Small Cell Lung Cancer (SCLC)

2025 Update

Markey Study of SCLC TIME

Jing Di MDPhD

Thèrése Bocklage MD

UK Department of Pathology and Laboratory Medicine

Drs Jing Di and Thèrése Bocklage declare no conflicts of interest

Learning Objectives

- Enumerate Small Cell Lung Carcinoma (SCLC) Basics
- Discuss Molecular Variants of SCLC
- Describe Tumor Immune Microenvironment of SCLC
- Compare Precision Therapy Potential Approaches

Small Cell Lung Cancer (SCLC) Basics

Definition: a high-grade neuroendocrine carcinoma of the lungs that is composed of small round blue cells

- Immunohistochemistry often used: INSM-1, chromogranin, synaptophysin, CD56, keratins, TTF-1
- Can occur in combination with other lung cancer types

Incidence: 13-15% of all lung cancers (200,000 deaths annually worldwide)

Location: central airways

Staging:

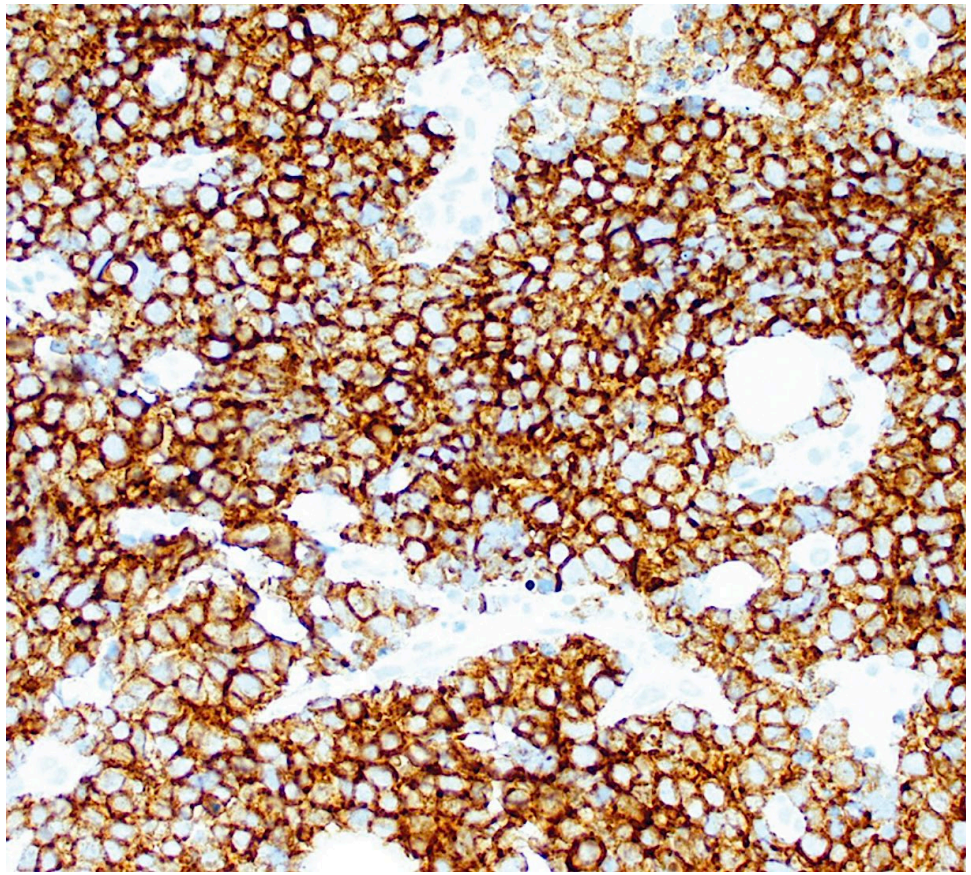
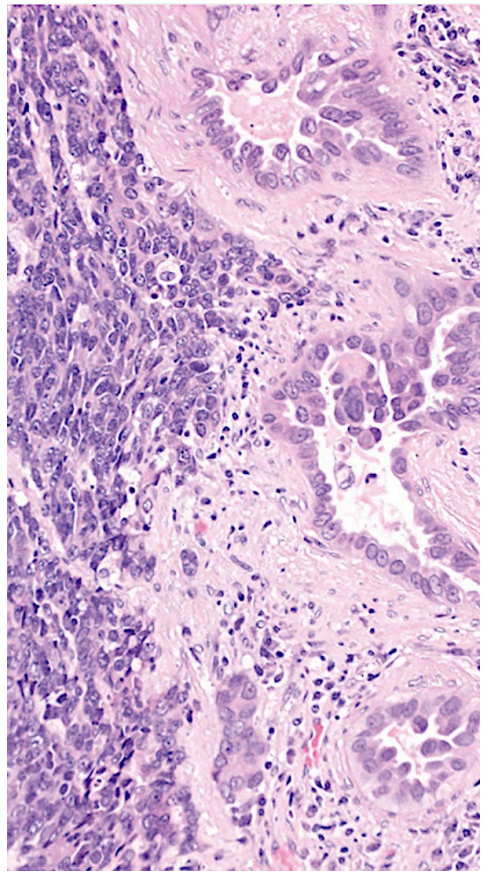
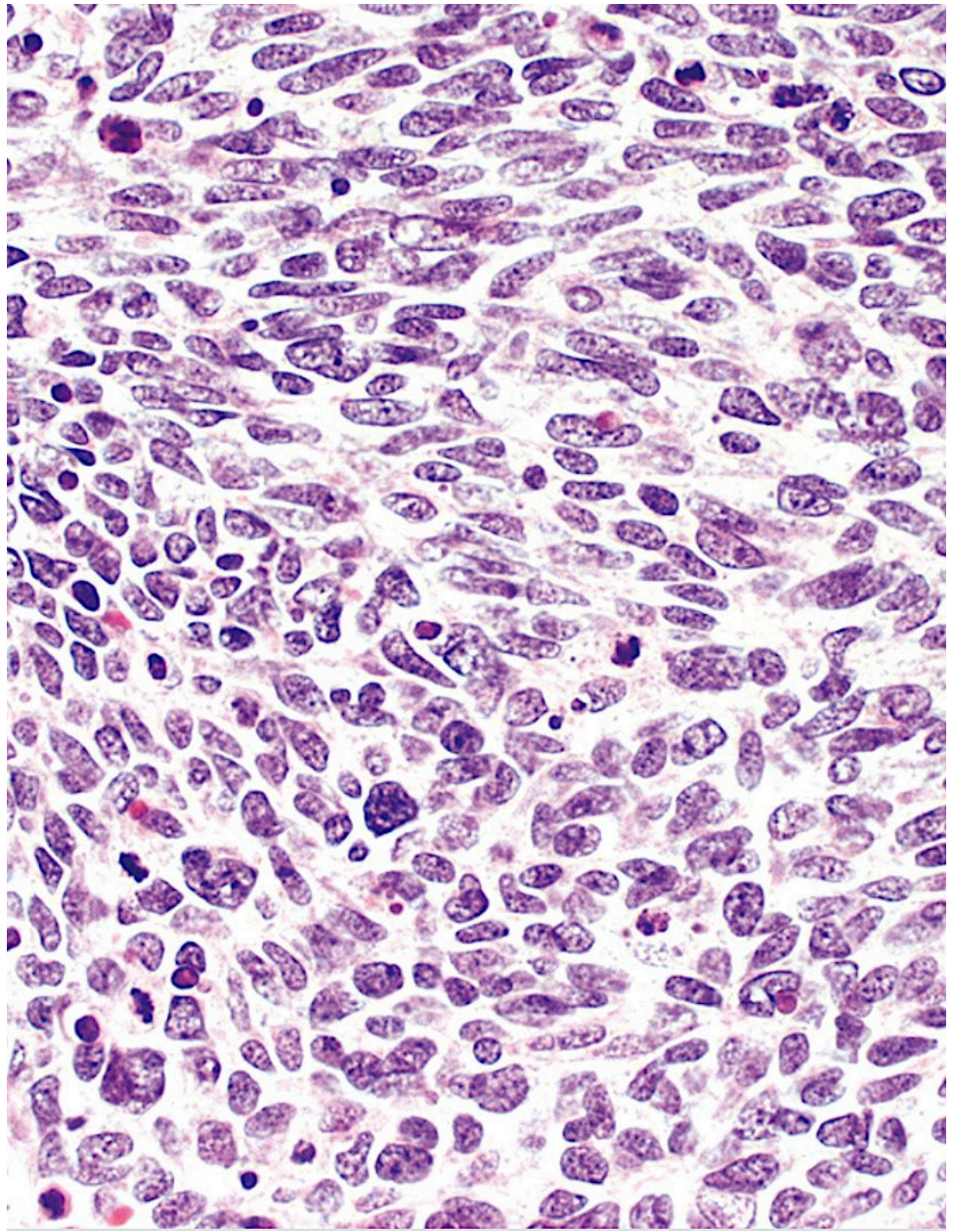
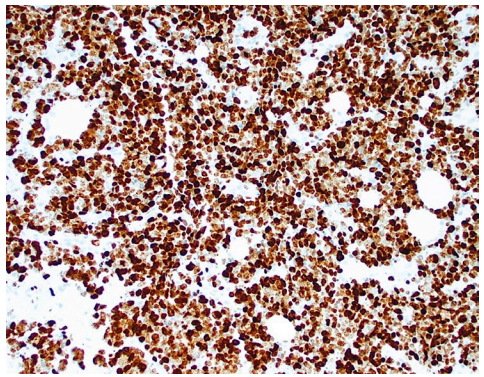
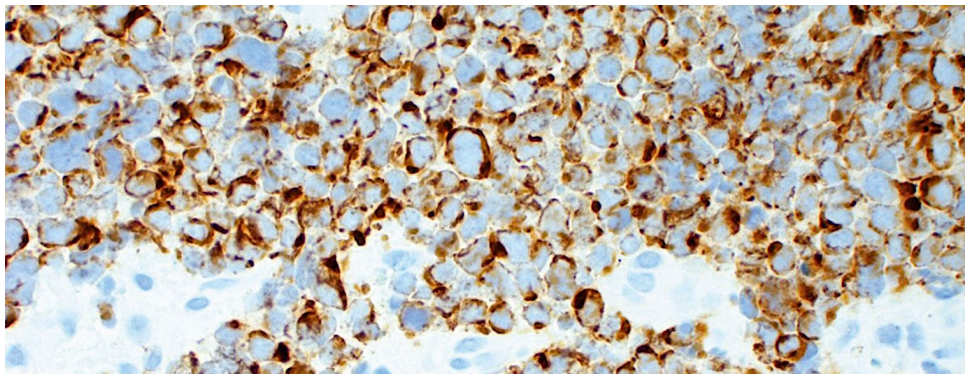
- Limited-Stage: AJCC TNM Stage I-III
- Extensive-Stage: AJCC TNM Stage IV
- Two thirds of patients have extra-thoracic metastases at presentation

