Clinical Laboratory Improvement Advisory Committee

FDA White Oak Campus

Tuesday, April 10, 2018

Implementation of Next-Generation Sequencing in Clinical and Public Health Laboratories

Background/Introduction

Ira M. Lubin, PhD, FACMG

Geneticist

Quality and Safety Systems Branch

Implementation of Next Generation Sequencing in Clinical Laboratories

- Background / Introduction — Ira M. Lubin, PhD, FACMG
- Public Health Perspective — Rebecca Hutchins, MS, MBA
- Clinical Laboratory Perspective — John D. Pfeifer, MD, PhD
- CLIAC Discussion

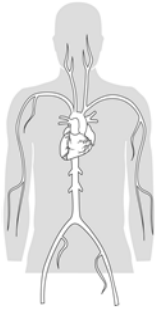
Background / Introduction

- Next-generation sequencing in clinical and public health applications
- Integrating next-generation sequencing into clinical laboratory testing
- Practice and regulatory framework for assuring the quality of clinical next-generation sequencing
- Proposed CLIAC Workgroup
- Questions for CLIAC

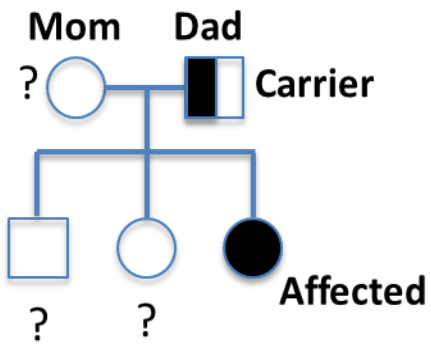
Next-Generation Sequencing in Clinical and Public Health Applications

Pediatrics:

- Rare Disease
- Developmental Disabilities

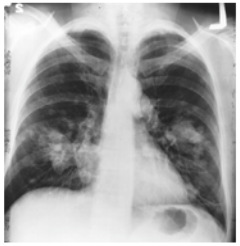


Chronic Disease



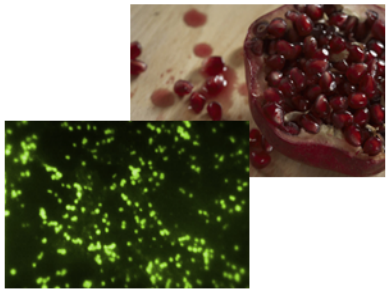
Screening:

- Carrier Screening



Cancer

Transplant



Pathogens:

- Identification
- Outbreak response
- Antibiotic resistance

What is the Landscape of Next-Generation Sequencing Offerings in the United States?

New York Clinical Laboratory Evaluation Program
https://www.wadsworth.org/regulatory/clep/approved-ldt

