# 临床分子诊断实验室的质量管理

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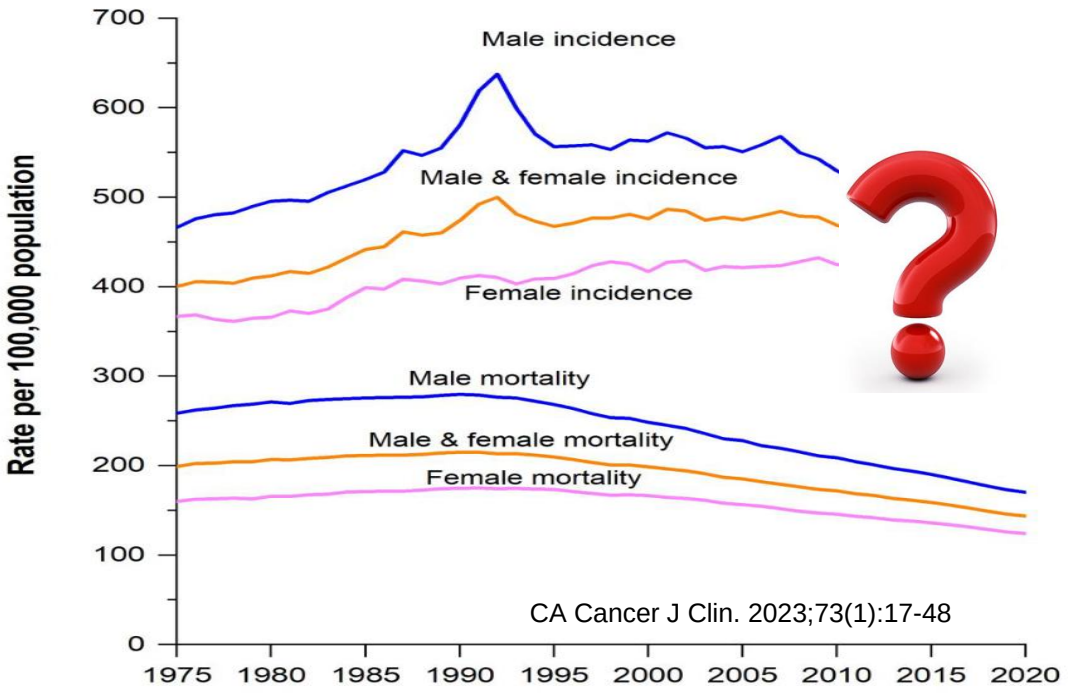
# 基因检测与肿瘤风险评估

| Males | | | Females | | |
|---|---|---|---|---|---|
| Prostate | 288,300 | 29% | Breast | 297,790 | 31% |
| Lung & bronchus | 117,550 | 12% | Lung & bronchus | 120,790 | 13% |
| Colon & rectum | 81,860 | 8% | Colon & rectum | 71,160 | 8% |
| Urinary bladder | 62,420 | 6% | Uterine corpus | 66,200 | 7% |
| Melanoma of the skin | 58,120 | 6% | Melanoma of the skin | 39,490 | 4% |
| Kidney & renal pelvis | 52,360 | 5% | Non-Hodgkin lymphoma | 35,670 | 4% |
| Non-Hodgkin lymphoma | 44,880 | 4% | Thyroid | 31,180 | 3% |
| Oral cavity & pharynx | 39,290 | 4% | Pancreas | 30,920 | 3% |
| Leukemia | 35,670 | 4% | Kidney & renal pelvis | 29,440 | 3% |
| Pancreas | 33,130 | 3% | Leukemia | 23,940 | 3% |
| All Sites | 1,010,310 | 100% | All Sites | 948,000 | 100% |




CA Cancer J Clin. 2023;73(1):17-48

## Detection, Prevention, and Risk Reduction

NCCN Clinical Practice Guidelines in Oncology (NCCN Guidelines®) are posted with the latest update date and version number.

**Breast Cancer Risk Reduction**
Version: 2.2024

**Breast Cancer Screening and Diagnosis**
Version: 2.2024

**Colorectal Cancer Screening**
Version: 1.2024

**Genetic/Familial High-Risk Assessment: Breast, Ovarian, and Pancreatic**
Version: 3.2024

**Genetic/Familial High-Risk Assessment: Colorectal**
Version: 2.2023

**Lung Cancer Screening**
Version: 2.2024

**Prostate Cancer Early Detection**
Version: 2.2024

https://www.nccn.org/

| Gene | Breast Cancer Risk and Management (First primary) | Epithelial Ovarian Cancer Risk and Management | Pancreatic Cancer Risk and Management[13-22] and Other Cancer Risks |
|---|---|---|---|
| *BRCA1* | • **Absolute risk:** >60%[5,25-29]<br>• **Management:** See *BRCA* Pathogenic Variant-Positive Management<br>• **Strength of evidence of association with cancer:** Very strong (with predisposition to triple-negative disease)<br><br>**Male breast cancer**<br>• **Absolute risk:** 0.2%–1.2% by age 70 y[30,31]<br>• **Strength of evidence of association with cancer:** Strong | • **Absolute risk:** 39%–58%[33]<br>• **Management:** See *BRCA* Pathogenic Variant-Positive Management<br>• **Strength of evidence of association with cancer:** Very strong | **Pancreatic cancer**<br>• **Absolute risk:** ≤5%[31]<br>• **Management:** Screen P/LP variant carriers with a family history of pancreatic cancer, see PANC-A.<br>• **Strength of evidence of association with cancer:** Strong<br><br>**Prostate cancer**<br>• **Absolute risk:** 7%–26%[34]<br>• **Management:** See *BRCA* Pathogenic Variant-Positive Management |
| | Comment: See GENE-B for reproductive implications/ recessive disease. | | |
| *BRCA2* | • **Absolute risk:** >60%[5,21-25]<br>• **Management:** See *BRCA* Pathogenic Variant-Positive Management<br>• **Strength of evidence of association with cancer:** Very strong<br><br>**Male breast cancer**<br>• **Absolute risk:** 1.8%–7.1% by age 70 y[30,31,32]<br>• **Strength of evidence of association with cancer:** Strong | • **Absolute risk:** 13%–29%[33]<br>• **Management:** See *BRCA* Pathogenic Variant-Positive Management<br>• **Strength of evidence of association with cancer:** Very strong | **Pancreatic cancer**<br>• **Absolute risk:** 5%–10%[31]<br>• **Management:** Screen P/LP variant carriers with a family history of pancreatic cancer, see PANC-A.<br>• **Strength of evidence of association with cancer:** Very strong<br><br>**Prostate cancer**<br>• **Absolute risk:** 19%–61%[34,35]<br>• **Management:** See *BRCA* Pathogenic Variant-Positive Management<br><br>**Melanoma**<br>• See *BRCA* Pathogenic Variant-Positive Management |
| | Comment: See GENE-B for reproductive implications/ recessive disease. | | |

# 肺癌的筛查与风险评估

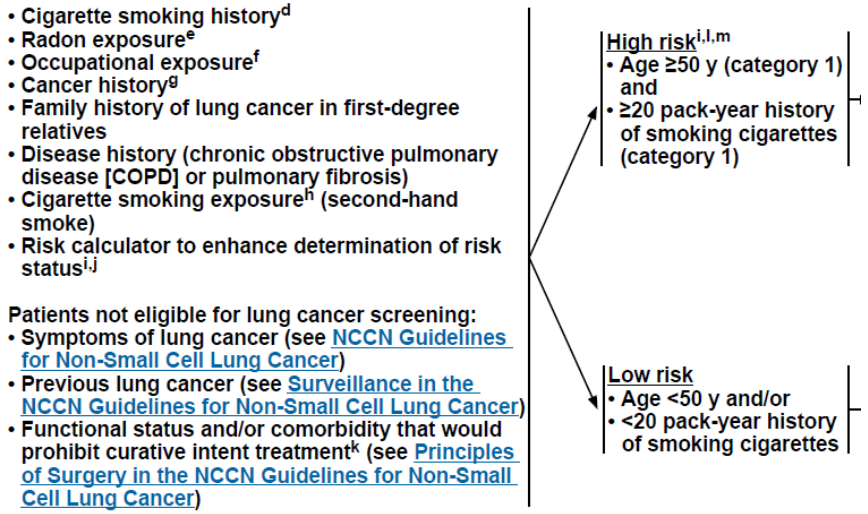
- 年龄≥50岁
- 具有以下任一危险因素者:

**高危人群**

  - (1)吸烟

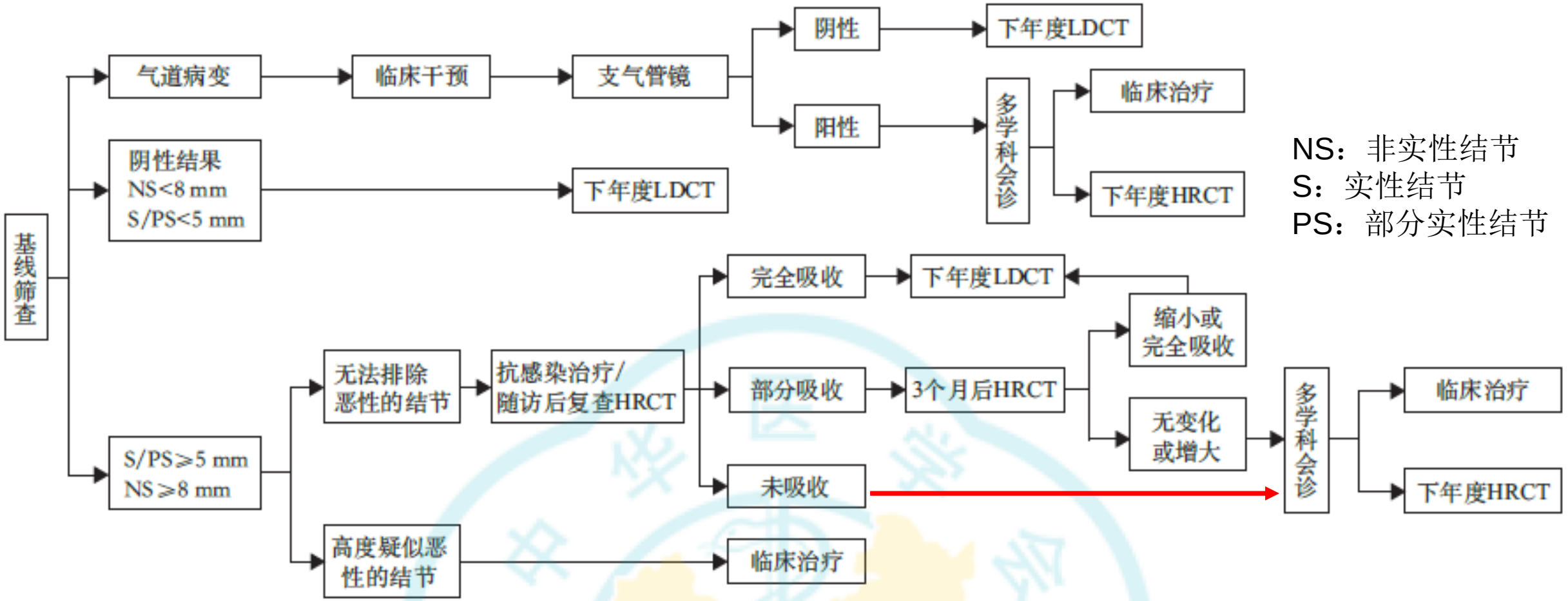
**筛查与戒烟结合**

    - ✓ ≥400年支(或20包年),
    - ✓ 或曾经吸烟≥400年支(或20包年),戒烟时间<15年
  - (2)有环境或高危职业暴露史(如石棉、铍、铀、氡等接触者)
  - (3)合并慢性阻塞性肺气肿、弥漫性肺纤维化或既往有肺结核病史者
  - (4)既往罹患恶性肿瘤或有肺癌家族史者,尤其一级亲属家族史
  - (5) 此外,还需考虑被动吸烟、烹饪油烟以及空气污染等因素




https://brocku.ca/lung-cancer-screening-and-risk-prediction/risk-calculators/?

