CONTRIBUCION DE LAS TECNICAS DE BIOLOGIA MOLECULAR EN LOS TUMORES METASTATICOS DE ORIGIN DESCONOCIDO

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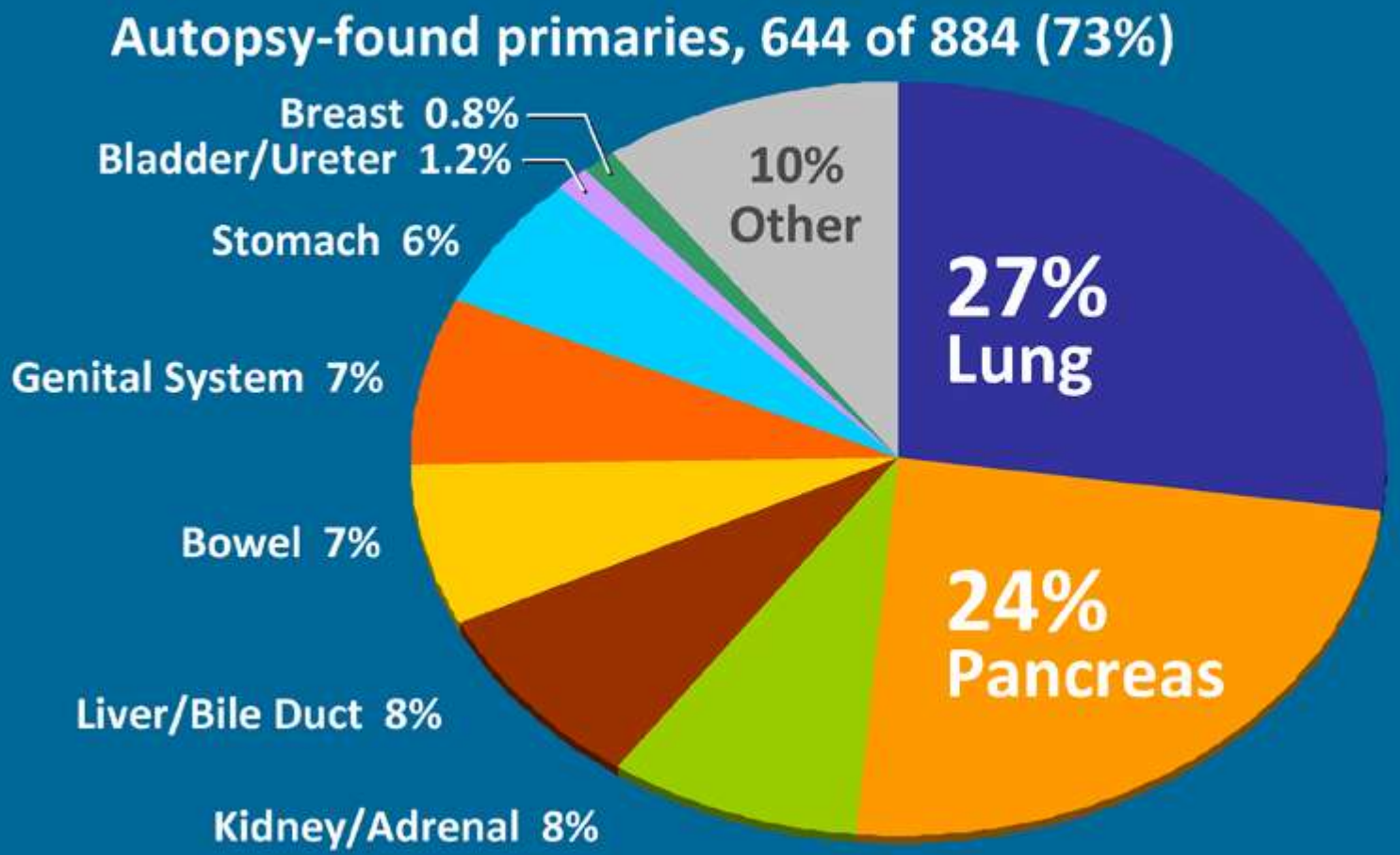
CANCER UNKNOWN PRIMARY

- 3-5 % of diagnosed cancers
- Variability depending of centers
- 50% adenocarcinomas
- Immunohistochemistry helps in 30-70%
- Metastases to liver (25%) and bone (25%)
- Prognosis depends of identification of primary site.

Primary Sites Determined at Autopsy in 884 Patients With Unknown Primary Cancer



Pentheroudakis G, Golfinopoulos V, Pavlidis N. Switching benchmarks in cancer of unknown primary: from autopsy to microarray. *Eur J Cancer.* 2007;43:2026-2036.

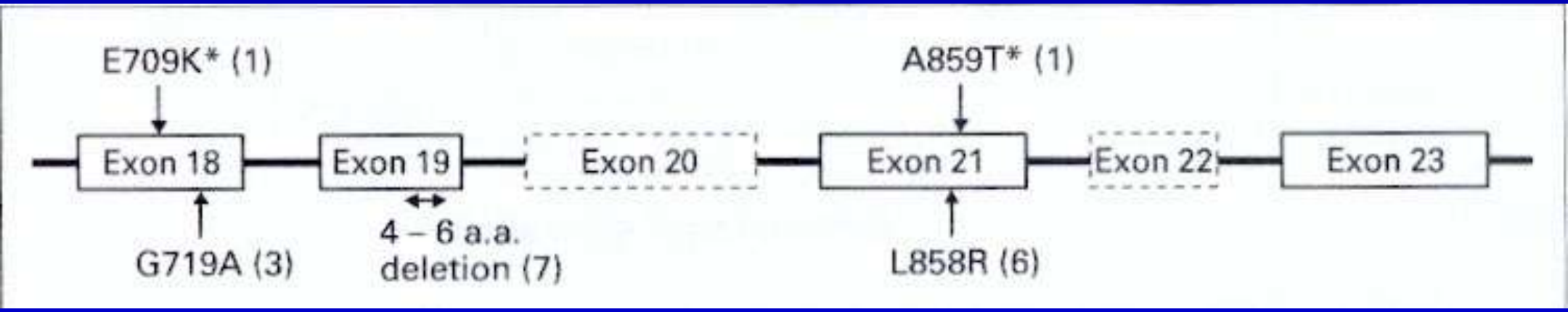
B **Sensitivity and specificity of each marker in first round of immunohistochemistry**

Marker	Tumors of	% Sensitivity	% Specificity
PSA	Prostate	100	99
TTF1	Lung	91	98
CDX2	Colon	83	96
CDX2	Colon and stomach	56	98
CK20	Colon	68	91
CK20	Colon, stomach and pancreas	36	97
GCDFP-15	Breast	54	96
ER	Breast and ovary	74	95
CA125	Ovary and pancreas	88	88
Mesothelin	Ovary and pancreas	85	85
Lysozyme	Stomach and pancreas	65	69
CK7	(Stomach and pancreas) versus colon	72	96