81 Laboratories
(searched 3/23/2018)

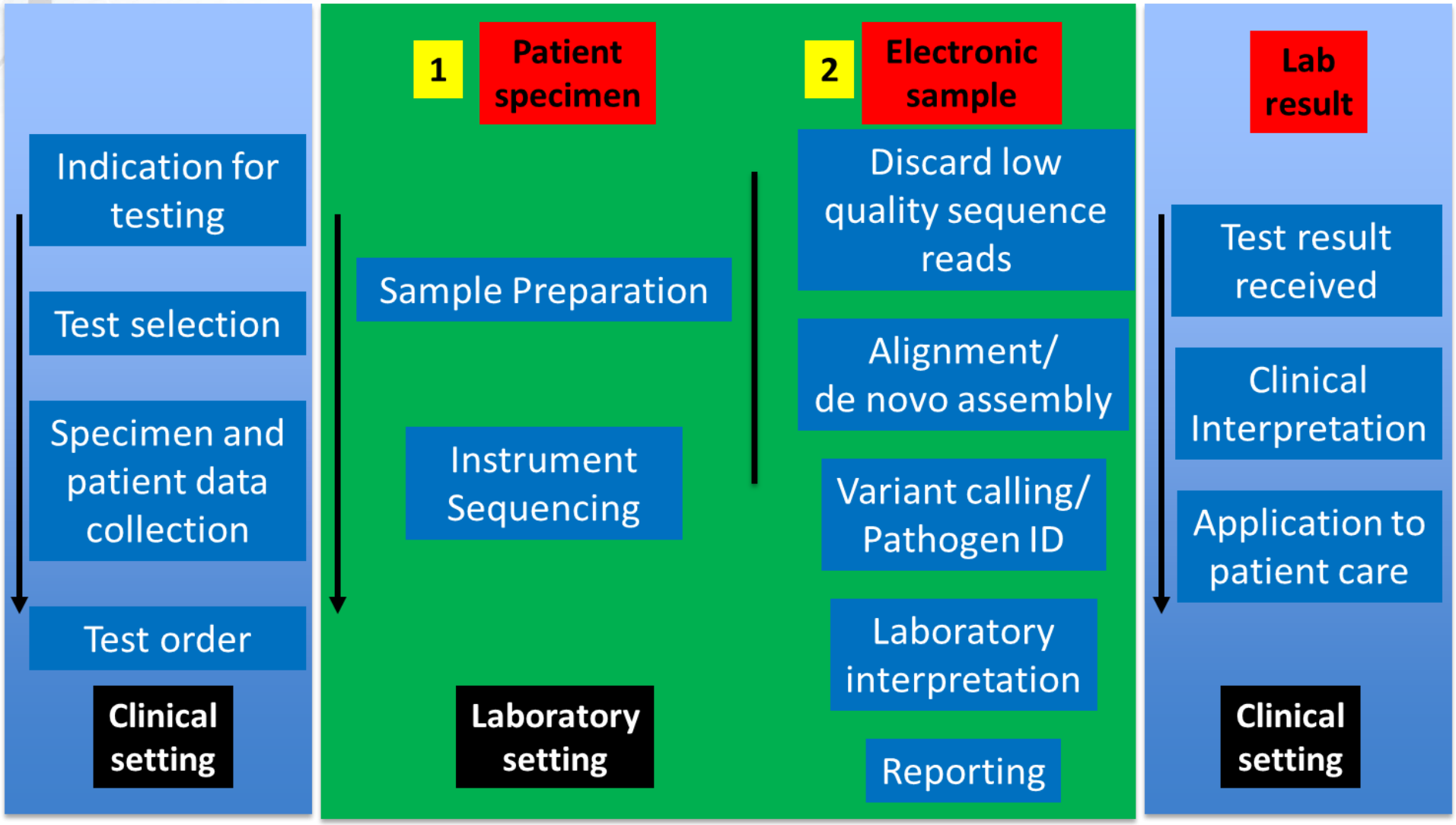
College of American Pathologists Accreditation Data (per organizational contact)

331 laboratories
(provided 3/16/2018)