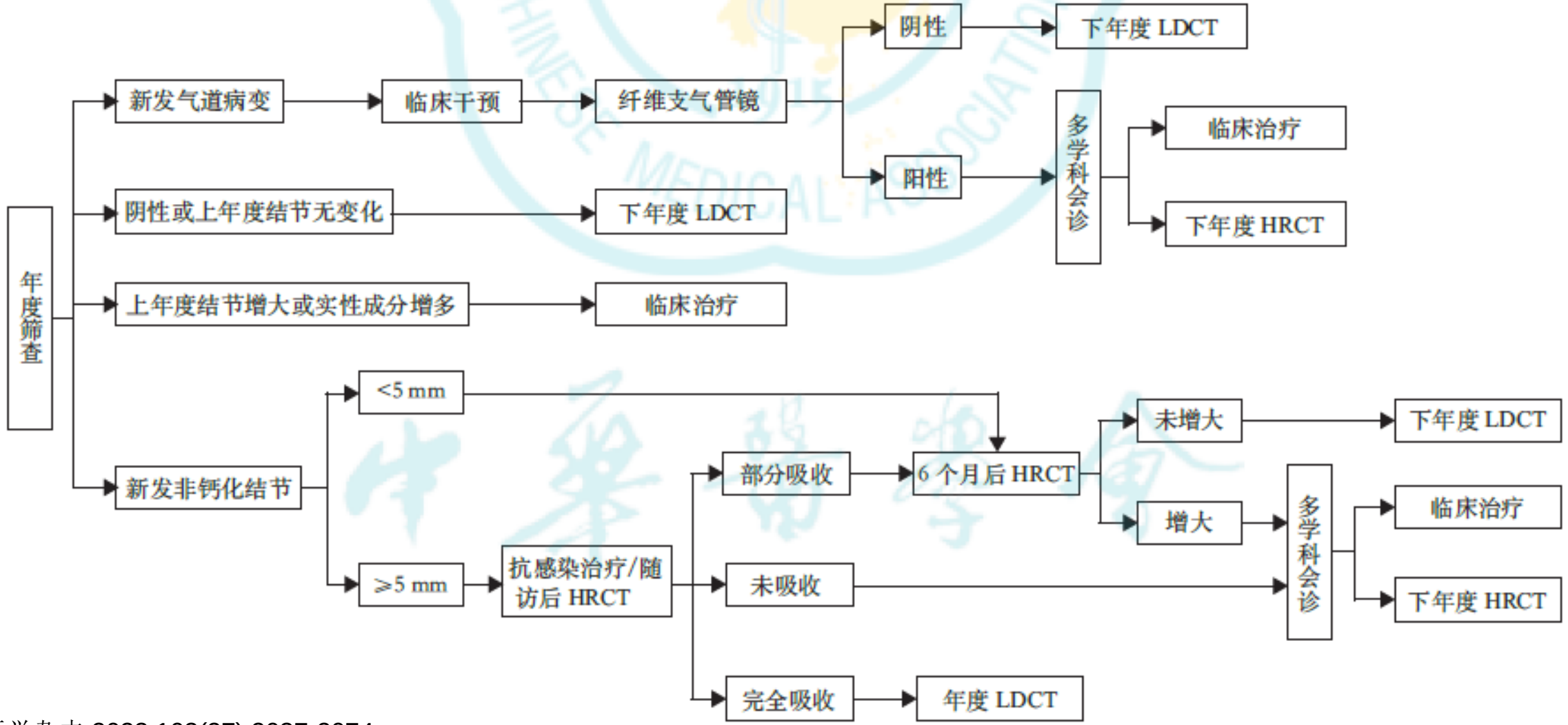
# 基线筛查流程及结节管理




NS：非实性结节
S：实性结节
PS：部分实性结节

中华医学杂志,2023,103(27):2037-2074

# 年度筛查流程及结节管理




中华医学杂志,2023,103(27):2037-2074

# 如何选择筛查指标

## 2A类推荐证据

4.个人肿瘤史：既往罹患其他恶性肿瘤者可能携带异常基因突变，基因突变可增加肺癌的发病风险[25]。对于肺癌基因筛查的研究仍在进行中[26]。

5.一二级亲属肺癌家族史：一级亲属被诊断为肺癌的个体患肺癌的风险明显升高[27]。有肺癌家族史的人群可能存在可遗传的肺癌易感位点[28]。

| 推荐类别 | 循证医学证据级别 |
| --- | --- |
| 1类推荐证据 | 级别最高，专家组一致推荐 |
| 2A类推荐证据 | 级别稍低，专家组一致推荐 |
| 2B类推荐证据 | 级别低，部分专家推荐 |
| 3类推荐证据 | 专家分歧较大 |

## 3类推荐证据

对于可疑的气道病变，建议采用支气管镜进一步检查。对于重度吸烟的患者，条件允许的情况下，可行荧光支气管镜检查[36]。人工智能辅助技术可降低CT影像读片的压力，并在一定程度上提高肺部结节诊断的准确性[37]。通过外周血循环肿瘤细胞、外泌体、自身抗体、肿瘤游离DNA、微小RNA（microRNA）等手段进行肺癌筛查的方法仍在探索中。辅助检测手段和LDCT筛查的联合应用可在一定程度上提高筛查的效果[38-41]。

中华医学杂志,2023,103(27):2037-2074

# 家族性癌症易感综合征

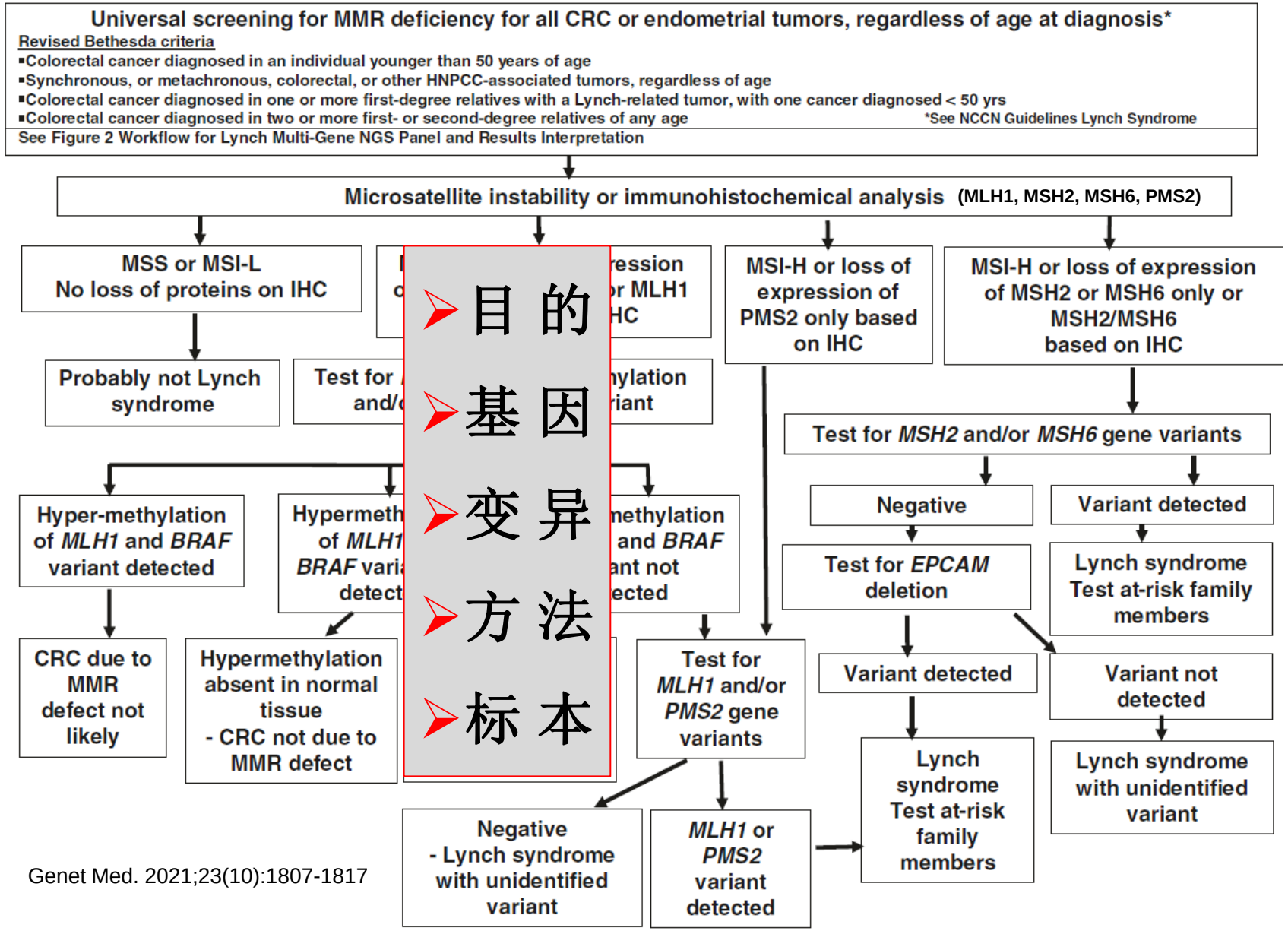
https://www.cancer.gov/publications/pdq/information-summaries/genetics/overview-hp-pdq#_123

