



# WHO 2015: Small sample diagnosis in lung cancer

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**Inserm**

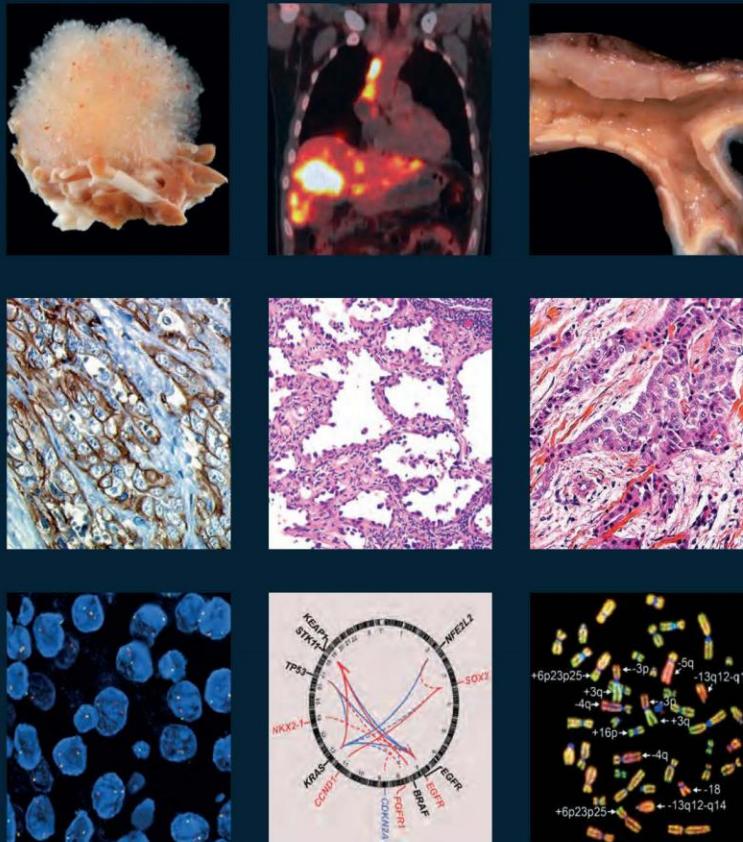
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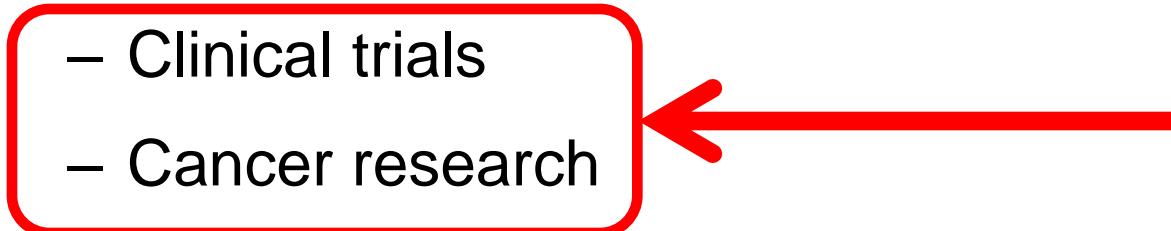
# WHO Classification of Tumours of the Lung, Pleura, Thymus and Heart

Edited by

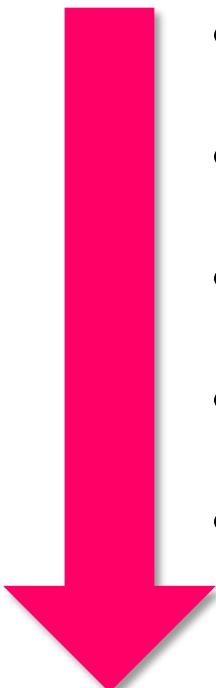
William D. Travis, Elisabeth Brambilla, Allen P. Burke, Alexander Marx, Andrew G. Nicholson



# What is a WHO classification?

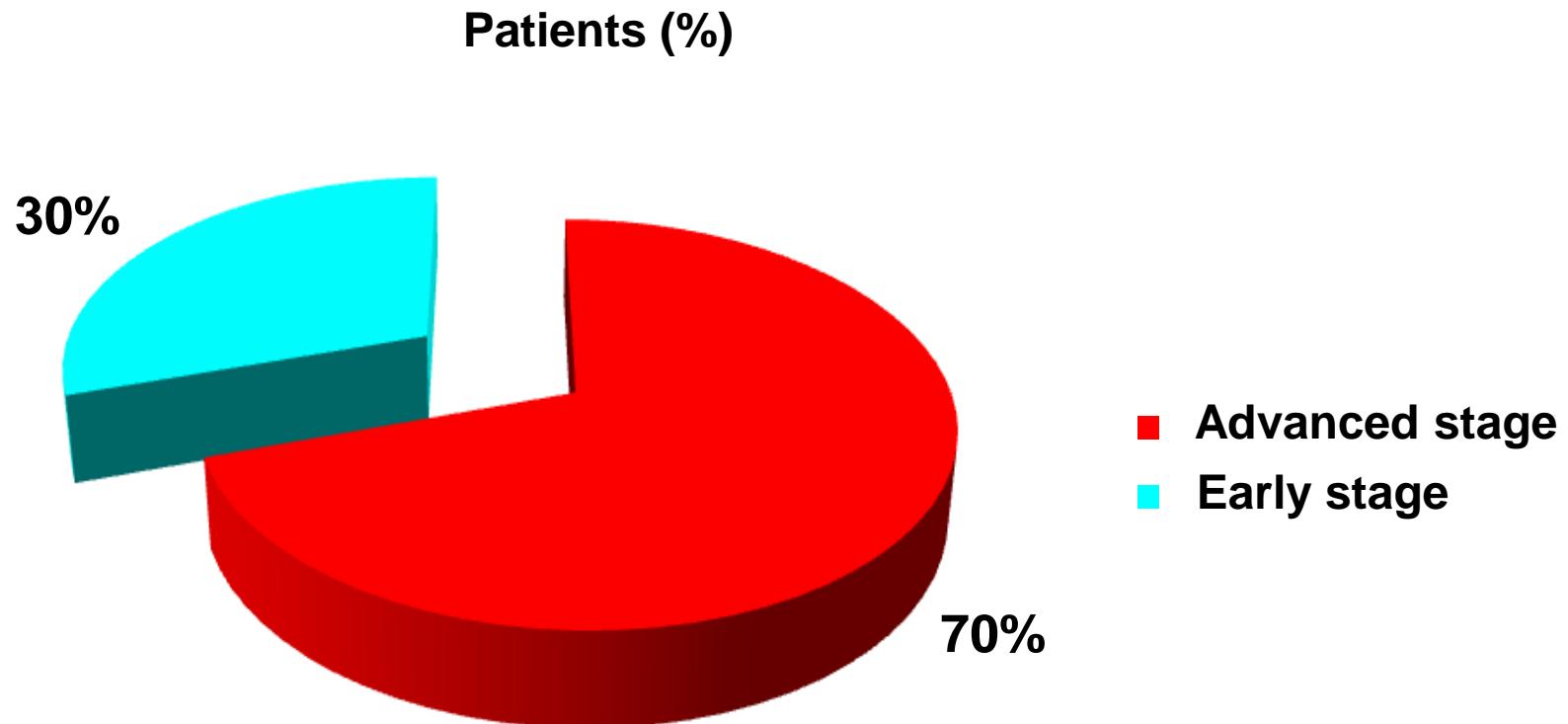
- A pathological and genetic classification and grading of human tumours designed to be accepted and used worldwide
  - Provides standard criteria for
    - Pathology diagnosis
    - Clinical practice
    - Cancer registration
    - Epidemiological studies
    - Clinical trials
    - Cancer research
- 

# **Increasing complexity in diagnostic tool**

- 1967 WHO
  - 1981 WHO
  - 1999 WHO
  - 2004 WHO
  - 2015 WHO
- 
- H&E
  - H&E & mucin
  - H&E, EM & IHC
  - H&E, EM, IHC & genetics
  - H&E, cytology, IHC, mucin, genetics, radiology

**INCREASING RELEVANCE FOR  
PERSONALISED MEDICINE**

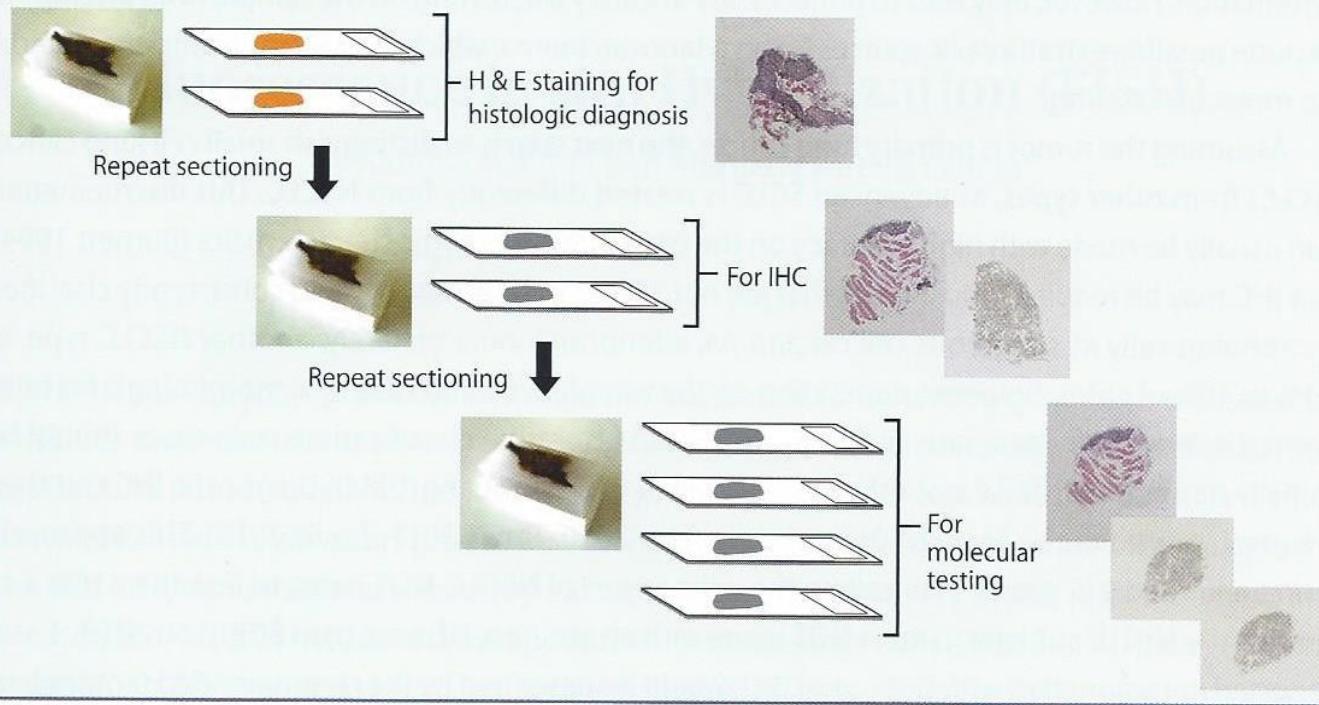
# Decreasing sample size for diagnosis: 70% of patients are diagnosed at an advanced stage (small biopsies)