MOLECULAR APPROACHES IN TUMORS OF UNKNOWN ORIGIN

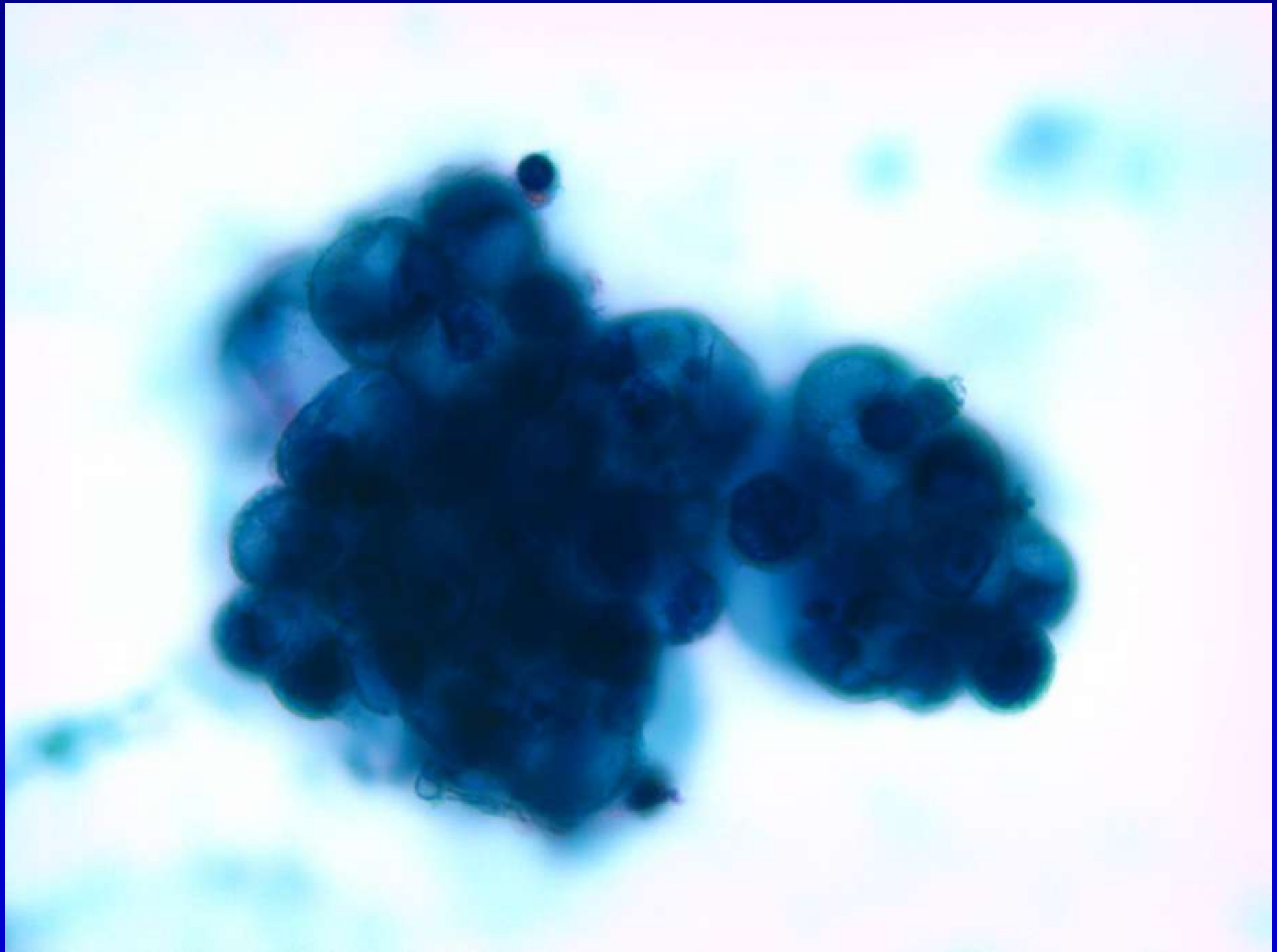
- ONE GENE
- MULTIPLE GENES

LUNG CANCER AND EGFR MUTATIONS

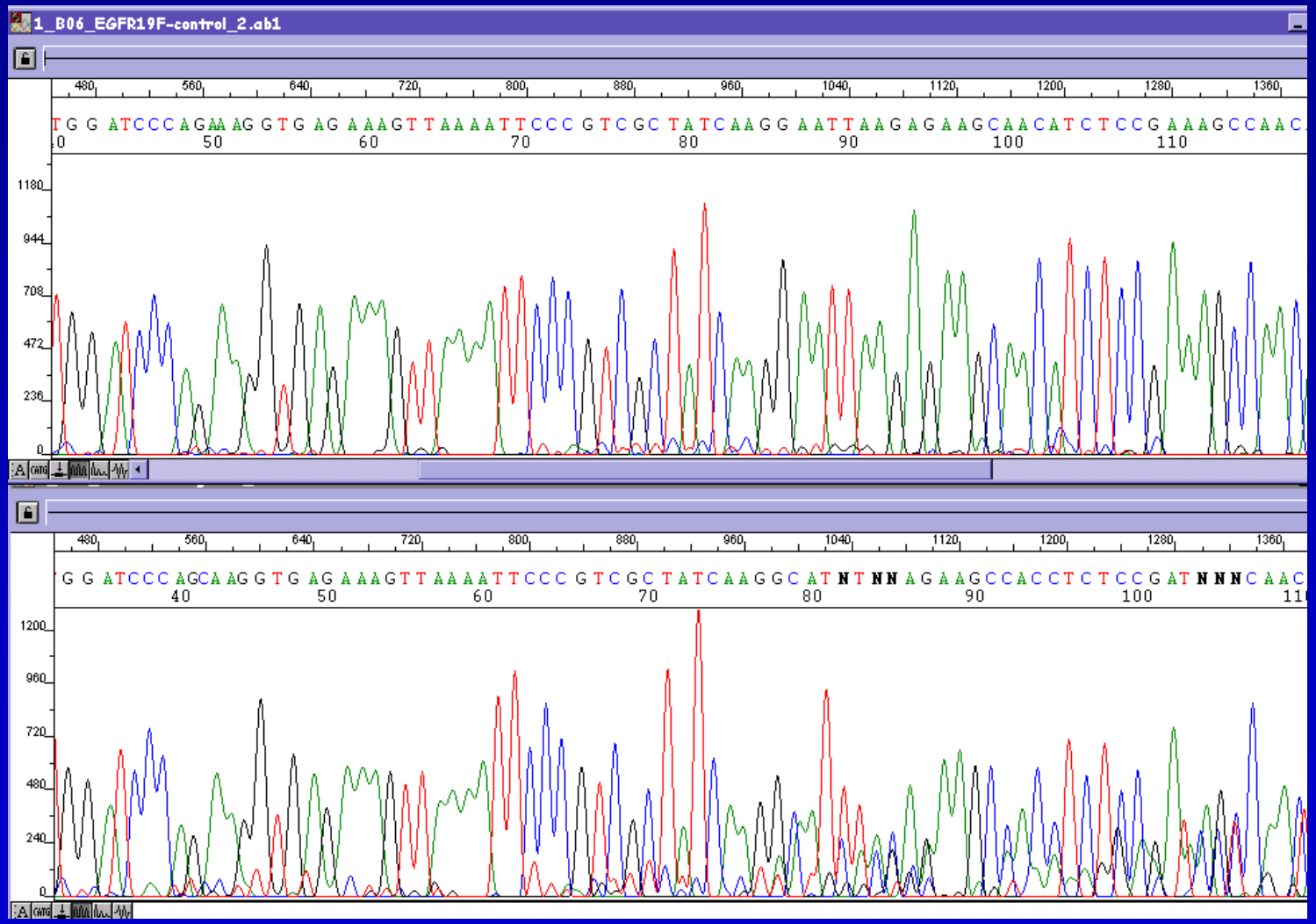


Clinical History

- **83 year old man**
- **Disseminated cancer (liver and lung). Pleural Effusion**
- **Pleural Cytology: Positive for malignancy, consistent with adenocarcinoma. TTF-1 negative**



