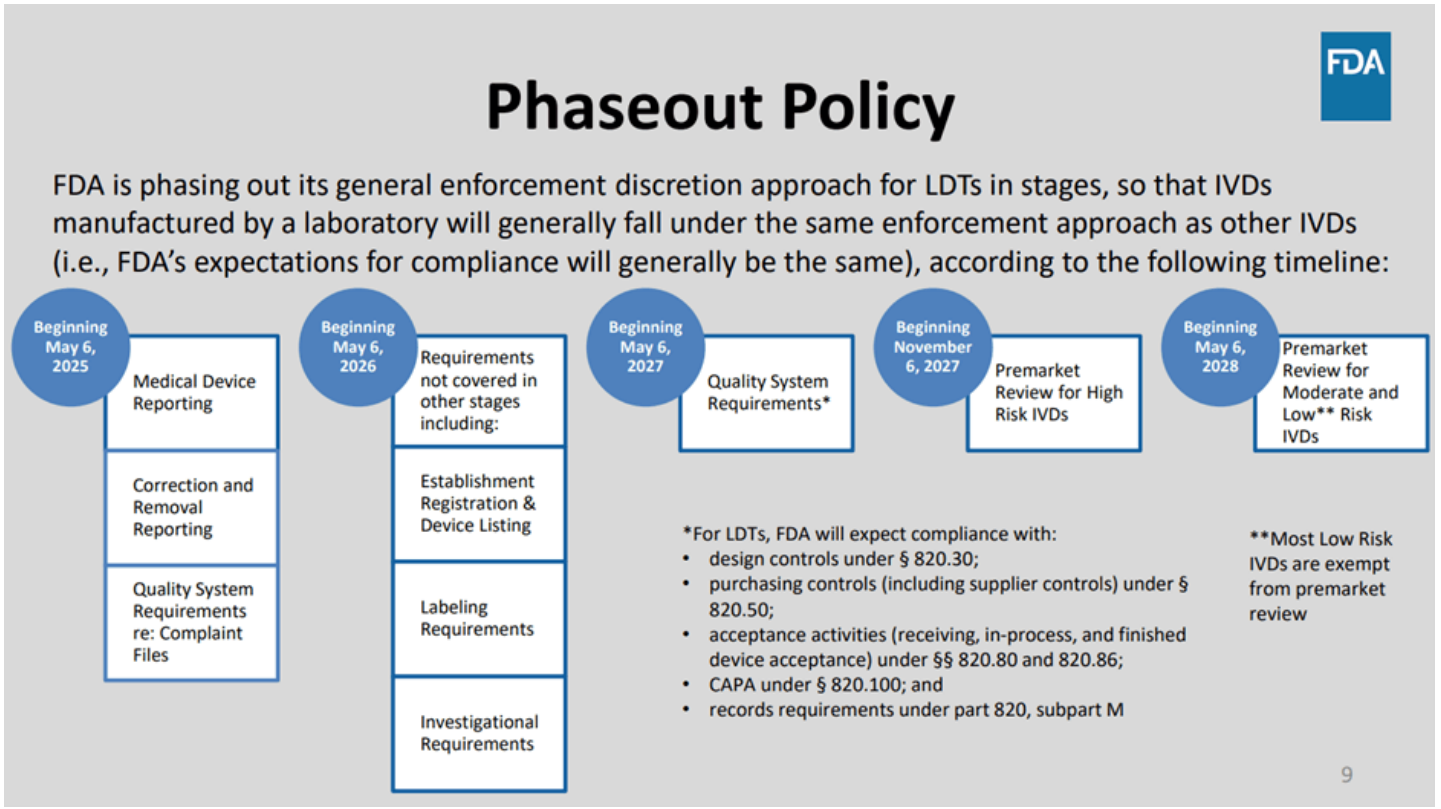
Examples

- Analytical Instrument Verification Example
- Employee Training Verification Checklist Example
- Guidelines for Verification and Validation of Laboratory Methods (Minnesota)
- Method Validation-Verification Summary Report Example (Fairfax County, VA)
- Method Verification Template Example (Indiana)
- Microbiology MALDI-TOF Validation Supplemental Checklist (New York CLEP)
- Microbiology NAAT Checklist (New York CLEP)
- NGS Method Validation SOP Example (NGS Quality Initiative)
- Validation and Verification SOP Example (Texas)
- Validation Plan Example (Washington)
- Validation Summary Report Example (Washington)
- Verification Plan Example (Washington)