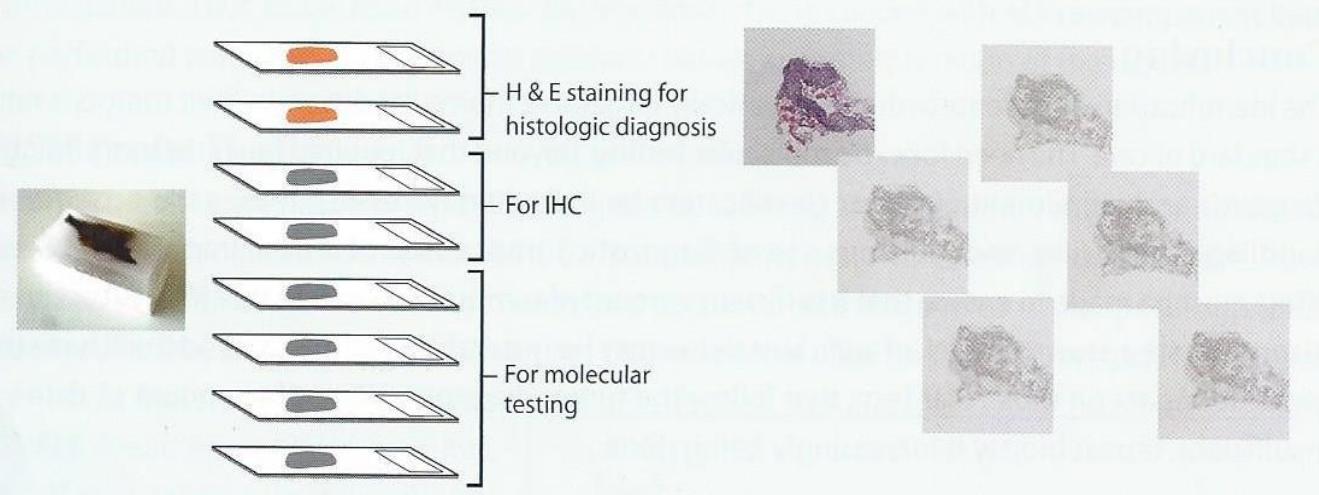
# 2015 WHO classification: impact on management of advanced lung cancer patients

- Criteria/terminology for small biopsies and cytology
- More accurate histological subtyping
- Strategic management of small tissues
- Streamlined workflow for molecular testing
- Need for local multidisciplinary team

## Common procedure to date



## The procedure in era of molecularly targeted drugs



# 2015 WHO Classification of resected lung adenocarcinoma

## Adenocarcinoma

- Lepidic adenocarcinoma<sup>e</sup>
- Acinar adenocarcinoma
- Papillary adenocarcinoma
- Micropapillary adenocarcinoma<sup>e</sup>
- Solid adenocarcinoma

- Invasive mucinous adenocarcinoma<sup>e</sup>

- Mixed invasive mucinous and nonmucinous adenocarcinoma

- Colloid adenocarcinoma

- Fetal adenocarcinoma

- Enteric adenocarcinoma<sup>e</sup>

- Minimally invasive adenocarcinoma

- Nonmucinous

- Mucinous

- Preinvasive lesions

- Atypical adenomatous hyperplasia

- Adenocarcinoma in situ<sup>e</sup>

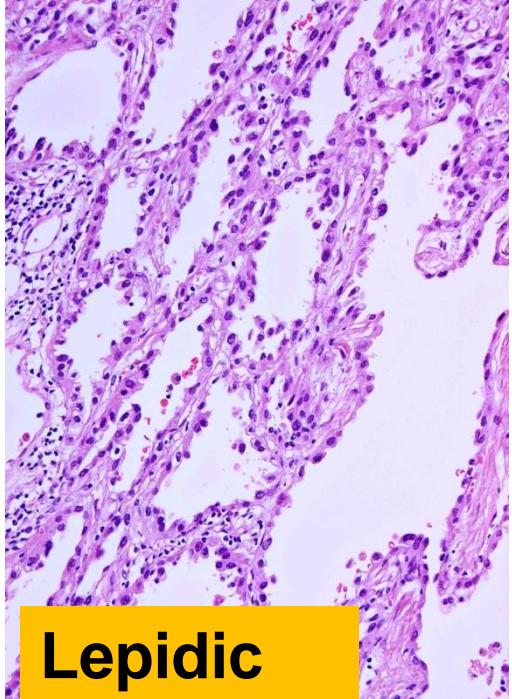
Features  
on small  
samples

Not diagnosed  
on small  
samples

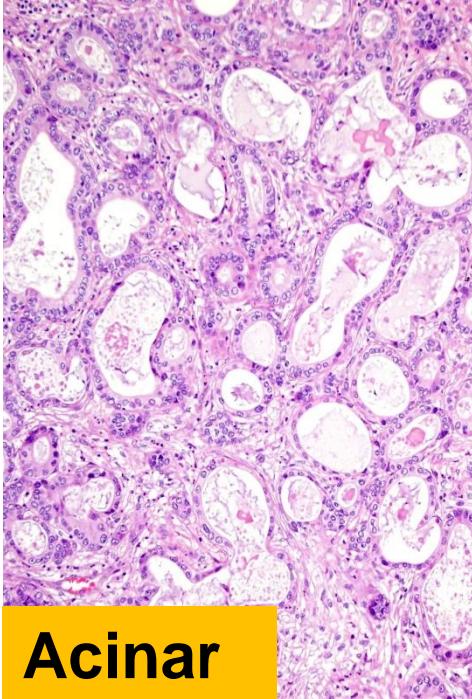
- Comprehensive histologic subtyping – 5% increments
- Solid ADC – mucin + or TTF-1 + (from large cell ca)

→ 100% 5y. survival expectancy after surgery

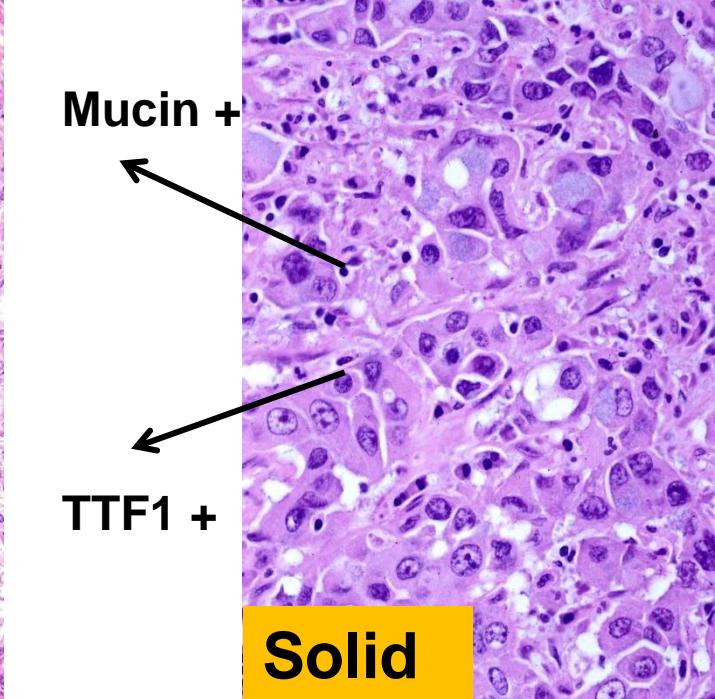
→ 100% 5y. survival  
→ Not to treat / Ct scan



