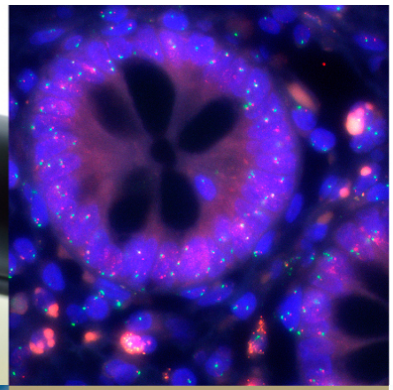
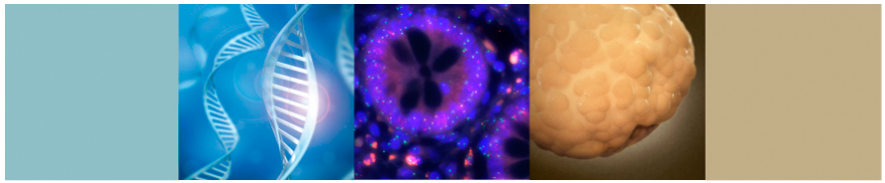






CARIS™
LIFE SCIENCES

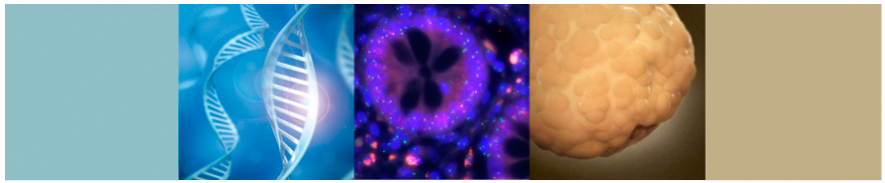
Malignant Peritoneal and Pleural Fluid Samples are adequate for Molecular Profiling

Presenter: Y. Erika Fong, MD
Co-author: Raheela Ashfaq, MD
November 6, 2011



Disclosure:

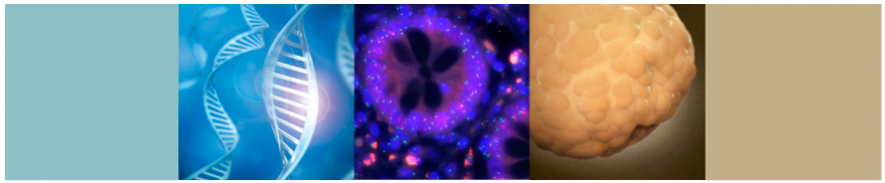
- Caris Life Sciences



CARIS LIFE SCIENCES

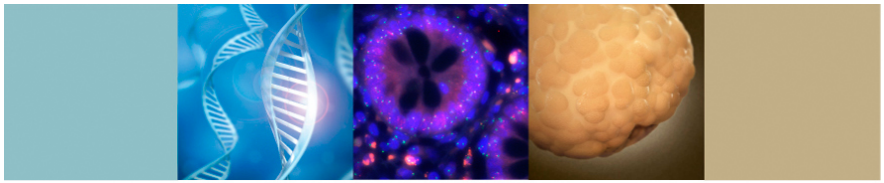
Introduction

- The diagnosis of a malignant effusion in the serosal cavities is a frequent event in the clinical setting of cancer
- Metastatic cancer cells may have unique characteristics that give them the ability to migrate from the primary tumor
- Since cancer patients often experience critical conditions, the analysis of the malignant fluid might be the only tissue sample available for these patients
- With the focus on targeted therapies, evaluation of different sample types for molecular studies is even more important

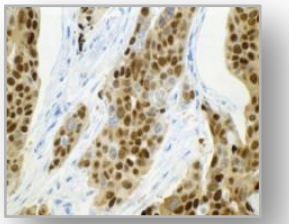


Introduction

- The Caris Target Now™ is proprietary evidence based molecular profiling system for solid tumors which provides specific and individualized molecular profiles for guidance of therapy in advanced stages and metastatic malignancies
- Associates therapeutic agents with potential benefit or potential lack of benefit, and may reveal treatments not previously considered

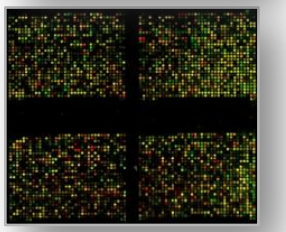


Caris Target Now Technologies



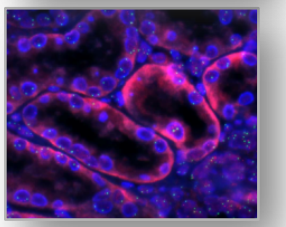
IHC

- Typically 18 predictive biomarkers
- Total of 30 IHCs – use depends on tumor type and progression



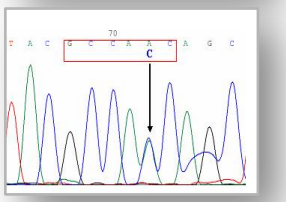
Microarray

- Looking at the over or under expression of the full genome of 24K gene targets, with reporting of 80 genes predicting response to therapies.



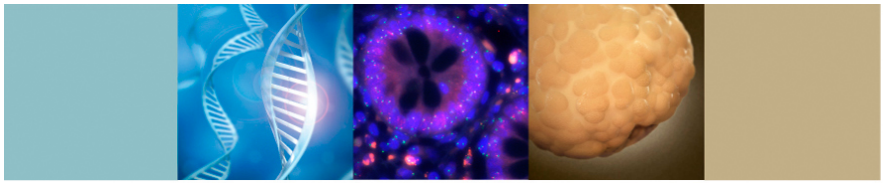
FISH

- Identifying gene copy number alterations in tumor tissue (HER2, EGFR, c-MYC, TOP2A, ALK, PIK3CA, cMET)



Mutational Analysis

- Identifying gene copy number mutations in tumor tissue (KRAS, BRAF, EGFR, c-KIT, PIK3CA)



Summary Report

Agents Associated WITH CLINICAL BENEFIT	Agents Associated With LACK OF CLINICAL BENEFIT
ON NCCN COMPENDIUM™	gemcitabine
erlotinib	irinotecan
cisplatin , carboplatin	doxorubicin, liposomal-doxorubicin, epirubicin
pemetrexed	lapatinib
OFF NCCN COMPENDIUM™	trastuzumab
fluorouracil	
gefitinib	
temozolomide	
calcitriol , cholecalciferol	
sunitinib , sorafenib	

CARIS TARGET NOW™
EVIDENCE-BASED MOLECULAR PROFILING SERVICE

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Patient Information	Specimen Information	Ordered By
Test Patient **Case Number:** TN11-111111 **Date Of Birth:** XX/XX/1949 **Sex:** Female **SSN:** XXX-XX-XXXX	**Primary Tumor Site:** Lung, NOS **Specimen Site:** Lung & Bronchus **Specimen Collected:** XX/XX/2011 **Specimen Received:** 07/08/2011 **Date Reported:** 07/21/2011	**Test Ordering Physician** **The Cancer Center** 1234 Main Street Dallas, TX 12345 123-456-7890

Caris Target Now Final Report

TN2011-06-17_0

Clinical History
Per the submitted surgical pathology report, the patient is a 61 year-old female with a history of adenosquamous carcinoma of lung.

Pathologic Diagnosis
Lower lobe, left lung, lobectomy: Invasive moderately differentiated adenosquamous cell carcinoma.