EGFR exon 19 E746-A750 del 15pb



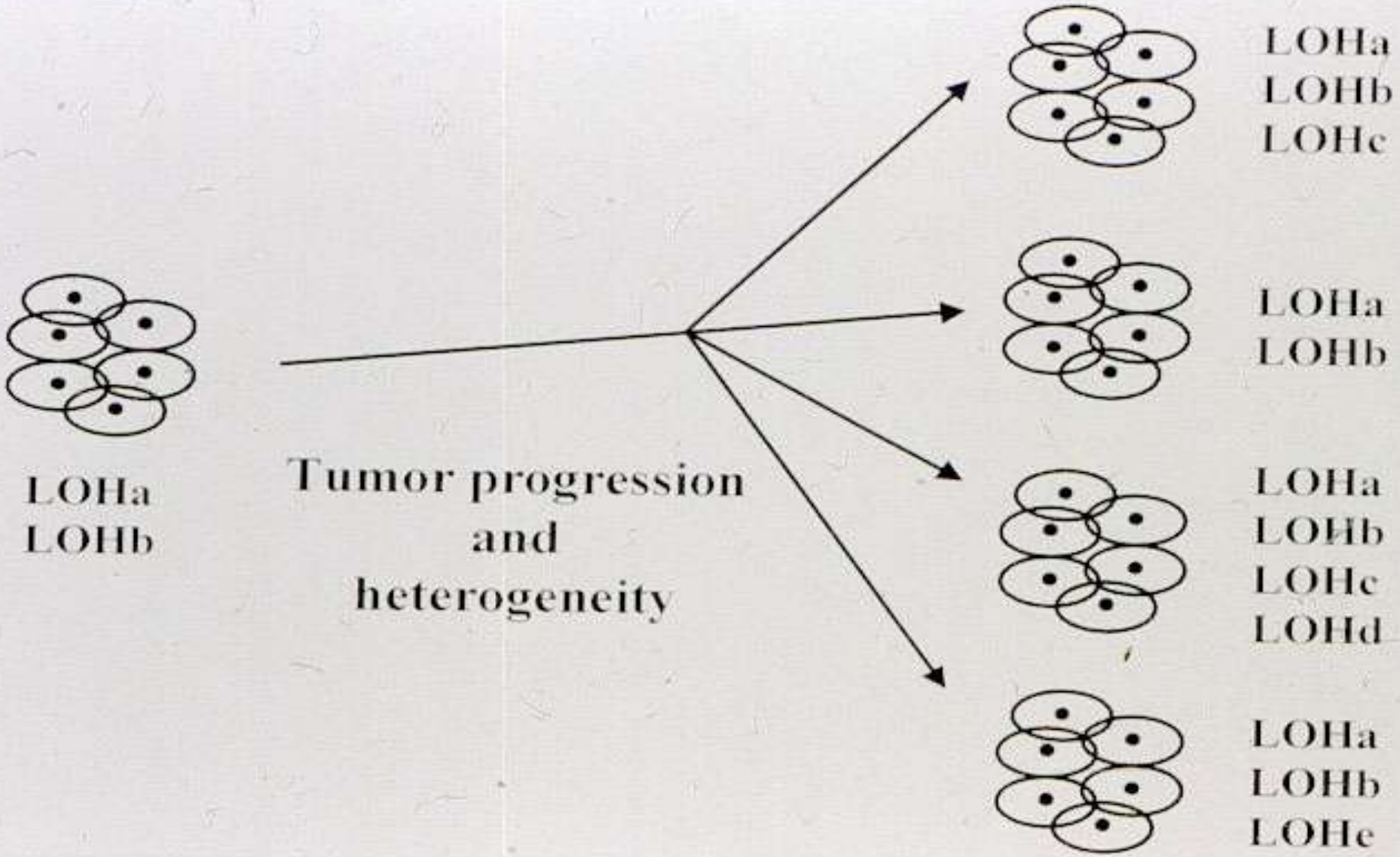
Diagnosis

Pleural metastasis from pulmonary adenocarcinoma with EGFR mutations. Treatment with EGFR inhibitors.

ASSESSMENT OF PRIMARY ORIGIN IN PATIENTS WITH A PREVIOUS HISTORY OF CANCER

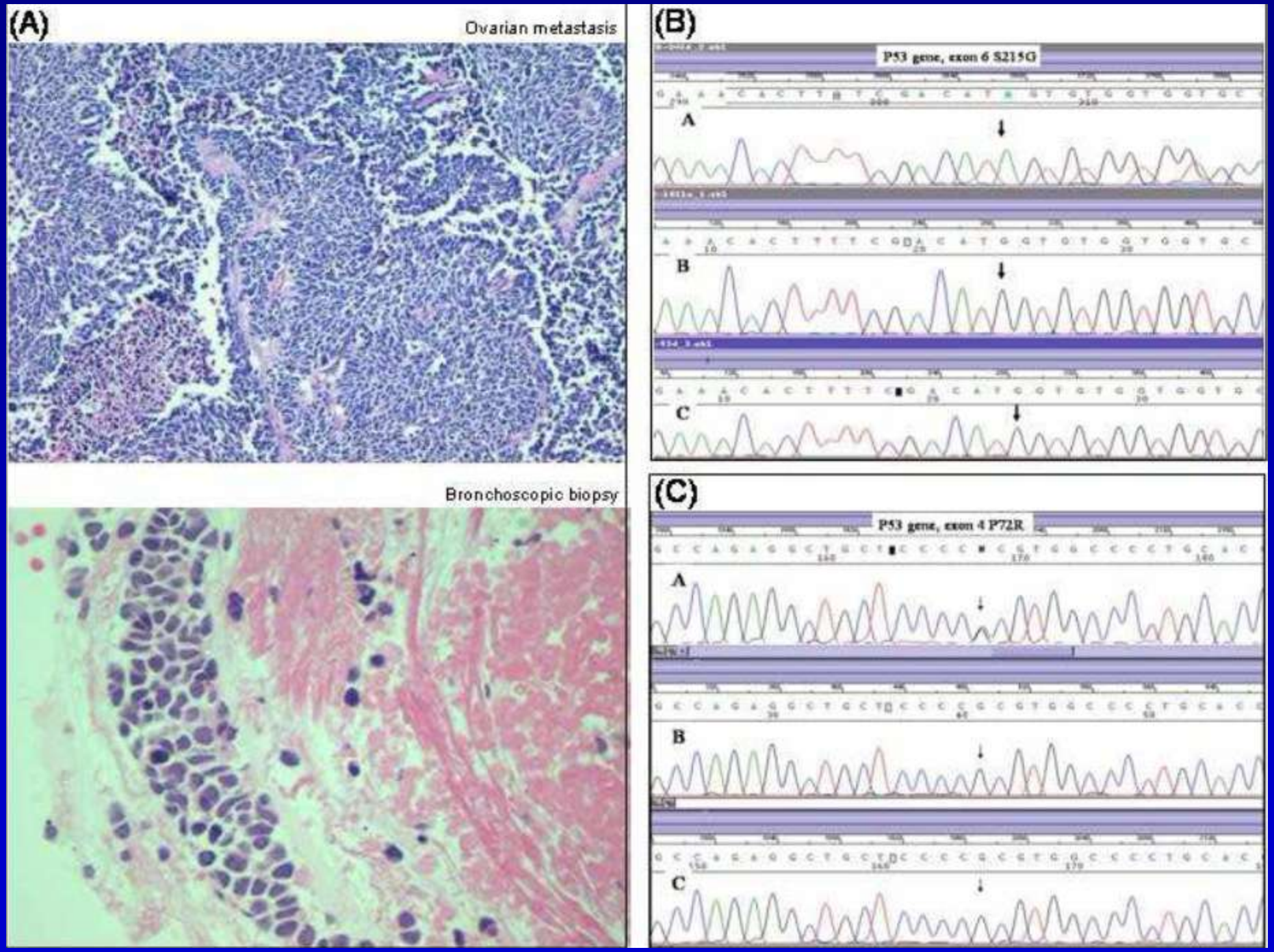
Independent vs Metastatic Tumors (clonality)

