The revised guidelines are in progress. These are not final Recommendations!

Advisory Panel review! Revisions!

Posting for public comment soon! Revisions!

Molecular Testing Guideline for Selection of Lung Cancer Patients- Guideline Revision & Update

Initiated in 2014
Writing began in 2015
This work is still in progress!
Current revisions underway!
Advisory Panel review!
Posting for public comment soon!
These are not final Recommendations!

Expert Panel Members

CO-CHAIRS
Philip T. Cagle, MD (CAP)
Yasushi Yatabe, MD, PhD (IASLC)
Neal Lindeman, MD (AMP)

EXPERT PANELISTS
Dara L. Aisner, MD, PhD
Maria E. Arcila, MD
Eric Bernicker, MD
Mary Beth Beasley, MD
Sanja Dacic, MD, PhD
Keith Kerr, MB ChB

David Kwiatkowski, MD, PhD
Marc Ladanyi, MD
Lynette Sholl, MD
Benjamin Solomon, MBBS, PhD
Erik Thunnissen, MD, PhD
Ming S. Tsao, MD

Steering Committee and Staff

• Steering Committee
  – Jan Nowak, MD, PhD (CAP)
  – Fred Hirsch MD, PhD (IASLC)
  – Neal Lindeman, MD (AMP)

• Staff
  – Christina Ventura, MLS(ASCP) (CAP)
  – Murry Wynes, PhD (IASLC)
  – Robyn Temple-Smolkin, PhD (AMP)
  – Lesley Souter, PhD (CAP)
  – Carol Calasacco, MLIS, SCT (ASCP) (CAP)
  – Pia Hirsch (IASLC)
  – Mrudula Pullambhatla, MS (AMP)

Patient Advocates

• Bonnie Addario
  The Bonnie J. Addario Lung Cancer Foundation

• Kim Norris
  Lung Cancer Foundation of America
Bethesda, February, 2016

Co-Chairs

Advisory Panel Members

Natasha Rekhtman, MD, PhD
Timothy Craig Allen, MD, JD
Marina Nikiforova, MD
Federico Cappuzzo, MD
Antonio Marchetti, MD, PhD
Juan-Sebastian Saldivar, MD
Marileila Varella Garcia, PhD
Edmund S. Cibas, MD
Dhananjay A. Chitale, MD
John Lafrate, MD, PhD
Suresh Ramalingam, MD
Lukas Bubendorf, MD
Yi-Long Wu, MD
Charles Powell, MD
Tony Mok, MD
Pasi A. Janne, MD, PhD
Julia A. Bridge, MD
Tetsuya Mitsudomi, MD, PhD
Paul Bunn, MD
Mark G Kris, MD

Contents

• Introduction
• New Key Questions
• Old Key Questions
KQ I. What other genes, previously not addressed, should be tested in lung adenocarcinoma?

- ROS1: 1-2% rearrangement
- RET: 1-2% rearrangement
- BRAF: 4% half are non-V600E
- MET: 3% exon 14 deletion and amplification
- ERBB2/HER2: 2%
- (KRAS: 30% mutation MEK inhibitors)

ROS1 Rearrangements in NSCLC

- Most auspicious; clinical trials
- Chromosomal rearrangements
- Approximately 1% to 2% of lung cancers with adenocarcinoma histology
- About 2,000 to 4,000 new cases of ROS1 positive lung cancer each year in the United States.

Late Breaking News

This past Friday afternoon March 11
US FDA approved Crizotinib for ROS1 positive advanced NSCLC

ROS1 and Crizotinib

- Lung cancers with ROS1 translocations respond to crizotinib (already approved for ALK positive lung cancers)
- Oncologists have been requesting ROS1 testing and treating ROS1 positive lung cancers with crizotinib
- Crizotinib has received FDA Breakthrough Therapy Designation Approval in ROS1 positive lung cancers.
Tests for ROS1 Rearrangements in NSCLC

NO GOLD STANDARD METHOD
• FISH
• Immunohistochemistry (IHC)
• Anchored multiplex PCR
• Reverse-transcriptase PCR
• Next generation sequencing

ROS 1 Considerations?

• To select patients for ROS1 targeted therapy, ROS1 testing on all lung adenocarcinoma patients?
• Maybe other cell types in never smokers, etc?
• Molecular or cytogenetic test—no specific method based on clinical trials?
• IHC for screening, but needs molecular or cytogenetic confirmation?

Related thought

• In addition to histological diagnosis, important to prioritize lung adenocarcinoma biopsy tissue for EGFR, ALK, and ROS1 testing

RET, BRAF, HER2/ERBB2, KRAS

• As with EGFR, ALK and ROS1, these are generally mutually exclusive with occasional exceptions
• Mostly or exclusively adenocarcinomas
• Clinical trials and/or potential drugs for each (KRAS may predict for downstream targets such as MEK)
• Literature evidence not to recommendation level??
RET Rearrangements in NSCLC

- Chromosomal rearrangements in about 1% to 2% of lung cancers with Adenocarcinoma histology

Tests for RET Rearrangements in NSCLC

- FISH
- Anchored multiplex PCR
- Next generation sequencing
- IHC-variable results; not widely accepted yet

BRAF Mutations in NSCLC

- BRAF mutations occur in about 5% of NSCLC
- In contrast to melanomas, only half of BRAF mutations in NSCLC are V600E mutations.
- Responses reported to V600E targeted drugs