Agents Associated WITH CLINICAL BENEFIT	Agents Associated With LACK OF CLINICAL BENEFIT
ON NCCN COMPENDIUM™	gemcitabine
erlotinib	irinotecan
cisplatin , carboplatin	doxorubicin, liposomal-doxorubicin, epirubicin
pemetrexed	lapatinib
OFF NCCN COMPENDIUM™	trastuzumab
fluorouracil	
gefitinib	
temozolomide	
calcitriol , cholecalciferol	
sunitinib , sorafenib	

Caris Target Now is an evidence-based molecular profiling service that associates biomarker status to agents with potential clinical benefit or potential lack of clinical benefit. Agents associated with clinical benefit are presented based on NCCN Compendium™ inclusion, relevance of tumor lineage, level of published evidence and strength of biomarker expression. The agents are not ranked in order of potential or predicted efficacy. The information in this report must be considered in conjunction with all other relevant information in respect of a given patient before determining the appropriate course of treatment. The agents identified may not be suitable for use with a particular patient and the report does not guarantee that any particular agent will be effective with the treatment of any particular condition. The selection of any, all or none of the matched agents resides with the discretion of the treating physician. Caris Life Sciences does not represent that any patient will be reimbursed or paid for by any healthcare provider or insurer.

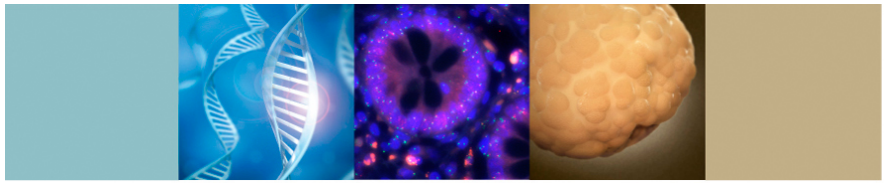
Caris Life Sciences has exercised all reasonable skill and care in the preparation of this report and believes that its findings will assist in the selection of appropriate treatments. Caris Life Sciences expressly excludes, all other representations, warranties, conditions and terms

**** FINAL REPORT ****

An expert oncology consultation can be arranged if a request is made through our Client Services Department at 1-800-901-5177.

Patient: Test Patient **TN11-111111** **Physician:** Test Ordering Physician

Caris Life Sciences / 4610 S. 44th Place / Phoenix, Arizona 85040 / Ph: 800.901.5177 / Fax: 866.479.4925 / CLIA 03D1019490
Caris Life Sciences / 4207 E Cotton Center Blvd / Phoenix, Arizona 85040 / Ph: 800.901.5177 / Fax: 866.479.4925 / CLIA 03D1064744
Caris Life Sciences / 6655 Macarthur Blvd / Irving, Texas 75039 / Ph: 800.901.5177 / Fax: 866.479.4925 / CLIA 45D0975010



Pilot Study Using Molecular Profiling of Patients' Tumors to Find Potential Targets and Select Treatments for Their Refractory Cancers

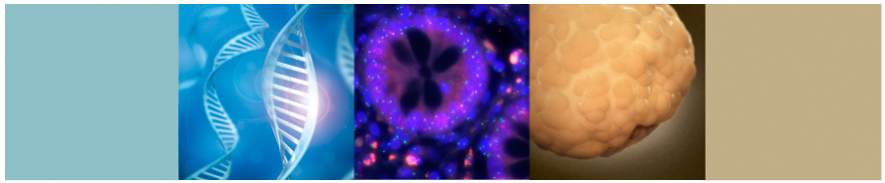
Primary Objective

- Compare progression free survival (PFS) for therapy selected by molecular profiling with PFS for the last line of therapy on which the patient progressed

If PFS_b/PFS_a ratio was $\geq$ 1.3, MP-selected therapy was defined as having benefit for patient.

PFS: length of time during and after treatment in which a patient is living with a disease that does not get worse

Von Hoff, D.D., "Pilot Study Using Molecular Profiling of Patients' Tumors to Find Potential Targets and Select Treatments for Their Refractory Cancers", Journal of Clinical Oncology, Published Online October 4, 2010: 10.1200/JCO.2009.26.5983 ; Temple, R. *Clinical Measurement in Drug Evaluation*. Ningano W. Thicker GT, eds. John Wiley and Sons Ltd: 1995; Von Hoff, D.D. c 1999; Dhani et al. *Clinical Cancer Research*. 2009; 15: 118-123.

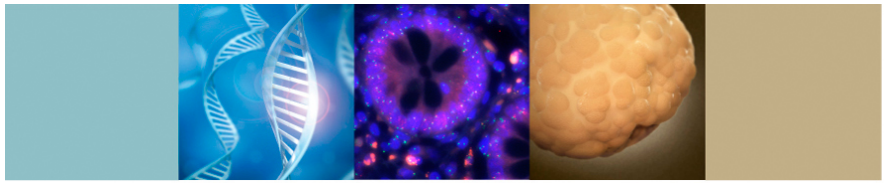


Results: Primary Endpoints

- 27% of patients had PFS ratio > 1.3
- 95% confidence interval (CI): 17% - 38%
- P = 0.007

Tumor Type	Total Treated	Number with PFS Ratio > 1.3	%
Breast	18	8	44
Colorectal	11	4	36
Ovarian	5	1	20
Miscellaneous*	32	5	16
	66	18	27

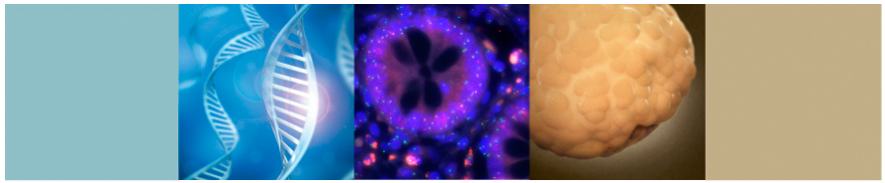
*Miscellaneous tumor types with PFS ratio > 1.3 included lung, cholangiocarcinoma, mesothelioma, eccrine sweat glands, and GIST (gastric).



Results: Overall Survival

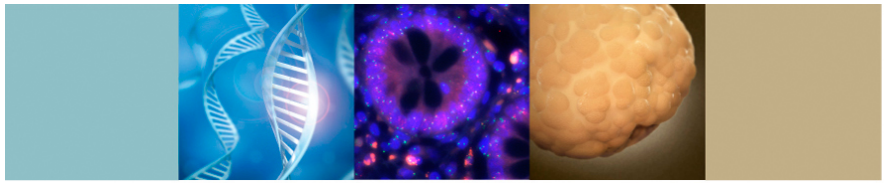
	N	Median OS (months)
Patients with PFS ≥ 1.3 (responders)	18	9.7
Patients who did not respond to molecular-profiling-selected treatments (non responders)	48	3.2
All patients who received molecular profiling (responders + non responders)	66	5.0
Patients whose treatment was not selected by molecular profiling	40	3.2

Patients with PFS ≥ 1.3 had longer OS by 6.5 months compared to non responders and patients whose treatment was not selected by molecular profiling



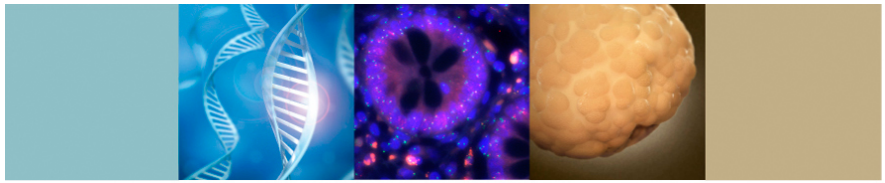
Study Conclusions

- Molecular profiling identified agents that would not have been the oncologist's first choice (0% correlation)
- Results support use of molecular profiling as means to successfully identify new treatment targets for patients with metastatic tumors
- Molecular profiling suggested regimens resulted in longer PFS in 27% of patients
- Longer PFS was demonstrated in patients with different histological types of tumors



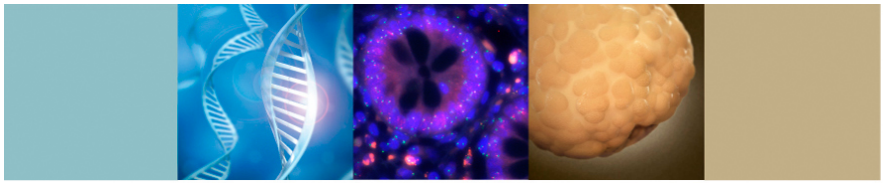
Objective

- The purpose of this study is to evaluate the feasibility of molecular profiling in pleural and peritoneal fluids