## Familial Cancer Susceptibility Syndromes

| | | |
|---|---|---|
| Basal Cell Nevus Syndrome, Gorlin Syndrome, Gorlin-Goltz Syndrome, or Nevoid Basal Cell Carcinoma Syndrome (Genetics of Skin Cancer) | Gastric Cancer, Diffuse and Lobular Breast Cancer (Genetics of Breast and Gynecologic Cancers) | Peutz-Jeghers Syndrome (Genetics of Colorectal Cancer; Genetics of Breast and Gynecologic Cancers) |
| Birt-Hogg-Dubé Syndrome (Birt-Hogg-Dubé Syndrome) | Hyperparathyroidism, Familial (Genetics of Endocrine and Neuroendocrine Neoplasias) | Polyposis, Familial Adenomatous and Attenuated Familial Adenomatous Polyposis (Genetics of Colorectal Cancer) |
| Bloom Syndrome (Genetics of Skin Cancer) | Li-Fraumeni Syndrome (Genetics of Breast and Gynecologic Cancers) | Polyposis, Familial Juvenile (Genetics of Colorectal Cancer) |
| Breast/Gynecologic Cancers, Hereditary (Genetics of Breast and Gynecologic Cancers) | Medullary Thyroid Cancer, Familial (Genetics of Endocrine and Neuroendocrine Neoplasias) | Polyposis, Hereditary Mixed (Genetics of Colorectal Cancer) |
| Brooke-Spiegler Syndrome (Genetics of Skin Cancer) | Melanoma, Hereditary (Genetics of Skin Cancer) | Polyposis, MUTYH-Associated (Genetics of Colorectal Cancer) |
| Carney-Stratakis Syndrome (Genetics of Endocrine and Neuroendocrine Neoplasias) | Muir-Torre Syndrome (Genetics of Skin Cancer) | Polyposis, Serrated (Genetics of Colorectal Cancer) |
| Colon Cancer, Hereditary Nonpolyposis or Lynch Syndrome (Genetics of Colorectal Cancer) | Multiple Endocrine Neoplasia Type 1 (Genetics of Endocrine and Neuroendocrine Neoplasias) | Prostate Cancer, Hereditary (Genetics of Prostate Cancer) |
| Cowden Syndrome and PTEN Hamartoma Tumor Syndromes (Genetics of Breast and Gynecologic Cancers; Genetics of Colorectal Cancer; Genetics of Skin Cancer) | Multiple Endocrine Neoplasia Type 2A, 2B (Sipple Syndrome) (Genetics of Endocrine and Neuroendocrine Neoplasias) | Renal Cell Cancer, Hereditary with Uterine Leiomyomas (Hereditary Leiomyomatosis and Renal Cell Cancer; Genetics of Skin Cancer) |
| Dyskeratosis Congenita (Zinsser-Cole-Engman Syndrome) (Genetics of Skin Cancer) | Multiple Familial Trichoepithelioma (Genetics of Skin Cancer) | Renal Cell Cancer, Hereditary Papillary (Hereditary Papillary Renal Carcinoma) |
| Epidermodysplasia Verruciformis (Genetics of Skin Cancer) | Oculocutaneous Albinism (Genetics of Skin Cancer) | Rothmund-Thomson Syndrome (Genetics of Skin Cancer) |
| Epidermolysis Bullosa (Genetics of Skin Cancer) | Oligopolyposis (Genetics of Colorectal Cancer) | Von Hippel-Lindau Disease (Von Hippel-Lindau Disease) |
| Familial Cylindromatosis (Genetics of Skin Cancer) | Paraganglioma, Hereditary (Genetics of Endocrine and Neuroendocrine Neoplasias) | Werner Syndrome (Genetics of Skin Cancer) |
| Fanconi Anemia (Genetics of Skin Cancer; Genetics of Breast and Gynecologic Cancers) | Pheochromocytoma, Hereditary (Genetics of Endocrine and Neuroendocrine Neoplasias) | Xeroderma Pigmentosum (Genetics of Skin Cancer) |

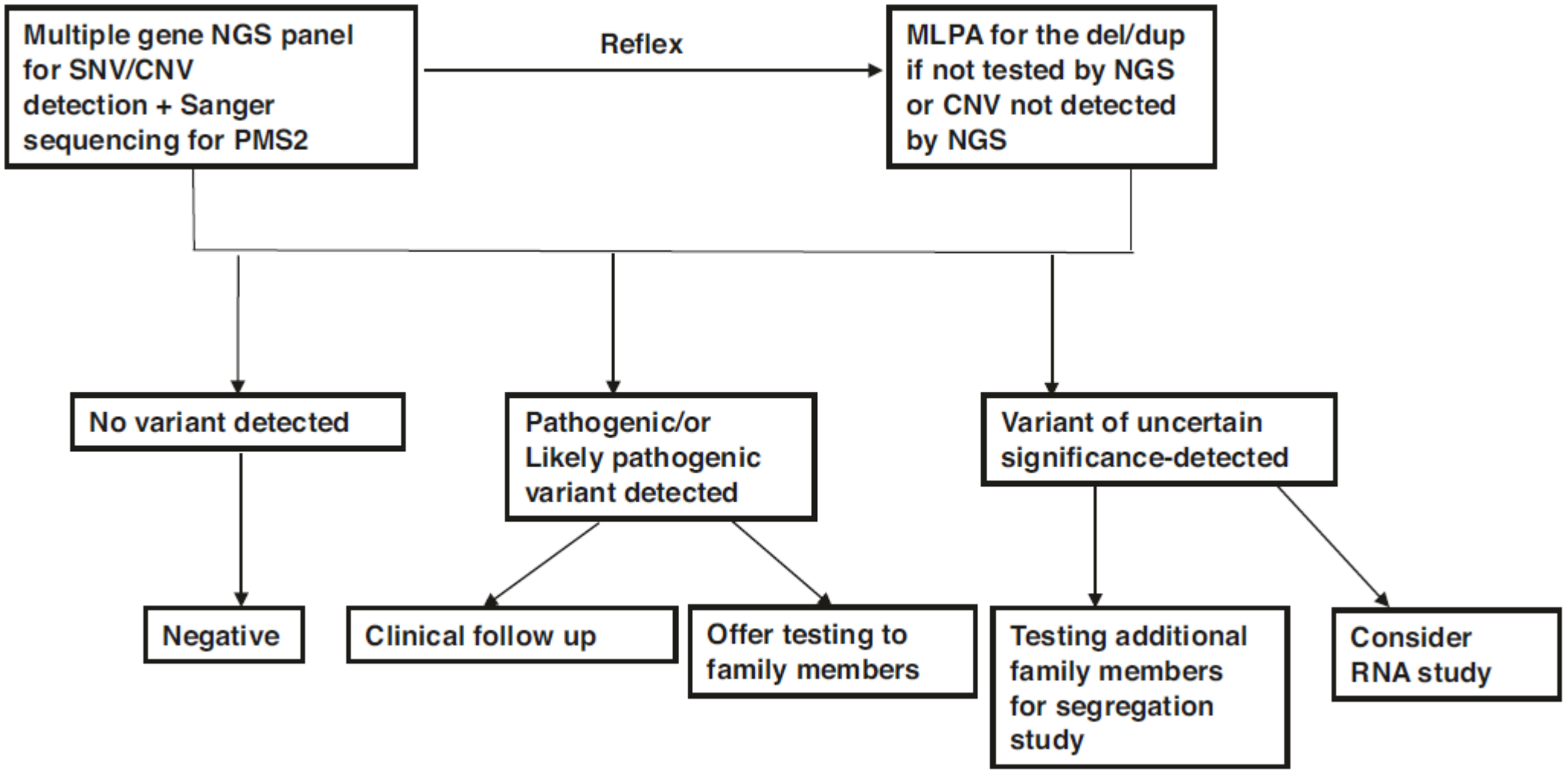
# 结直肠癌或子宫内膜癌MMR缺失筛查

- Patient meets the Amsterdam II criteria or the revised Bethesda guidelines. MSI testing or IHC for the causative gene products in the tumor tissue to confirm MSI-H or the bsence of mismatch repair proteins
- Presence of synchronous or metachronous colorectal cancer or other LS-related tumor regardless of age
- Colorectal cancer in an individual under 60 years of age exhibiting tumor-infiltrating lymphocytes
- Colorectal cancer at any age, plus colorectal cancer or LS related tumor diagnosed before the age of 50 in at least one first-degree relative
- Colorectal cancer at any age, plus colorectal cancer or LS related tumor diagnosed at any age in two or more first-degree or second-degree relatives




Genet Med. 2021;23(10):1807-1817

# 林奇综合征相关的检测指标




Genet Med. 2021;23(10):1807-1817

# CNAS-CL02:2023 医学实验室质量和能力认可准则

## 前言

医学实验室对于患者医疗至关重要，其在伦理和监管范畴内开展活动，并明确医疗服务提供者对患者的责任。这些活动及时开展以满足所有患者及负责患者医疗的人员的需求，包括：检验申请的安排，患者准备，患者识别，样品采集、运送、患者样品的处理，选择符合预期用途的检验，样品检验，样品储存，以及后续的解释、报告和建议。可能还包括向患者提供结果、安排急诊检测和通知危急结果。

## 3.20 医学实验室

以提供诊断、监测、管理、预防和治疗疾病或评估的相关信息为目的，对来自人体的材料进行检验（3.8）的实体。

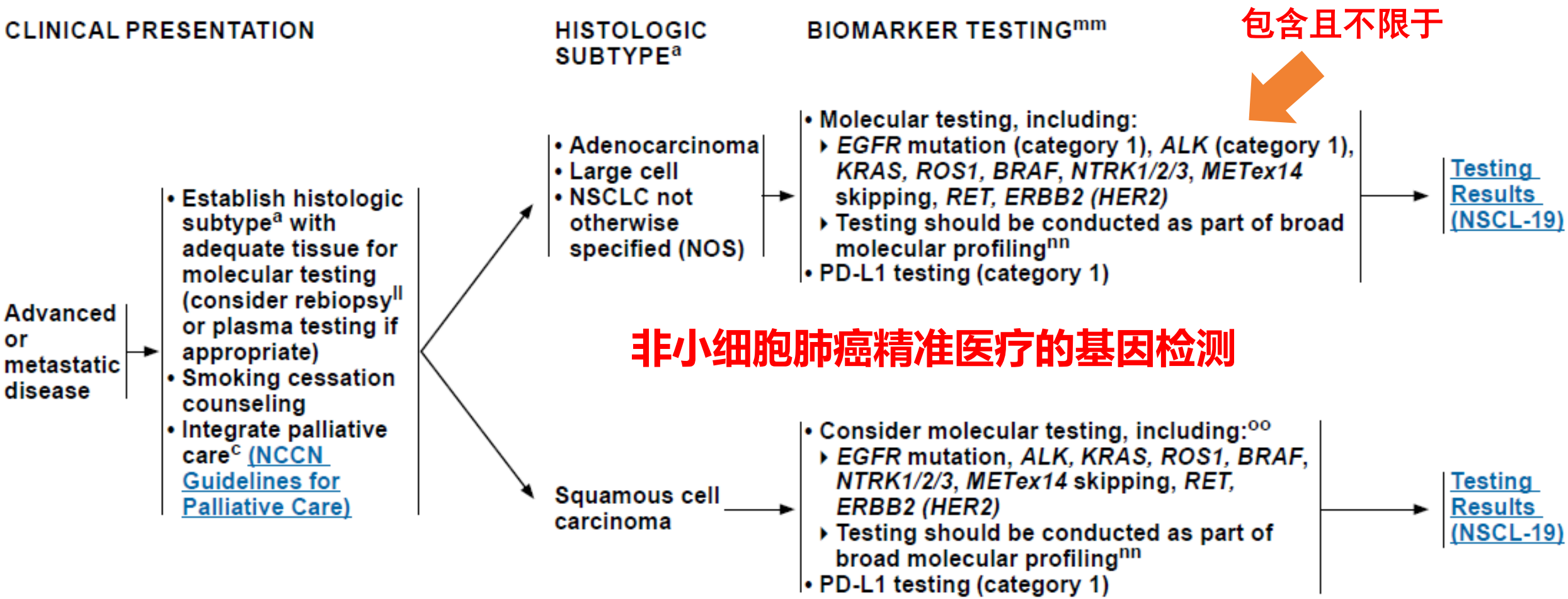
注 1：该类实验室也可提供涵盖检验各方面的咨询，包括合理选择项目，结果解释及进一步检查的建议。