Treatment:

- Platinum-based agent + topoisomerase inhibitor either combined with surgery or radiation (or both) or systemic ICIs, predominantly depending on disease stage

Outcomes: 5-year OS < 7%

Horvath et al *Curr Opin Oncol* 2024

WHO BLUE BOOK IMAGES OF SCLC

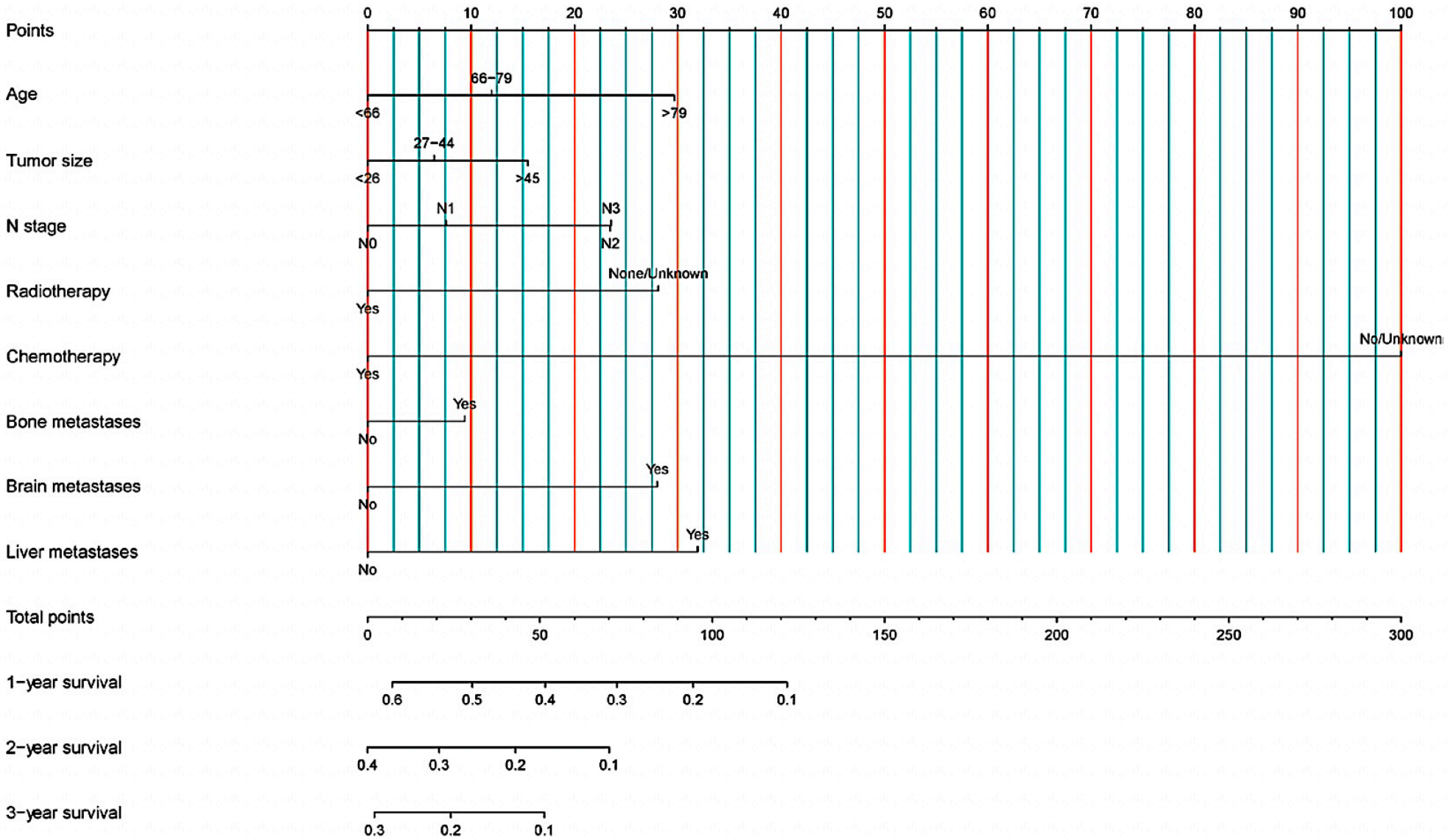


Figure 3. Nomogram predicted 1-, 2-, and 3-year OS for DM-SCLC with 8 available factors. DM-SCLC = small-cell lung cancer, OS = overall survival.

Qie S et al *Medicine* 2022

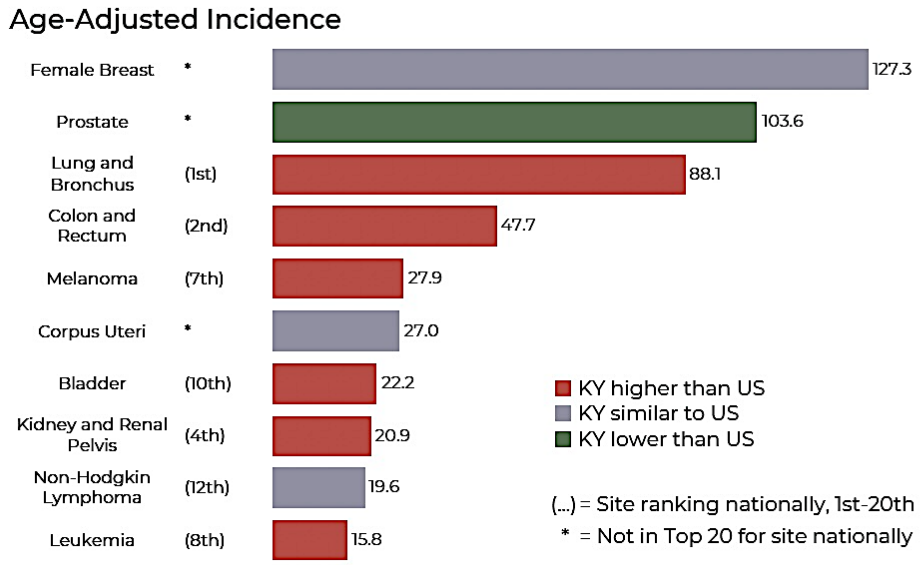
SCLC in Kentucky

Table 1 Distribution of Lung Cancer Histologies, 2012 to 2016, Appalachian Kentucky; Non-Appalachian Kentucky; All Kentucky; US (SEER)