Material and Methods

- A computer search was conducted to retrospectively identify malignant fluid samples or cell blocks from January 2009 to April 2011



1

Bottle of Fluid

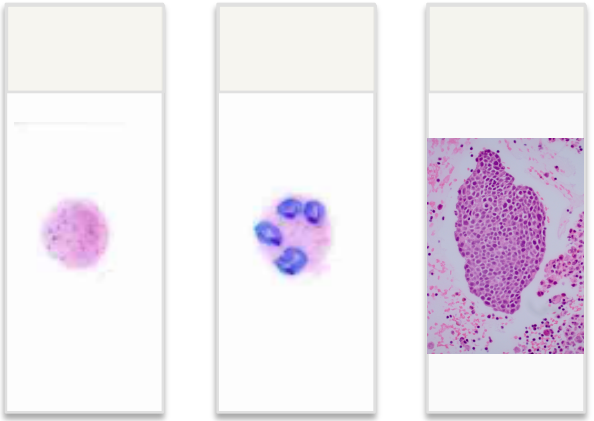
Centrifuge

Pallet

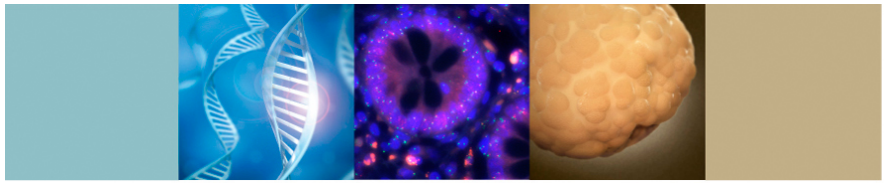
Paraffin Block

2

Paraffin Block

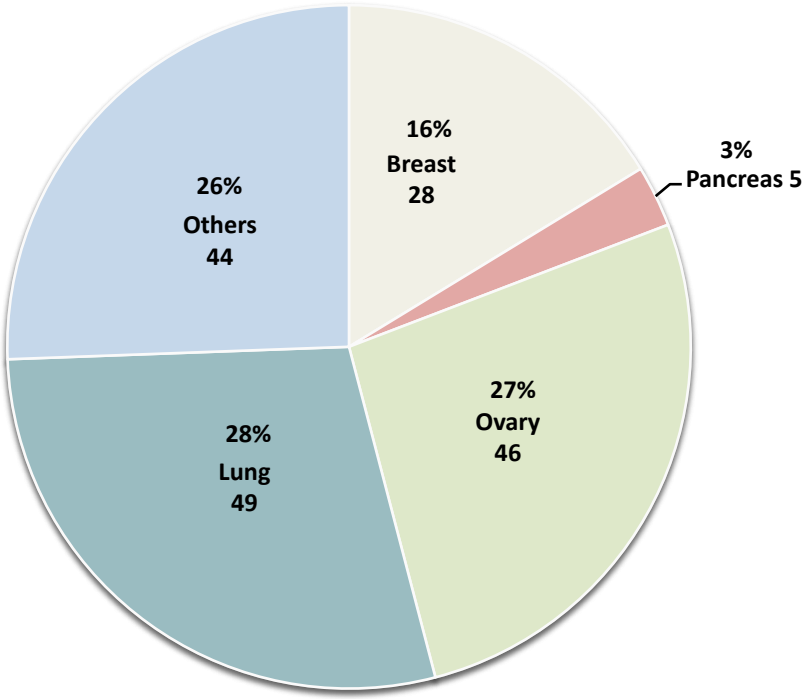


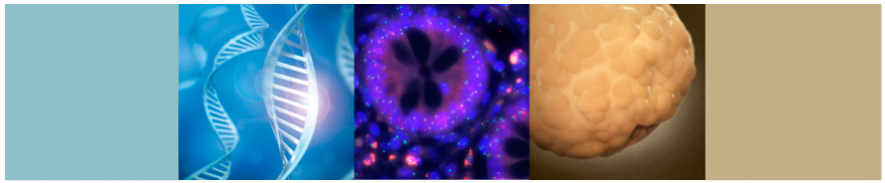
H&E Slides



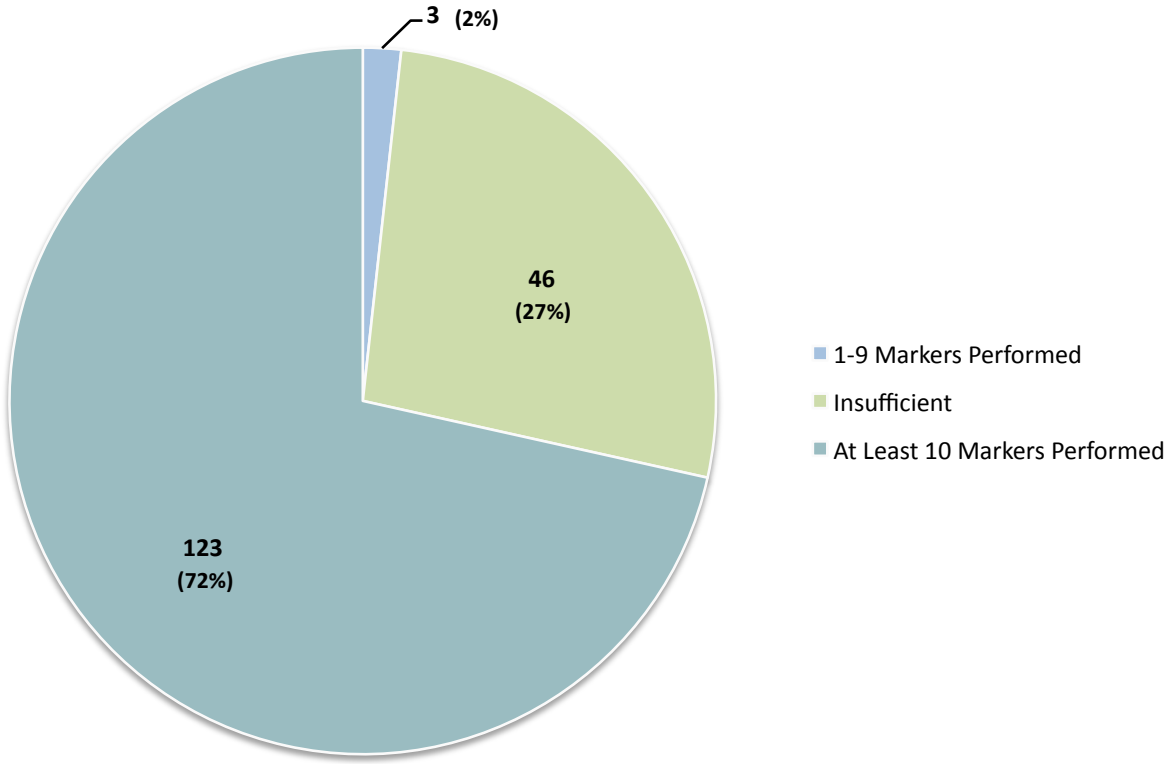
Results

172 Samples of peritoneal and pleural fluids

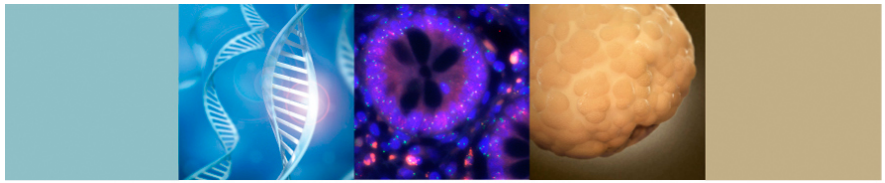




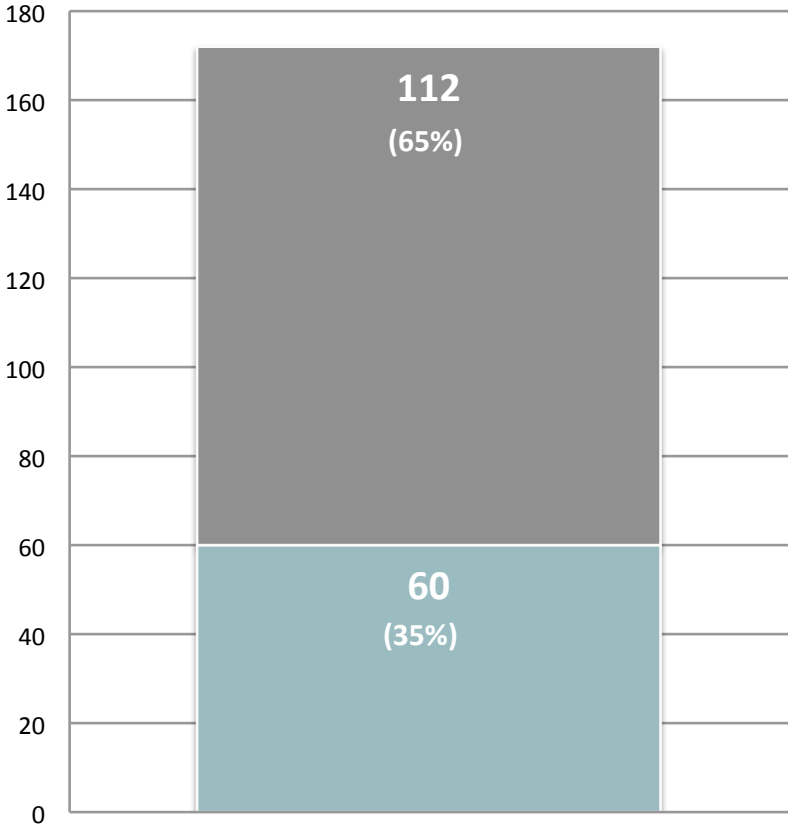
IHC



Based on 172 Samples of peritoneal and pleural fluids



Microarray



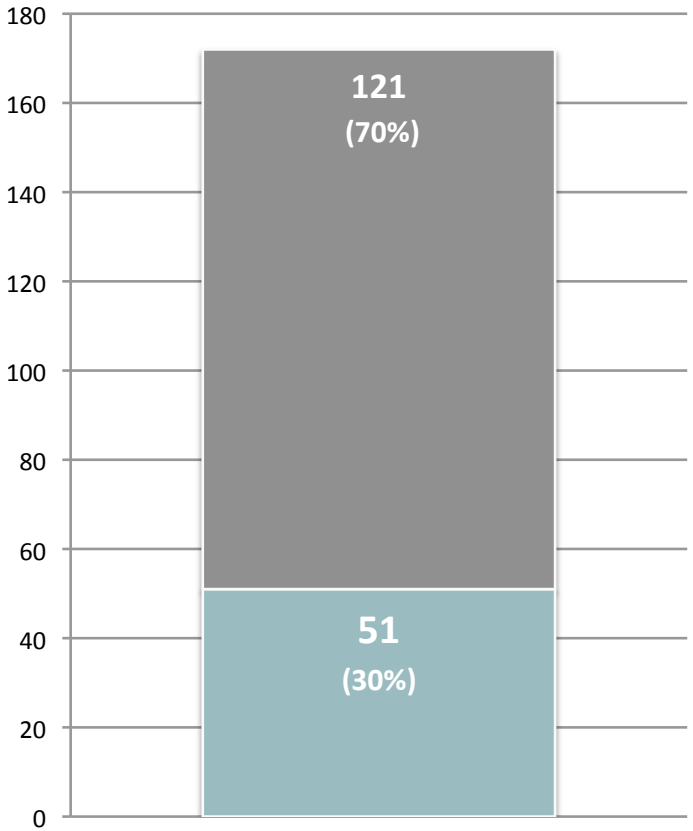
- Not performed
- Microarray Performed

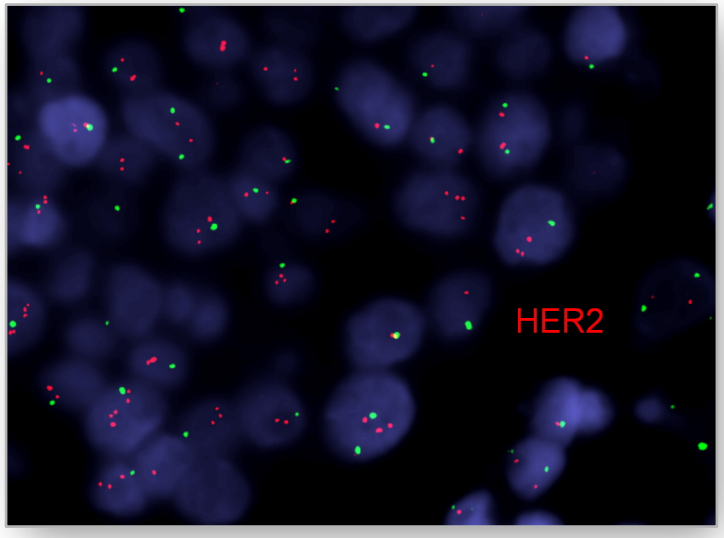
Microarray Analysis of RNA Expression on Paraffin Blocks Tissue

Gene	Ratio	Expression*	Significant Result
PDGFRB	0.03	Under Expressed	
KIT	0.05	Under Expressed	
PTGS2	0.09	Under Expressed	
IGFBP5	0.12	Under Expressed	
EGFR	0.12	Under Expressed	
SPARC	0.18	Under Expressed	
GNRH1	0.19	Under Expressed	
MET	0.19	Under Expressed	
GART	0.22	Under Expressed	✓
PDGFRA	0.24	Under Expressed	
HIF1A	0.24	Under Expressed	
VDR	0.26	Under Expressed	
CDA	0.27	Under Expressed	
SSTR5	0.27	Under Expressed	
PGR	0.27	Under Expressed	
ASNS	0.28	Under Expressed	
NFKB2	0.28	Under Expressed	
SIK2	0.36	Under Expressed	
SRC	0.50	Under Expressed	
DHFR	0.56	Under Expressed	✓
MSH2	0.57	Under Expressed	