Tests for BRAF Mutations in NSCLC

- Sanger sequencing and other molecular
- Next generation sequencing
- IHC not established for NSCLC
  - Only half of BRAF mutations in NSCLC are V600E
<table>
<thead>
<tr>
<th><strong>ERBB2/HER2 in NSCLC</strong></th>
<th><strong>KRAS Mutations in NSCLC</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td>• Amplification or overexpression of HER2 well known predictive biomarker for breast cancer</td>
<td></td>
</tr>
<tr>
<td>• HER2 activation in lung cancer is associated with mutations, mostly insertions in exon 20, which are independent of HER2 gene amplification</td>
<td></td>
</tr>
<tr>
<td>• These mutations are not seen in breast cancer.</td>
<td></td>
</tr>
<tr>
<td>• HER 2 mutations are found in 2% of lung adenocarcinomas</td>
<td></td>
</tr>
<tr>
<td>• 30% Adenocarcinomas</td>
<td></td>
</tr>
<tr>
<td>• No approved direct KRAS inhibitors hence, testing for KRAS has not been useful as a predictive biomarker for a KRAS inhibitor</td>
<td></td>
</tr>
<tr>
<td>• But recent reports of response to MEK inhibitors downstream of KRAS</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th><strong>RET, BRAF,HER2/ERBB2, KRAS Considerations</strong></th>
<th><strong>MET in NSCLC</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td>• Adenocarcinomas, maybe other cell types in never smokers, etc?</td>
<td></td>
</tr>
<tr>
<td>• Not indicated as a routine stand-alone assay outside the context of a clinical trial?</td>
<td></td>
</tr>
<tr>
<td>• Can be part of larger testing panels performed either</td>
<td></td>
</tr>
<tr>
<td>– Initially? or</td>
<td></td>
</tr>
<tr>
<td>– when routine EGFR, ALK, and ROS1 testing are negative?</td>
<td></td>
</tr>
<tr>
<td>• De novo MET amplification has been associated with profound responses to therapy with crizotinib.</td>
<td></td>
</tr>
<tr>
<td>• Recently, splicing variants and insertion-deletion mutations leading to MET exon 14 deletion have been found in about 3% of lung Adenocarcinomas</td>
<td></td>
</tr>
<tr>
<td>• Lead to MET activation and response to crizotinib and cabozantinib.</td>
<td></td>
</tr>
</tbody>
</table>
MET in NSCLC
• MET amplification can be seen in tumors with MET exon 14 deletion
• MET protein overexpression appears to correlate with MET amplification, although MET IHC is controversial as a biomarker for MET-targeted therapies
• Squamous cell as well as adenocarcinoma

Tests for MET Activation in NSCLC
• Mutational analysis – exon 14
  – Sequencing
  – NGS
• Copy number
  – FISH, CISH
  – NGS
• IHC?

MET Considerations
• Not indicated as a routine stand-alone assay outside the context of a clinical trial?
• Can be part of larger testing panels performed either
  – Initially? or
  – when routine EGFR, ALK, and ROS1 testing are negative?

KQ II. Is immunohistochemistry reliable for screening for ALK translocations?
FDA Approval 6-12-2015

VENTANA ALK (D5F3) CDx Assay is intended for the qualitative detection of the ALK protein in FFPE NSCLC tissue stained with a BenchMark XT automated staining instrument. It is indicated as an aid in identifying patients eligible for treatment with XALKORI® (crizotinib).

ALK IHC Considerations

- When testing for ALK rearrangements, IHC as an equivalent alternative to FISH for adenocarcinoma
- May identify some cases not readily recognized by FISH

KQ III. In patients who are undergoing treatment with targeted tyrosine kinase inhibitors, what are the types and rates of secondary resistance?

Secondary resistance considerations

- Molecular tests with appropriate performance characteristics for detection of secondary mutations
- EGFR T790M mutation testing to select patients for EGFR T790M mutation targeted therapy
- ALK mutational testing to select patients for ALK secondary resistance mutation targeted therapy
KQ IV. Are there biomarkers that are predictive of clinical outcome in squamous and small cell carcinomas?

Squamous cell carcinoma Considerations
- Testing if squamous cell not excluded; especially if never smoker, young age, etc
- Insufficient evidence for new specific markers?

EGFR Inhibiting Monoclonal Antibodies for Squamous Cell Carcinoma
- EGFR copy number (FISH) as potential biomarker
- Cetuximab (Erbitux)
- Portrazza (Necitumumab) Eli Lilly
  - FDA approved with chemo in advanced squamous cell carcinoma

V. What are the clinical performance characteristics of circulating DNA/CTC in plasma when used for diagnosis of primary lung adenocarcinoma or relapse?
**cfDNA and CTC Considerations**

- In general, insufficient evidence for recommendation?
- Maybe cfDNA when insufficient biopsy tissue for EGFR testing?
- May consider cfDNA for T790M mutation testing when progression on EGFR TKIs (follow up biopsy if the result is negative)?

**Old Key Questions (from 2013 guideline)**

- Has new data emerged to warrant changing the original recommendations?

**Contents**

- Introduction
- New Key Questions
- Old Key Questions

**NGS Considerations**

- Strengthen previous statement
- Benefits vs availability
- Multiplexed genetic sequencing preferred over multiple single-gene tests for selecting lung adenocarcinomas for targeted therapy beyond EGFR, ALK, and ROS1—specific circumstances
Cytology Specimen Revision Considerations

- Previously, cell blocks preferred over cytology smears
- Cytology smears or cell blocks, both suitable for lung cancer biomarker molecular testing.

Introduction References


ROS1 References

RET References


BRAF References


MET References


ERBB2/HER2 References

KRAS references


KRAS references


Squamous Cell Carcinoma References


Squamous Cell Carcinoma References


ALK IHC References