Lepidic



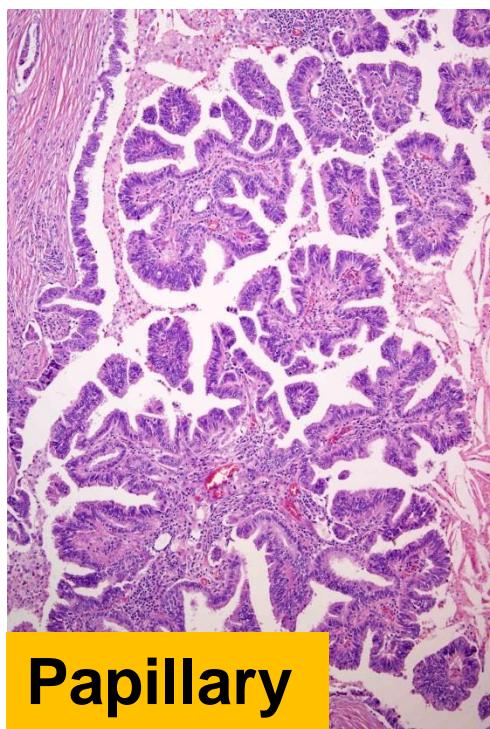
Acinar



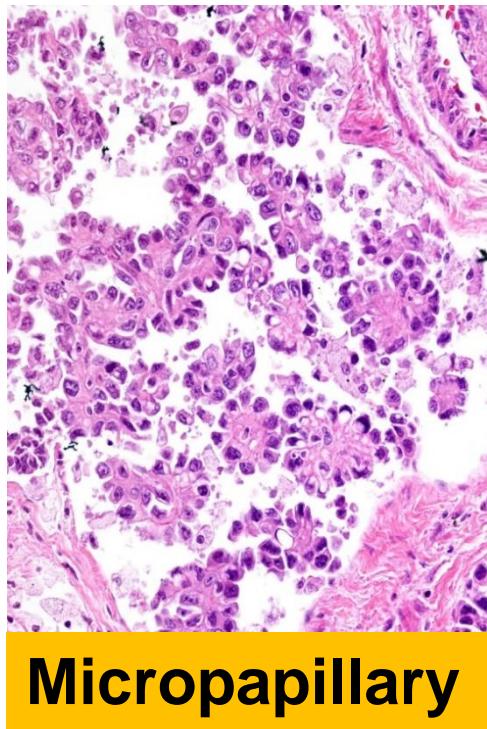
Solid



If pure , an  
invasive  
pattern cannot  
be excluded

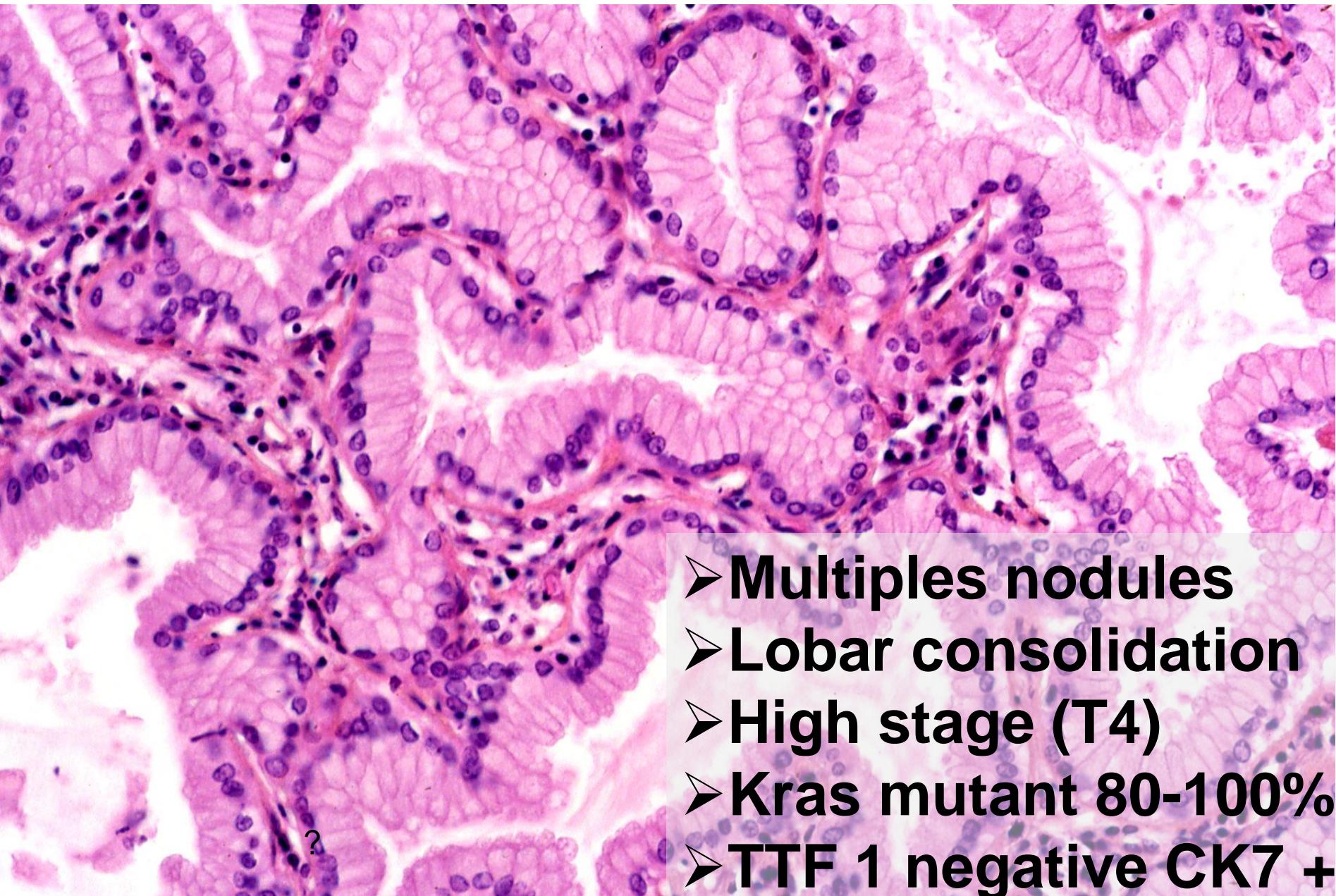


Papillary



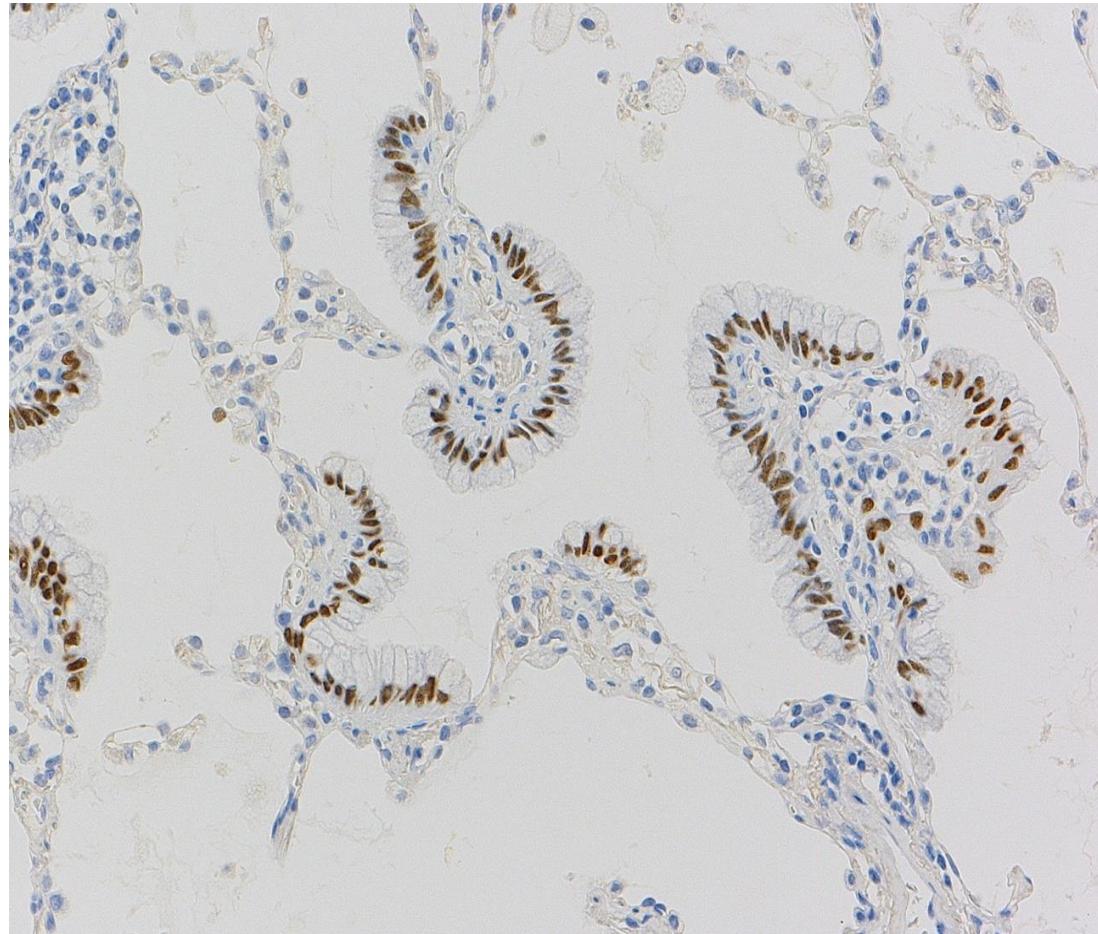
Micropapillary

# Variant : Invasive mucinous adenocarcinoma



- Multiples nodules
- Lobar consolidation
- High stage (T4)
- Kras mutant 80-100%
- TTF 1 negative CK7 +

# HNF4- $\alpha$ : a marker of invasive mucinous adenocarcinoma

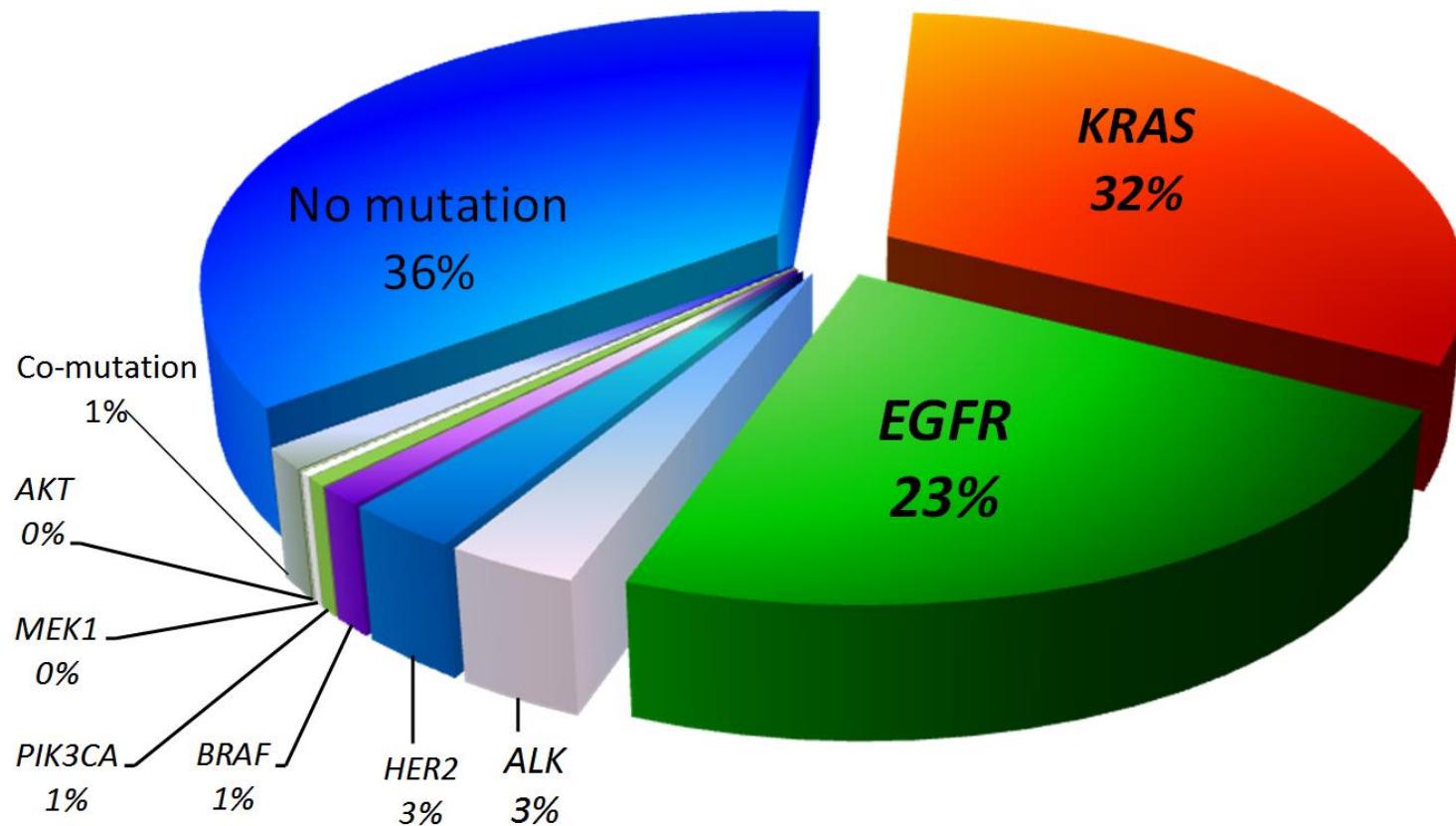


Sugano M et al: Am J Surg Pathol 37:211-8, 2013

# Therapeutic advances have impacted the need for more accurate histological diagnosis and molecular testing on small samples