OGFR	0.59	Under Expressed	
CD52	0.60	Under Expressed	
GSTP1	0.62	Under Expressed	
AR	0.62	Under Expressed	
VHL	0.63	Under Expressed	✓
RRM1	0.64	Under Expressed	✓
BCL2	0.65	No Change	
TOP2B	0.66	No Change	
ERCC1	0.68	No Change	
ERBB2	0.68	No Change	
FYN	0.71	No Change	
RAF1	0.73	No Change	
BRCA2	0.76	No Change	
ERCC3	0.79	No Change	
PTEN	0.80	No Change	
PDGFC	0.80	No Change	
YES1	0.88	No Change	
MLH1	0.90	No Change	
RXRB	0.92	No Change	
IGFBP4	0.92	No Change	
ADA	1.02	No Change	
NFKBIA	1.11	No Change	
SSTR1	1.15	No Change	
TYMS	1.29	No Change	
KDR	1.31	No Change	
MGMT	1.34	No Change	
CES2	1.36	No Change	
RXRG	1.45	No Change	
FLT1	1.47	No Change	
TOP1	1.54	No Change	
NFKB1	1.54	No Change	
LYN	1.55	Over Expressed	
ABCG2	1.59	No Change	
TK1	1.62	Over Expressed	
DNMT3B	1.70	Over Expressed	
DNMT1	1.71	Over Expressed	
RARA	1.71	Over Expressed	
BRCA1	1.75	Over Expressed	
HDAC1	1.77	Over Expressed	
HSP90AA1	1.81	Over Expressed	
DNMT3A	1.83	Over Expressed	
SSTR4	1.99	No Change	
TXNRD1	2.07	Over Expressed	
PGP	2.24	Over Expressed	
TNF	2.32	Over Expressed	
DCK	2.47	Over Expressed	✓
FOLR2	2.53	Over Expressed	
RRM2B	2.56	Over Expressed	
TOP2A	2.82	Over Expressed	✓
IL2RA	2.88	Over Expressed	
BIRC5	2.99	Over Expressed	
LCK	2.99	Over Expressed	
PARP1	3.10	Over Expressed	
ECGF1	3.33	Over Expressed	
HCK	3.62	Over Expressed	
CD33	3.86	Over Expressed	
VEGFA	4.00	Over Expressed	
ZAP70	5.48	Over Expressed	
ESR1	6.50	Over Expressed	✓
RRM2	8.92	Over Expressed	