# 临床分子诊断项目来源依据

- 专业性的临床诊疗指南
- 专业性的临床专家共识
- 权威机构发布的检测策略

- NMPA批准的项目
- 实验室自建的项目

# 基于药物基因组学的非小细胞肺癌精准医疗




方法学：NGS、RNA based NGS、Sanger测序、IHC、FISH、SMAP、SnaPshot、PCR，MassARRAY

National Comprehensive Cancer Network®

NCCN Guidelines Version 2.2023
Non-Small Cell Lung Cancer

# NSLC靶向治疗与分子诊断

**MOLECULAR AND BIOMARKER-DIRECTED THERAP**

***EGFR* Exon 19 Deletion or Exon 21 L858R**
- First-line therapy
  - Afatinib[1]
  - Erlotinib[2]
  - Dacomitinib[3]
  - Gefitinib[4,5]
  - Osimertinib[6]
  - Erlotinib + ramucirumab[7]
  - Erlotinib + bevacizumab[c] (nonsquamous)[8]
- Subsequent therapy
  - Osimertinib[9]

***EGFR* S768I, L861Q, and/or G719X**
- First-line therapy
  - Afatinib[1,10]
  - Erlotinib[2]
  - Dacomitinib[3]
  - Gefitinib[4,5]
  - Osimertinib[6,11]
- Subsequent therapy
  - Osimertinib[9]

***EGFR* Exon 20 Insertion Mutation**
- Subsequent therapy
  - Amivantamab-vmjw[12]
  - Mobocertinib[13]

***KRAS* G12C Mutation**
- Subsequent therapy

***ALK* Rearrangement**
- First-line therapy
  - Alectinib[16,17]
  - Brigatinib[18]
  - Ceritinib[19]
  - Crizotinib[16,20]
  - Lorlatinib[21]
- Subsequent therapy
  - Alectinib[22,23]
  - Brigatinib[24]
  - Ceritinib[25]
  - Lorlatinib[26]

***ROS1* Rearrangement**
- First-line therapy
  - Ceritinib[27,28]
  - Crizotinib[29]
  - Entrectinib[30]
- Subsequent therapy
  - Lorlatinib[31]
  - Entrectinib[30]

***BRAF* V600E Mutation**
- First-line therapy
  - Dabrafenib/trametinib[32]
  - Dabrafenib[32]
  - Vemurafenib
- Subsequent therapy
  - Dabrafenib/trametinib[33,34]

不同的基因不同的治疗方案

非小细胞肺癌临床诊疗指南（NCCN V2.2023）

## BRAF V600E & BRAF V600E/D/K

***BRAF V600E Mutations***

BRAF (v-Raf murine sarcoma viral oncogene homolog B) is a ser

同一基因不同的变异相同治疗方案

The

adenocarcinoma; it is the most common of the *BRAF* point mutations when considered across all tumor types.[190,243] Rare *BRAF* mutations include p.V600K, p.V600D, and other mutations. Patients with *BRAF* p.V600E mutations typically either currently or previously smoked cigarettes, whereas those with *EGFR* mutations or *ALK* rearrangements typically have never smoked.[244] Mutations in *BRAF* typically do not overlap with *EGFR* mutations, *MET*ex14 skipping mutations, *RET* rearrangements, *ALK* rearrangements, or *ROS1* rearrangements.[190,191] Testing for *BRAF* mutations is recommended in patients with metastatic nonsquamous NSCLC. Testing may be considered in patients with metastatic NSCLC squamous cell carcinoma because *BRAF* mutations also occur in squamous cell NSCLC, although at a lower rate than nonsquamous NSCLC.[139,140,190,191] Real-time PCR, Sanger sequencing, and NGS are the

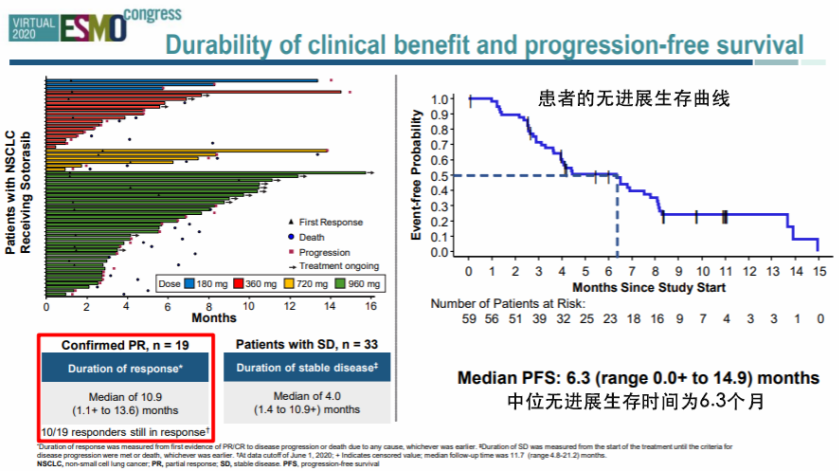
同一基因不同的变异不同治疗方案

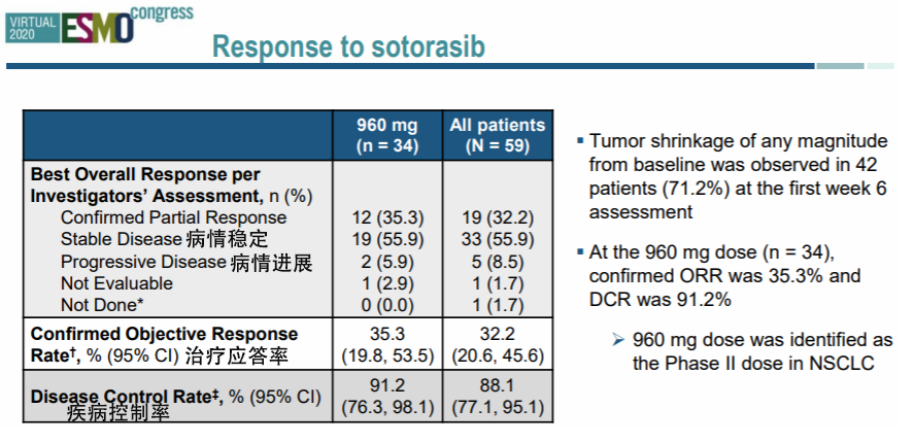
# 针对KRAS基因突变的口服小分子靶向药

| 抑制剂名称 | Ras | H-ras | N-ras | K-ras | Ral | 其他靶点 |
|---|---|---|---|---|---|---|
| Lonafarnib (SCH66336) | | ++++ | +++ | +++ | | |
| BI-3406 | | | | ++ | | |
| Kobe2602 | + | | | | | |
| BAY-293 | | | | ++ | | |
| KRpep-2d | | | | ++++ | | |
| ARS-853 (ARS853) | | | | | | |
| Zoledronic acid (ZOL 446) | | | | | | |
| MCP110 | √ | | | | | Raf-1 |
| MRTX-1257 | | | | √ | | KRAS dependent ERK phosphorylation |
| Oncrasin-1 | | | | √ | | RNA polymerase II,PKCι |
| KY1220 | √ | | | | | Wnt/β-catenin |
| Antineoplaston A10 | √ | | | | | |
| K-Ras-IN-1 | | | | | | |
| Sotorasib (AMG510) racemate | | | | | | |
| Adagrasib (MRTX849) | | | | | | |
| Sotorasib (AMG510) | | | | √ | | |
| ARS-1620 | | | | √ | | |
| Fendiline hydrochloride | | | | √ | | L-type calcium channel |
| Zoledronic acid monohydrate | √ | | | | | Rho |
| Kobe0065 | | √ | | | | |
| BQU57 | | | | | √ | |
| Salirasib | √ | | | | | PPMTase |
| Deltarasin | | | | √ | | PDEδ |
| K-Ras(G12C) inhibitor 9 | | | | √ | | |
| K-Ras(G12C) inhibitor 6 | | | | √ | | |
| K-Ras(G12C) inhibitor 12 | | | | √ | | www.selleck.cn |
| 6H05 | | | | √ | | |

**KRAS& KRAS/HRAS/NRAS**

**不同的基因同一治疗方案**




Clin Cancer Res. 2020;26(22):5962-5973
Clin Cancer Res. 2020;26(7):1633-1643
N Engl J Med. 2020;383(13):1207-1217