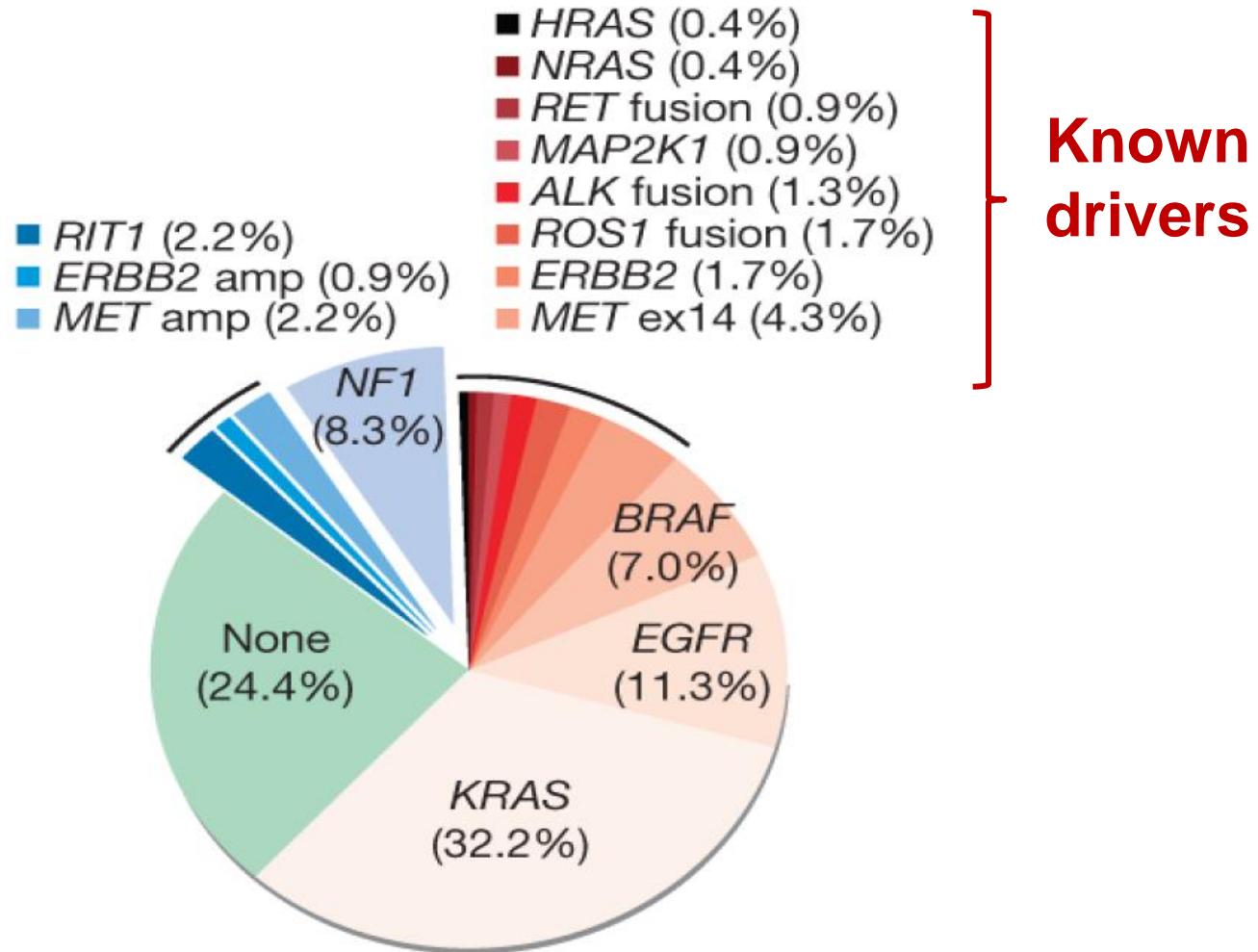
- **Predictive of response**
  - *EGFR* mutation (adenocarcinoma) – EGFR TKIs
  - Adenocarcinoma or NSCLC-NOS – pemetrexed
  - *ALK* fusion/ *ROS1* fusion, Met Ex.14 Del.  
(adenocarcinoma) – crizotinib/ceritinib /Cabozantinib
- **Predictive of toxicity**
  - Bevacizumab – contraindicated for life-threatening haemorrhage in squamous cell carcinoma

# Driver mutations found in 65% of adenocarcinoma specimens



# Adenocarcinoma: new candidate drivers activating the RTK/RAS/RAF pathway

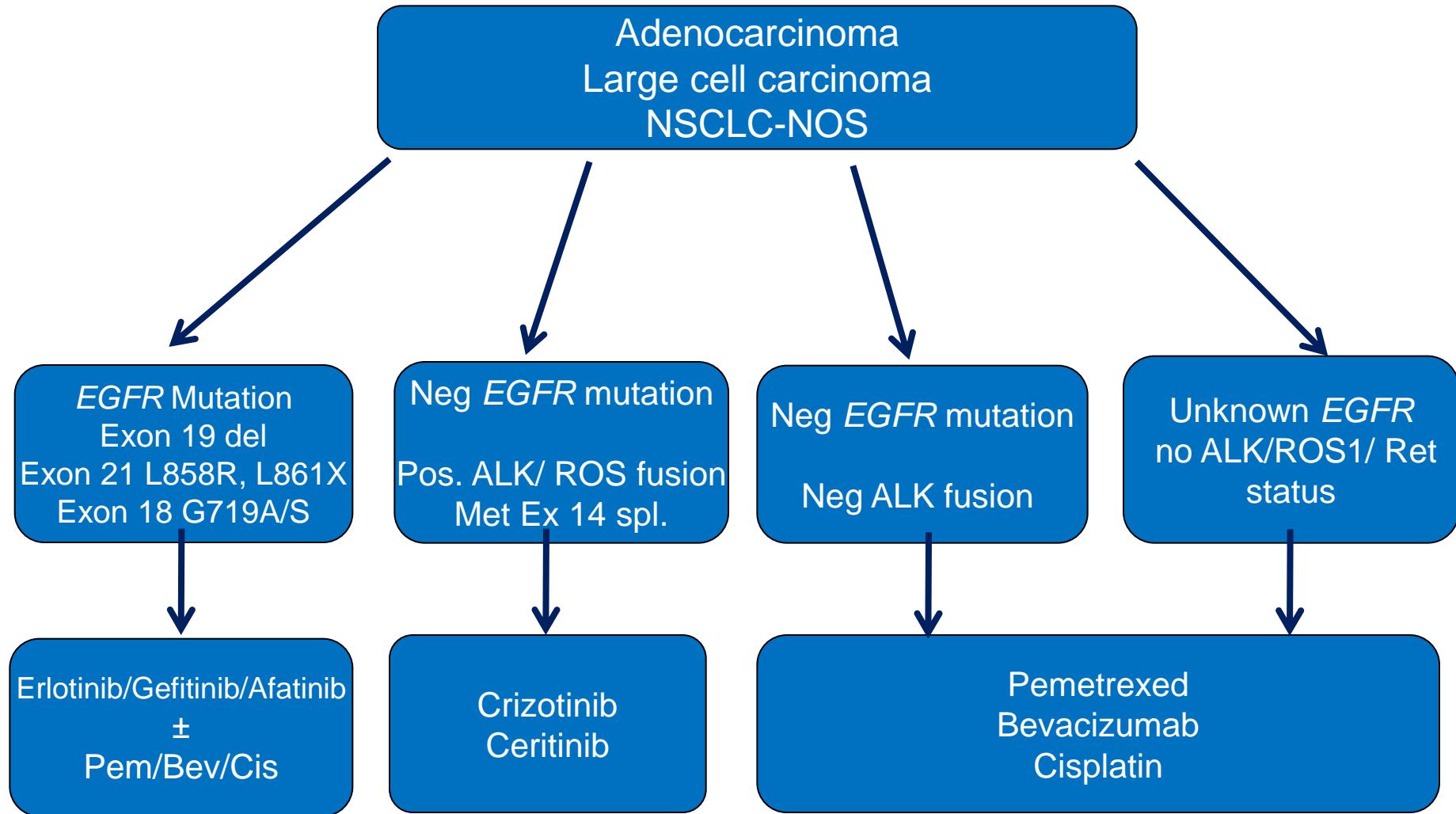
New candidate drivers {



# Driver mutations are targets for molecular-based therapy

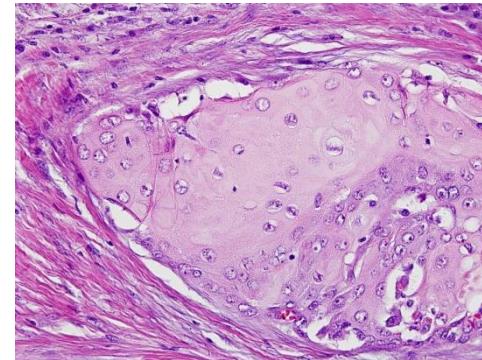
Target	Drug
<i>EGFR</i>	Erlotinib/gefitinib Afatinib
<i>ALK</i> fusions	Crizotinib Ceritinib
<i>BRAF</i> V600E	Dabrafenib
<i>ROS1</i> fusions	Crizotinib
<i>RET</i> fusions	Cabozantinib
<i>MET</i> splice site Exon 14 mutations	Cabozantinib (and crizotinib)

# Treatment of advanced NSCLC is based on histology and genetics



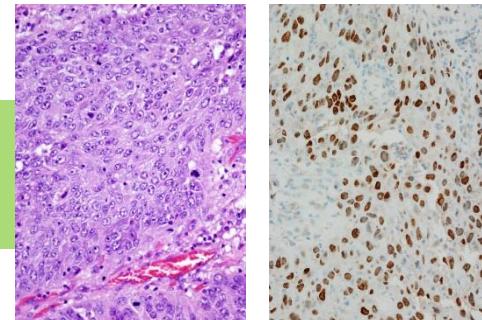
# WHO 2015 classification for squamous cell carcinoma

