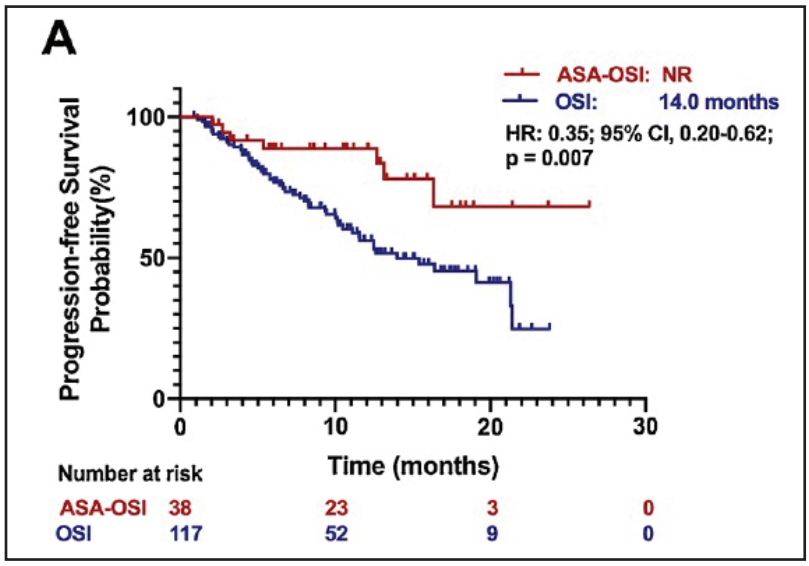
CIRCULOGENE
www.circulogene.com
info@circulogene.com
855-614-7083

77-Year-Old female with metastatic EGFR-mutated lung cancer

In this patient with an EGFR exon 21 L858R mutated lung adenocarcinoma, the identified co-mutations and PD-L1 expression drive nuanced therapeutic decisions.

At age 77, a simple cancer treatment with an oral EGFR TKI or single-agent immune checkpoint blockade with PD-L1 > 50% would appear ideally kind and gentle. However, the co-mutations of the TP53 exon 5 and KDR Q472H greatly impact the benefit of simple EGFR TKI therapy alone. KDR Q472H co-mutations have been associated with primary resistance to an EGFR TKI. Even when responsive, TP53 mutations portend a much shorter PFS and OS. However, this adverse prognostic and predictive clinical behavior of these co-mutations may be clinically mitigated. TP53 mutations are associated with elevated VEGFA levels, which can be a resistance pathway in EGFR-mutated lung cancers. The kinase insert domain receptor (KDR) mutation encodes VEGFR-2 function, and these mutations are also associated with elevated VEGFA levels. Studies have shown dual EGFR and VEGF blockade can reverse secondary and overcome primary EGFR TKI resistance. Given these co-mutations and tumor biology, a dual EGFR and VEGF blockade with either bevacizumab or ramucirumab is a very compelling best-treatment approach in this setting.



The PD-L1 expression also carries a treatment decision impact—not with immune checkpoint blockade but with the choice of the EGFR TKI. In the FLAURA trial, osimertinib was associated with an unchanged 18.9/18.4-month PFS irrespective of PD-L1 expression of ≥ 1% or < 1%. However, in the comparator arm of either gefitinib or erlotinib, the PFS was impacted and decreased from 10.9 months to 6.9 months with PD-L1 expression.

Another emphasis with this case is the importance of knowing EGFR and ALK status before embarking upon immune-based therapies in lung cancer. These two targetable driver mutations do not benefit from immune checkpoint blockade, with FDA indications for first-line immune-based therapy mandating negative EGFR and ALK findings. Charging forward with immune-based therapies without knowing these molecular findings can lead to missing very effective targeted therapy and stepping forward with ineffective therapy with heightened pulmonary toxicity. The right treatment matters and time matters. Plasma and tissue molecular findings are complementary and can achieve the right treatment at the right time. Tissue NGS alone will miss one-third of targetable driver mutations. Both are needed to completely identify potential driver mutations/fusions. Drawing the plasma NGS at the time of tissue diagnosis and sending tissue for NGS immediately after the pathologist confirms a malignant diagnosis is the approach CIRCULOGENE advocates to ensure your patient is getting the right treatment in the quickest possible time from their cancer diagnosis.