# CNAS-CL02:2023 医学实验室质量和能力认可准则

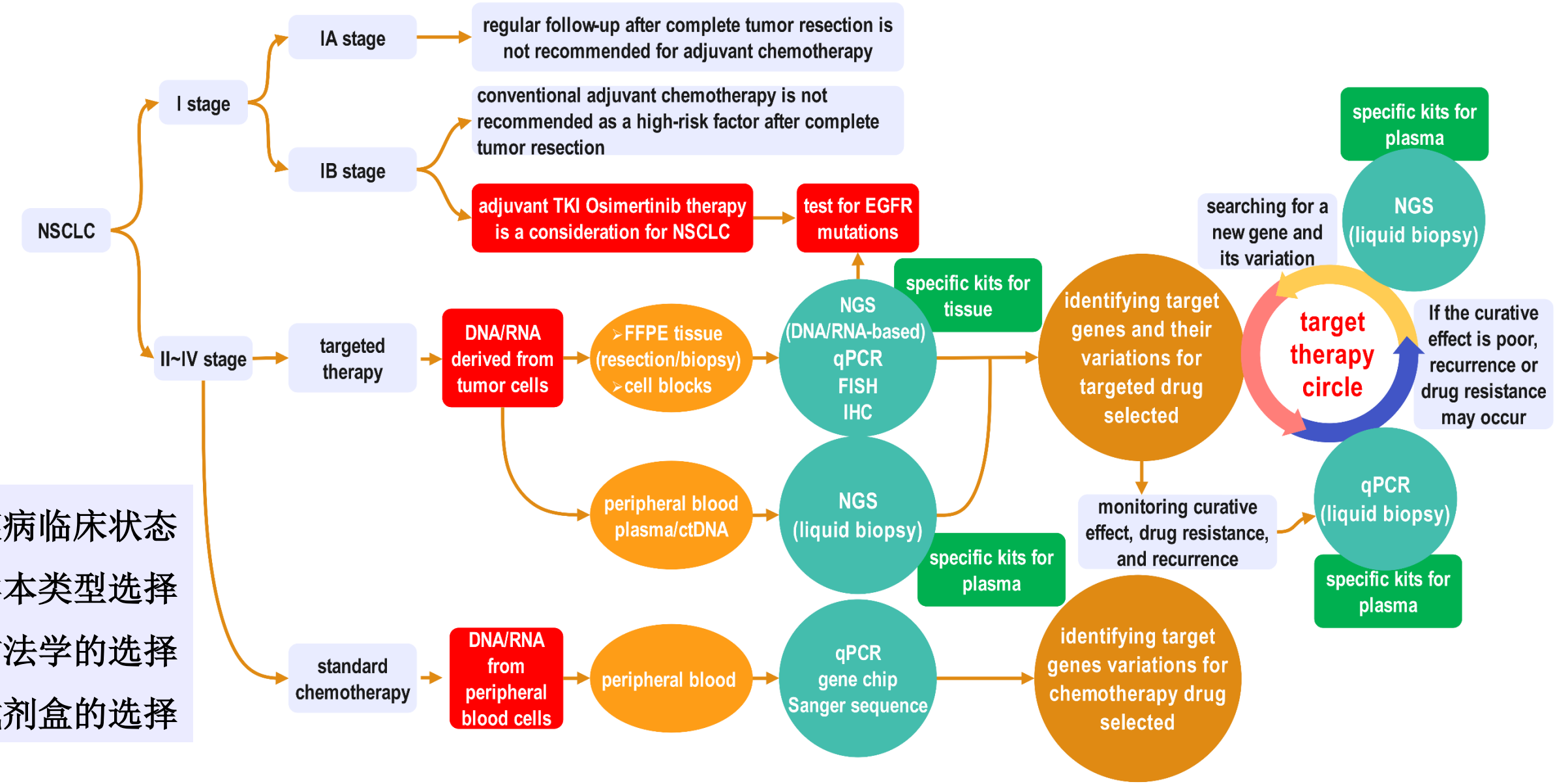
## 咨询活动

实验室管理层应确保提供适当的实验室建议和解释，并满足患者和用户的需求。

适用时，实验室应建立协议与实验室用户**进行沟通**，包括：

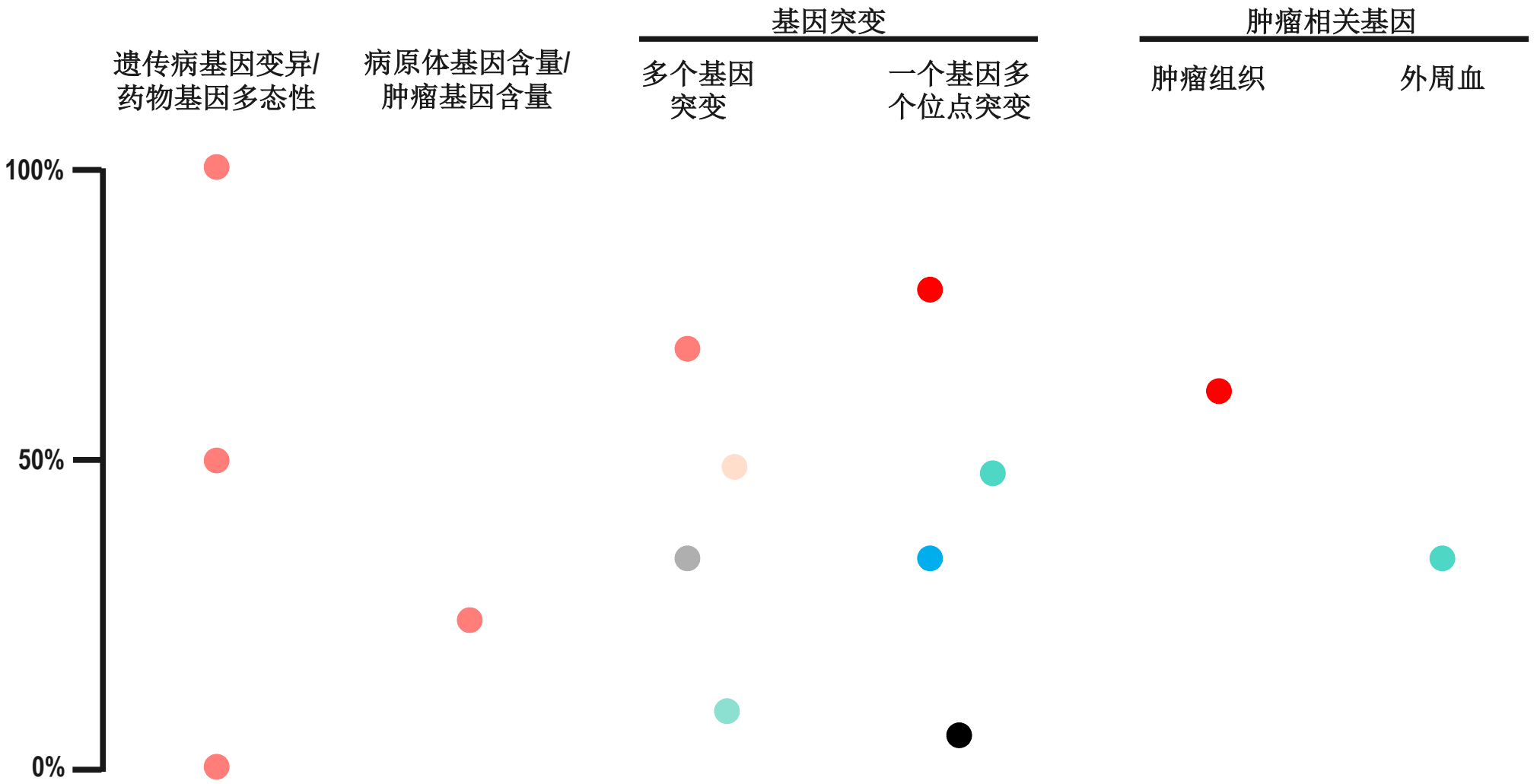
- 为选择和使用检验提供意见，包括所需**样品类型**、**检验方法**的临床**适应症**和**局限性**，以及要求**检验的频率**；
- 为检验结果的**解释**提供专业判断；
- 促进实验室检验的**有效利用**；
- 就科学及事务性工作提供意见，例如样品不符合**可接受标准**的情况。

# NSCLC精准医疗与临床分子诊断




- 疾病临床状态
- 样本类型选择
- 方法学的选择
- 试剂盒的选择

# 标本中核酸含量示意图


基因突变
肿瘤相关基因
遗传病基因变异/
药物基因多态性
病原体基因含量/
肿瘤基因含量
多个基因
突变
一个基因多
个位点突变
肿瘤组织
外周血
100%
50%
0%

# 分子诊断检验程序性能验证指南

# CNAS-GL039:2019

注：鉴于实际临床工作中进行分子诊断的样本类型（如进行原位杂交的样本有血液、羊水穿刺、肿瘤组织等）以及预期用途差别较大，而不同样本类型对性能验证的要求和难易程度差别较大，建议结合实际情况酌情选择与之相符合的性能验证方案。

# CNAS-GL039:分子诊断检验程序性能验证指南

## 4.2 性能验证的参数

如果检验程序适用样本类型包括血清与血浆，实验室在临床检测时同时使用血清与血浆，应进行**血清与血浆结果一致性的验证**。在肿瘤靶向基因检测时，如果检验程序适用样本类型包括除肿瘤组织/细胞以外的样本（如血浆），应进行与**肿瘤组织结果一致性的验证**。

# HCV RNA定量检测的要求

## 丙型肝炎病毒核糖核酸测定试剂技术审查指导原则

2.1 最低检出限与定量限的确定。

建议使用国际参考品/国家参考品进行梯度稀释并多次检测，将具有95%阳性检出率的病毒水平作为最低检出限。至少应不高于国家参考品最低检出限50IU/ml的要求。根据循证医学中有关对HCV感染者进行抗病毒治疗效果及预后评估的需要，企业可根据自身产品性能情况和临床诊疗指南设定检测下限以符合临床需求。定量限应高于或等于检出限，将多次（至少20次）测

| 产品名称 | 丙型肝炎病毒核酸检测试剂盒（PCR-荧光探针法） |
|---|---|
| 核酸提取技术 | 超顺纳米磁珠法 |
| 样本类型 | 血清、血浆 |
| 检测下限 | 25 IU /mL |
| 线性范围 | 50-1.0×10$^8$ IU/mL |
| 覆盖基因型 | 1-6型基因型 |
| 内标 | 内标全程参与核酸提取和扩增 |
| 已获证书 | NMPA |

**EASL recommendations on treatment of hepatitis C: Final update of the series**☆

HCV RNA detection and quantification in serum or plasma should be made by a sensitive assay with a lower limit of detection of ≤15 IU/ml **(A1)**.

## 丙型肝炎防治指南(2022年版)

采用敏感检测方法(检测下限≤15 IU/mL)进行血清或血浆HCV RNA定量检测。如果敏感的HCV RNA检测不可进行时,可使用检测下限≤1 000 IU/mL的HCV RNA检测试剂,如果HCV RNA检测仍然低于检测线,建议再使用敏感试剂进行检测确认。

## Diagnosis and Monitoring of Hepatic Injury. I. Performance Characteristics of Laboratory Tests

Clin Chem. 2000;46(12):2027-49

EDTA and sodium citrate plasma are preferred specimens for HCV RNA tests. Heparinized plasma is inhibitory for many nucleic acid amplification assays, and serum specimens provide suboptimal stability unless serum is frozen soon after specimen collection. HCV RNA is very susceptible to degradation by the high activities of RNase present in blood; therefore, serum specimens for HCV RNA should be centrifuged as soon as possible after clot formation. If centrifugation is performed immediately,<10% of HCV RNA is lost even if the plasma or serum is not separated from the formed elements for up to 6h.