LDT Regulation

FDA Final Rule – May 6, 2024

- Phased approach to discontinue enforcement discretion of LDTs
- LDTs will be treated as *in vitro* diagnostic products (IVDs)
- Laboratories (including PHLs) will now be considered manufacturers of IVDs
  - LDTs will require FDA review and approval for use



APHL

APHL LDT Regulation Resources

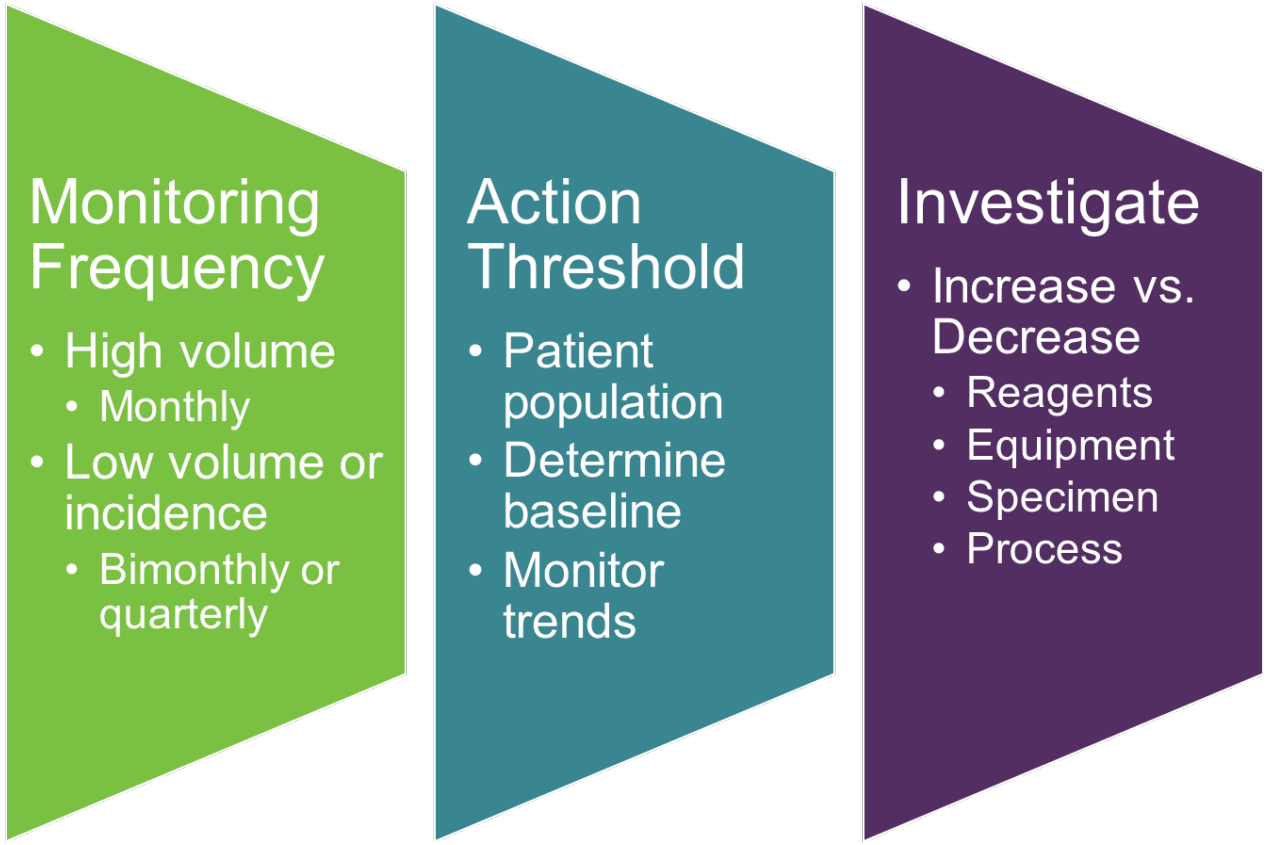
https://www.aphl.org/policy/Pages/LDT-members.aspx