- Cromosome X (HUMARA)
- LOH
- Mutation analysis
- Viral Detection


LOHa
LOHb
Tumor progression
and
heterogeneity
LOHa
LOHb
LOHc
LOHa
LOHb
LOHa
LOHb
LOHc
LOHd
LOHa
LOHb
LOHe

Clinical History

- **54 year old woman**
- **Small cell carcinoma of the lung. Metastatic work-up negative. A combined treatment with chemotherapy (cisplatin and VP16) and radiotherapy was administered. Prophylactic cranial irradiation.**
- **Left ovarian mass, 3 months ofter finishing treatment**


(A)
Ovarian metastasis
Bronchoscopic biopsy
(B)
P53 gene, exon 6 S215G
A
B
C
(C)
P53 gene, exon 4 P72R
A
B
C

Diagnosis

Ovarian metastasis of small cell carcinoma of the lung

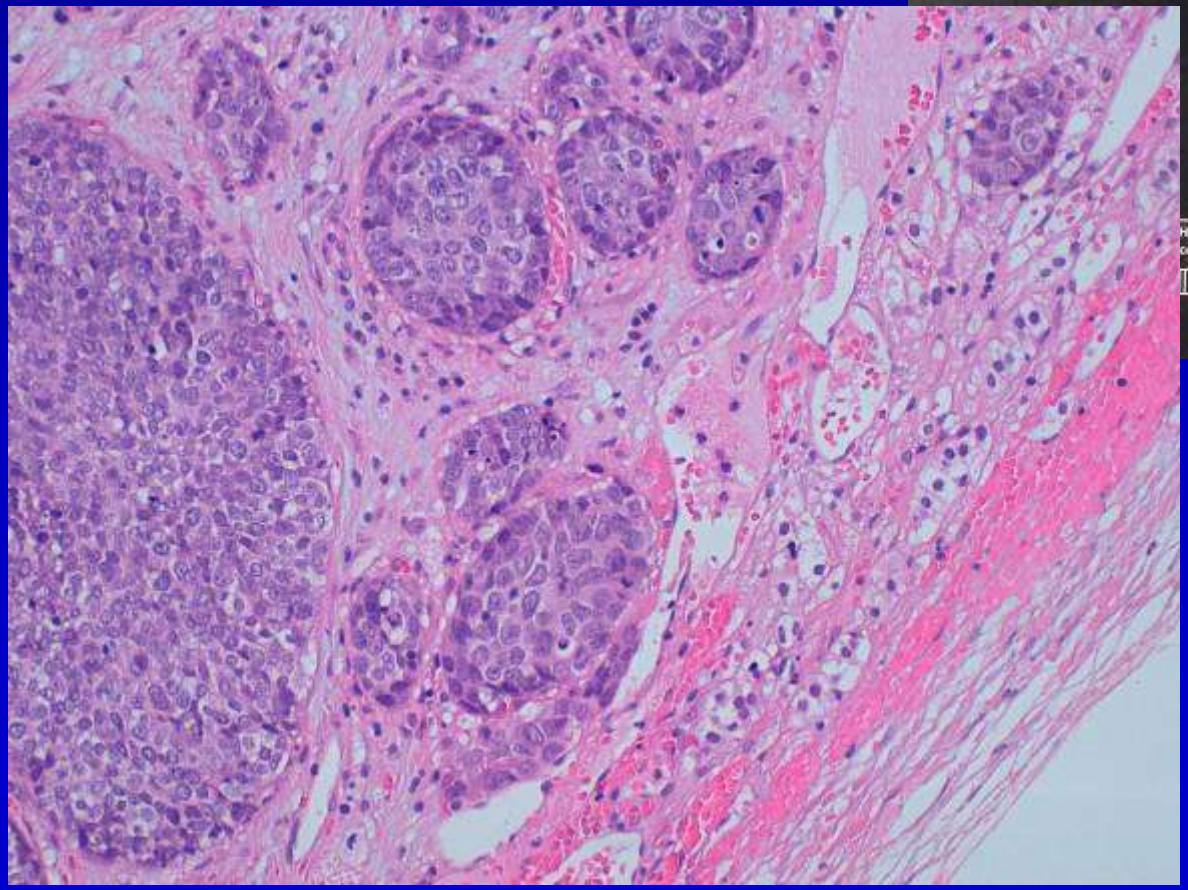
Independent vs Metastatic Tumors (clonality)

- Cromosome X (HUMARA)
- LOH
- Mutation analysis
- Viral Detection

Clinical History

- 71 year old woman
- Cervical cancer, two years ago.
- Adrenal Tumor


HUAV-HSM-UdL PATOLOGIA
09/1424



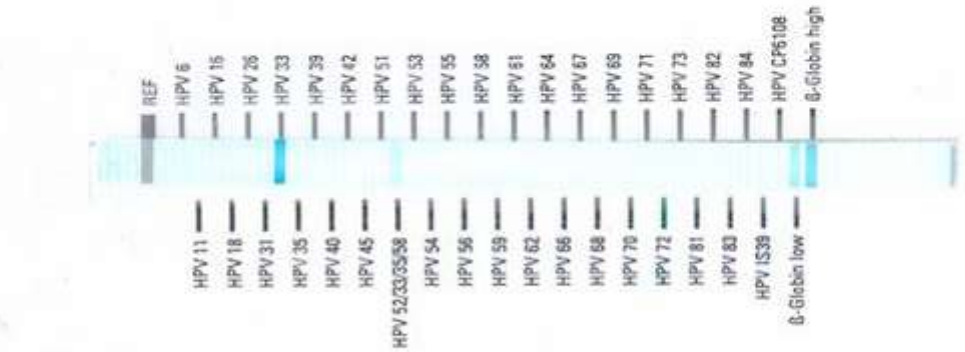
2009G000208

2009B001424 congelado

LINEAR ARRAY HPV Genotyping Test

Roche

Reference Guide/ Referenzleitfaden/ Guide de références/
Guide di riferimento/ Guía de referencia/ Guia de Referência/
Referenceguide/ Referensguide