**Quantitative detection of hepatitis C virus RNA with a solid-phase signal amplification method: definition of optimal conditions for specimen collection and clinical application in interferon-treated patients**

Hepatology. 1994;19(6):1337-41

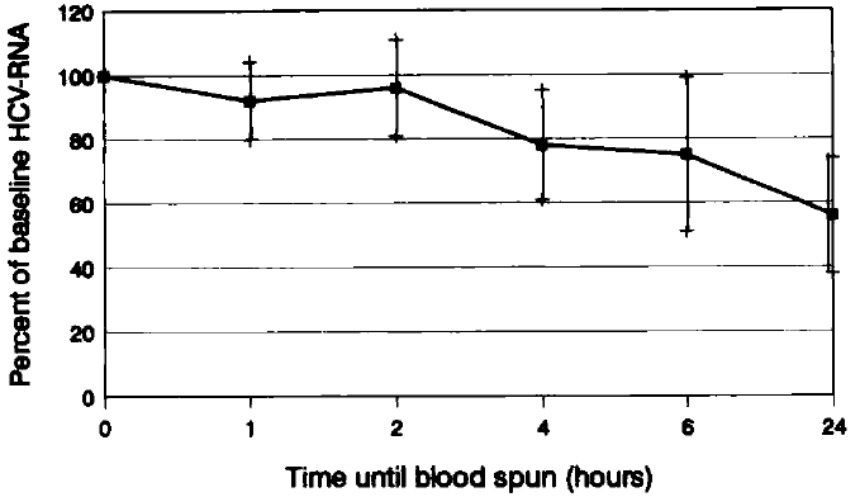
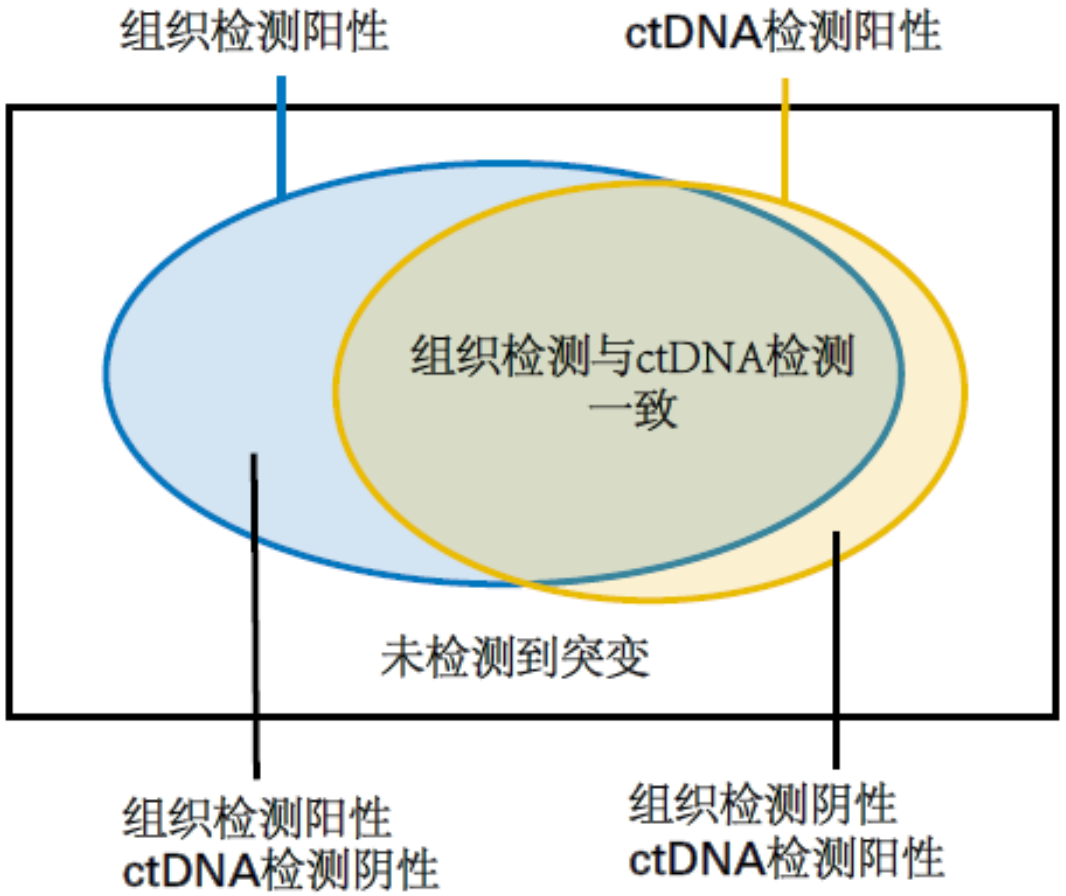
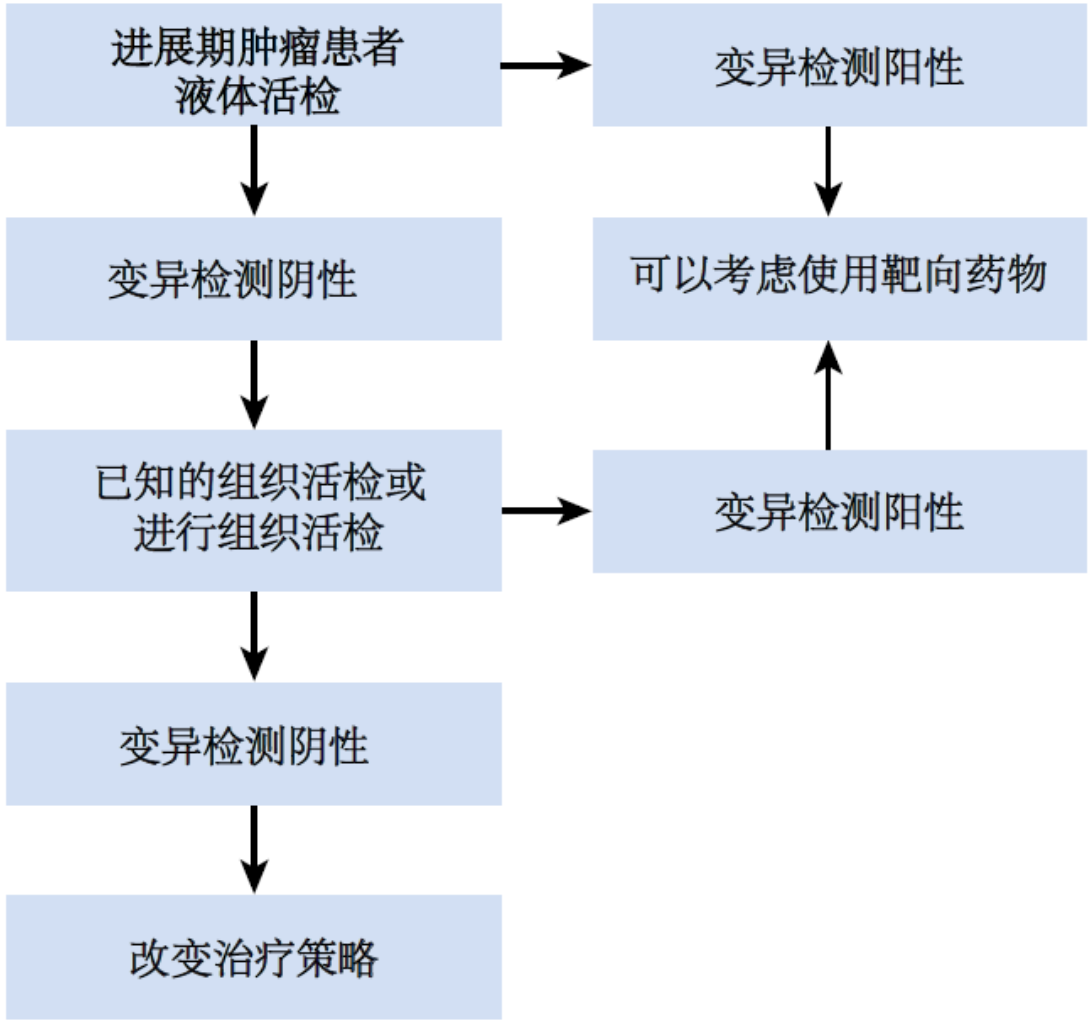



**TABLE 1. Percentage loss of HCV RNA as measured with the bDNA assay, related to conditions of serum preparation**

| Blood processing | 0 hr | 1 hr | 2 hr | 4 hr | 6 hr | 12 hr | 24 hr |
|---|---|---|---|---|---|---|---|
| Unspun | 0% | 3.4% | 2.2% | 14.2% | 31.8% | ND | 49.4% |
| Immediately separated: RT | 0% | ND | 4.1% | ND | ND | 16.6% | ND |
| Immediately separated: 4° C | 0% | ND | 6.9% | ND | ND | 6.0% | ND |
| Spun, left on clot: RT | 0% | 9.5% | 7.7% | 10% | 4.8% | ND | 15.5% |
| Spun, left on clot: 4° C | 0% | 4.4% | 5.2% | 5.9% | 8.3% | ND | 0% |
| Spun, SST: RT | 0% | 3.1% | 2.9% | 0% | 2.0% | ND | 6.6% |
| Spun, SST: 4° C | 0% | 1.2% | 0% | 0% | 0% | ND | 0% |

# 组织活检与液体活检的比较




J Clin Oncol. 2018;36(16):1631-1641

# 标本的选择

NCCN National Comprehensive Cancer Network®
NCCN Guidelines Version 2.2023
Non-Small Cell Lung Cancer

**If repeat biopsy is not feasible, plasma biopsy testing should be considered**

Formalin-fixed paraffin-embedded (FFPE) material is suitable for most molecular analyses, except bone biopsies that were previously treated with acid decalcifying solutions. Non-acid decalcification approaches may be successful for subsequent molecular testing. While many molecular pathology laboratories currently also accept cytopathology specimens such as cell blocks, direct smears, or touch preparations, laboratories that do not currently do so are strongly encouraged to identify approaches to testing on non-FFPE cytopathology specimens.

不能替代组织检查

"Plasma Cell-Free/Circulating Tumor DNA Testing:

- Cell-free/circulating tumor DNA testing should not be used in lieu of a tissue diagnosis.
- Some laboratories offer testing for molecular alterations examining nucleic acids in peripheral circulation, most commonly in processed plasma (sometimes referred to as 'liquid biopsy').
- Studies have demonstrated cell-free tumor DNA testing to generally have very high specificity, but significantly compromised sensitivity, with up to 30% false-negative rate.

假阴性率可达30%

- Standards for analytical performance characteristics of cell-free tumor DNA have not been established, and in contrast to tissue-based testing, no guidelines exist regarding the recommended performance characteristics
- Cell-free tumor DNA testing can identify alterations that are unrelated to a lesion of int sis of indeterminate potential (CHIP).
- The use of cell-free/circulating tumor DNA testing can be considered in specific clinica
  - ◊ If a patient is medically unfit for invasive tissue sampling
  - ◊ In the initial diagnostic setting, if following pathologic confirmation of a NSCLC diag molecular analysis, cell-free/circulating tumor DNA should be used only if follow-up tissue-bas in which an oncogenic driver is not identified"

液体活检的条件
A. 不能组织活检的
B. 组织活检标本量不足的
C. 疾病进展监测

**Plasma-based testing should be considered at progression on EGFR TKIs for the T790M mutation.**
**If plasma-based testing is negative, tissue-based testing wit ded.**
**Practitioners may want to consider scheduling the biopsy c**

液体标本阴性时，应考虑组织标本
需考虑液体标本与组织标本检测的顺序

## 分析前考虑的因素

- 项目选择
- 标本类型
- 技术平台

### 4.2 性能验证的参数

实验室应根据**检测项目的预期用途**以及**生产制造商声明**，选择对检测结果质量有重要影响的参数进行验证。**不同技术平台、样本类型以及预期用途**不同时， 所需验证的性能指标宜有所侧重