Quality Indicators & Metrics

Quality Indicators for the TB Laboratory

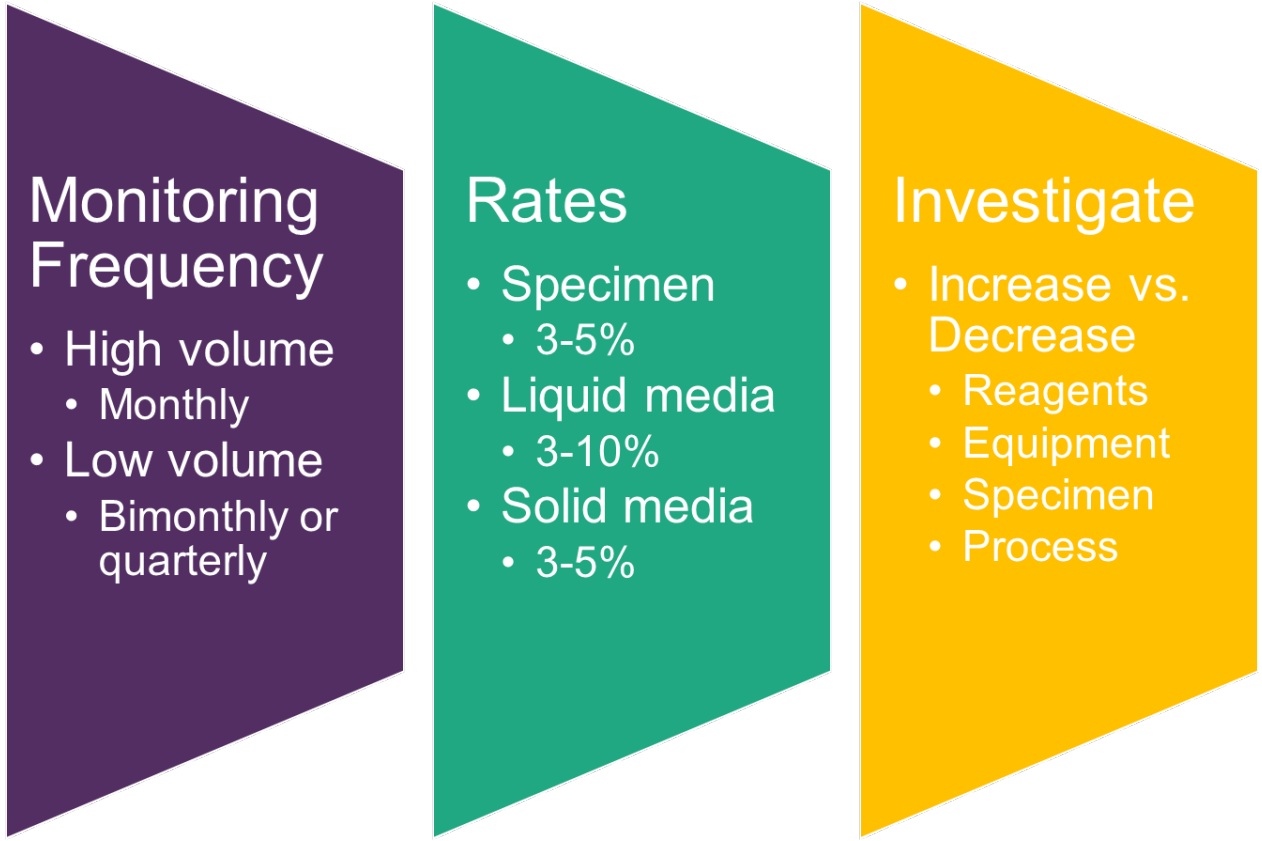
Culture Positivity Rates

- Percent (%) of cultures reported as positive for:
  - All species: # of cultures with positive result / total # of final cultures
  - MTBC: # of cultures positive for MTBC / total # of final cultures
  - NTM: # of cultures positive for NTMs / total # of final cultures