- Keratinising



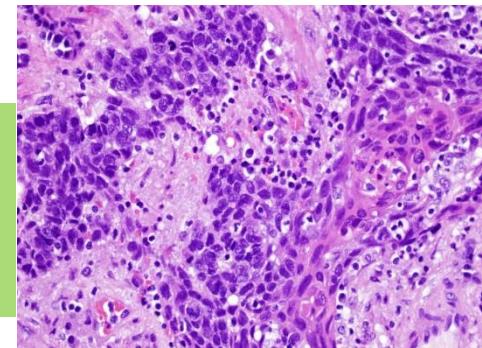
- Non-keratinising

IHC P40 positive,  
TTF-1 negative

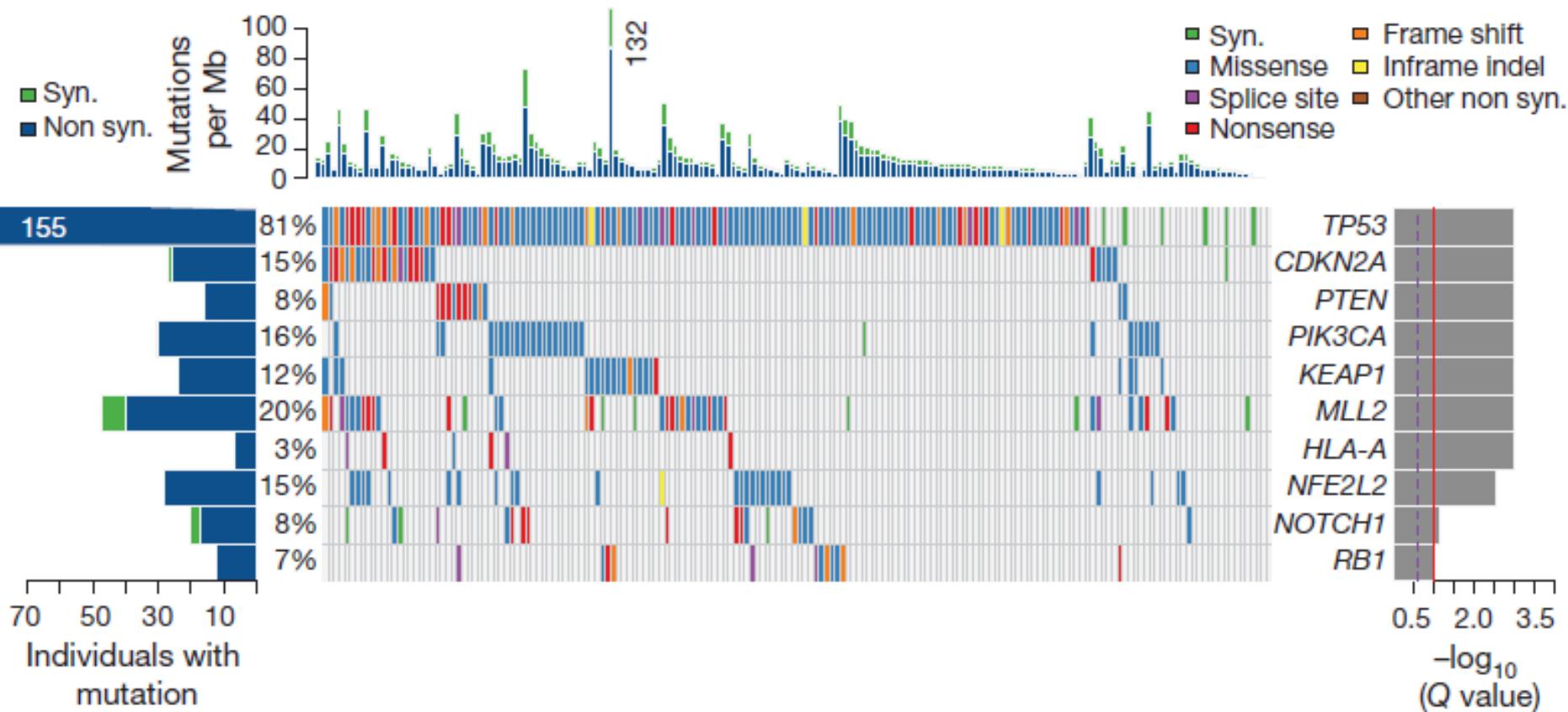


- Basaloid

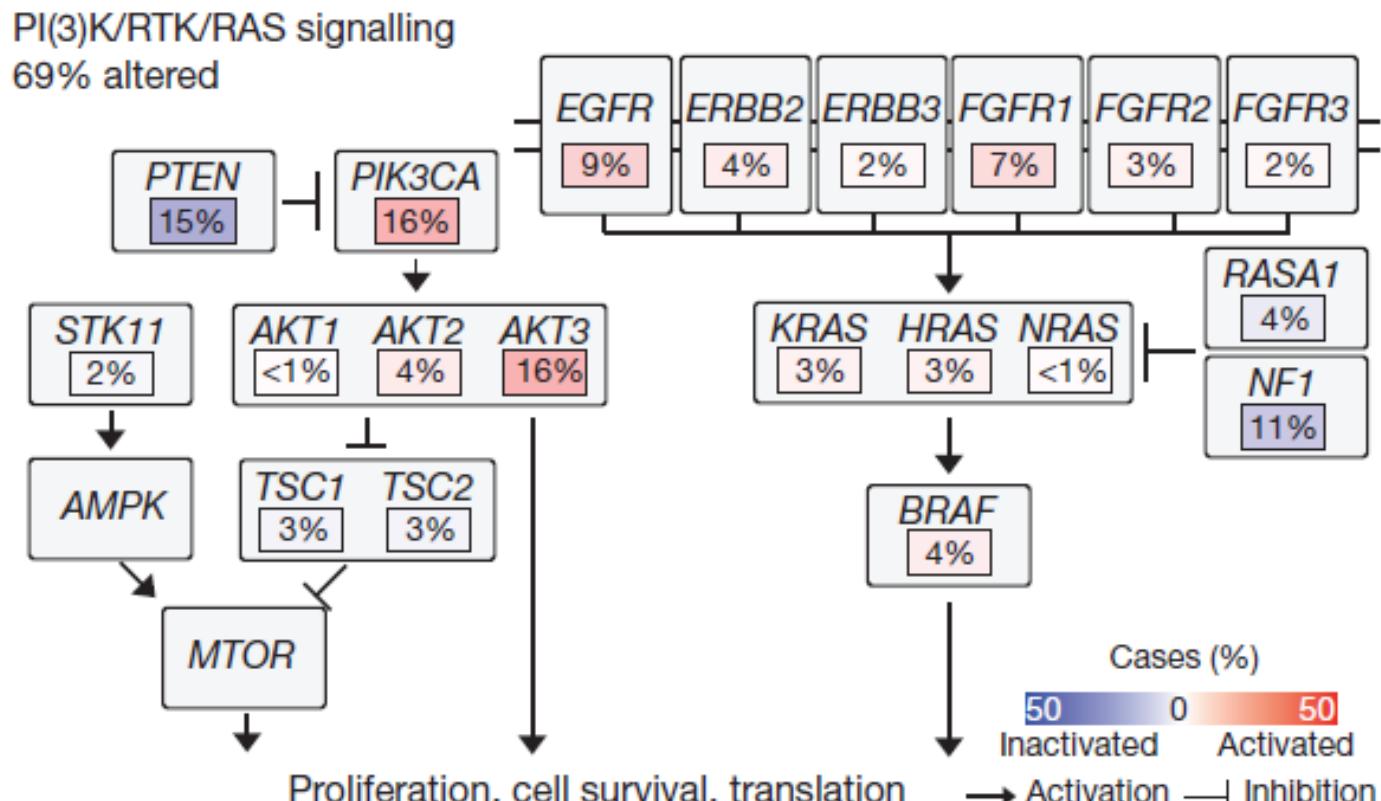
IHC (+p40, -TTF1 & NE  
markers)  
r/o LCNEC & SCLC



# Significant mutated genes in squamous cell carcinoma



# Alterations in targetable oncogenic pathways in 69% of lung SQSC



## Alteration pattern



# **Subtyping of morphologically undifferentiated non small cell lung carcinoma (formerly large cell carcinoma)**

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## **Adenocarcinoma solid subtype**

- Positive for TTF1 or mucin
- Negative for P40/P63, CK5/6

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## **Non-keratinising squamous cell carcinoma**

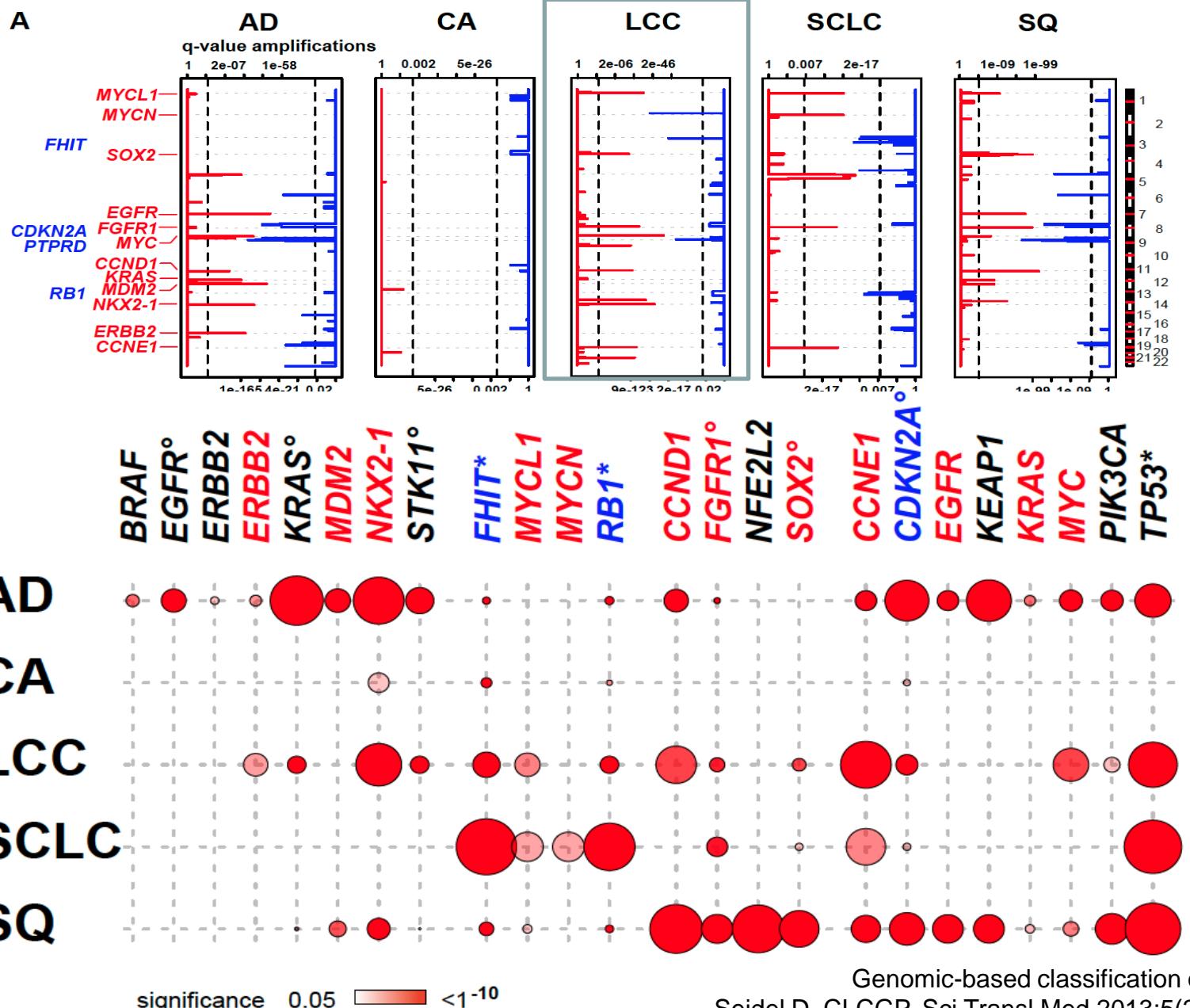
- Diffusely positive for P40/P63, and/or CK5/6
- Negative for TTF1 or mucin

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## **Adenosquamous carcinoma**

- Positive for both ADC and SCC in distinct populations

# Driver mutations and genomic profiles are histology-related



# 2015 WHO terminology for small biopsies/cytology

2015 WHO resections	Small biopsy/cytology
ADENOCARCINOMA Lepidic Acinar Papillary Micropapillary Solid	<p><i>Morphological adenocarcinoma patterns clearly present:</i> Adenocarcinoma, describe identifiable patterns present</p>
No 2004 WHO counterpart – most will be solid adenocarcinomas	<p><i>Morphological adenocarcinoma patterns not present (supported by special stains; i.e TTF-1 +; p40 -):</i> Non-small cell carcinoma, favour adenocarcinoma</p>
SQUAMOUS CELL CARCINOMA Keratinising Non-keratinising Basaloid	<p><i>Morphologic squamous cell patterns clearly present:</i> Squamous cell carcinoma</p>
No 2004 WHO counterpart	<p><i>Morphological squamous cell patterns not present (supported by stains; i.e. p40+, TTF-1 -):</i> Non-small cell carcinoma, favour squamous cell carcinoma</p>
LARGE CELL CARCINOMA	<p>Non-small cell carcinoma, not otherwise specified (NOS)</p>