Genetic Testing Registry
https://www.ncbi.nlm.nih.gov/gtr/

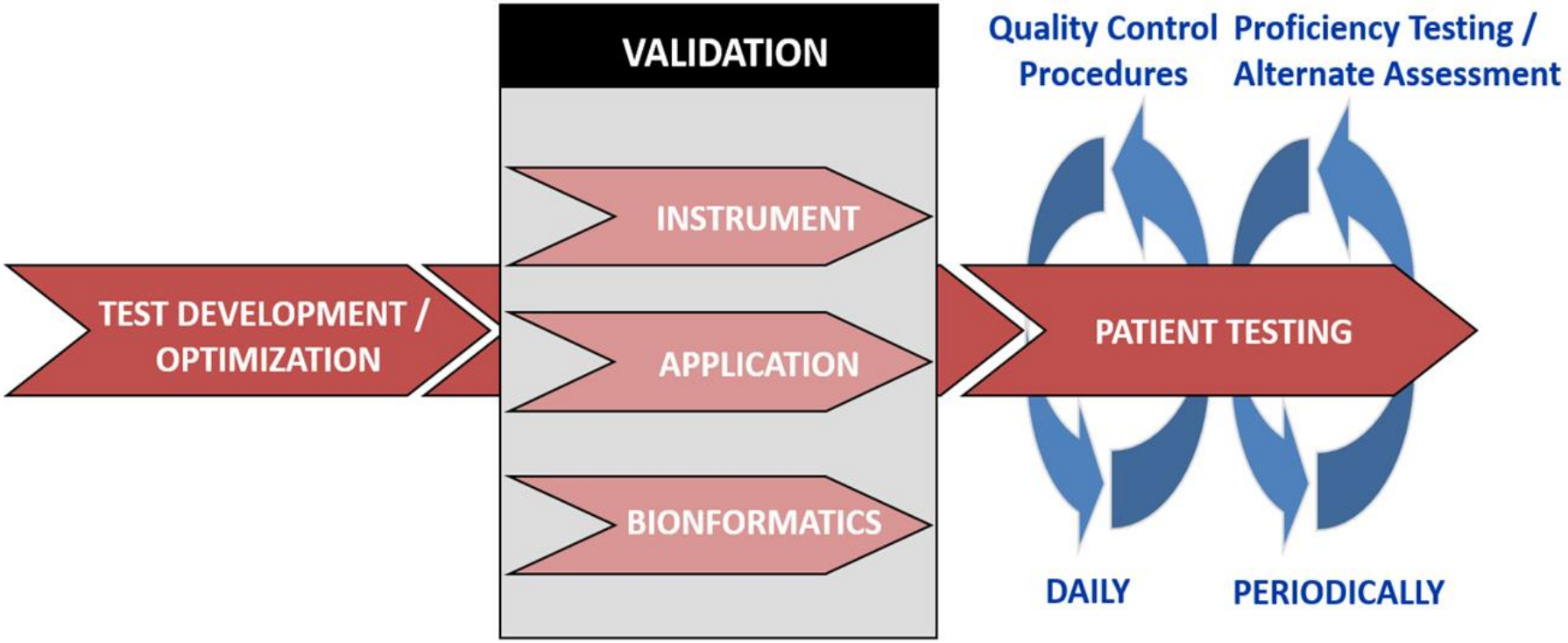
80 Laboratories
(Provided 3/12/2018)

Workflow: Next Generation Sequencing



Framework for the Implementation of Clinical NGS Testing

As of 2018, the majority of clinical NGS tests are developed within the laboratory that they are offered.



Gargis et al., Nat Biotechnol. 2012;30:1033

Practice and Regulatory Framework for Assuring the Quality of Clinical Next-Generation Sequencing

Applying the CLIA regulations to clinical next-generation sequencing is challenging due to the novelty and complexity of the technology, and new paradigms for data analyses

- **Code of Federal Regulations**. **The Clinical Laboratory Improvement Amendments (CLIA).** 42 CFR Part 493 sec. 1256.
- **Interpretive guidelines***: https://www.cms.gov/Regulations-and-Guidance/Guidance/Manuals/Downloads/som107ap_c_lab.pdf*

Applying CLIA to Next-Generation Sequencing in the Clinical Setting

- **Test validation** ***(CLIA general requirements at §493.1253)***
- **Quality control procedures** ***(CLIA general requirements at §493.1256; unidirectional workflow for molecular amplification at §493.1101 (a)(3))***
  - **Quality control materials (physical and electronic)?**
  - **Use of external data to support analyses?**
- **Test ordering** ***(CLIA general requirements at §493.1241)***
- **Result Reporting** ***(CLIA general requirements at §493.1291)***
- **Proficiency testing** ***(CLIA does not specify PT requirements for NGS, so alternative performance assessment requirements at §493.1236(c) apply)***
- **Personnel competencies** **(Subpart M; No specific requirements)**
- **Bioinformatics pipeline quality assessment** **(No specific requirements)**

Other issues:

- **Distributive testing – single test / multiple sites**
- **Reporting of secondary findings**
- **Reanalysis**