Roche Molecular Systems, Inc.
Branchburg, NJ USA

04392124001-02

Diagnosis

Cervical Carcinoma metastatic to the adrenal gland

MOLECULAR APPROACHES IN TUMORS OF UNKNOWN ORIGIN

- ONE GENE
- MULTIPLE GENES

Commercial Tests for Cancer of Unknown Origin

- Quest-Lab Corp (92 gene PCR test for 39 cancer types)
- Agendia (microarray-based test for 43 cancer types)
- Pathworks Diagn (microarray-based test for 15 cancer types, representing 60 different morphologies)
- Veridex (PCR-based test for 6 cancer types)
- CancerTYPE-bioTheranostics (PCR-based test for 39 cancer types)
- Rosetta Genomics (PCR-based test on miRNA for 25 cancer types)

Pathwork™ Tissue of Origin Test

A microarray-based test
Measures the expression of >1600 genes
Helps identify Uncertain Primary Cancers
Currently, 15 tissue types representing 60 different morphologies
Test in development; now under review at the FDA

TISSUE OF ORIGIN	SIMILARITY SCORE	Low SIMILARITY SCORE High
Non-small Cell Lung	60.2	
Bladder	12.9	
Breast	9.0	
Ovarian	7.0	
Kidney	2.5	
Soft Tissue Sarcoma	1.9	
Colorectal	1.7	
Gastric	1.0	
Pancreas	1.0	
Thyroid	0.9	
Germ Cell	0.5	
Hepatocellular	0.5	
Melanoma	0.5	
Non-Hodgkin's Lymphoma	0.3	
Prostate	0.2	

Cancer metastático de origen desconocido (Inmunohistoquímica versus Patología Molecular en 73 casos)

- **73 casos, que se presentan como metástasis de cáncer de origen incierto o controvertido (103 casos evaluados)**
- **Pathwork Tissue of Origin Test en tejido congelado**
- **Immunohistoquímica (CK7, CK20, CK19, PSA, Thyrogl, TTF1, GCDF-15, Mamoglobin, ER, PR, WT1, CDX2, villin, PAX2, HepPar 1, Glypican, CD-10, Inhibin, S-100, Melan-A, HMB-45)**

Cancer metastático de origen desconocido (Analisis con Pathwork en 73 casos)

		Patologia Molecular		**Total**
		+	**-**	
I H Q	**+**	**49**	**11**	**62**
	-	**10**	**2**	**11**
Total		**59**	**14**	**73**

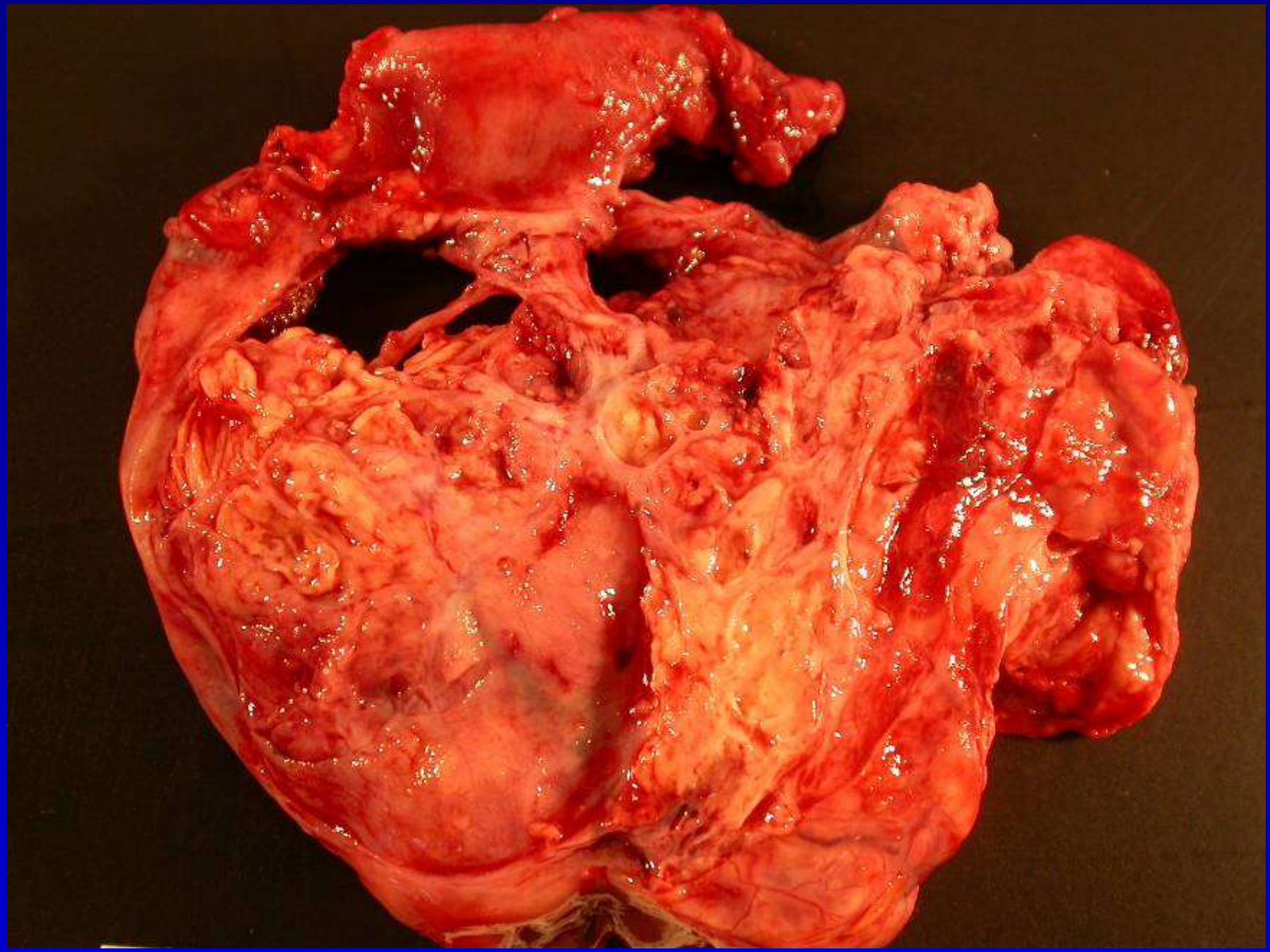
Cancer metastático de origen desconocido (Inmunohistoquímica versus Patología Molecular en 73 casos)