	SEER Cases (%)	All KY Cases (%)	χ^2 *P* Value[a]	Non-Appalachian KY Cases (%)	Appalachian KY Cases (%)	χ^2 *P* Value[b]
Male and female			< .0001			< .0001
Adenocarcinoma	111,886 (44.7)	6,098 (32.4)		4,234 (34.1)	1,864 (29.0)	
Squamous	48,239 (19.3)	4,794 (25.4)		3,054 (24.6)	1,740 (27.0)	
Other	51,502 (20.6)	4,051 (21.5)		2,592 (20.9)	1,459 (22.7)	
Small cell	29,176 (11.7)	3,262 (17.3)		2,084 (16.8)	1,178 (18.3)	
Neuroendocrine	9,452 (3.8)	634 (3.4)		440 (3.5)	194 (3.0)	
Male			< .0001			.0003
Adenocarcinoma	51,664 (40.8)	3,056 (29.9)		2,072 (31.6)	984 (26.7)	
Squamous	29,702 (23.5)	3,048 (29.8)		1,883 (28.7)	1,165 (31.6)	
Other	26,969 (21.3)	2,291 (22.4)		1,435 (21.9)	856 (23.3)	
Small cell	14,332 (11.3)	1,553 (15.2)		982 (15.0)	571 (15.5)	
Neuroendocrine	3939 (3.1)	289 (2.8)		184 (2.8)	105 (2.9)	
Female			< .0001			< .0001
Adenocarcinoma	60,222 (48.7)	3,042 (35.4)		2,162 (37.0)	880 (32.0)	
Squamous	18,537 (15.0)	1,746 (20.3)		1,171 (20.0)	575 (20.9)	
Other	24,533 (19.8)	1,760 (20.5)		1,157 (19.8)	603 (21.9)	
Small cell	14,844 (12.0)	1,709 (19.9)		1,102 (18.8)	607 (22.0)	
Neuroendocrine	5513 (4.5)	345 (4.0)		256 (4.4)	89 (3.2)	

Brainson et al *Clin Lung Ca* 2021

Figure 1. Top 10 Cancer Incidence in Kentucky



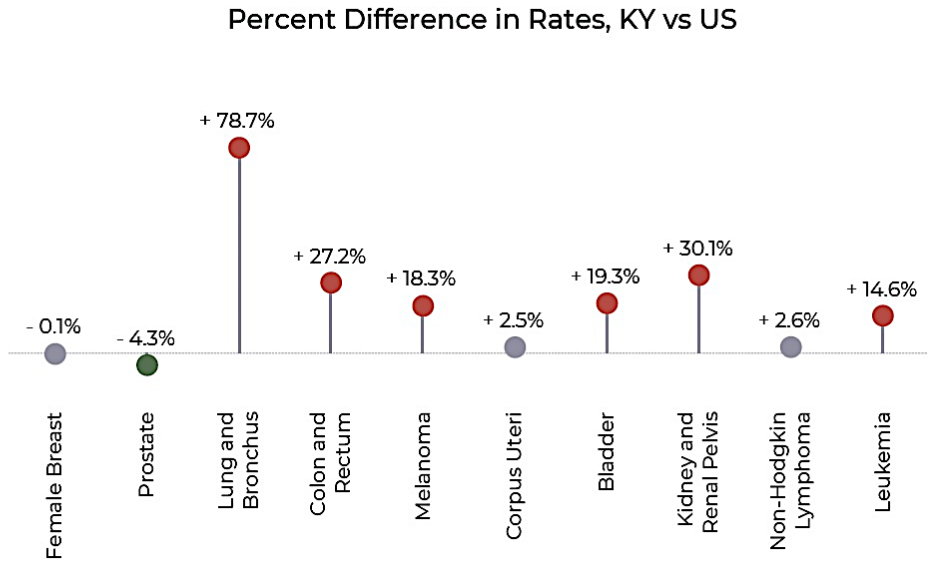
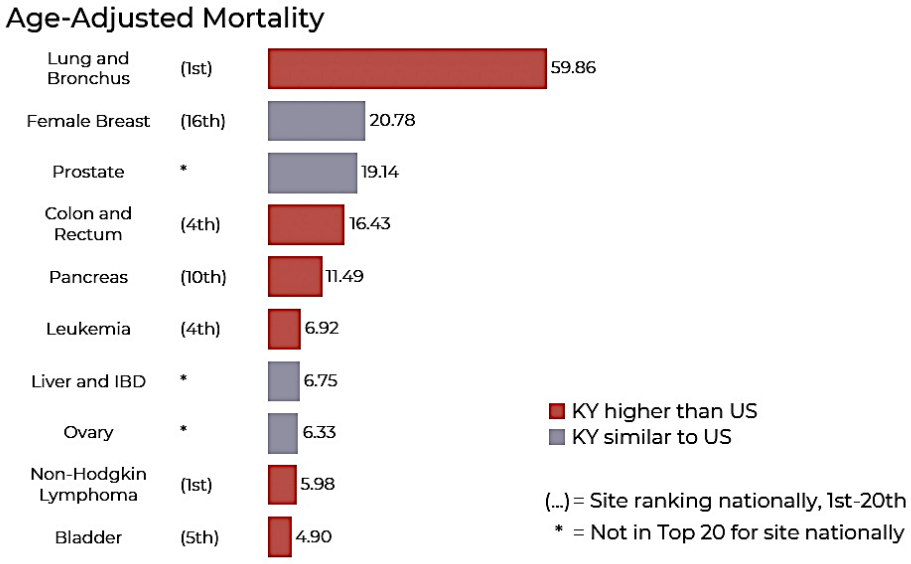
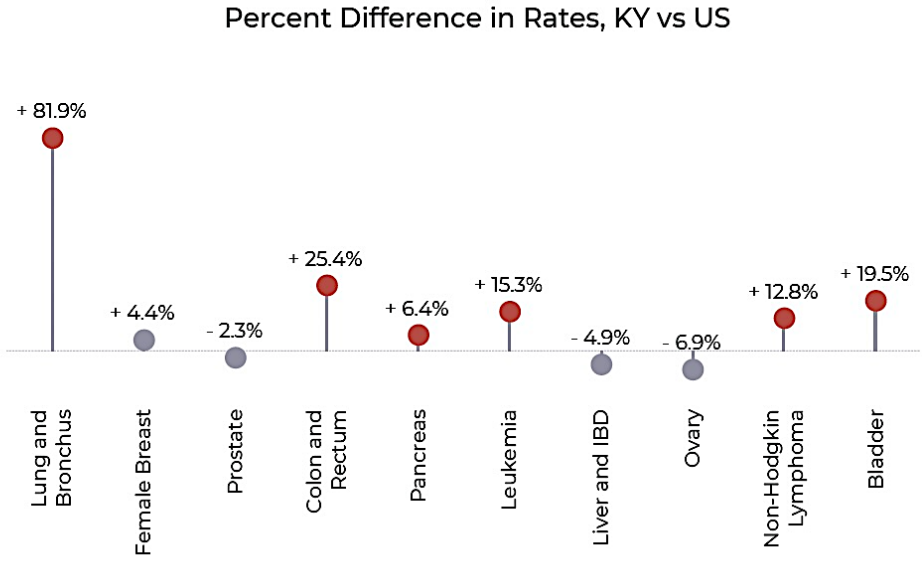
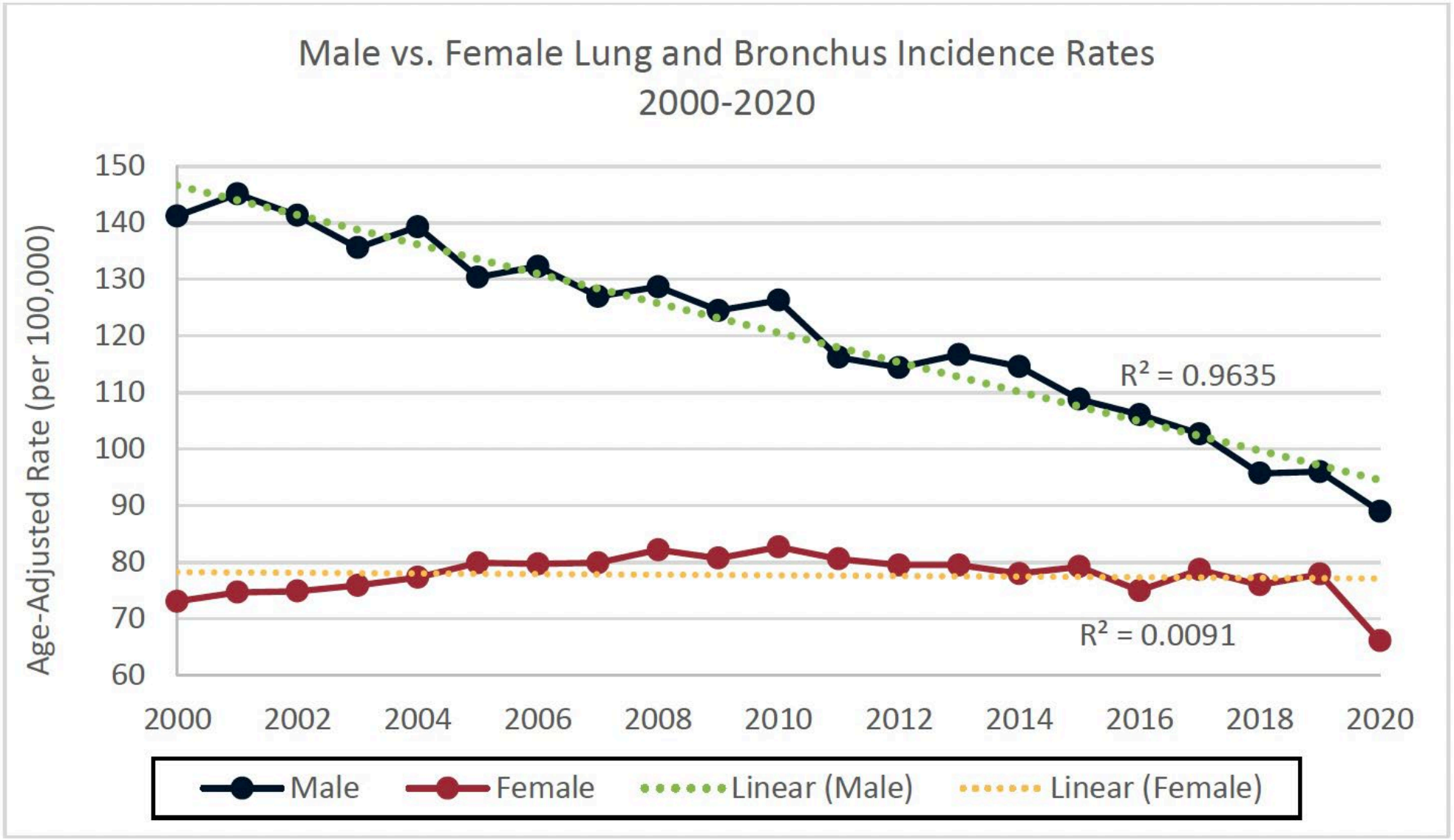