CNAS-GL039:分子诊断检验程序性能验证指南

### 4.3 性能验证的判断标准

实验室应根据临床需求选择经确认的符合预期用途的检验程序。实验室性能验证结果的判断标准是厂商或研发者在试剂盒或检测系统说明书中声明的性能

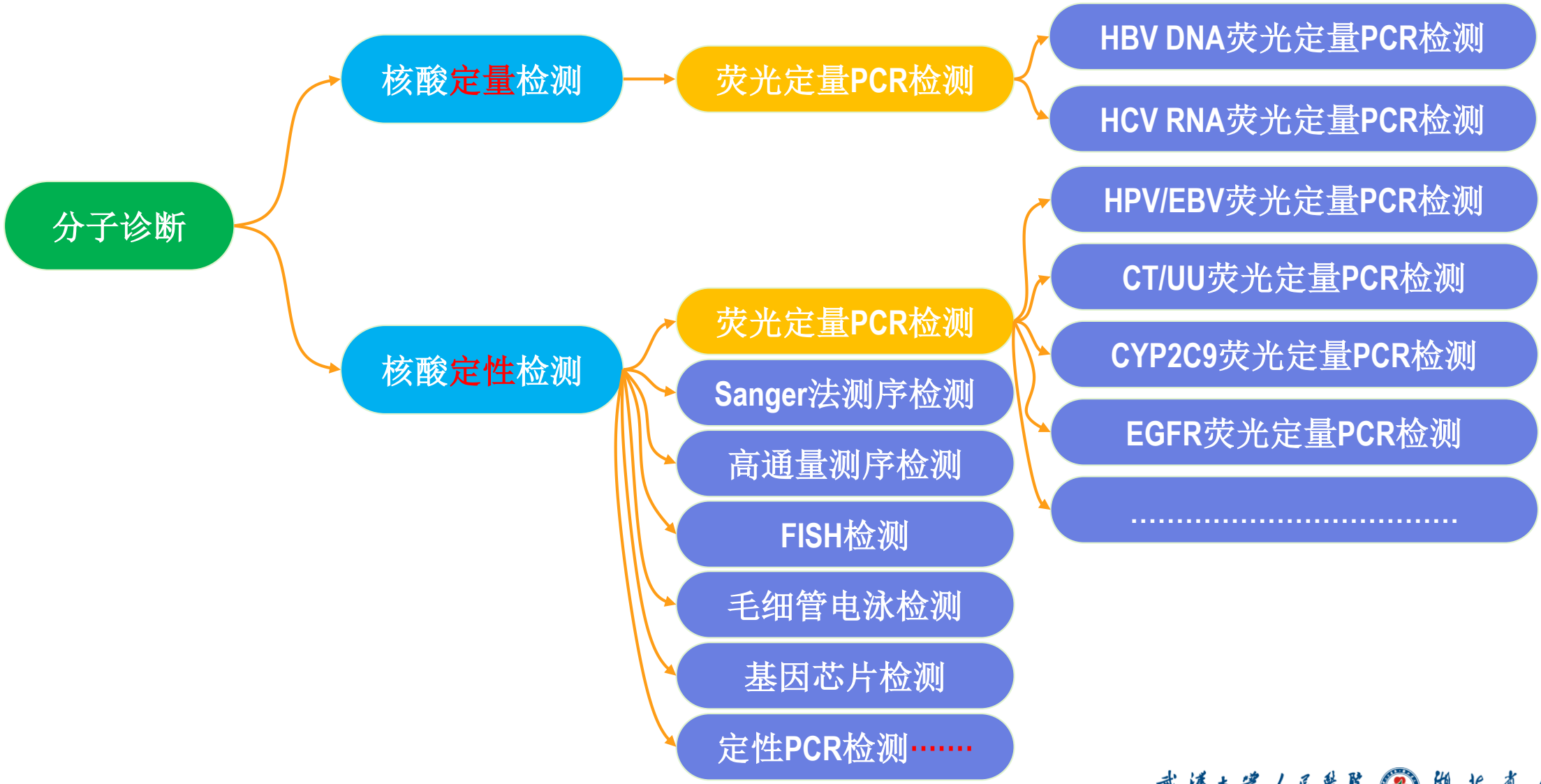


指标。

**7.2.1通用要求** 检验申请、样品采集、运送、储存等检验前活动宜参考《全国临床检验操作规程》以及相关国家/行业标准的要求，如GB/T 42060、WS/T 348、WS/T 359、WS/T 402、WS/T 640、WS/T 661、WS/T794等。

CNAS-CL02-A001:2023医学实验室质量和能力认可准则的应用要求

# 分子诊断技术与临床应用



# 性能验证的参数（CNAS-GL039）

分子诊断检验程序的性能参数主要包括PCR定性和定量检测、Sanger测序、二代基因测序(NGS)和原位杂交等：

- **PCR定性检测** 选择验证的性能指标宜包括方法符合率、检出限、抗干扰能力、交叉反应等。Sanger测序和NGS选择验证的性能指标宜包括方法符合率和检出限等。原位杂交技术应依据样本类型和预期用途，选用适宜的性能指标进行验证，如基于完整细胞的原位杂交宜选用分析敏感性和特异性，基于组织的宜选用方法符合率

**本文件适用于医学实验室采用的经确认的检验程序**

# 定性项目的性能验证

6.1.1 方法符合率

6.1.1.1 验证要求

通过与参比方法进行比较。参比方法包括但不限于：金标准方法、行业公认方法、经验证性能符合要求满足临床预期用途的方法（如：通过 ISO15189 认可实验室使用的相同检测方法）。

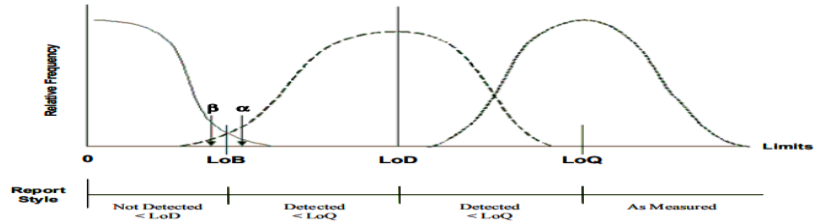
6.1.2 检出限

6.1.2.1 验证要求

所用检验程序在厂家试剂使用说明书等有声明检出限时，检测项目在有标准物质时，或以定量形式表达定性结果时，应进行检出限的验证。

# CNAS-GL037:2019 临床化学定量检验程序性能验证指南

## 4.2 性能验证的参数



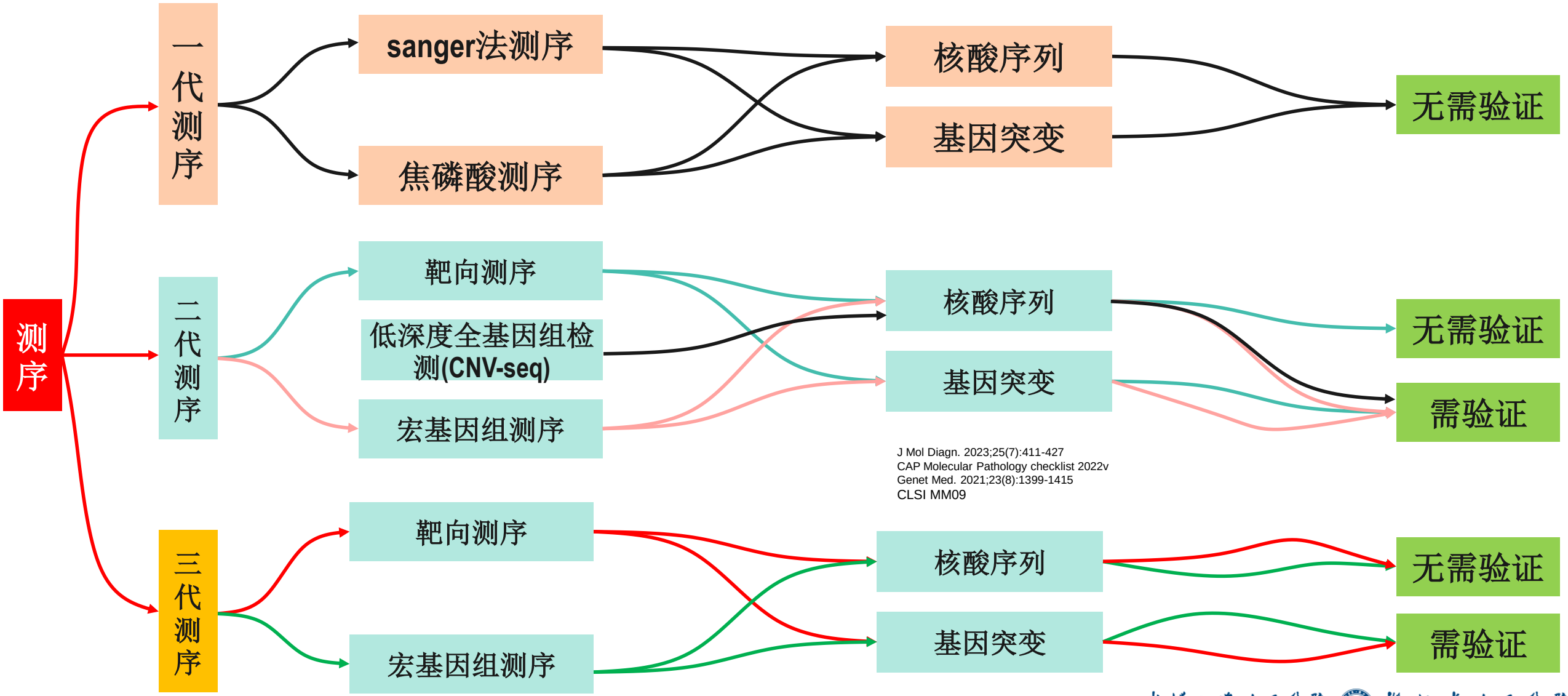

临床化学定量检验程序的分析性能参数一般包括：测量正确度、测量精密度（含测量重复性和测量中间精密度）、测量不确定度、分析特异性（含干扰物）、分析灵敏度、检出限和定量限、线性区间（可报告区间）等。实验室应根据不同检验项目的预期用途，选择对检验结果质量有重要影响的参数进行验证。