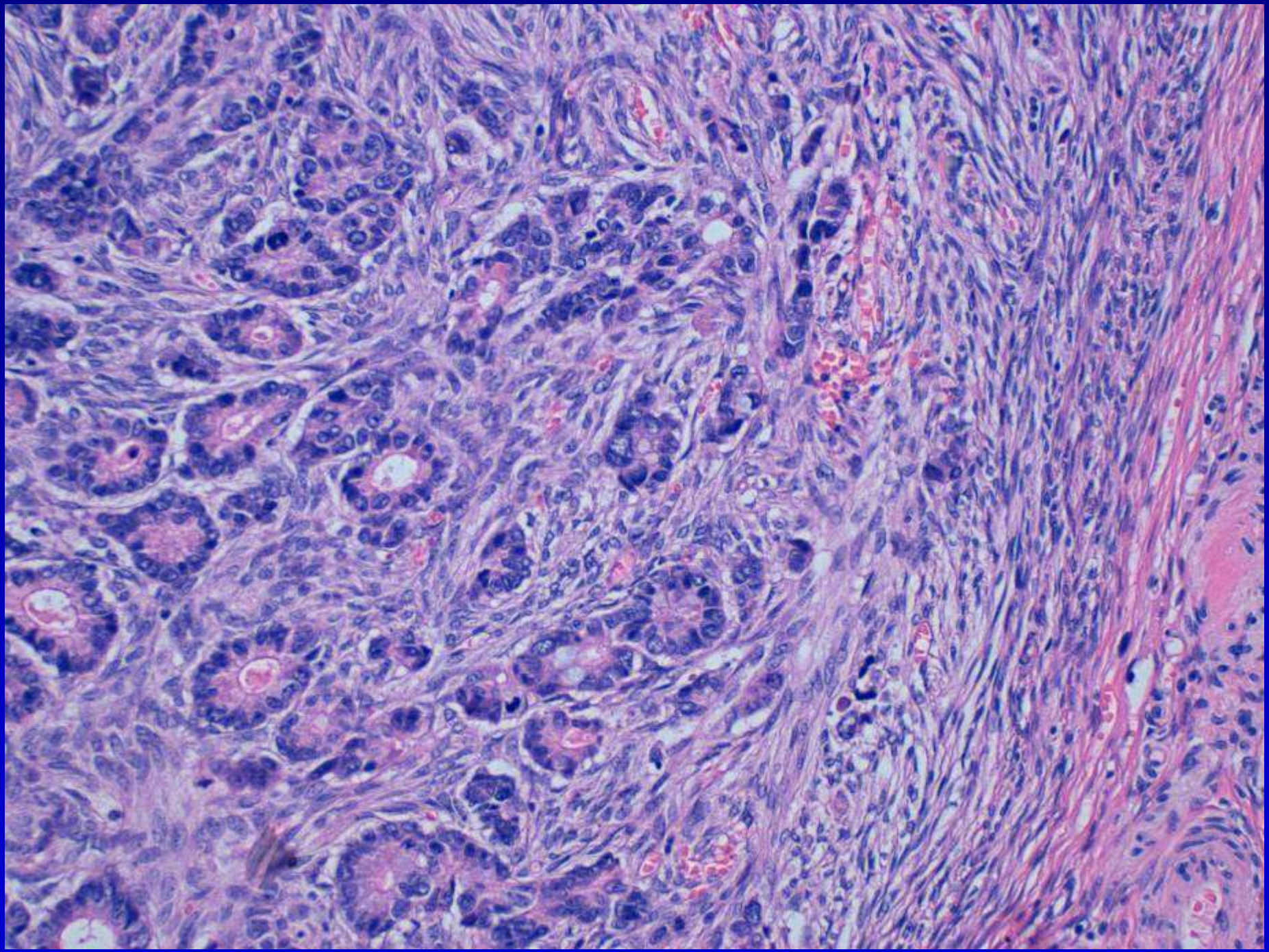
Los principales problemas a los que se plantea la técnica molecular son:

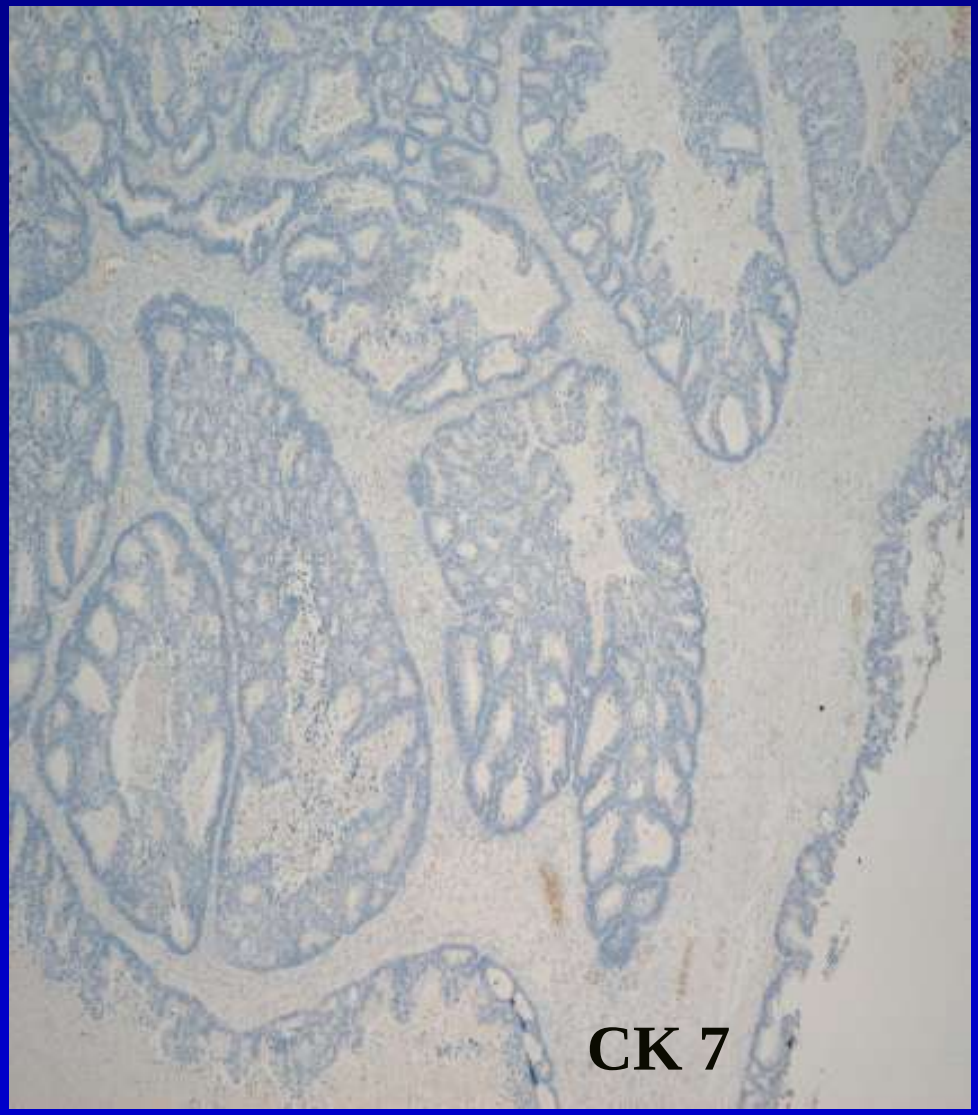
1) **Dificultades técnicas inherentes al sistema de microarrays**
2) **Dificultad de trabajar con muestras congeladas**
3) **Problemas de "ruido de fondo" por la población normal acompañante**
4) **Metástasis de cánceres primarios, que no están incluidos entre los que el sistema evalúa**
5) **Problemas específicos en tipos histológicos precisos.**