Figure 2. Top 10 Cancer Mortality in Kentucky







KY Dept for Public Health, Lung Cancer Program 2023 Annual Report

General Genetics of SCLC (High TMB, Low LOH)

Petty WJ and Paz-Arez L *Jama Oncology 2023*

GENE	Alteration Type	Frequency	Normal Protein Function
TP53	Biallelic inactivation	75%-90%	Stress response protein involved in regulation of cell cycle arrest, apoptosis, senescence, DNA repair and metabolism shifts
RB1	Biallelic inactivation	60%-90%	Negative regulator of cell cycle Stabilizes chromatin structure
CREBBP and EP300	Co-occurring sequence variations	Common	Histone acetyltransferases involved in transcriptional coactivation of many transcription factors
NOTCH genes	Sequence variations	Common	Cell fate specification, differentiation, proliferation and survival via NOTCH signaling pathway
TP73 or RLF::MYCL1	Fusions	Uncommon (7%)	TP73 is a member of the p53 family of transcription factors Fusion found in 7% of SCLC-A and acts as met. driver
MYC genes	Amplification	16%	Nuclear phosphoproteins involved in cell cycle progression, apoptosis and cell transformation
SOX2	Amplification	27%	Transcription factor involved in embryogenesis, cell fate and stem-cell maintenance in CNS
FGFR1	Amplification	Uncommon (6%)	TK + FGF receptor involved in mitogenesis and differentiation

Variants of SCLC

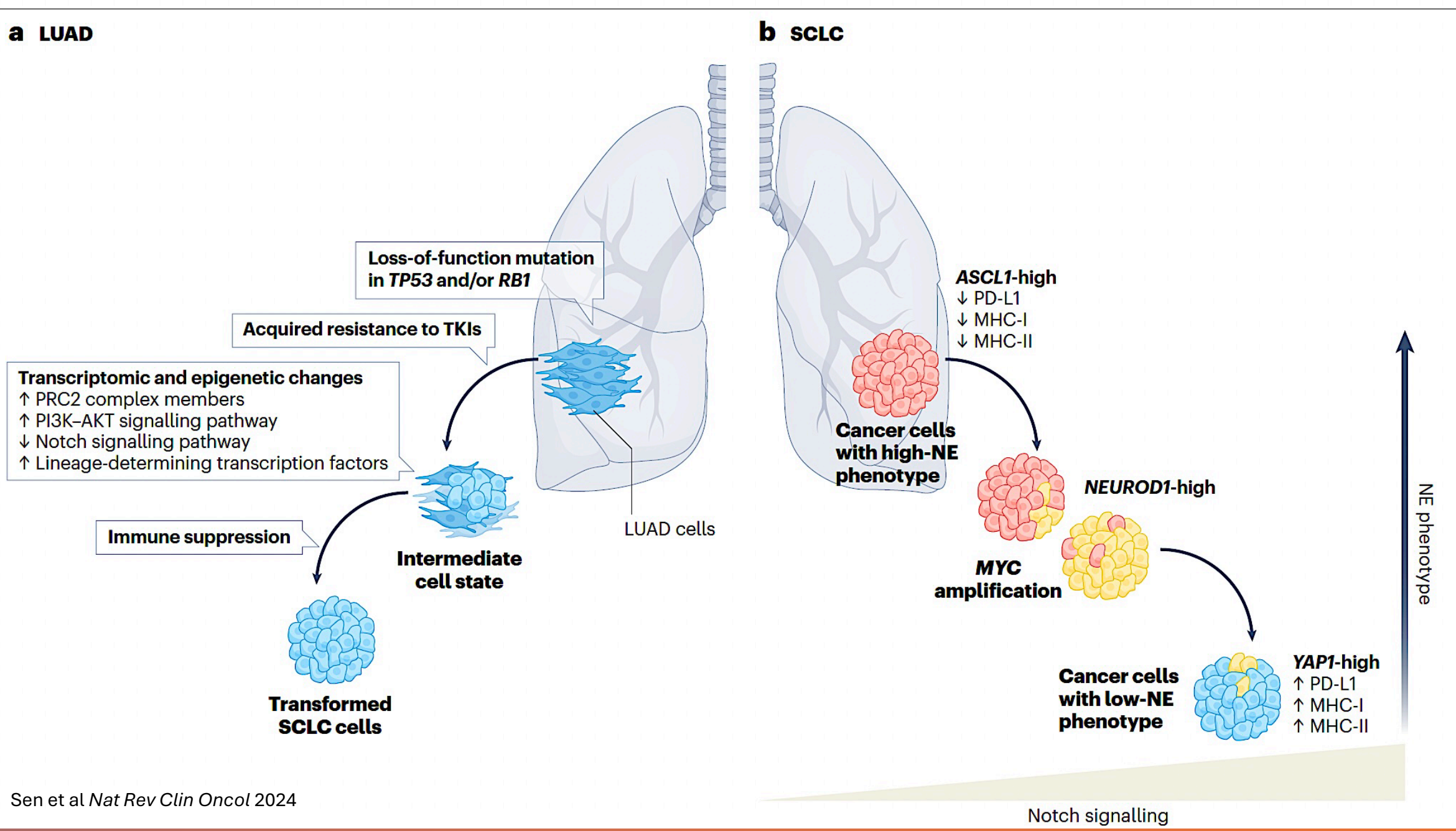
SCLC-A (40%)	• ASCL-1 + NE markers expressed
SCLC-N (20%)	• NEUROD1 + NE markers expressed
SCLC-AN (?)	• Dual ASCL-1 and NEUROD1 expression + NE markers
SCLC-P (6%)	• POU2F3 expressed but no NE markers
SCLC-I (20%)	• Inflammatory gene signature; YAP1 +/-; no NE markers