Next Generation Sequencing: Standardization of Clinical Testing

Assuring the quality of next-generation sequencing in clinical laboratory practice

To the Editor:
We direct your readers' attention to the principles and guidelines (**Supplementary Guidelines**) developed by the Next-generation Sequencing: Standardization of Clinical Testing (Nex-StoCT) workgroup. These guidelines represent initial steps to ensure that results from tests based on next-generation sequencing (NGS) are reliable and useful for clinical decision making. The US Centers for Disease Control and Prevention (CDC) convened this national workgroup, which collaborated to define platform-independent approaches for establishing technical process elements of a quality management system (QMS) to assure the analytical validity and compliance of NGS tests with existing regulatory and professional quality standards. The workgroup identified and addressed gaps in

The workgroup recommendations are summarized in **Table 1**. Although the workgroup focused on detection of DNA sequence variations associated with heritable human disorders, many of the principles and recommendations described are also relevant to the application of NGS to other areas of laboratory medicine, including the diagnosis, prognosis and treatment of cancer and infectious-disease testing.

Validation is the process of establishing analytical performance specifications for a clinical test system developed in house to confirm that the system is suitable for its intended use[1]. During the validation process, the laboratory must demonstrate that the assay functions as expected and provides

Table 1 Selected workgroup recommendations for establishing NGS test s clinical use

Requirements for test establishment	Objective	NGS-specific recommendations[a]
Validation	Document reliability of the platform, test, and informatics pipeline before testing of patient specimens	•Platform validation: establish that the system pro sequence analysis across the genomic regions tar •Test validation: establish that the system correct disease-associated (and other) variants in target genome (**Supplementary Guidelines**, section 4). •Informatics pipeline validation: establish that th reliably analyze platform data to produce an acc

Gargis et. al. Nat. Biotechnol. 2012; 30:1033-1036+ Supplemental

Principles and Recommendations for Standardizing the Use of the Next-Generation Sequencing Variant File in Clinical Settings

Ira M. Lubin,* Nazneen Aziz,[†‡] Lawrence J. Babb,[§] Dennis Ballinger,[¶] Himani Bisht,[‖] Deanna M. Church,[**††] Shaun Cordes,[¶] Karen Eilbeck,[‡‡] Fiona Hyland,[§§] Lisa Kalman,* Melissa Landrum,[††] Edward R. Lockhart,* Donna Maglott,[††] Gabor Marth,[¶¶] John D. Pfeifer,[‖‖] Heidi L. Rehm,[***] Somak Roy,[†††] Zivana Tezak,[‖] Rebecca Truty,[¶] Mollie Ullman-Cullere,[‡‡‡] Karl V. Voelkerding,[§§§] Elizabeth Worthey,[¶¶¶] Alexander W. Zaranek,[‖‖‖****] and Justin M. Zook[††††]

Lubin IM et al. J Mol Diagn. 2017;19:417-426

Good laboratory practice for clinical next-generation sequencing informatics pipelines

To the Editor:
We report principles and guidelines (**Supplementary Note**) that were developed by the Next-Generation Sequencing: Standardization of Clinical Testing II (Nex-StoCT II) informatics workgroup, which was first convened on October 11–12, 2012, in Atlanta, Georgia, by the US Centers for Disease Control and Prevention (CDC; Atlanta, GA). We present recommendations are summarized in **Table 1**, and detailed in the guidelines presented in the **Supplementary Note**.

Currently, most clinical NGS tests are offered as laboratory-developed tests (LDTs), which are tests designed, manufactured and used within a single laboratory. These tests use commercially available sequencing platforms to generate raw sequence data that are subsequently

Gargis et al. Nat. Biotechnol. 2015;33: 689-693 + Supplemental

CDC facilitated the development of the first consensus guideline for clinical next-generation sequencing.