Based on 172 Samples of peritoneal and pleural fluids

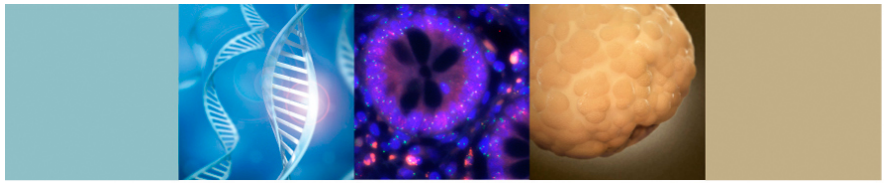
FISH



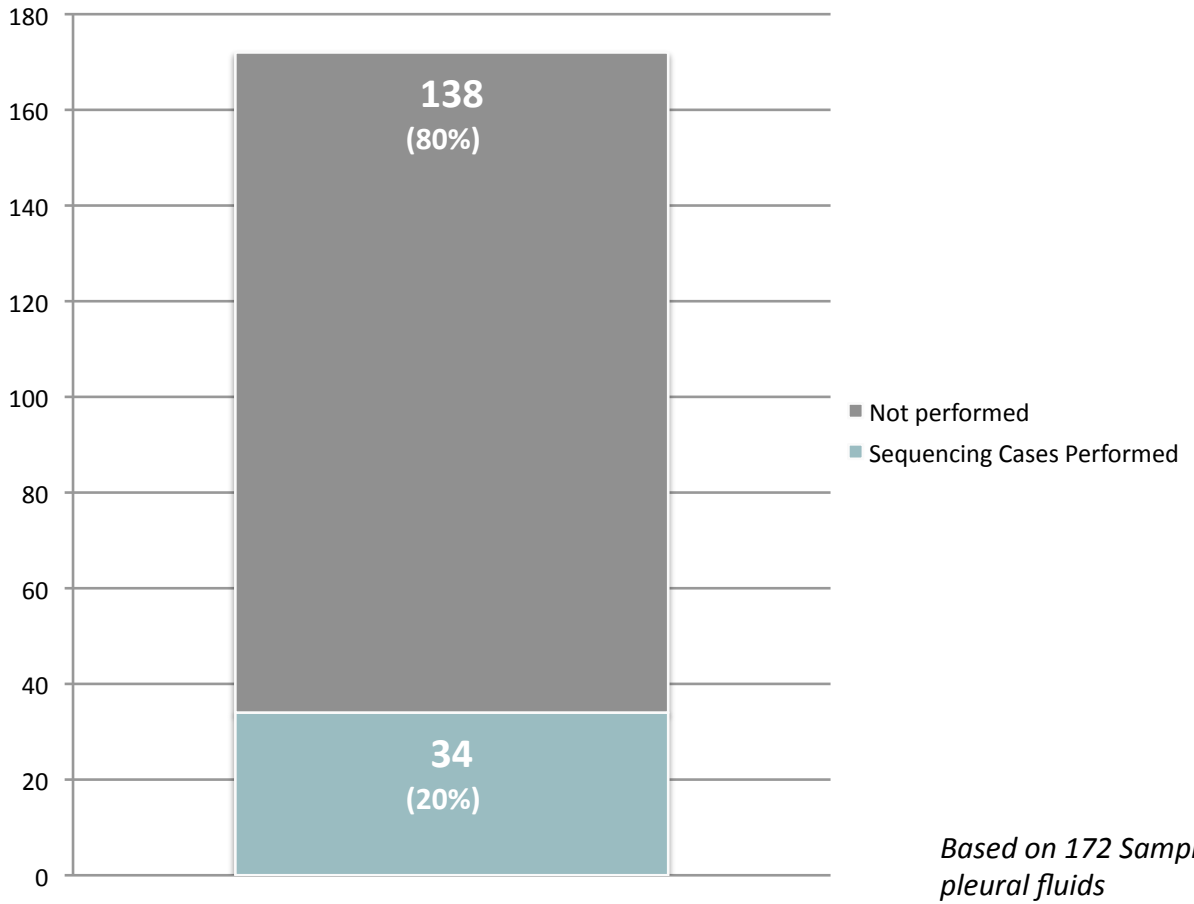


HER2/NEU FISH

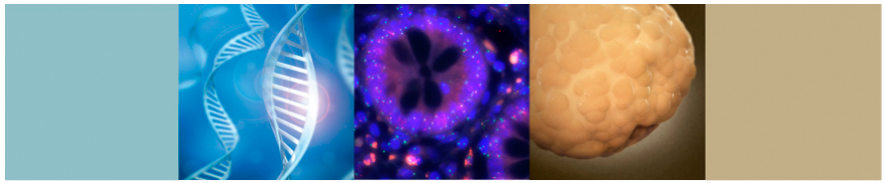
Based on 172 Samples of peritoneal and pleural fluids