Carlisle and Leal *Cancer*, 2023

SCLC-A (40%)	• ASCL-1 + NE markers expressed • Could be two distinct subsets, A-alpha and A-delta; the latter may respond to ICIs
SCLC-N (20%)	• NEUROD1 + NE markers expressed
SCLC-AN (?)	• Dual ASCL-1 and NEUROD1 expression + NE markers
SCLC-P (6%)	• POU2F3 expressed but no NE markers
SCLC-I (20%)	• Inflammatory gene signature; YAP1 +/-; no NE markers

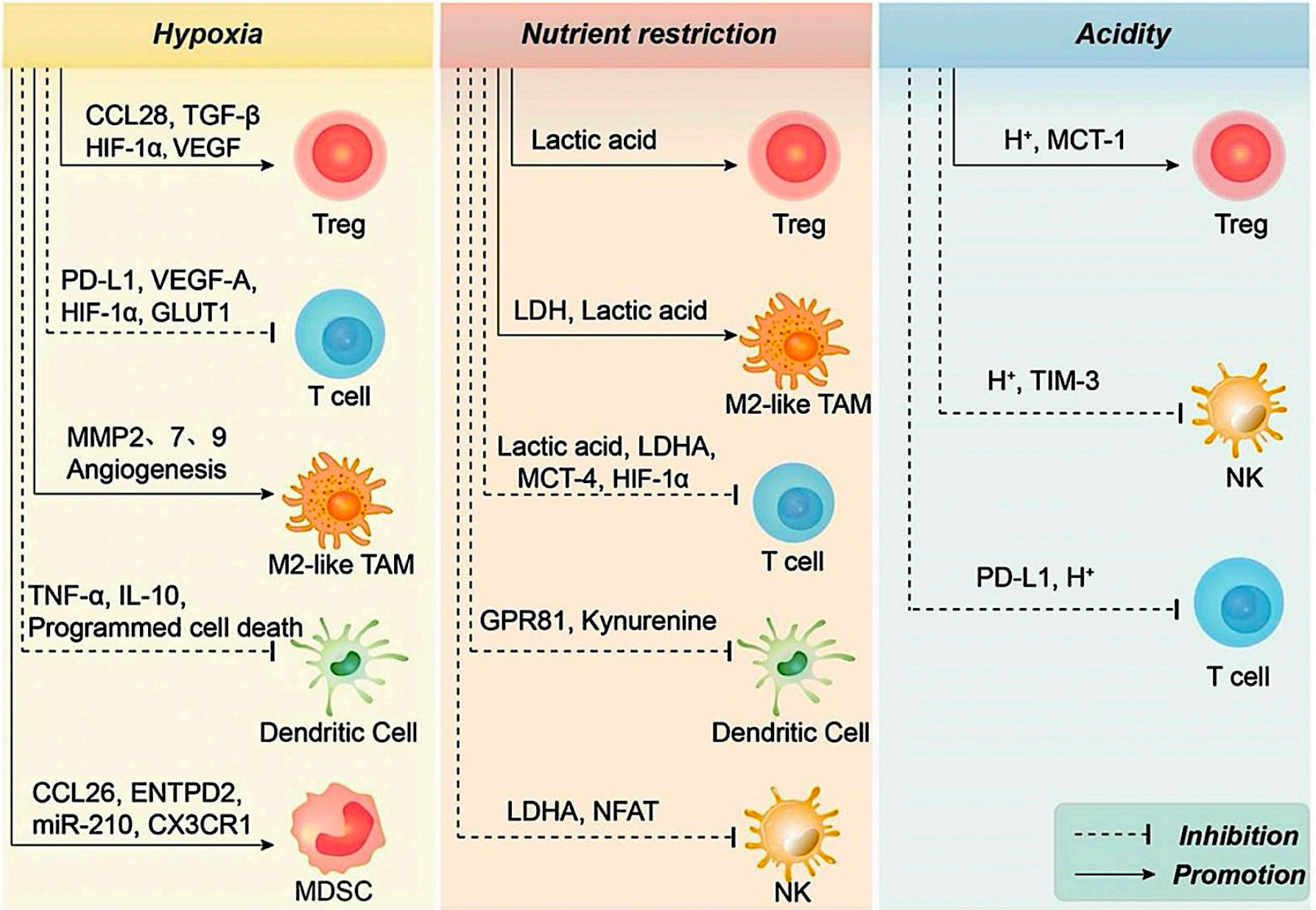
High numbers of stem cell like tumor cells expressing PLCG2 can occur across all types with a very cold TIME and worse OS. Chan et al , *Cancer Cell* 2021

Carlisle and Leal *Cancer*, 2023



Sen et al *Nat Rev Clin Oncol* 2024

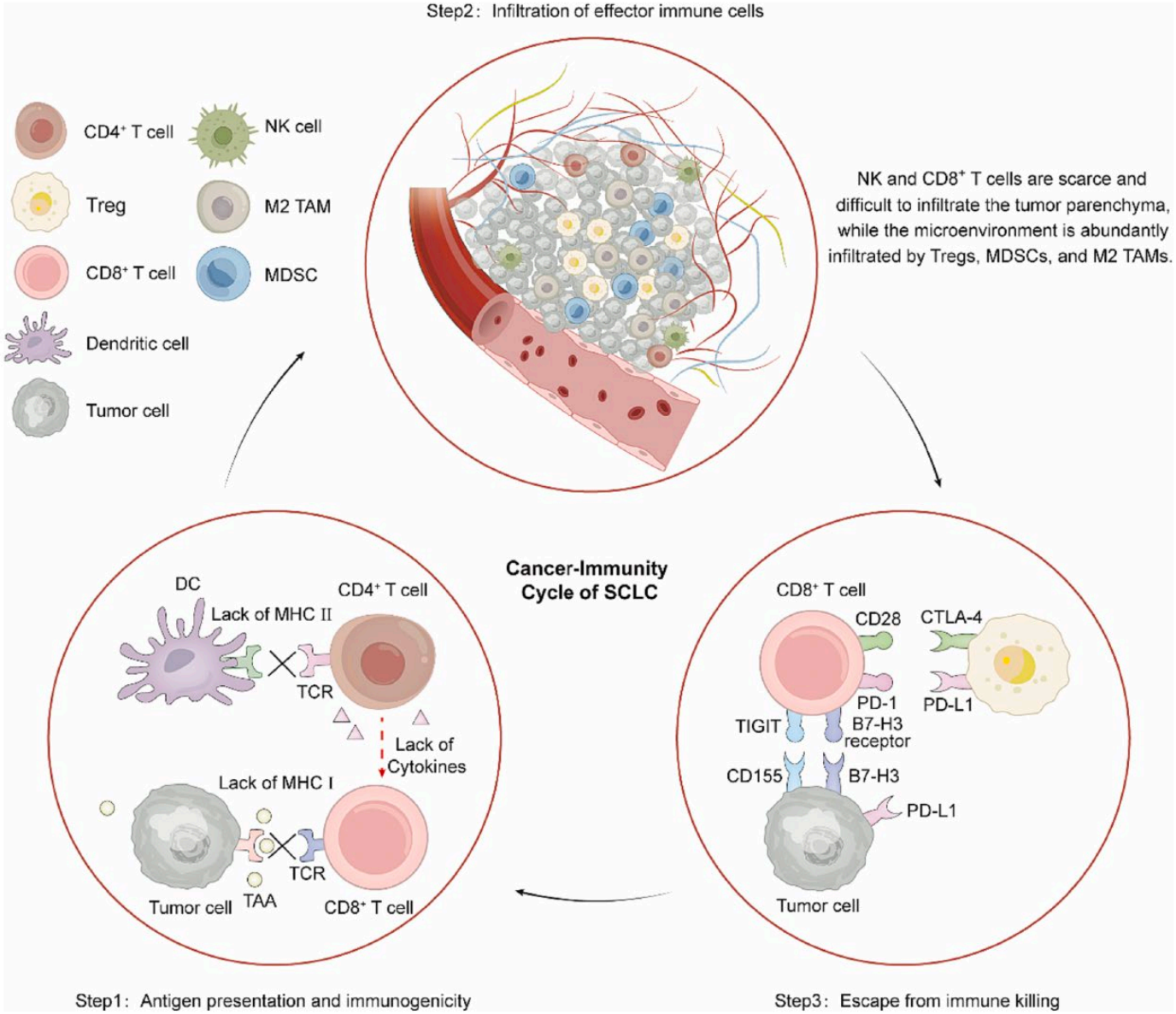
SCLC TME Directly Affects TIME



Li, Qiao *Sem Ca Biol* 2022

Tumor Immune Microenvironment of SCLC

Chen et al *Cancer Treatment Reviews* 2023



Precision Therapy for SCLC
New Approaches

SCLC-A	SCLC-N	SCLC-P	SCLC-I
• DLL3 I's • BCL-2 I's • HDAC I's • LSD1 I's • CAR-T-Cell Rx	• c-Myc I's • AURKA I's • ADI-PEG20 • Seneca Valley Virus	• PARP I's • IGF-1R I's • Nucleoside analogs • Lurbinectidin	• ICI's • mTOR I's • CDK4/6 I's • PLK1 I's