# Microscopy

**Previous NSCLC  
NOS: 20–40%**

**2015**

**classification**

Squamous cell carcinoma

**20–30%**

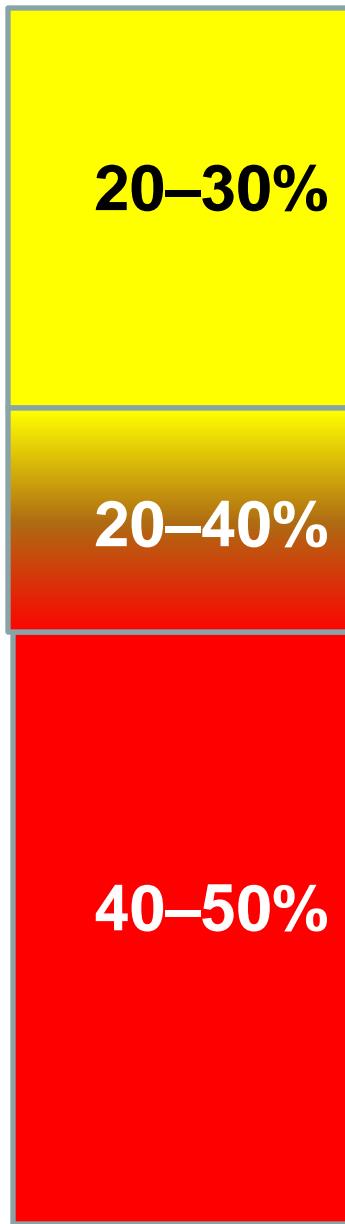
NSCLC NOS

**20–40%**

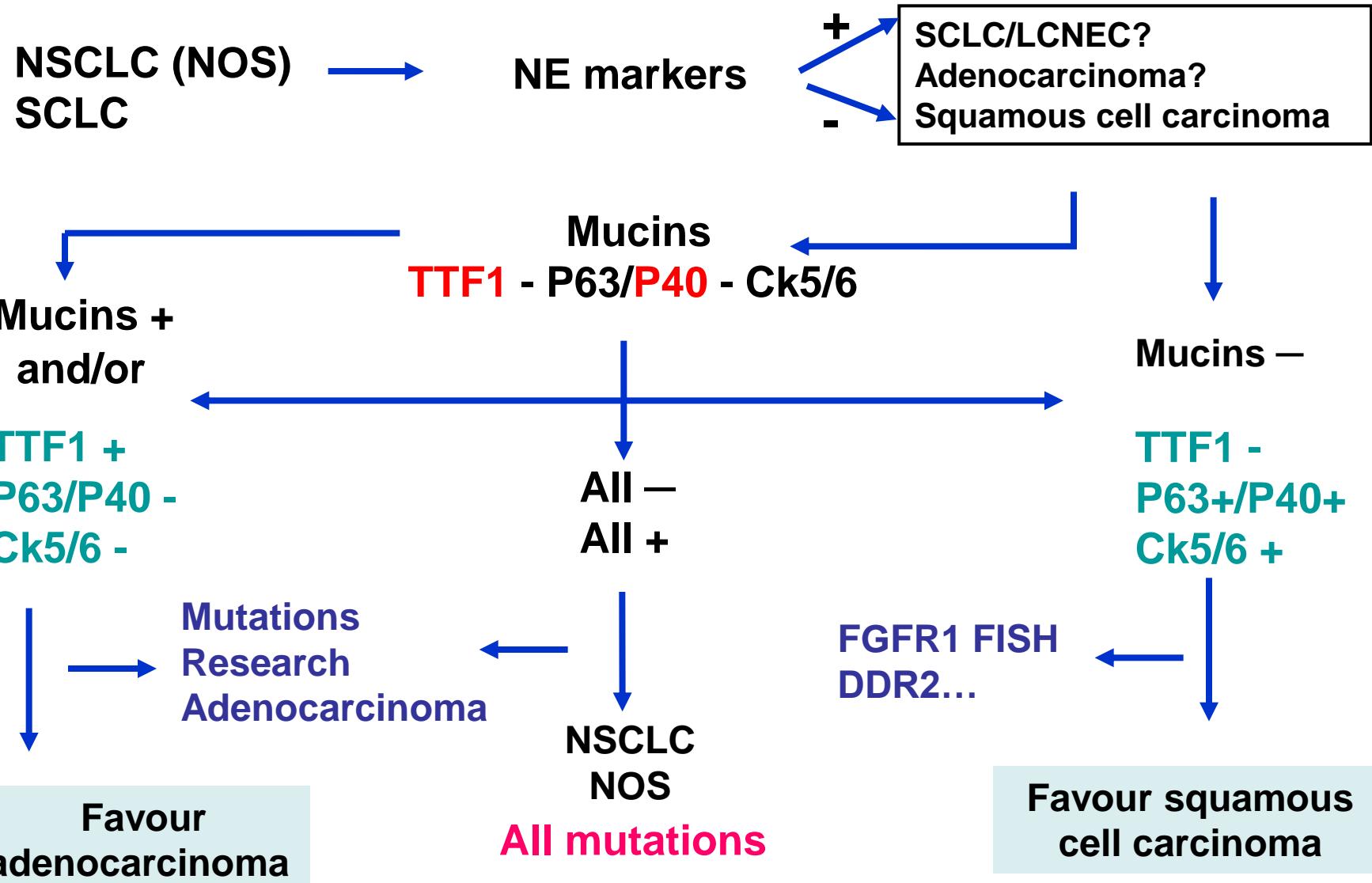
Adenocarcinoma

**40–50%**

**NSCLC NOS  
objective <5%**

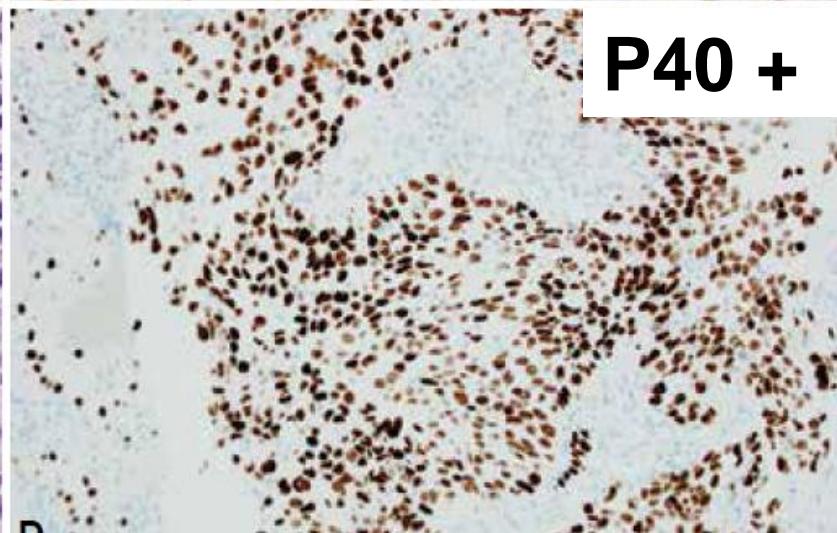
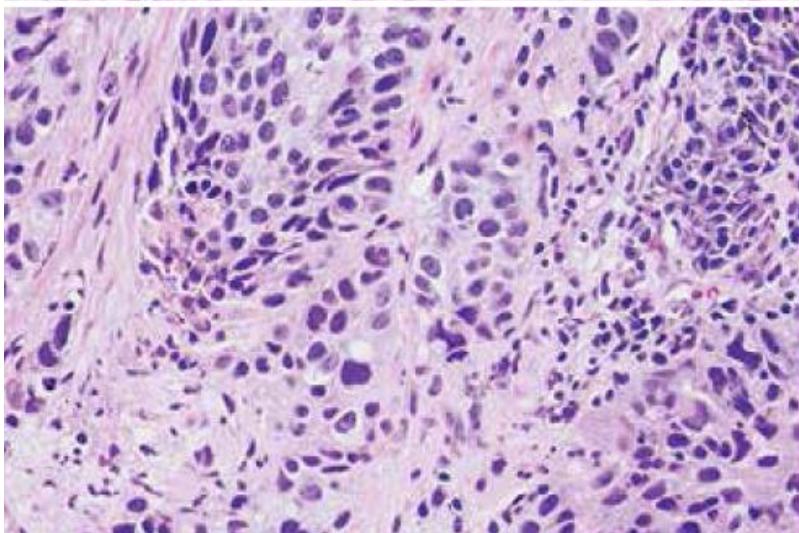
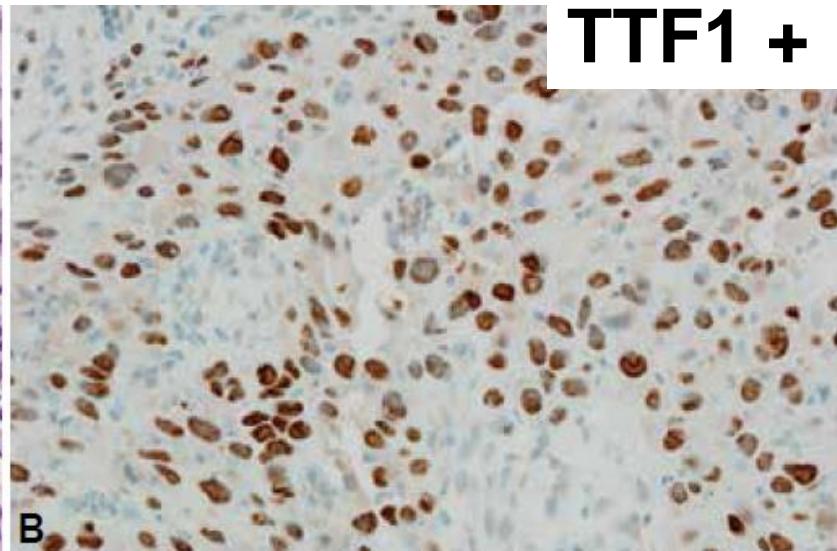
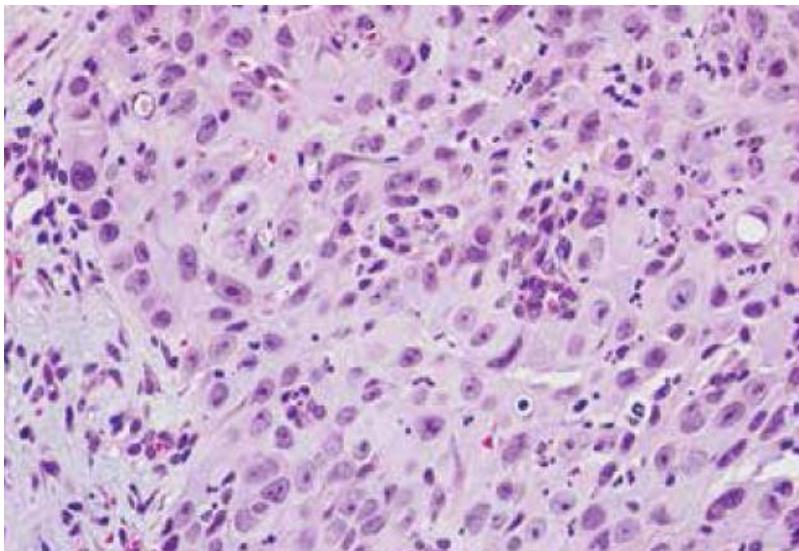