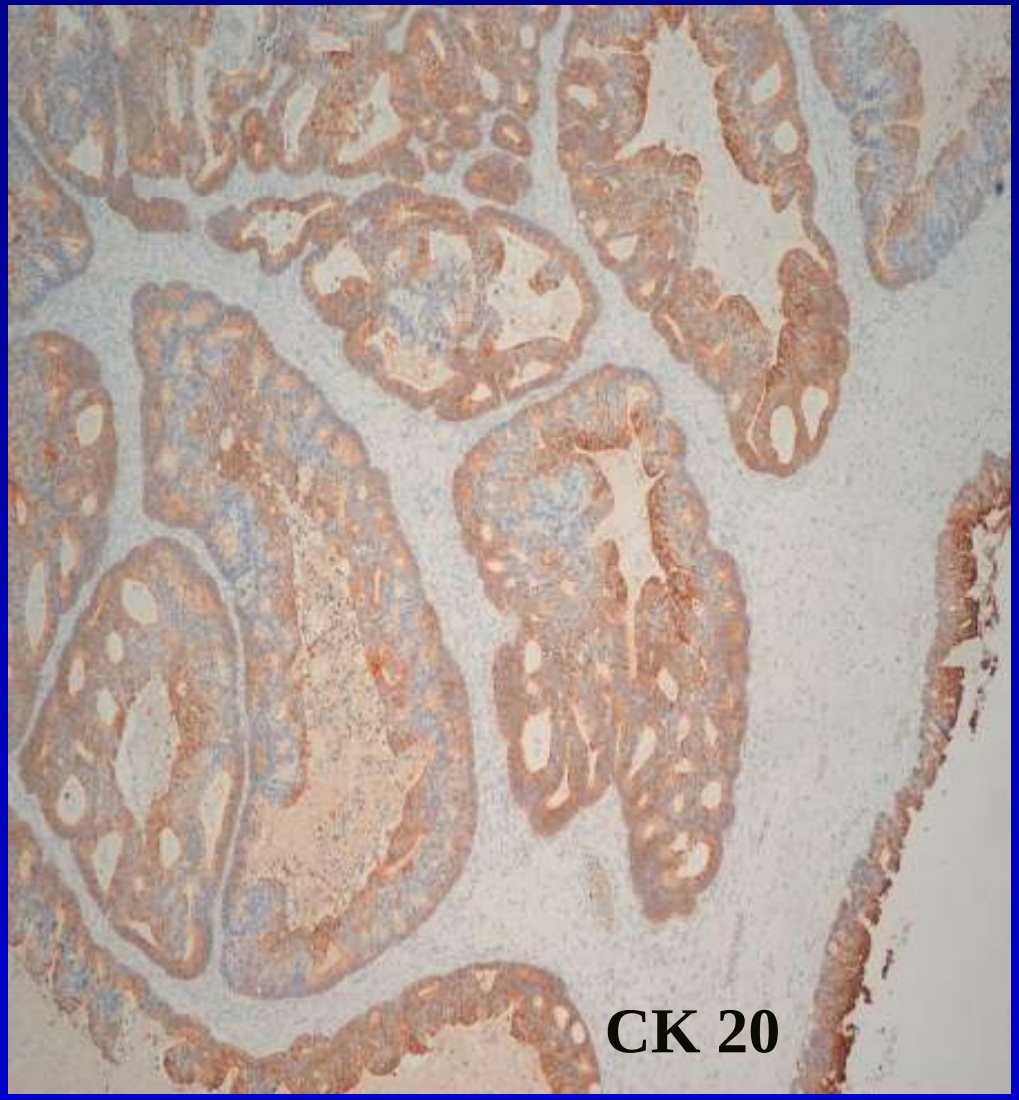
Clinical History

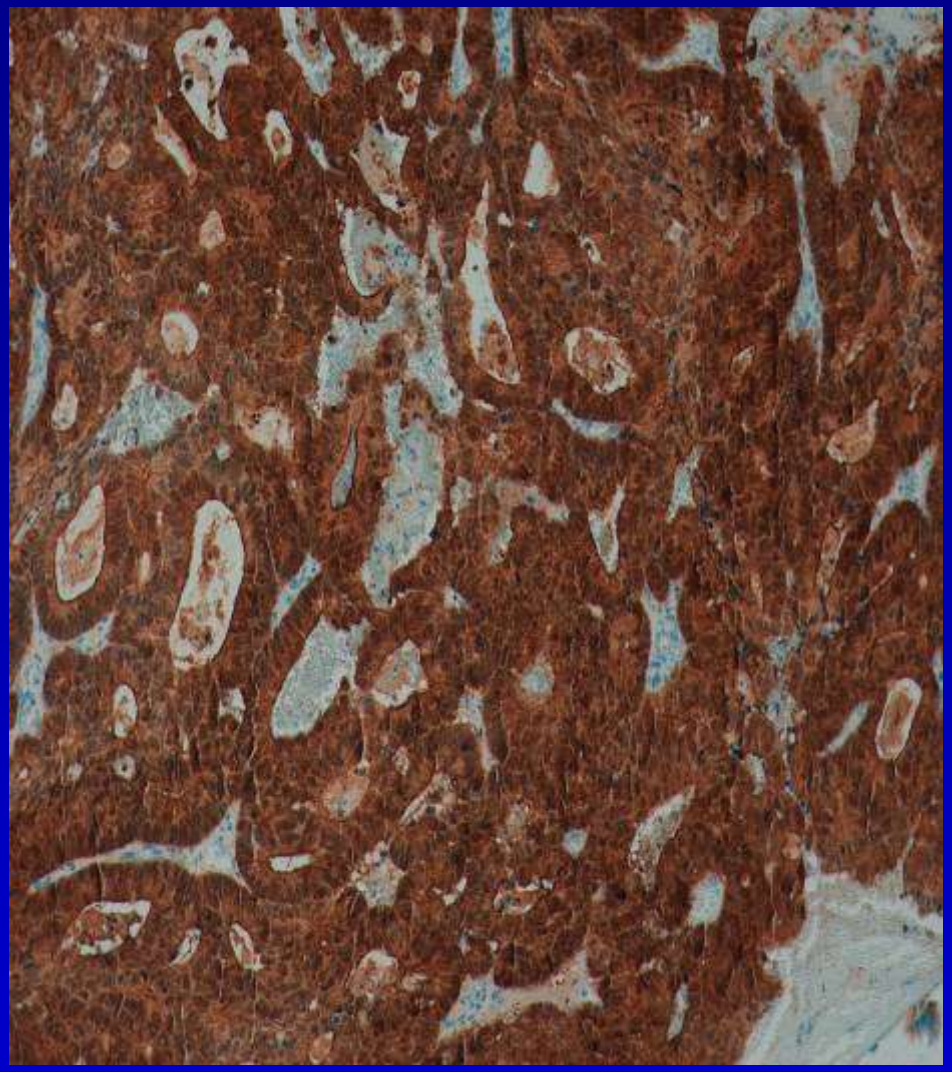
- 69 year old woman
- Unilateral ovarian tumor
- Colon cancer, several years ago