IMMUNOTHERAPY TEST RESULTS		FDA GUIDANCE	RNA TEST RESULTS	
PD-L1 EXPRESSION	Positive (≥50%)	Pembrolizumab	ALK GENE FUSION	Not Detected
			ROS1 GENE FUSION	Not Detected

bPCR TEST RESULTS EGFR mutation: (+L858R) KRAS mutation: Not detected BRAF mutation: Not detected

TP53	p.V157I; c.469G>A Exon 5	5.7%
KDR	p.Q472H; c.1416A>T	4.1%

Another intriguing clinical tidbit is a recent study showing improved PFS with concurrent use of aspirin with osimertinib independent of TP53 mutation and PD-L1 status.

Case Study Prepared by Doctor Paul Walker
Chief Medical Officer, Former Director of Thoracic Oncology at East Carolina University

Sources:
- Clin Cancer Res. May 15 2009 15 (10) 3600-3609. DOI:10.1158/1078-0432.CCR-08-2568
- Masago et al. BMC Cancer (2015) 15:908. DOI 10.1186/s12885-015-1925-2
- X. Liu et al. Lung Cancer. 2020 Nov;149:33-40. DOI: 10.1016/j.lungcan.2020.08.023
- 2019 International Association for the Study of Lung Cancer. Published by Elsevier Inc.
- 2015 American Association for Cancer Research. DOI: 10.1158/0008-5472.CAN-14-2305

Real Patients, Real Results

60-Year-Old female with metastatic EGFR-mutated lung cancer

EGFR p.T790M; c.2369C>T (Exon 20) 10.9% Osimertinib

GENE	ALTERATION	MUTANT FRACTION	FDA TARGETED THERAPIES (lung cancer)	FDA TARGETED THERAPIES (for other indications)	CLINICAL TRIALS (DETAILS BELOW)
RB1	p.R455*; c.1363C>T	7.2%	None		3
TP53	p.I254T; c.761T>C (Exon 7)	5.3%	None		6
TP53	p.R156C; c.466C>T (Exon 5)	4.2%	None		6

ADDITIONAL THERAPEUTIC INFORMATION (see pg 4)
Chemotherapy

RB1 DESCRIPTION
The protein encoded by this gene is a negative regulator of the cell cycle and was the first tumor suppressor gene found. The encoded protein also stabilizes constitutive heterochromatin to maintain the overall chromatin structure. The active, hypophosphorylated form of the protein binds transcription factor E2F1. Defects in this gene are a cause of childhood cancer retinoblastoma (RB), bladder cancer, and osteogenic sarcoma. [provided by RefSeq, Jul 2008]

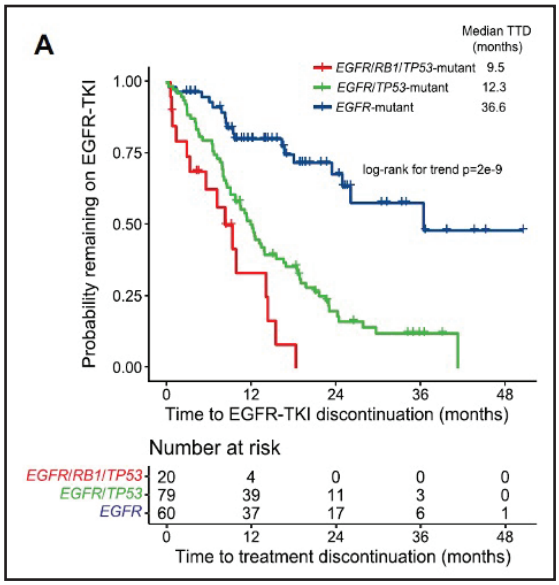
TP53 DESCRIPTION
This gene encodes a tumor suppressor protein containing transcriptional activation, DNA binding, and oligomerization domains. The encoded protein responds to diverse cellular stresses to regulate expression of target genes, thereby inducing cell cycle arrest, apoptosis, senescence, DNA repair, or changes in metabolism. Mutations in this gene are associated with a variety of human cancers, including hereditary cancers such as Li-Fraumeni syndrome. Alternative splicing of this gene and the use of alternate promoters result in multiple transcript variants and isoforms. Additional isoforms have also been shown to result from the use of alternate translation initiation codons from identical transcript variants (PMIDs: 12032546, 20937277). [provided by RefSeq, Dec 2016]