Horvath et al *Curr Opin Oncol* 2024

Liquid Biopsy Potential Biomarkers in SCLC

- Circulating tumor DNA (ctDNA)
- Circulating tumor cells (CTCs)
- Serum neuronal autoantibodies (SNAAs)
- Inflammatory hematologic parameters
- Blood tumor mutation burden (bTMB)

MCC SCLC Study
Background and Hypothesis

Survival beyond three years occurs in 5-10%. Exceptional survival may be attributable to an enhanced anti-tumoral immune response, although small cell carcinoma is generally described as an “immune desert” or as immersed in an immunosuppressive tumor immune microenvironment (TIME).

We posited that specific TIME features in primary and matched metastatic SCLC significantly affect survival. Furthermore, TIME features could inform optimal immunotherapy selection, tailored to an individual’s specific immune microenvironmental conditions.

Methods: Two Key Parts

Study Component	Details
Quantifying Tumor Immune Environment	Identified SCLC patients from the Kentucky Cancer Registry
	Created two cohorts: 12 expected survivors (<14 months) and 12 exceptional survivors (>36 months)
	Measured 78 immune-oncology proteins using NanoString GeoMX and Lunaphore COMET platforms
Histologic Stratification	Four pathologists independently reviewed samples to classify cases into survival groups (blinded data)

Patients

Feature	Expected	Exceptional
Total Number	12	12
Male	4	4
Female	8	8
Average Age	62	59
Limited Stage (LS)	4	10
Extended Stage (ES)	3	0
Standard Rx	4	10
ImmunoRx	1	0

Specimens

Feature	Expected	Exceptional
Lymph Node	11	10
Primary Tumor	1	2
Block Years/Age	10.42	10.75

Number and Types of Immuno-Oncologic Proteins Assessed (GeoMx panel)

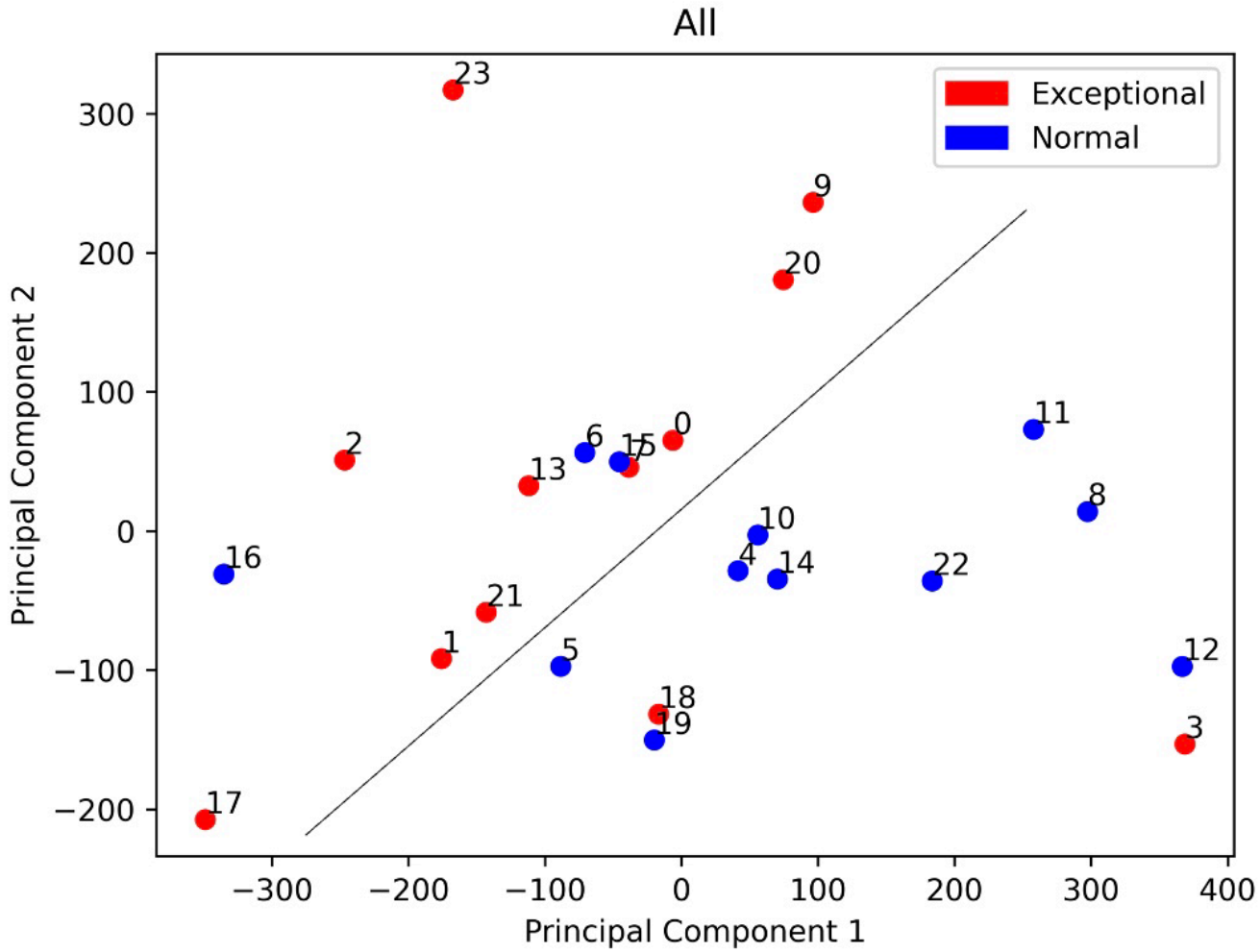
Cytokines and Chemokines	T-cell markers	Macrophage markers	Myeloid cells	Antigen presentation*
16	9	4	10	6

Checkpoint*	Myeloid activation	T-cell activation*	Tumor Proteins	Apoptosis
11	5	9	7	3

* = contains one or more significant discriminators of survival

Category	Probe Names
Cytokines and Chemokines	IFNG, IL12B, IL15, IL6, CXCL10, CXCL9, CCL5, TNF
T-cell Markers	CD3E, CD4, CD8A, FOXP3, GZMB, TBX21
Macrophage Markers	CD68, ARG1, CSF1R
Myeloid Cells	ITGAM, ITGAX, CSF1R
Antigen Presentation	HLA-DQ, HLA-DRB, HLA-E, **CD74**
Checkpoint	PDCD1 (PD-1), **CD274 (PD-L1),** CTLA4, PDCD1LG2 (PD-L2), LAG3, TIGIT
Myeloid Activation	CD40, CD86, CSF1R, ICAM1, CD276 (B7-H3)
T-cell Activation	CD3E, CD4, CD8A, **CD27**, CD28, CD2, CD5, CD7, CD154 (CD40LG)
Tumor Proteins	EPCAM, BCL2, PTEN, STAT3, CTNNB1, KRAS, MYC
Apoptosis	FAS, BCL2, CASP3
Cell cycle regulation	**CCND1**

Results of GeoMx PCA



Groupings significantly differentiate between exceptional and expected survival

- Chi-squared= 8.2238
- p = 0.004135

Caveats:

- Small patient number (24)
- Near significance in age difference with older patients in expected survival group; p = 0.07672 (Mann-Whitney)

Differences in Expression of Immune Regulatory Proteins Expression

- CD27 and CD274 have nonlinear correlation with survival (0.35 and 0.49, respectively, on a scale of 0-1).
- **With a cutoff of CD27<35.69, 8 have short survival and 1 has long survival (*P*=0.003163).**
- **With a cutoff of CD274<30, 6 have short survival and 1 has long survival.**
- In both cases, the incorrectly classified person in the 2nd oldest individual (4194).