## 4.3 性能验证的判断标准

注：如果验证结果符合制造商或研发者声明的性能指标，但不满足实验室制定的判断标准，结果不可接受。

# 测序结果是否需要验证

MOL.35850 NGS Confirmatory Testing

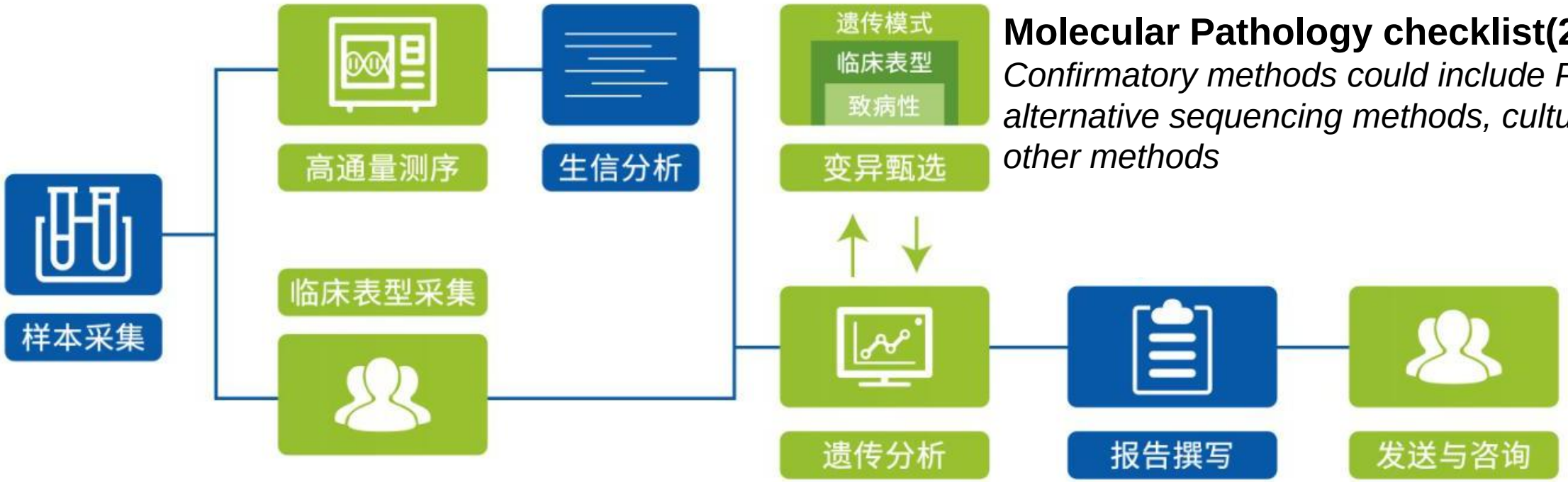



J Mol Diagn. 2023;25(7):411-427
CAP Molecular Pathology checklist 2022v
Genet Med. 2021;23(8):1399-1415
CLSI MM09

# 检测结构变异常用方法比较

| | Variant Type | | | |
|---|---|---|---|---|
| | Copy Number Variants (CNVs) | Balanced Structural Variants (SVs) | Regions of Homozygosity (ROH) | Repeat Expansions (REs) |
| Karyotype | ✓ | ✓ | | |
| Chromosomal Microarray | ✓ | | ✓ | |
| Gene Panels | ✓ | | | |
| Exome Sequencing | ✓ | | ✓ | |
| Virtual Panels | ✓ | | | |
| Low-Pass Genome Sequencing | ✓ | | ✓ | |
| Genome Sequencing | ✓ | ✓ | ✓ | ✓ |
| Mate-Pair Sequencing | ✓ | ✓ | | |
| Optical Mapping | ✓ | ✓ | ✓ | ✓ |
| Long-Read Sequencing | ✓ | ✓ | ✓ | ✓ |

# 低深度全基因组检测(CNV-seq)检测流程




**Molecular Pathology checklist(2022)**

*Confirmatory methods could include PCR, alternative sequencing methods, culture, or other methods*

# CNAS-CL02-A001:2023医学实验室质量和能力认可准则的应用要求

## 人才队伍建设

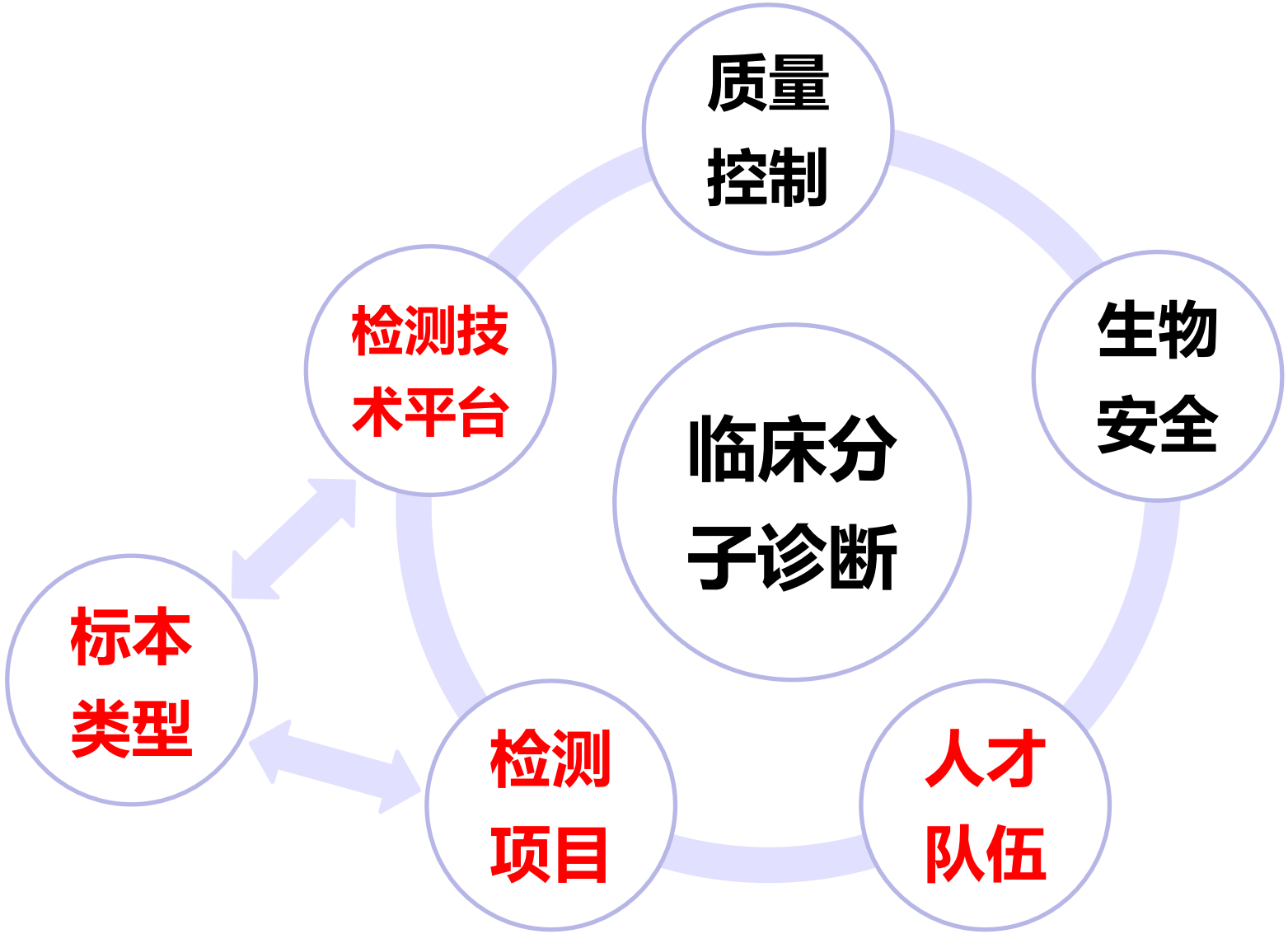
- 丰富的背景知识
- 报告解读能力
- 检测建议能力
- 临床沟通能力

6.2.2 **能力要求** 基因变异检测报告签发人员应通过参加相关领域的培训或学术交流等继续教育活动，**熟悉行业规范、指南以及专家共识**，了解**基因变异检测技术和临床应用**的最新进展

7.4.1 **结果报告** 应定期评审并**更新基因变异**检测报告中提供给用户参考的**分子变异临床意义和用药信息**，确保其准性

7.4.1.7**报告附加信息** 适用时，报告内容还应包括方法的局限性、检测结果临床意义的**简要解读、进一步检测建议**；肿瘤分子病理报告内容还应包括检测样品中肿瘤细胞的含量。

# 分子诊断实验室：全流程全方位的质量管理




## CNAS-GL050:2021
## 质量指标

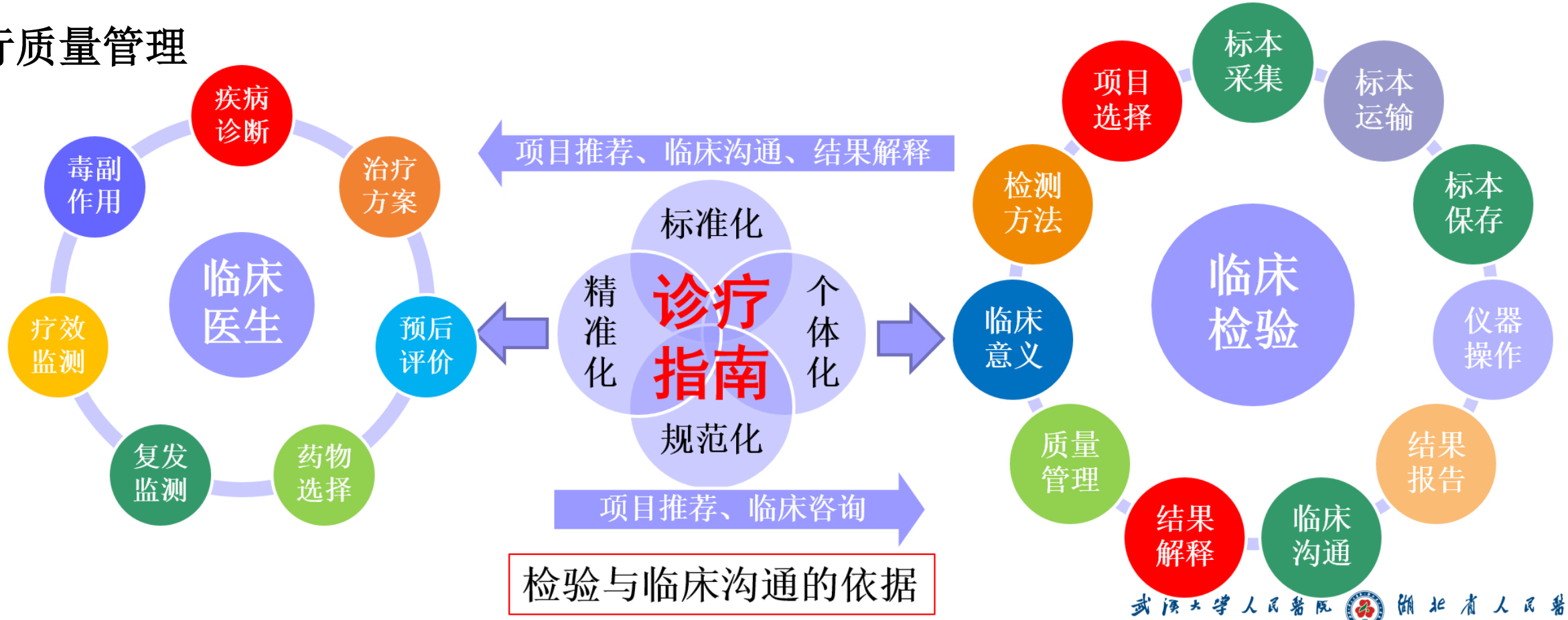
实验室应建立**质量指标**以监控和评估检验前、检验和检验后过程中的关键环节，例如：不合格标本率、室间质评合格率、报告及时率、投诉处理率等

## CNAS CL02:2023
## 人员--通用要求

实验室应有足够数量**有能力的人员**开展其活动

# 基于临床疾病诊疗指南下的检验服务能力建设

一个合适的临床分子诊断实验室质量管理，首先应满足临床需求，其次应从人员能力、检测平台、检测项目及内容、标本类型、质量控制和生物安全等多维度进行质量管理



# 感谢您的聆听！