# Diagnosis on small biopsies (70%) minimise the use of NSCLC (NOS)



# NSCLC: biopsies

Favour adenocarcinoma

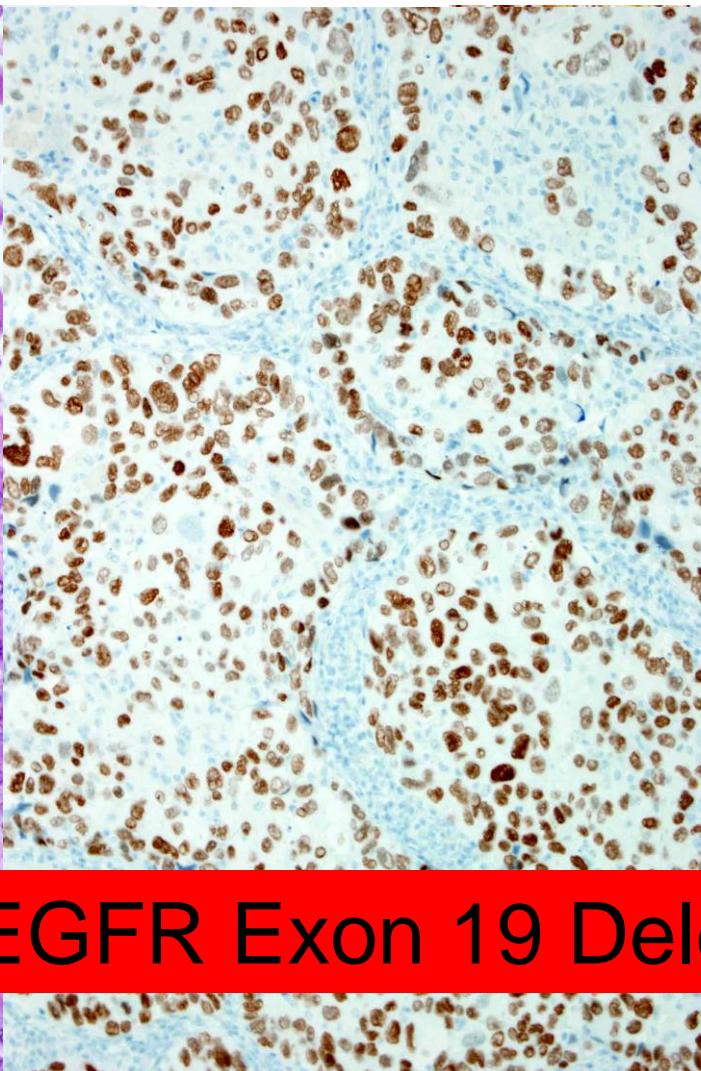
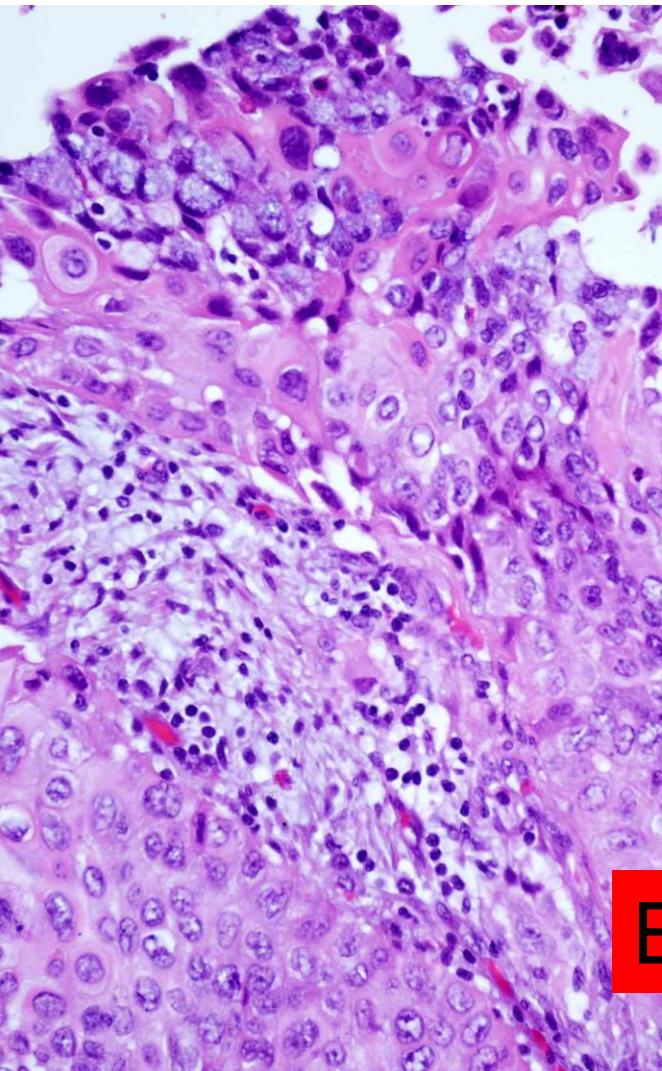


Favour squamous cell carcinoma

# Adenocarcinoma, solid pseudosquamous

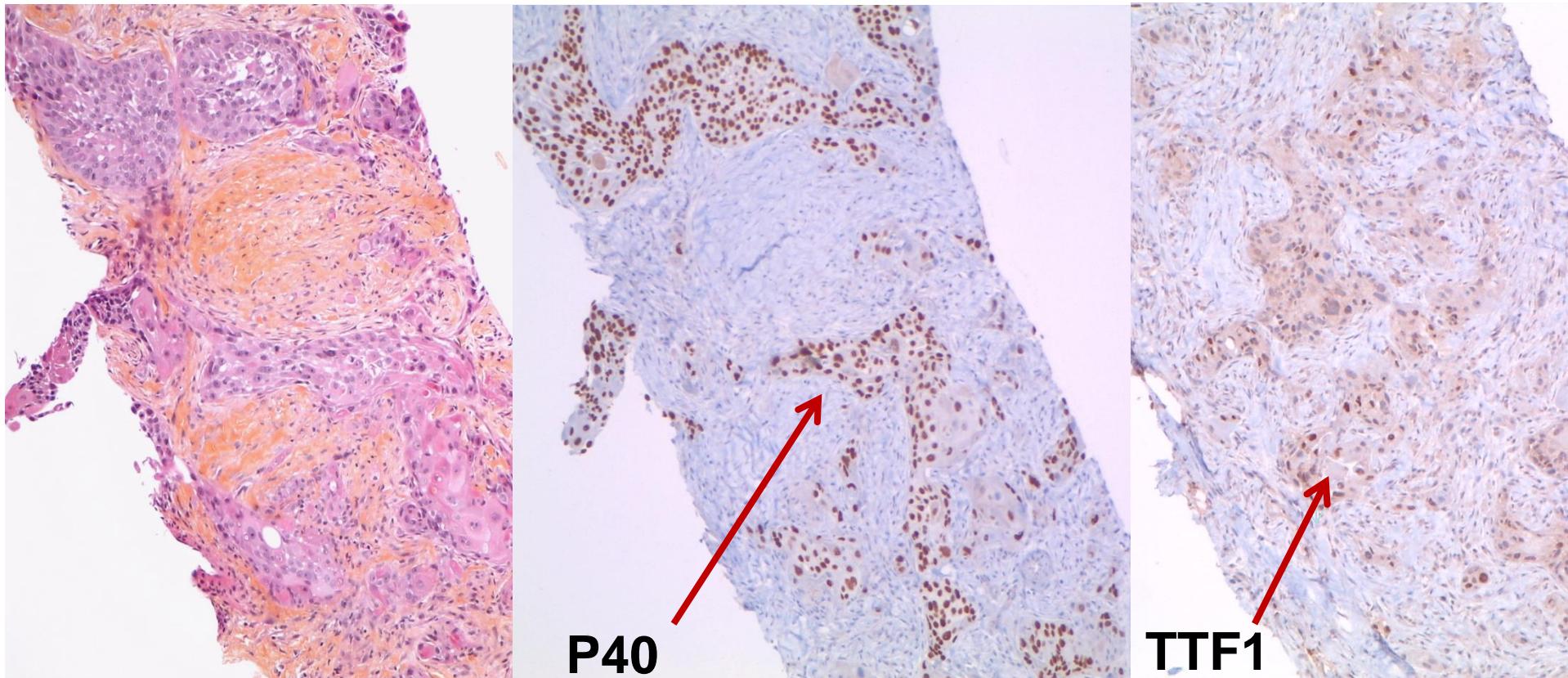
TTF-1

Mucicarmine



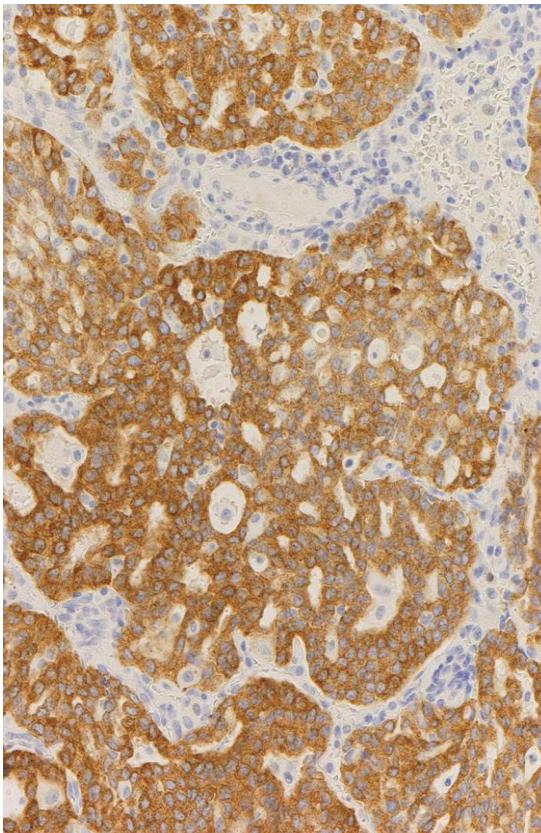
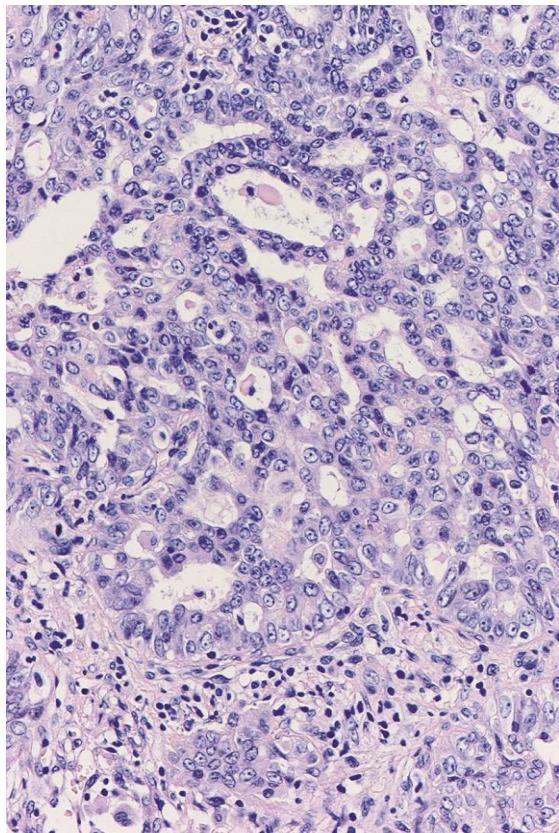
EGFR Exon 19 Deletion