Contamination Rates

- Percent (%) of cultures:
  - Specimen: # of cultures reported as “contaminated” / total # of cultures reported
  - Liquid media: # of inoculated tubes discarded / total # of tubes inoculated
  - Solid media: # of solid media (slants/plates) discarded / total # of solid media inoculated



Smear Status Rates

- Percent (%) of smears reported as positive
  - # of AFB smears reported as positive / total # of AFB smears performed
- Percent (%) of positive cultures that are smear positive (sensitivity)
  - # of smear positive specimens, culture + for MTBC / total # of cultures + for MTBC
  - # of smear positive specimens, culture + for all species / total # of cultures + for all species
- Correlation of smear positive specimens & culture positive specimens (specificity)
  - # of smear positive specimens, culture + for all species / total # of smear positive specimens

Smear Status Rates

Monitoring Frequency

- High volume
  - Monthly
- Low volume or incidence
  - Bimonthly or quarterly

Action Thresholds

- Patient population
- Determine baseline
- Monitor trends
- Specificity 90-98%

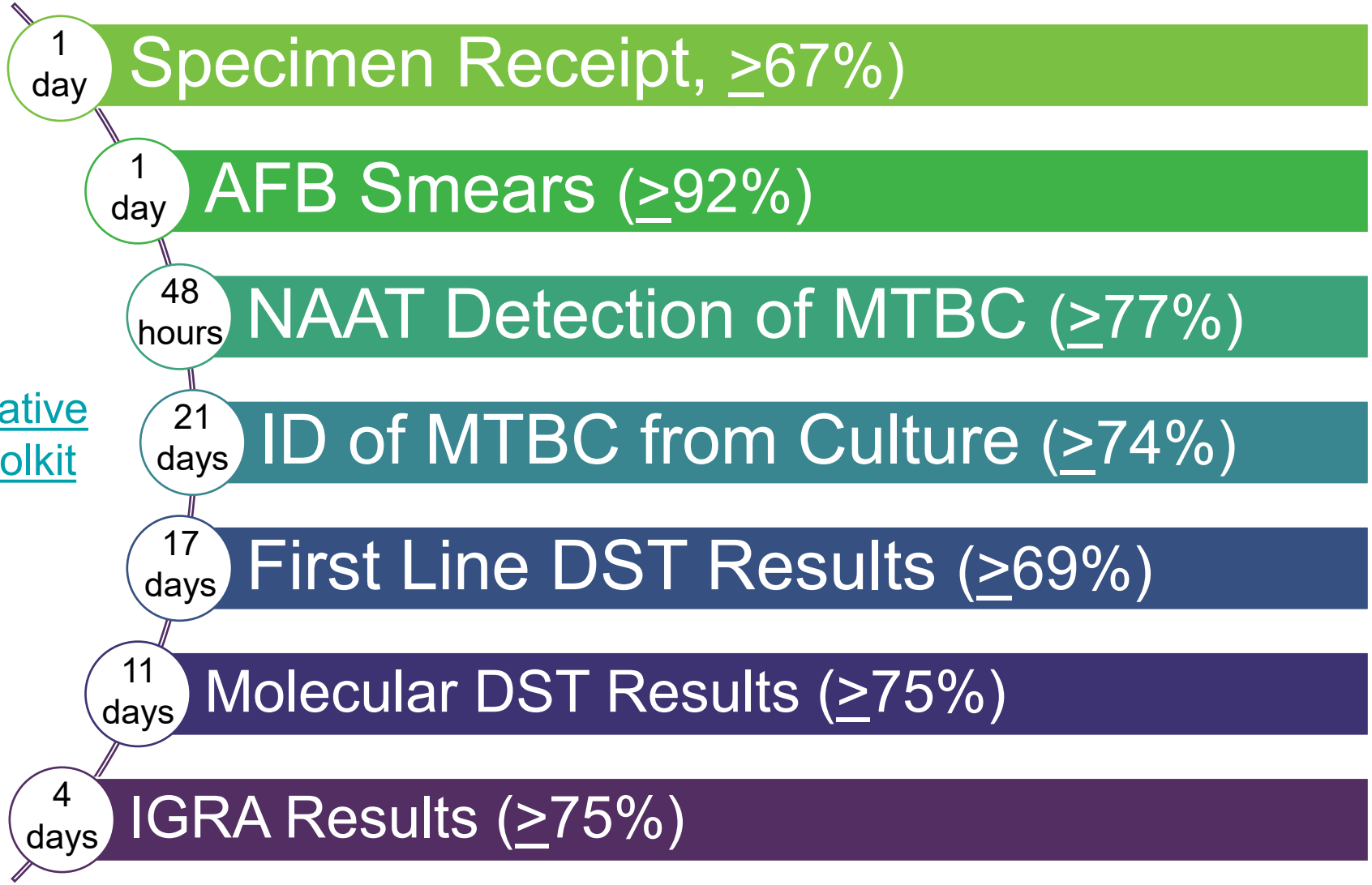
Investigate

- Increase vs. Decrease
  - Reagents
  - Equipment
  - Human error
  - Specimen
  - Treatment status
  - Process

Turn-Around Times (TAT)

- Laboratories should measure their own TAT and develop realistic, laboratory-specific goals
- Monthly monitoring (high volume), bimonthly or quarterly monitoring (low volume)
- Receipt of specimen in the laboratory
  - # of specimens received within TAT / total # of specimens received
- AFB smears
  - # of results reported within TAT / total # of AFB smear results reported
- Detection of MTBC in clinical specimens using NAAT
  - # of NAAT positive patients reported within TAT / total # of NAAT positive patients
- ID of MTBC from culture
  - # of MTBC results reported within TAT / total # of MTBC identifications reported
- First line DST
  - # of DST results reported within TAT / total # of DST results reported

CDC Target Turn-Around Times (TAT)



[APHL Cooperative Agreement Toolkit]