Sequencing

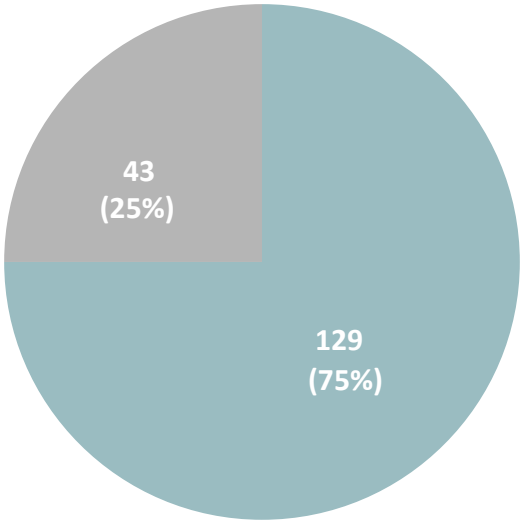


Based on 172 Samples of peritoneal and pleural fluids

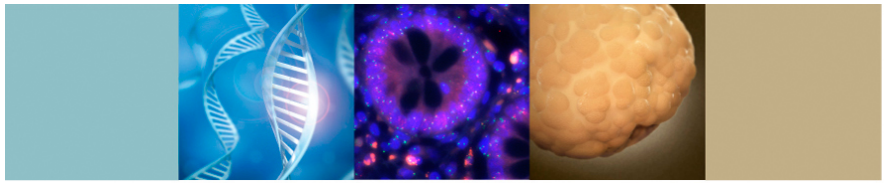


Results

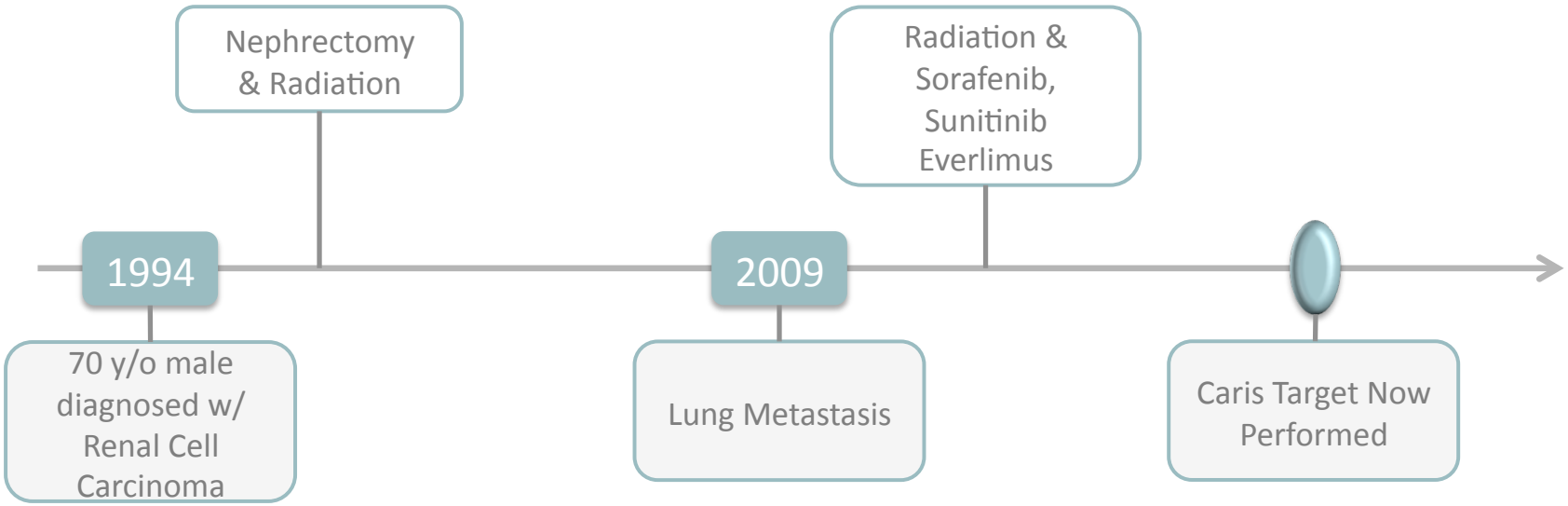
- Combined results of predictive markers from these various platforms were able to provide information on therapeutic guidance for associated clinical benefit or lack of clinical benefit for various therapies in 129 of the 172 cases (75%)



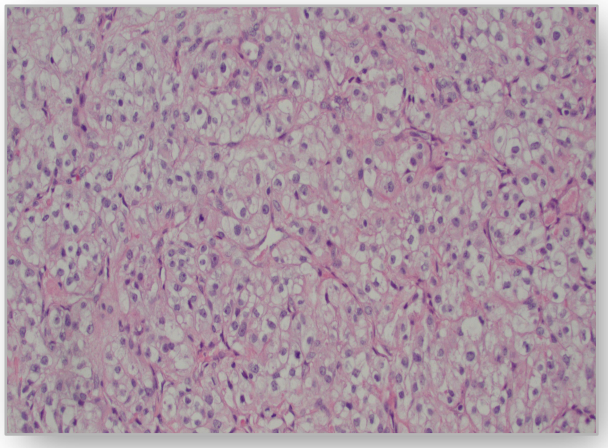
Based on 172 Samples of peritoneal and pleural fluids