# Mutant EGFR adenocarcinoma resistant to TKI

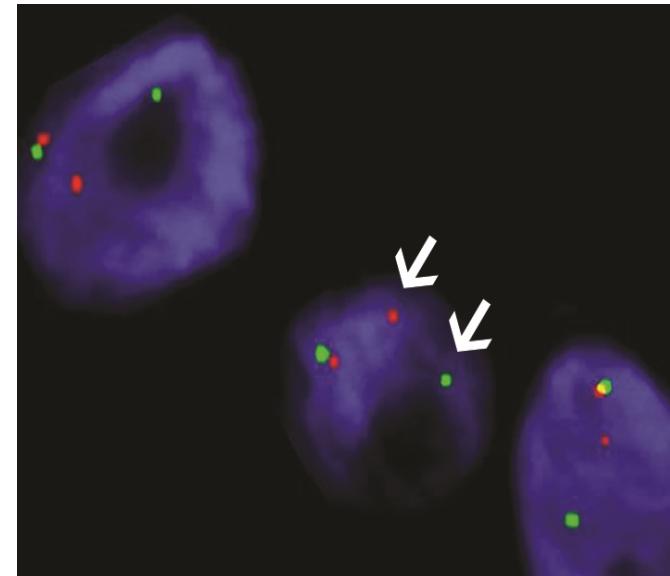


Switch to (or therapeutic selection of ) a squamous carcinoma phenotype → adeno-squamous carcinoma  
90 % ADC / 10% SCC on original resected tumor

# Adenocarcinoma: *ALK* Fusion



ALK IHC (D5F3  
ou 5A4)



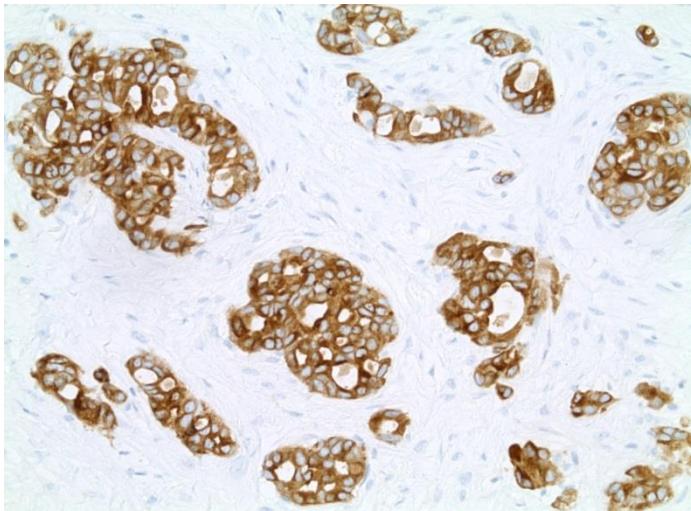
ALK  
FISH

# Adenocarcinoma : ROS1 fusion

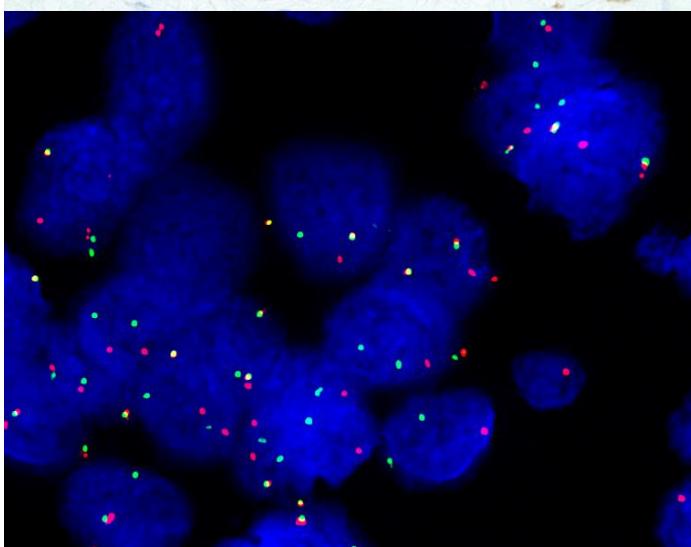
**ROS1 IH 2+ compared to ROS1 FISH :100% sensitivity ; 96,2% specificity**

121 ADC triple negative (*EGFR ,RAS,ALK*): 9 cases *ROS1* rearranged (7.5%)

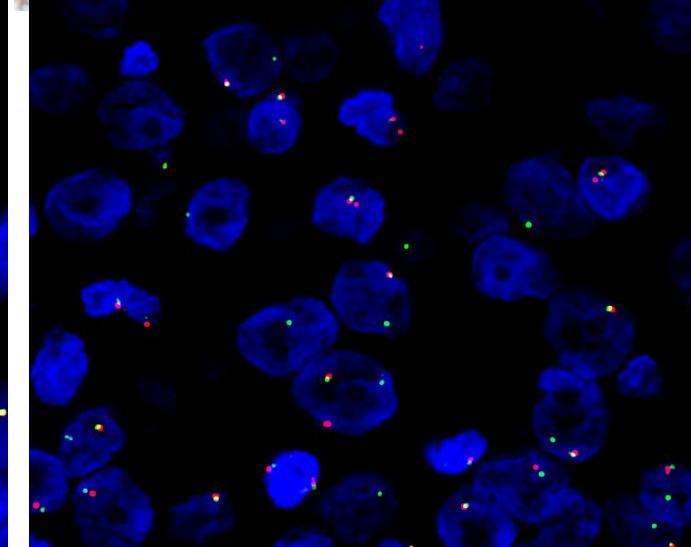
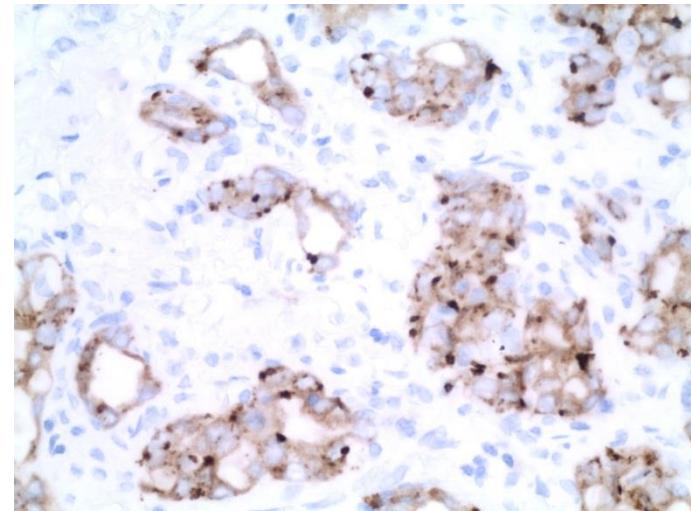
ROS1  
IH+++



ROS1  
FISH +



ROS1  
IH++  
Golgi



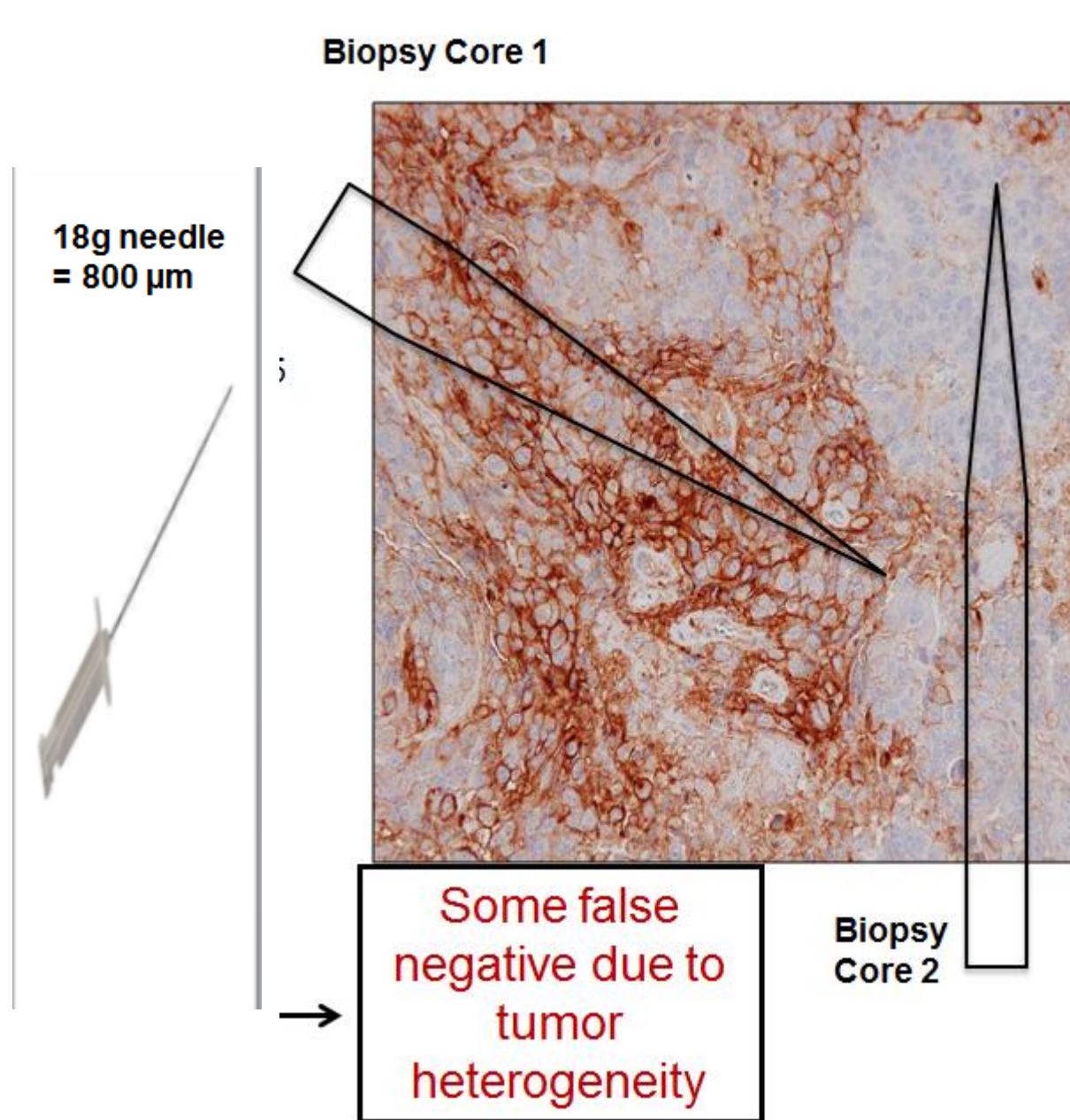
ROS1  
FISH +

# New biomarkers recommended or validated for FDA-approved drugs

- Most consist of a single genetic mutation, gene amplification, or translocation/rearrangement
- FGFR1 amplification in squamous cell carcinoma<sup>1</sup>
  - Predictive value for FGFR inhibitors questioned ?
- PDL1 recommended, in course of validation, for immune checkpoint blockade<sup>2-3</sup>
  - Predictor of survival benefit for pembrolizumab, nivolumab in non-squamous carcinoma and atezolizumab

1. Weiss J et al. Sci Transl Med 2010;2(62):62ra93
2. Kroemer M et al. Transfus Clin Biol 2015;22(3):132-40
- 3-Garon E et al. N;E.J.M .2015 372,21

# Pitfalls with heterogeneous markers :PDL1



# **Summary: diagnosis on small samples, implications for clinical practice**

- WHO 2015 adenocarcinoma classification now requires histopathology and molecular testing for diagnosis, treatment , clinical trials and genetic research .
- Apply new strategies for diagnosis and biomarkers to small biopsies and cytology .
- Minimise the use of small biopsies for diagnostic purposes, with maximum of 3 IHC markers to maximise that for biomarkers studies .
- Reassign most “ non small cell lung carcinomas-NOS ” to other histological classes according to 2 markers (TTF1, P40): restrict NSCLC/ NOS to tumour lacking both morphological or IHC differentiation .

# 2015 WHO editors



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