Genes that are either correlated with survival or have survival correlated with expression (higher is better. Range: 0-1)

Expression Correlated with Survival

- **CD274 (0.4914)**
- **CD27 (0.3829)**
- **CCND1 (0.3636)**
- **CD74 (0.35)**
- VSIR (0.175)
- POLR2A (0.1636)
- CTLA4 (0.15)
- FNGR1 (0.1386)
- VEGFA (0.0914)
- B2M (0.08571)
- PECAM1 (0.08)
- OAZ1 (0.08)
- IDO1 (0.055)
- TNFRSF9 (0.04143)

Survival Correlated with Expression:

- PECAM1 (0.0563)
- CXCL10 (0.0348)
- **HLA-DRB (0.009093)**

CD27

- **CD27**: Receptor on T/B cells enhancing immune response.
- **CD70**: Ligand that activates CD27, boosting **T cell survival & cytokine production**.
- **CD70 is overexpressed in SCLC**, aiding immune evasion & T cell exhaustion.
- **Our Study**: **Low CD27 expression = Shorter survival in SCLC patients**.
- **Mechanism**: CD27-CD70 signaling strengthens **anti-tumor immunity** but prolonged activation can exhaust T cells.
- **Clinical Relevance**:
  - **CD27 as a prognostic marker** (higher CD27 = better survival).
  - **Potential therapy**: CD27 activation + immune checkpoint inhibitors (PD-1/PD-L1) for better tumor response.

CD74

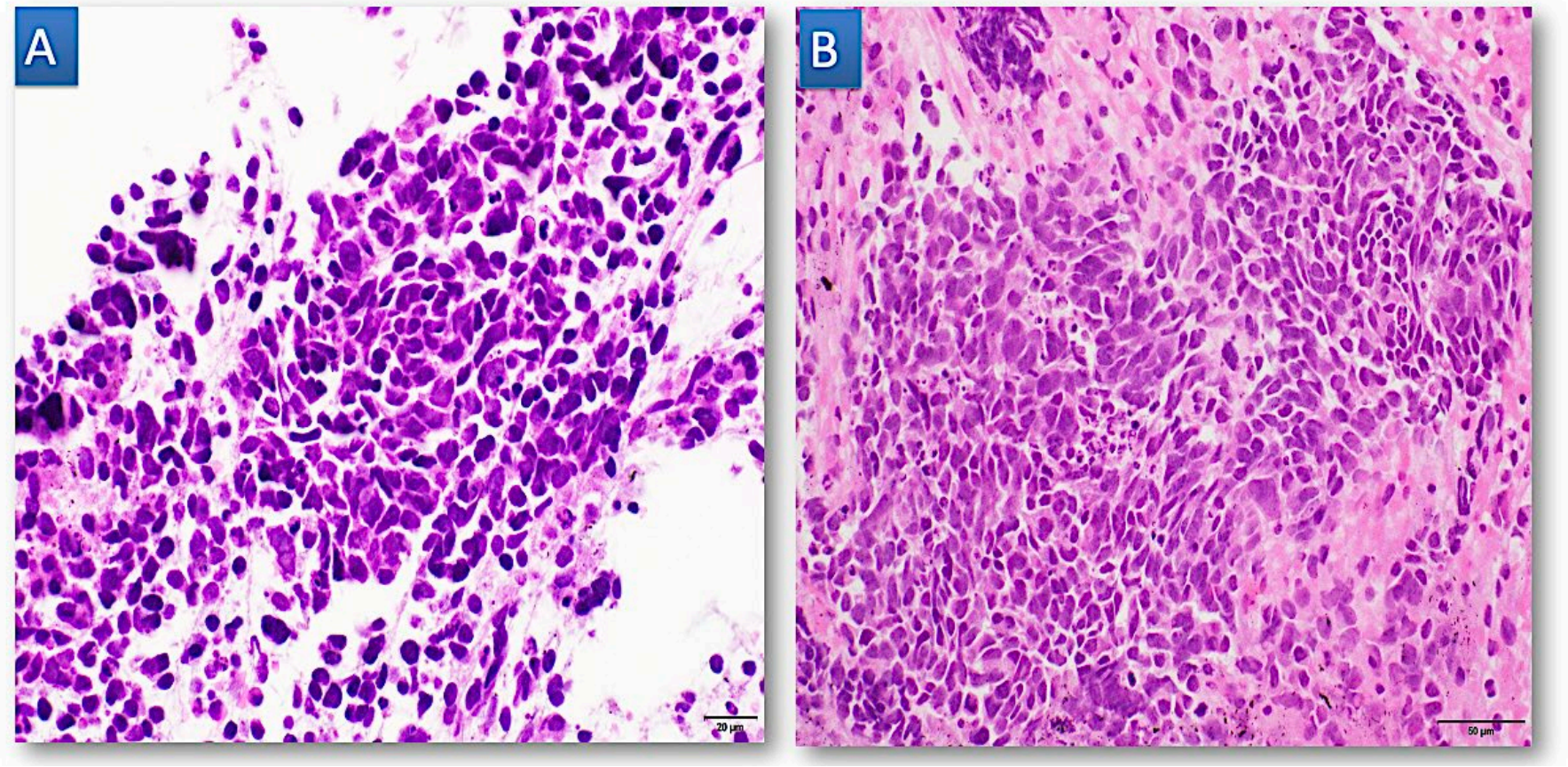
- **CD74**: A transmembrane protein that acts as an MHC class II chaperone and plays a role in immune regulation.
- **CD74 is overexpressed in SCLC**, facilitating tumor survival and immune evasion.
- **Mechanism**:
  - Regulates antigen presentation and **macrophage migration inhibitory factor (MIF) signaling**.
  - Promotes **tumor proliferation and resistance to apoptosis**.
- **Our Study**: **High CD74 expression is linked to poor prognosis in SCLC patients**.
- **Clinical Relevance**:
  - **CD74 as a prognostic marker**: High expression correlates with **worse survival**.
  - **Potential therapy**: Targeting CD74-MIF interactions may **enhance anti-tumor immunity** and **improve treatment response**.

CD274 and CCND1

- **CD274 (PD-L1):**
  - An immune checkpoint protein that inhibits T cell activation.
  - **Overexpressed in SCLC**, allowing tumors to evade immune detection.
  - **Clinical Relevance**:
    - PD-L1 expression predicts response to **immune checkpoint inhibitors**.
    - Blocking PD-L1 restores T cell-mediated anti-tumor activity.
- **CCND1 (Cyclin D1):**
  - A regulator of the cell cycle, driving the **G1-to-S phase transition**.
  - Amplified or overexpressed in SCLC, contributing to **uncontrolled tumor cell proliferation**.
  - **Clinical Relevance**:
    - CCND1 amplification is associated with **aggressive tumor behavior**.
    - Targeting Cyclin D1-CDK4/6 could **halt tumor progression**.

Histologic Stratification Results

Histomorphology groupings of cases devised independently by pathologists showed a significant correlation with survival in one of four personally-devised tumor stratification systems (P = 0.014)



Representative FFPE cell block samples of SCLC independently scored by four pathologists.

A. Expected Case #8
B. Exceptional Case #9

Histologic Stratification as Prognostic Biomarker

Pathologist D's Approach

DIVISION	GROUP 1	GROUP 2
SET A. Nuclear Uniformity and Size	Smaller nuclei, more uniform (usually higher N/C ratio and molding): #0, #1, #4, #8, #10, #12, #13, #14, #15, #20, #22	Bigger nuclei, more variable (usually lower N/C ratio and less molding): #2, #3, #6, #7, #9, #17, #18, #19, #23
SET B. Apoptosis Degree in Best Preserved Nests	Higher levels of apoptosis (often associated with Group 1 above): #1, #2, #3, #5, #8, #12, #14, #15, #20	Lower levels of apoptosis: #0, #6, #7, #10, #11, #13, #17, #18, #19 Intermediate group: #4, #9, #21, #22, #23
SET C. Combination of SET A and SET B criteria	Small, uniform nuclei + high apoptosis: #1, #8, #14, #15, #16 and probably #20	Larger more variable nuclei with low apoptosis: #6, #7, #17, #18, #19 INTERMEDIATE Group: (intermediate apoptosis): #4, #9, #21, #22, #23
SET D. Mitotic rate only	Higher mitoses: #4, #5, #7, #11, #14, #17, #18, #23	Lower mitoses: #0, #1, #2, #3, #6, #8, #9, #15, #22 Intermediate group: #10, #12, #13, #19, #20
Unable to evaluate due to scant tumor	#16	Poor preservation but most likely belongs to SET A, Group 1 and SET B Group 1.
Unusual Cases	#21	Peculiar pale nuclei larger with diminished molding. May be pale due to degeneration. Unable to assess N/C ratio.