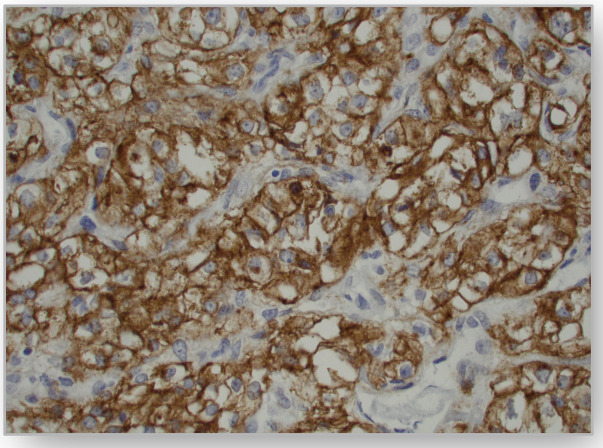
Case #1



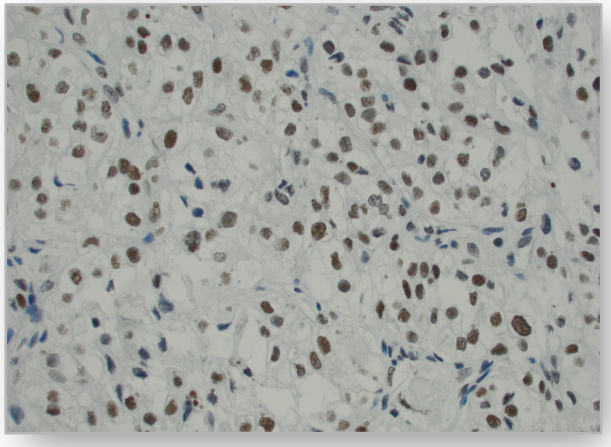
Case #1



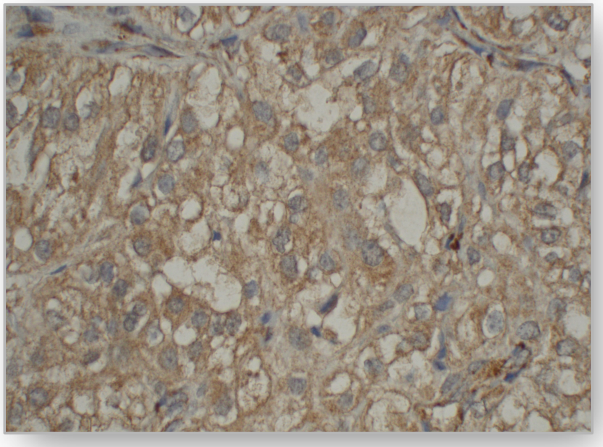
HE



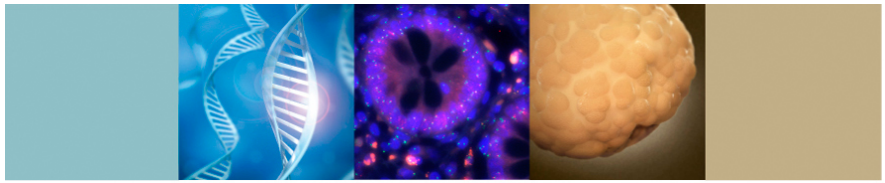
EGFR IHC



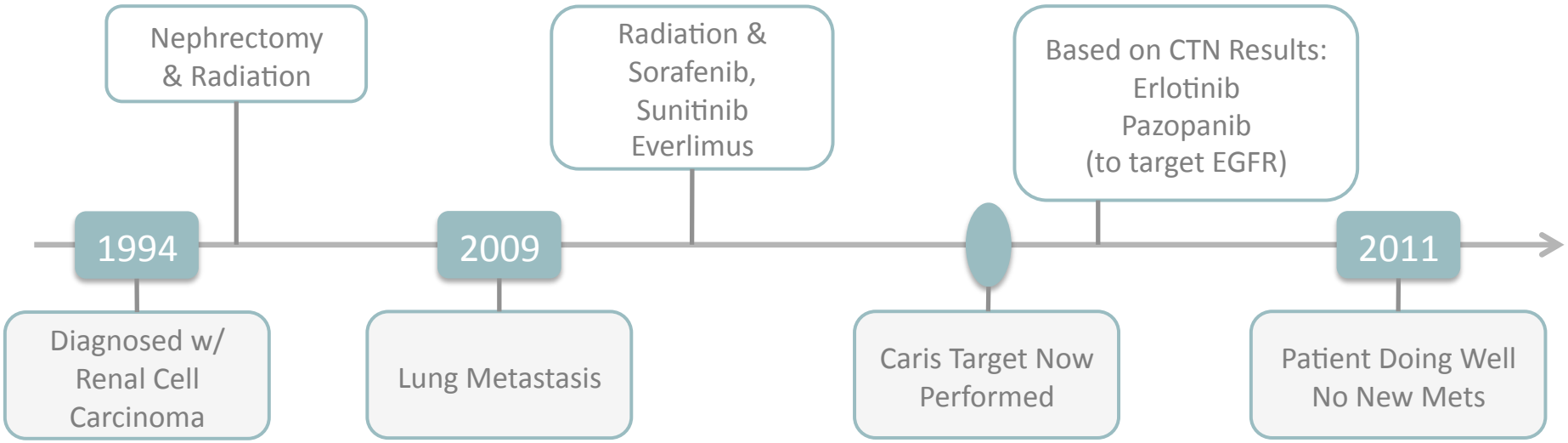
TOPO1

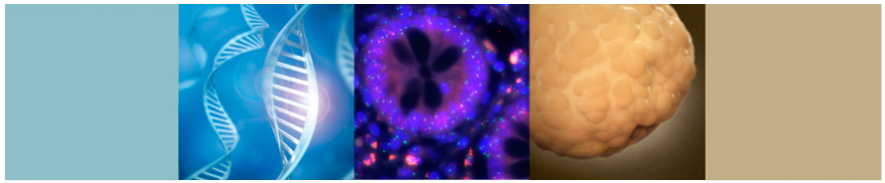


SPARC IHC



Case#1





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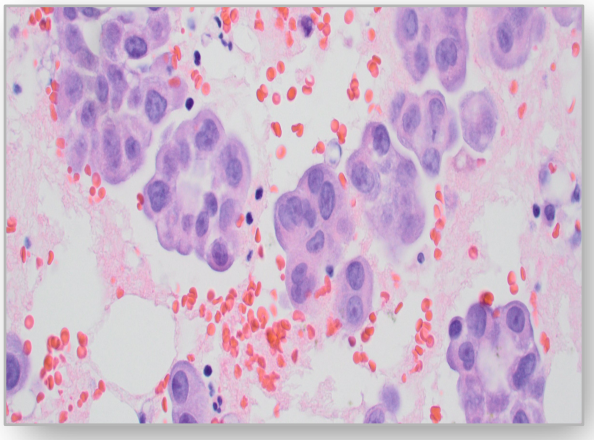
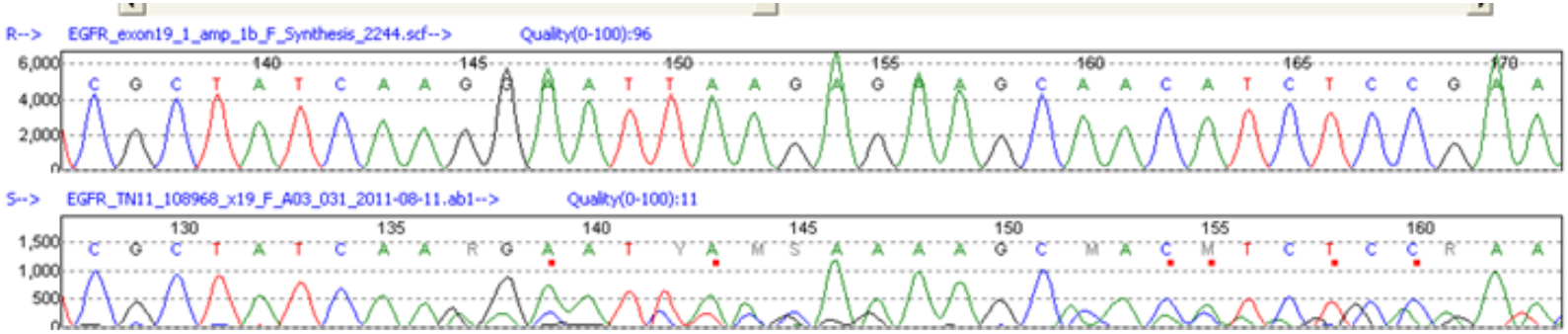
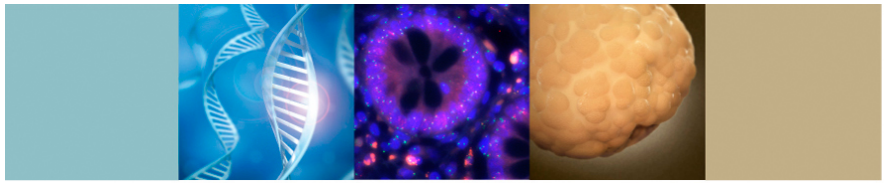
Case #2: 59 year old female with history of metastatic lung cancer

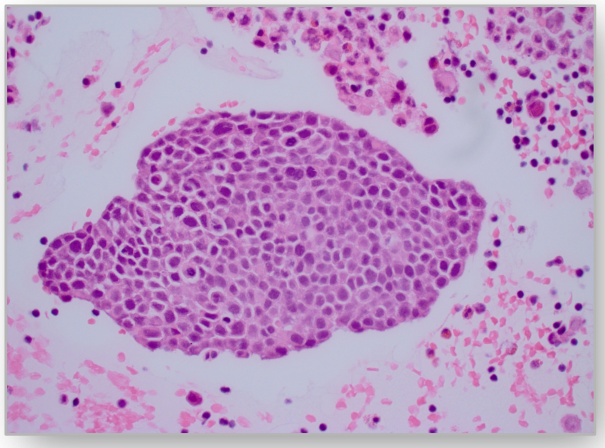


H/E

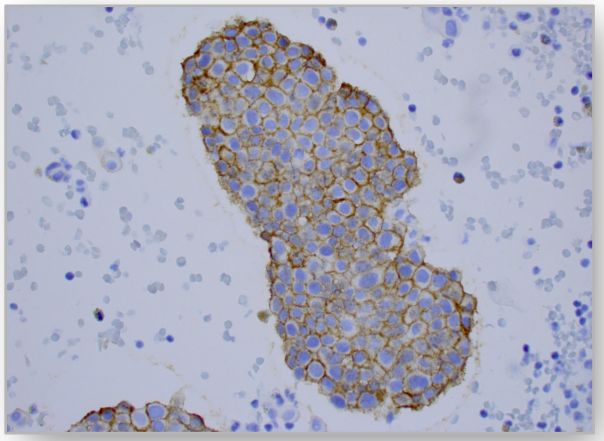




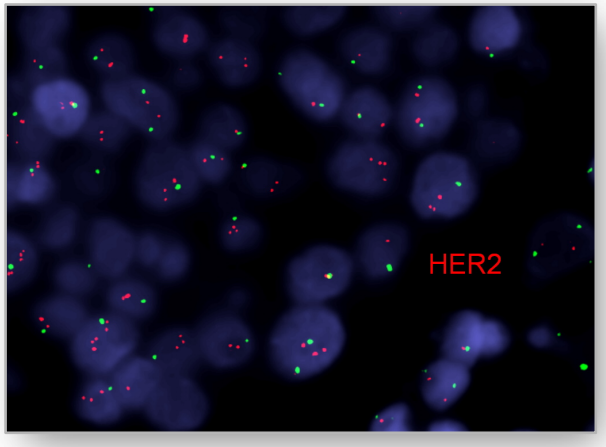
Case # 3: 48 year old female with history of metastatic breast cancer