The Food and Drug Administration: Draft Guidance for Next-Generation Sequencing

Contains Nonbinding Recommendations

Draft - Not for Implementation

Infectious Disease Next Generation Sequencing Based Diagnostic Devices: Microbial Identification and Detection of Antimicrobial Resistance and Virulence Markers

Draft Guidance for Industry and Food and Drug Administration Staff

DRAFT GUIDANCE

This draft guidance document is being distributed for comment purposes only.

Document issued on: May 13, 2016

Contains Nonbinding Recommendations
Draft – Not For Implementation

Use of Standards in FDA Regulatory Oversight of Next Generation Sequencing (NGS)-Based In Vitro Diagnostics (IVDs) Used for Diagnosing Germline Diseases

Draft Guidance for Stakeholders and Food and Drug Administration Staff

DRAFT GUIDANCE

This draft guidance document is being distributed for comment purposes only.

Document issued on July 8, 2016.

You should submit comments and suggestions regarding this draft document within 90 days of

Contains Nonbinding Recommendations
Draft – Not For Implementation

Use of Public Human Genetic Variant Databases to Support Clinical Validity for Next Generation Sequencing (NGS)-Based In Vitro Diagnostics

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You should submit comments and suggestions regarding this draft document within 90 days of publication in the *Federal Register* of the notice announcing the availability of the draft guidance. Submit electronic comments to http://www.regulations.gov. Submit written

A Sampling of Professional Guidance

CLINICAL AND LABORATORY STANDARDS INSTITUTE

February 2014

MM09-A2

Nucleic Acid Sequencing Methods in Diagnostic Laboratory Medicine; Approved Guideline—Second Edition

ACMG PRACTICE GUIDELINES | Genetics inMedicine

ACMG clinical laboratory standards for next-generation sequencing

Heidi L. Rehm, PhD[1,2], Sherri J. Bale, PhD[3], Pinar Bayrak-Toydemir, MD, PhD[4], Jonathan S. Berg, MD[5], Kerry K. Brown, PhD[6], Joshua L. Deignan, PhD[7], Michael J. Friez, PhD[8], Birgit H. Funke, PhD[1,2], Madhuri R. Hegde, PhD[9] and Elaine Lyon, PhD[5]; for the Working Group of the American College of Medical Genetics and Genomics Laboratory Quality Assurance Committee

European Journal of Human Genetics (2016) 24, 2–5

EJHG Open

POLICY

Guidelines for diagnostic next-generation sequencing

Gert Matthijs[1,8], Erika Souche[1,8], Marielle Alders[2], Anniek Corveleyn[1], Sebastian Eck[3], Ilse Feenstra[4], Valérie Race[5], Erik Sistermans[3], Marc Sturm[6], Marjan Weiss[3], Helger Yntema[4], Egbert Bakker[7], Hans Scheffer[4] and Peter Bauer[6]

We present, on behalf of EuroGentest and the European Society of Human Genetics, guidelines for the evaluation and validation of next-generation sequencing (NGS) applications for the diagnosis of genetic disorders. The work was performed by a group of laboratory geneticists and bioinformaticians, and discussed with clinical geneticists, industry and patients' representatives, and other stakeholders in the field of human genetics. The statements that were written during the elaboration of the guidelines are presented here. The background document and full guidelines are available as supplementary material. They include many examples to assist the laboratories in the implementation of NGS and accreditation of this service. The work and ideas presented by others in guidelines that have emerged elsewhere in the course of the past few years were also considered and are acknowledged in the full text. Interestingly, a few new insights that have not been cited before have emerged during the preparation of the guidelines. The most important new feature is the presentation of a 'rating system' for NGS-based diagnostic tests. The guidelines and statements have been applauded by the genetic diagnostic community, and thus seem to be valuable for the harmonization and quality assurance of NGS diagnostics in Europe.

