THE ROLE OF Ki-67 IN PULMONARY NEUROENDOCRINE TUMORS

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2015 WHO CLASSIFICATION
NEUROENDOCRINE TUMORS

- **Small cell carcinoma**
  - Combined SCLC
- **Large cell neuroendocrine carcinoma**
  - Combined LCNEC
- **Carcinoid tumor**
  - Typical carcinoid
  - Atypical carcinoid
  - Preinvasive: Diffuse idiopathic pulmonary neuroendocrine cell hyperplasia
LUNG NE TUMOR FREQUENCY

- NON-NE CARCINOMAS: 75-80.0%
- SCLC: 15-20.0%
- LCNEC: 3.0%
- CARCINOID: 1-2.0%
- ATYPICAL CARCINOID: 0.1-0.2%
2015 WHO CLASSIFICATION
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  - Typical carcinoid
  - Atypical carcinoid
LUNG CARCINOMA

SMALL CELL VS NON SMALL CELL CA

SCLC 15.0%

NSCLC 85.0%
Pulmonary neuroendocrine (carcinoid) tumors: European Neuroendocrine Tumor Society expert consensus and recommendations for best practice for typical and atypical pulmonary carcinoids

M. E. Caplin1*, E. Baudin2, P. Ferolla3, P. Filosso4, M. Garcia-Yuste5, E. Lim6, K. Oberg7, G. Pelosi8, A. Perren9, R. E. Rossi1,10 & W. D. Travis11 the ENETS consensus conference participants†

Evidence Based Recommendations:
Systematic review of literature
International Multidisciplinary Panel

LUNG NE TUMORS: SURVIVAL

Survival Functions

5 yr
TC: 97%
AC: 51.6%
LCNEC: 15.5%
SCLC: 12.2%

Travis WD et al, in preparation. 515 AFIP Cases: TC-92; AC-128, LCNEC – 154, SCLC – 141; p<0.0001
Ki-67

- Ki-67 or MKI67 antigen identified by monoclonal antibody Ki-67, is a 359-kD non-histone nuclear protein with short half-life, which is encoded by the 15 exon-spanning \textit{MKI67} gene mapping to chromosome 10q26.2.

- Plays an essential role in control and timing of cell proliferation

- Name from the city of Kiel, Germany where the antibody was first raised and the number 67 from the clone position in the original 96-well plate generated immunizing mice with nuclei of the lymphoma cell line L428
DATA IN LUNG FAR BEHIND GI/PANCREAS

- No comparative studies evaluating different methods for Ki-67 in lung NE tumors
- Most papers: Ki-67 LI in hot spot areas taking into account all nuclear signals
- Counting at least 2000 consecutive tumor cells in hot spot at 40X or 2mm² – possible in areas of counting mitoses
- In small bx/cytology – evaluate all tumor cells
- Eye ball estimation differs little for experienced pathologists
# 2017 WHO CLASSIFICATION OF PANCREATIC NE NEOPLASMS

<table>
<thead>
<tr>
<th>Tumor Type</th>
<th>Ki-67 Index</th>
<th>Mitotic Index</th>
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<tbody>
<tr>
<td><strong>Well Differentiated NEN</strong></td>
<td></td>
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<tr>
<td>NET G1</td>
<td>&lt;3%</td>
<td>&lt;2/10 HPF</td>
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<tr>
<td>NET G2</td>
<td>3-20 %</td>
<td>2-20/10 HPF</td>
</tr>
<tr>
<td>NET G3</td>
<td>&gt;20%</td>
<td>&gt;20/10 HPF</td>
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<tr>
<td><strong>Poorly Differentiated NEN</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NE Carcinoma (NEC) G3</td>
<td>&gt;20%</td>
<td>&gt;20/10 HPF</td>
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<tr>
<td>Small cell type</td>
<td></td>
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<tr>
<td>Large cell type</td>
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<tr>
<td><strong>Mixed NE-NonNE neoplasm (MiNEN)</strong></td>
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2017 WHO CLASSIFICATION OF PANCREATIC NE NEOPLASMS

- Ki-67 index based on at least 500 cells in areas of higher nuclear labeling (“hot spots”); mitoses in 50 HPF, (2.0 mm²) in areas of higher density and expressed per 10 HPF (2.0 mm²)

- The final grade is based on whichever index (mitotic rate or Ki-67) places the tumor in the highest grade category.