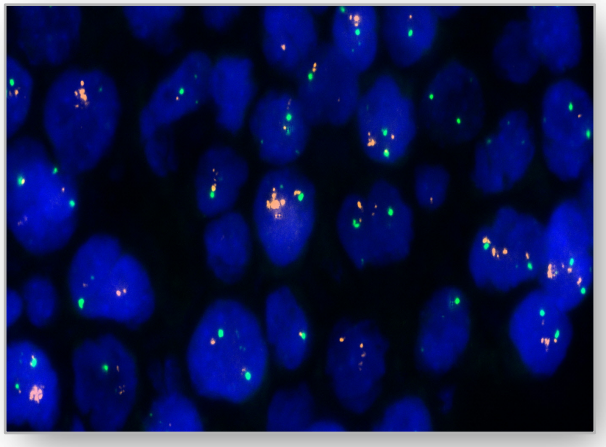
HE



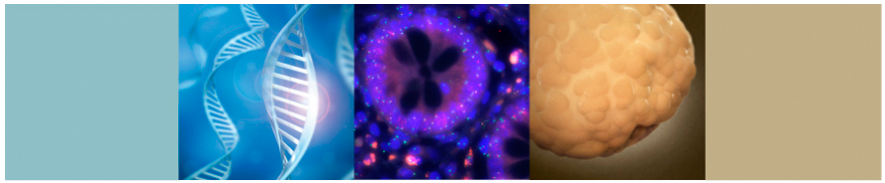
HER2/NEU IHC



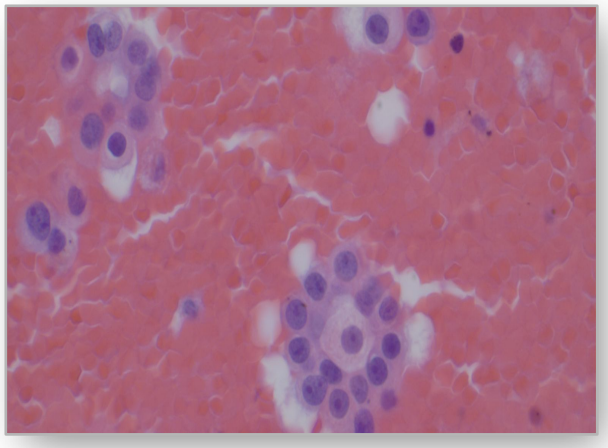
HER2/NEU FISH



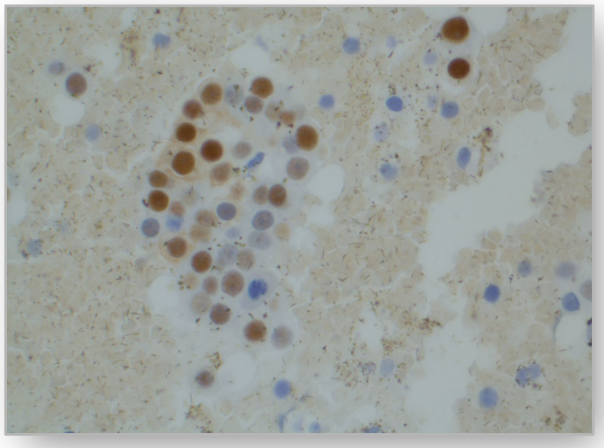
TOPO2 FISH



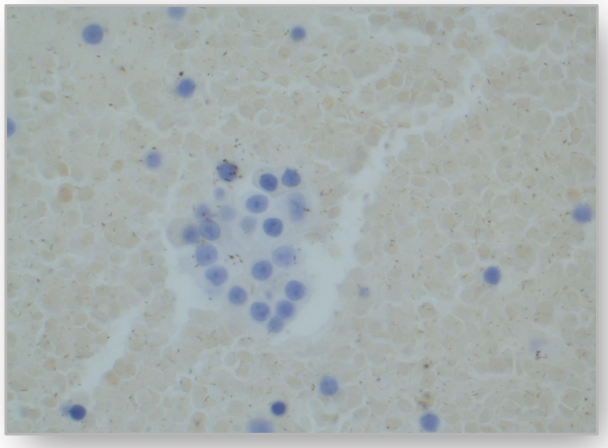
Case #4: 61 year old female with history of metastatic breast cancer



HE



ER IHC



PR IHC

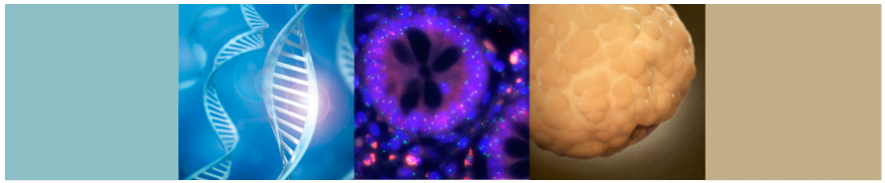
Microarray Analysis of RNA Expression on Paraffin Blocks Tissue

Gene	Ratio	Expression*	Significant Result
PDGFRB	0.03	Under Expressed	
KIT	0.05	Under Expressed	
PTGS2	0.09	Under Expressed	
IGFBP5	0.12	Under Expressed	
EGFR	0.12	Under Expressed	
SPARC	0.18	Under Expressed	
GNRH1	0.19	Under Expressed	
MET	0.19	Under Expressed	
GART	0.22	Under Expressed	✓
PDGFRA	0.24	Under Expressed	
HIF1A	0.24	Under Expressed	
VDR	0.26	Under Expressed	
CDA	0.27	Under Expressed	
SSTR5	0.27	Under Expressed	
PGR	0.27	Under Expressed	
ASNS	0.28	Under Expressed	
NFKB2	0.28	Under Expressed	
SIK2	0.36	Under Expressed	
SRC	0.50	Under Expressed	
DHFR	0.56	Under Expressed	✓
MSH2	0.57	Under Expressed	
OGFR	0.59	Under Expressed	
CD52	0.60	Under Expressed	
GSTP1	0.62	Under Expressed	
AR	0.62	Under Expressed	
VHL	0.63	Under Expressed	✓
RRM1	0.64	Under Expressed	✓

Gene	Ratio	Expression*	Significant Result
BCL2	0.65	No Change	
TOP2B	0.66	No Change	
ERCC1	0.68	No Change	
ERBB2	0.68	No Change	
FYN	0.71	No Change	
RAF1	0.73	No Change	
BRCA2	0.76	No Change	
ERCC3	0.79	No Change	
PTEN	0.80	No Change	
PDGFC	0.80	No Change	
YES1	0.88	No Change	
MLH1	0.90	No Change	
RXRB	0.92	No Change	
IGFBP4	0.92	No Change	
ADA	1.02	No Change	
NFKBIA	1.11	No Change	
SSTR1	1.15	No Change	
TYMS	1.29	No Change	
KDR	1.31	No Change	
MGMT	1.34	No Change	
CES2	1.36	No Change	
RXRG	1.45	No Change	
FLT1	1.47	No Change	
TOP1	1.54	No Change	
NFKB1	1.54	No Change	
LYN	1.55	Over Expressed	
ABCG2	1.59	No Change	

Gene	Ratio	Expression*	Significant Result
TK1	1.62	Over Expressed	
DNMT3B	1.70	Over Expressed	
DNMT1	1.71	Over Expressed	
RARA	1.71	Over Expressed	
BRCA1	1.75	Over Expressed	
HDAC1	1.77	Over Expressed	
HSP90AA1	1.81	Over Expressed	
DNMT3A	1.83	Over Expressed	
SSTR4	1.99	No Change	
TXNRD1	2.07	Over Expressed	
PGP	2.24	Over Expressed	
TNF	2.32	Over Expressed	
DCK	2.47	Over Expressed	✓
FOLR2	2.53	Over Expressed	
RRM2B	2.56	Over Expressed	
TOP2A	2.82	Over Expressed	✓
IL2RA	2.88	Over Expressed	
BIRC5	2.99	Over Expressed	
LCK	2.99	Over Expressed	
PARP1	3.10	Over Expressed	
ECGF1	3.33	Over Expressed	
HCK	3.62	Over Expressed	
CD33	3.86	Over Expressed	
VEGFA	4.00	Over Expressed	
ZAP70	5.48	Over Expressed	
ESR1	6.50	Over Expressed	✓
RRM2	8.92	Over Expressed	

Microarray

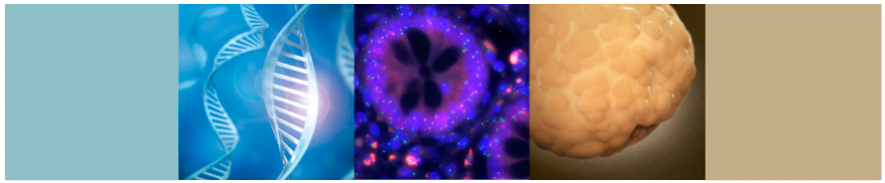


Conclusion:

- Molecular profiling of malignant effusions offers additional opportunities for testing when other tissue samples, such as needle core biopsy or tumor resection, are not available

- Molecular profiling of effusion samples can provide insight into the molecular characteristics of malignant cells

- Molecular profiling of malignant effusion can provide information to create targeted therapies for cancer



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Thanks