European Journal of Human Genetics (2016) 24, 2–5; doi:10.1038/ejhg.2015.226; published online 28 October 2015

The Journal of Molecular Diagnostics, Vol. 17, No. 6, November 2015

ELSEVIER

SPECIAL ARTICLE

Next-Generation Sequencing for Infectious Disease Diagnosis and Management

A Report of the Association for Molecular Patholog

Martina I. Lefterova,[*,†] Carlos J. Suarez,[*,†] Niraz Banaei,[*,‡] and Benjamin A. Pinsky[*,‡]

Applications of Clinical Microbial Next-Generation Sequencing

A report from the American Academy of Microbiology

CAP Laboratory Improvement Programs

College of American Pathologists' Laboratory Standards for Next-Generation Sequencing Clinical Tests

Nazneen Aziz, PhD; Qin Zhao, PhD; Lynn Bry, MD, PhD; Denise K. Driscoll, MS, MT(ASCP)SBB; Birgit Funke, PhD; Jane S. Gibson, PhD; Wayne W. Grody, MD; Madhuri R. Hegde, PhD; Gerald A. Hoeltge, MD; Debra G. B. Leonard, MD, PhD; Jason D. Merker, MD, PhD; Rakesh Nagarajan, MD, PhD; Linda A. Palicki, MT(ASCP); Ryan S. Robetorye, MD; Iris Schrijver, MD; Karen E. Weck, MD; Karl V. Voelkerding, MD

STATE OF THE ART

Next Generation Sequencing Implementation Guide

Best practices for evaluating single nucleotide variant calling methods for microbial genomics

Nathan D. Olson[1*], Steven P. Lund[1], Rebecca E. Colman[1], Jeffrey T. Foster[4†], Jason W. Sahl[1,4], James M. Schupp[5], Paul Keim[1,4], Jayne B. Morrow[1], Marc L. Salit[1,5] and Justin M. Zook[1]

SPECIAL ARTICLE

Guidelines for Validation of Next-Generation Sequencing—Based Oncology Panels

A Joint Consensus Recommendation of the Association for Molecular Pathology and College of American Pathologists

NEW YORK STATE OF OPPORTUNITY. Department of Health

ANDREW M. CUOMO, Governor | HOWARD A. ZUCKER, M.D., J.D., Commissioner | SALLY DRESLIN, M.S., R.N., Executive Deputy Commissioner

APHL ASSOCIATION OF PUBLIC HEALTH LABORATORIES

UPDATED AND REVISED — March 2016

Oncology – Molecular and Cellular Tumor Markers

"Next Generation" Sequencing (NGS) guidelines for somatic genetic variant detection

The following describes requirements for the development of procedures and the establishment of performance (validation) of assays for the detection of somatic genetic variants by Next

Summary

- **Next generation sequencing is continuing to integrate into diverse medical disciplines.**
- **Bioinformatics (computational analysis) comprises a significant component of the test.**
- **Communication between the laboratory professional (s) and clinician(s) is vital toward understanding of the available testing, limitations, and test findings**
- **Clinical NGS testing is covered under CLIA but application of the laboratory requirements may not be sufficiently defined.**

Proposed CLIAC NGS Workgroup

Proposed Charge

Provide input to CLIAC for consideration in developing recommendations to CDC, CMS, and FDA for assuring the quality of next generation sequencing in clinical laboratory settings

Proposed Workgroup Tasks

- **Identify challenges in applying the existing regulatory framework**
- **Identify challenges and gaps in guidance**
- **Consider and suggest strategies to address the identified gaps and challenges**
- **Consider and suggest strategies for assuring workforce competency**

Questions for CLIAC

1. What major gaps and issues do laboratories experience when implementing NGS-based testing for various applications?

2. What guidance is lacking for laboratories that perform NGS testing to address critical steps in the total testing process?

3. How can CDC, CMS, and FDA assist in filling NGS-related gaps and providing guidance to laboratories?

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For more information please contact Centers for Disease Control and Prevention

1600 Clifton Road NE, Atlanta, GA 30333
Telephone: 1-800-CDC-INFO (232-4636)/TTY: 1-888-232-6348
E-mail: cdcinfo@cdc.gov | Web: www.cdc.gov

The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the Centers for Disease Control and Prevention.