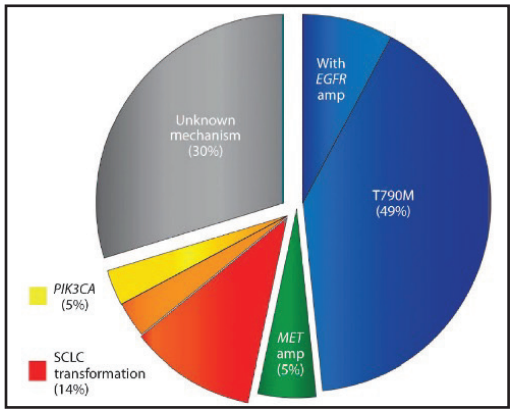
TP53 DESCRIPTION
This gene encodes a tumor suppressor protein containing transcriptional activation, DNA binding, and oligomerization domains. The encoded protein responds to diverse cellular stresses to regulate expression of target genes, thereby inducing cell cycle arrest, apoptosis, senescence, DNA repair, or changes in metabolism. Mutations in this gene are associated with a variety of human cancers, including hereditary cancers such as Li-Fraumeni syndrome. Alternative splicing of this gene and the use of alternate promoters result in multiple transcript variants and isoforms. Additional isoforms have also been shown to result from the use of alternate translation initiation codons from identical transcript variants (PMIDs: 12032546, 20937277). [provided by RefSeq, Dec 2016]

The plasma ctDNA in this patient identifies an EGFR T790M mutation but also RB1 and TP53 co-mutations. These co-mutations provide guidance for a treatment decision today but also direct a heightened clinical awareness of potential evolving tumor biology changes tomorrow.

An unexpected oncologic finding treating EGFR-mutated lung adenocarcinomas with an EGFR TKI has been a 10-15% clonal small cell transformation upon progression. The radiographic presentation is often classic bulky small cell lung cancer with CNS metastasis but can be an isolated recurrence. This appears consistent irrespective of the TKI used, including second- or first-line osimertinib. The EGFR mutation often persists but is no longer the driver oncogene in this setting. Aggressive chemotherapy is forced with transformed small cell median survivals of 10.9 months. De novo small cell histologies with an EGFR mutation have also been reported and appear different, with potential retained EGFR driver and TKI sensitivity.

There is now data to indicate that the finding of RB1 and TP53 co-mutations at the time of diagnosis can portend this aggressive transformation. In a study of 863 patients with EGFR-mutated lung adenocarcinomas evaluated by NGS molecular testing, 5% were EGFR/RB1/TP53 triple mutated. Small cell histology was ultimately seen in 25% of that molecular subset, either de novo or upon progression. Notably, none of the patients without baseline RB1 and TP53 co-mutations had small cell transformation. However, even in those without small cell transformation, the presence of RB1 and TP53 mutations had a much shorter time until progression and discontinuation of the EGFR TKI of only 9.5 months compared to 36.6 months in patients without these two co-mutations.





When RB1 and TP53 co-mutations are present at baseline, a heightened awareness of this small cell transformation potential is needed. PIK3CA mutations also frequently evolve. When present, T790M ctDNA mutations have cleared on osimertinib with SCLC progression, whereas other EGFR mutations persist. Although yet to be known, this shortened benefit of an EGFR TKI alone and the aggressive small cell transformation potential is very compelling for a combinatorial approach of upfront systemic chemotherapy with the EGFR TKI, clearly osimertinib in this setting.

Only with broad NGS testing would this be known. Narrow molecular testing, just looking at targetable driver mutations, would miss clinically impactful co-mutations. A liquid biopsy plasma NGS provides insight into a cancer's aggressive tumor biology that can make a difference for your patient.

Case Study Prepared by Doctor Paul Walker
Chief Medical Officer, Former Director of Thoracic Oncology at East Carolina University

Sources:
- 2019 Published by Elsevier Inc. on behalf of the International Association for the Study of Lung Cancer
- Science Translational Medicine, 23 March 2011 Vol 3 Issue 75 75ra26
-J Clin Oncol. 37:278-285. © 2018 by the American Society of Clinical Oncology
-J Clin Oncol. 35:3065-3074. © 2017 by the American Society of Clinical Oncology
-JAMA Oncol. 2018;4(11):1527-1534. DOI:10.1001/jamaoncol.2018.2969. Published online August 2, 2018