Note: All cases but one showed extensive geographic necrosis; I assessed instead apoptosis (individual cell necrosis) in the best preserved clusters of 15-20 tumor cells. One case was nearly 100% degenerated (#16) so limited for evaluation and one case had abundant tumor >> than the other cases (#20) with more spindled tumor nuclei than the others.

Histologic Stratification as Prognostic Biomarker

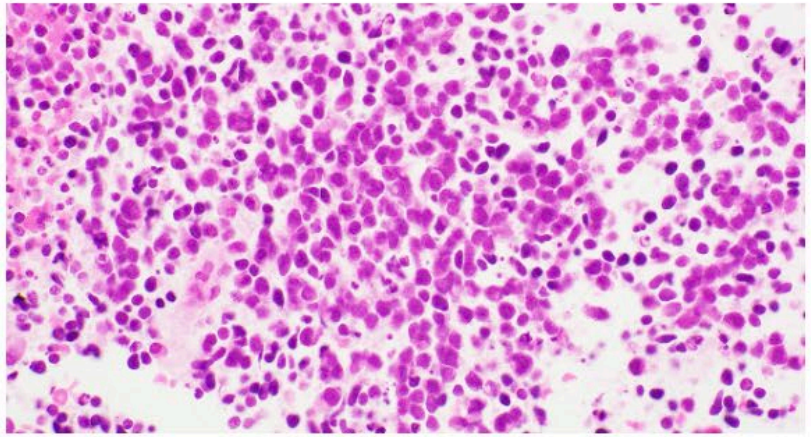
Pathologist D's Approach

DIVISION	GROUP 1	GROUP 2
SET A. Nuclear Uniformity and Size	Smaller nuclei, more uniform (usually higher N/C ratio and molding): #0, #1, #4, #8, #10, #12, #13, #14, #15, #20, #22	Bigger nuclei, more variable (usually lower N/C ratio and less molding): #2, #3, #6, #7, #9, #17, #18, #19, #23
SET B. Apoptosis Degree in Best Preserved Nests	Higher levels of apoptosis (often associated with Group 1 above): #1, #2, #3, #5, #8, #12, #14, #15, #20	Lower levels of apoptosis: #0, #6, #7, #10, #11, #13, #17, #18, #19 Intermediate group: #4, #9, #21, #22, #23
SET C. Combination of SET A and SET B criteria	Small, uniform nuclei + high apoptosis: #1, #8, #14, #15, #16 and probably #20	Larger more variable nuclei with low apoptosis: #6, #7, #17, #18, #19 INTERMEDIATE Group: (intermediate apoptosis): #4, #9, #21, #22, #23
SET D. Mitotic rate only	Higher mitoses: #4, #5, #7, #11, #14, #17, #18, #23	Lower mitoses: #0, #1, #2, #3, #6, #8, #9, #15, #22 Intermediate group: #10, #12, #13, #19, #20
Unable to evaluate due to scant tumor	#16	Poor preservation but most likely belongs to SET A, Group 1 and SET B Group 1.
Unusual Cases	#21	Peculiar pale nuclei larger with diminished molding. May be pale due to degeneration. Unable to assess N/C ratio.

Note: All cases but one showed extensive geographic necrosis; I assessed instead apoptosis (individual cell necrosis) in the best preserved clusters of 15-20 tumor cells. One case was nearly 100% degenerated (#16) so limited for evaluation and one case had abundant tumor >> than the other cases (#20) with more spindled tumor nuclei than the others.

Statistically Significant Pathologist Devised System (Pathologist A)

Usual Group



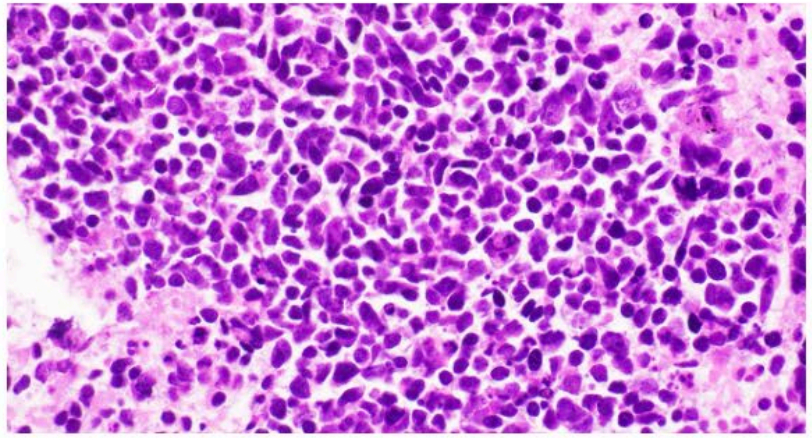
Usual group

0 – poorly preserved, hi necrosis, hi apoptosis, moderate crush, small size, few lymphs

1 – focal necrosis, moderate apoptosis, moderate crush, small size, few lymphs

2 – hi necrosis, hi apoptosis, moderate crush, intermediate size with larger cells, few lymphs

Atypical Group



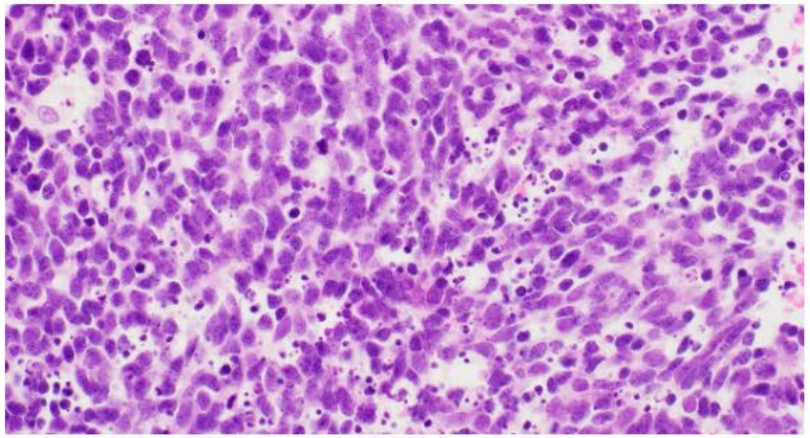
Atypical group

3 – little to no necrosis, little apop, mod crush, small size, little lymphs

4 – little to no necrosis, little apop, mod crush, small size with some spindle cells, little lymphs

8 – little necrosis, mod apop, mod crush, small size, mod lymphs

Indeterminate Group



Indeterminant

12 – little necrosis, hi apop, mod crush, small size and uniform, mild lymphs – favor atypical

15 – poorly preserved, focal necrosis, mod / hi apop, mod crush, small to intermediate in size, mod lymphs – favor usual

16 – hard, very little tumor, mostly crushed, mod necrosis, ? apop, hi crush, small size, no lymphs – favor usual

Study Conclusions

01

FFPE cell block specimens work for multiplex IF and subsequent image analysis.

02

Four immunoprotein expression levels related to the TIME correlate with survival in SCLC.

03

Partially successful, qualitative morphology stratification by pathologists offers potential for development of an immunotherapy predictive AI algorithm for SCLC.

Next Steps

Expand Case Numbers

Focus on CD27, CD274 and CD74 expression

Begin digital slide capture for AI and manual analysis

Summary

SCLC transcriptome subtypes are increasingly clinically relevant

SCLC TIME can potentially be ‘ignited’ to become more substantially responsive to immunotherapy

Blood biomarkers may join a comprehensive prognostic and predictive biomarker panel

Acknowledgements

Jing Di MD, PhD

- Ashish Maskey, MD
- Susanne Arnold, MD
- Christine Brainson, PhD
- Dava Piecoro, MD
- Emily Bachert, MD
- Kimberly Absher, MD
- Hafa Nebacche, MD
- Akif Guney, MD
- Justin Miller, PhD
- Thèrése Bocklage, MD