Drug Resistance Rates

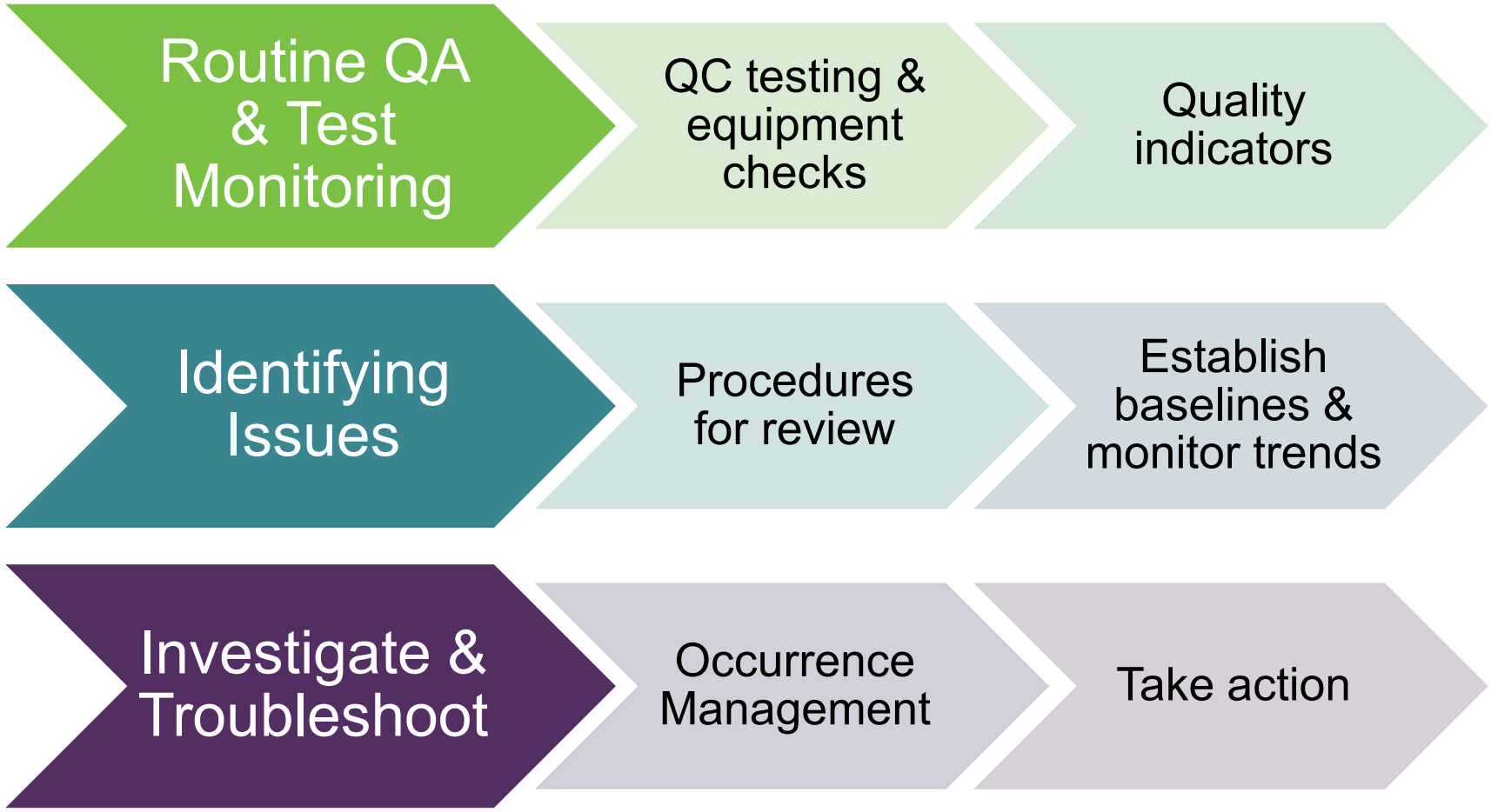
- First Line Drugs (RIF, INH, EMB, PZA)
- # of resistant isolates / total # isolates set up for DST
- Monitor monthly (high volume), bimonthly or quarterly (low volume)
- Action threshold is dependent on patient population/area
  - Determine baseline and monitor trends
- Investigate
  - Mixed cultures
  - Reagents, equipment, human error

PT Performance

- Thresholds vary by provider
  - Usually, 80% correct
- Some labs set their own internal requirements
- # of correct results / # of challenges
- Monitor with each challenge and trends year to year
- Investigate
  - Human error
  - Reagents
  - PT materials

Quality Investigations & Troubleshooting

Monitoring for Quality Issues



Investigate & Troubleshoot

Common Quality Issues

- Reagents & Equipment
  - QC failures (PZA)
  - Calibration issues & errors
- Specimen Submission/Handling
  - Collection & labeling
  - Transport conditions
- Testing Personnel
  - Human error
  - Competency lapse
- Test System & Quality Management

Steps to Investigate & Troubleshoot

- One Off vs. Repeat Issue
- Occurrence Management
  - Probability
    - Consider volume & frequency of testing
  - Severity
    - Consider the harm associated with issue
  - Recommended Course of Action
    - Corrective Action
    - Correction
    - No Action
  - Documentation of investigation & action

Occurrence Management

Recommended Course of Action			
	High severity	Moderate severity	Low severity/Unlikely Risk
High probability	Corrective action	Corrective action	Corrective action / Correction
Moderate probability	Corrective action	Corrective action / Correction	Correction / No action
Low/Rare probability	Corrective action / Correction	Correction	No action

Questions?

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