- For assessing Ki67, casual visual estimation (eyeballing) is not recommended; manual counting of printed images is suggested
Role of Ki-67 in Lung NET

- Are there relevant technical issues to Ki-67 IHC and evaluation of results?
- Is there a diagnostic role for Ki-67 LI in lung NET?
- Is there a prognostic role for Ki-67 LI?
- Is there an established role for Ki-67 LI in tumor grading?
- Is there a predictive role for Ki-67 LI in therapeutic decisions?

Typical and Atypical Pulmonary Carcinoid Tumor Overdiagnosed as Small-Cell Carcinoma on Biopsy Specimens

A Major Pitfall in the Management of Lung Cancer Patients

Giuseppe Pelosi, MD, * Jaime Rodriguez, MD, † Giuseppe Viale, MD, * and Juan Rosai, MD †

- Careful evaluation of H&E sections & Ki-67 index
- Carcinoid tumors <20% Ki-67
- SCLC >50% Ki-67

NE TUMORS: Ki-67

TC

AC

LCNEC

SCLC
NE TUMORS: Ki-67

KI-67 CAN BE USEFUL IN DIAGNOSIS OF NE TUMORS

TC - <5%
AC – 5-20%
LCNEC – 60-100%
SCLC – 80-100%
Multiple Ki-67 Reagents

- Polyclonal Ki-67
- Monoclonal MIB-1-3-5
- IND.64
- JG-67-2a
- Ki-S1
- Ki-S3
- Ki-S5
- Ki-S11
- Mib-1
Ki-67 IHC Problem Issues

- Technical issues to Ki-67 IHC and evaluation
- Different Abs – most use MIB-1
- Different Ab dilutions (1:25-1:1800)
- Different antigen retrieval methods
- Different specimen types: small bx, resection, cytology
Methods of Evaluation of Ki-67

- Hot spot vs Hot and Cold spot vs random
- Average vs mean labeling
- Staining extent (labeling index vs various categorical groupings)
- (4-8 fields at 20X or 40X; 2 mm² at 25X; random 1 mm²; 400-2000 cells)
  - Eyeball estimate
  - Manual counting (from printed image)
  - Digital Image Analysis (scanned slides)
- Quality of staining
- Reproducibility

Is there a diagnostic role for Ki-67 LI in lung NET?

- Yes as an aid in separating carcinoid tumors from high grade SCLC & LCNEC, particularly in small crushed biopsies
- Not as an aid in separating TC from AC where the main criteria are based on morphology alone
- Precise thresholds are not established for separating TC from AC

CRUSHED SCLC

AE1/AE3

CGA

Ki-67
LARGE CELL NEUROENDOCRINE CARCINOMA: CORE BIOPSY
LARGE CELL NEUROENDOCRINE CARCINOMA: CORE BIOPSY

CD56

Ki-67 >80%
LARGE CELL NEUROENDOCRINE CARCINOMA: CORE BIOPSY

CD56

Ki-67 >80%
Is there a prognostic role for Ki-67 LI?

- Possibly
- Ki-67 LI has been emerging as a promising prognostic factor in excised specimens of TC and AC
- However, existing data are conflicting and not conclusive to recommend Ki-67 LI as a prognostic marker in carcinoids

Is there an established role for Ki-67 LI in tumor grading

- No
- Grading in lung NET is inherent in the classification based on morphology, particularly mitoses and necrosis
  - TC – low grade
  - AC – intermediate grade
  - SCLC & LCNEC – high grade

Is there a predictive role for Ki-67 LI in therapeutic decisions?

- No
- Therapy is primarily based upon histologic classification (which includes grading) and stage
- No randomized clinical trials have been done to stratify patients according to Ki-67 LI in lung NE tumors

CONCLUSIONS

- Most accurate evaluation of Ki-67 LI is by the % of stained tumor cells on at least 500 cells in hot spot areas
- Lung NE tumors are classified by morphology, Ki-67 is not reliable to distinguish TC from AC
- Ki-67 is useful to distinguish carcinoids from SCLC or LCNEC
- GI/Pancreatic grading not recommended in lung
- More work is needed to understand the role of Ki-67 in pulmonary NET