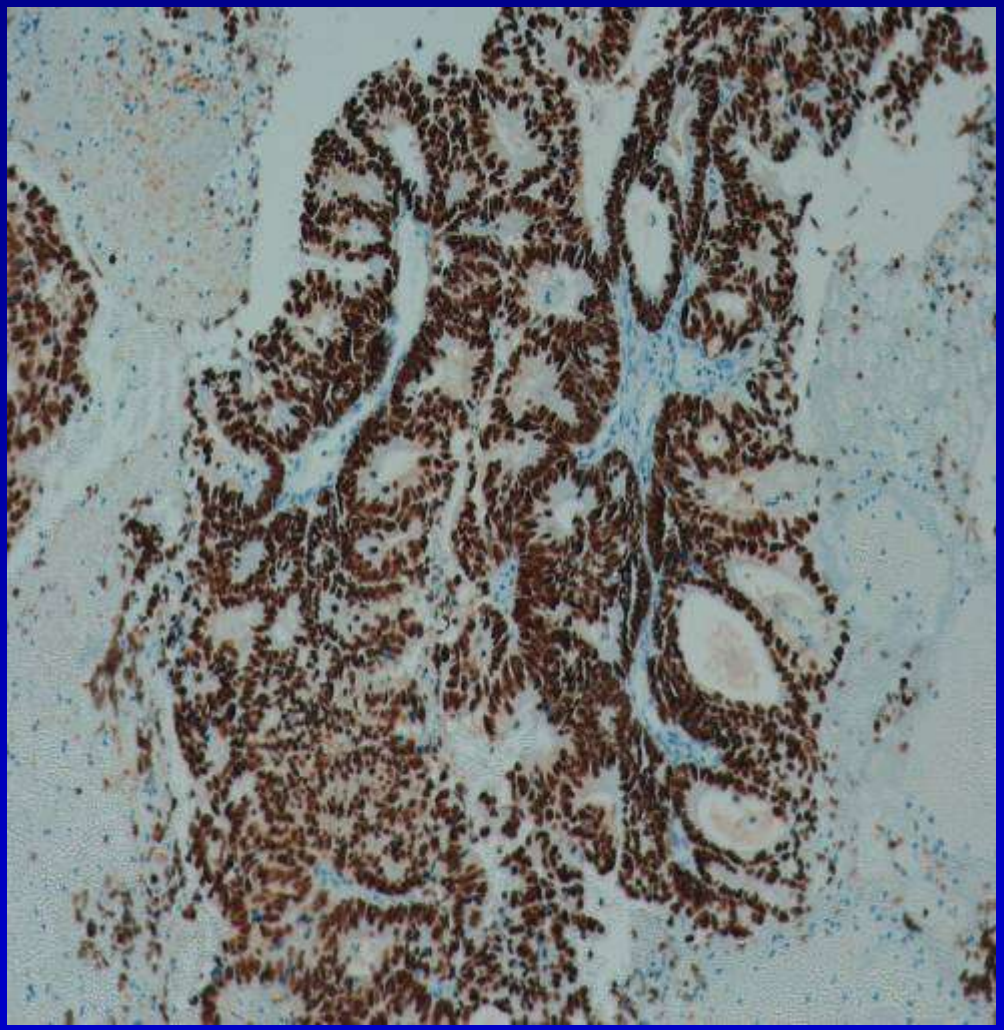



CK 7


CK 20

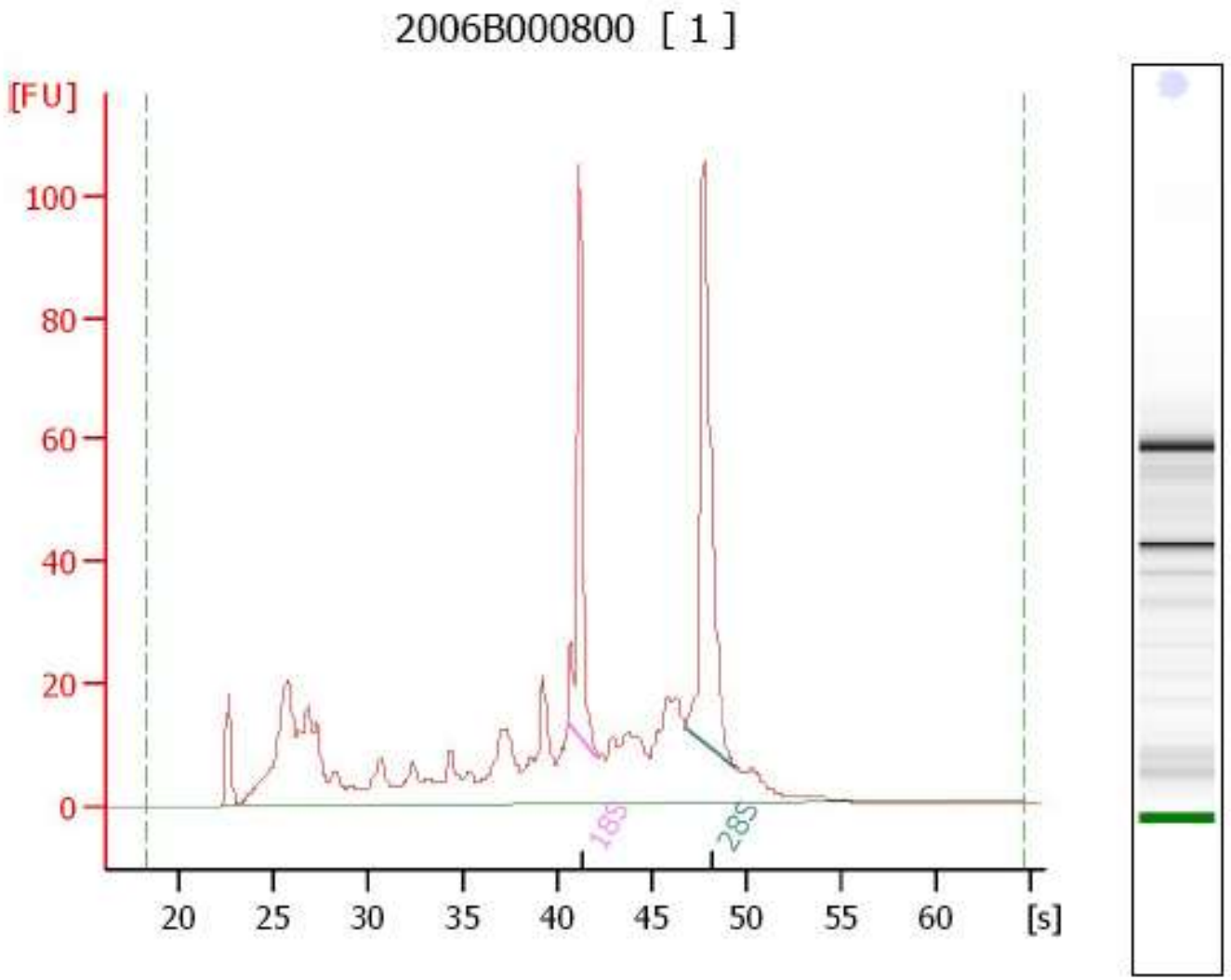


Villin



CDX 2

Electropherogram Summary



Test Report Date: 27Jul2009 11:46PDT
Specimen ID: MM1
Specimen Type: Frozen
File: MM1_20090723_134954.CEL

Pathwork Analysis ID: 29149
Pathchip Serial #: 06963
Version: TOOv3.1

Data Quality: Acceptable

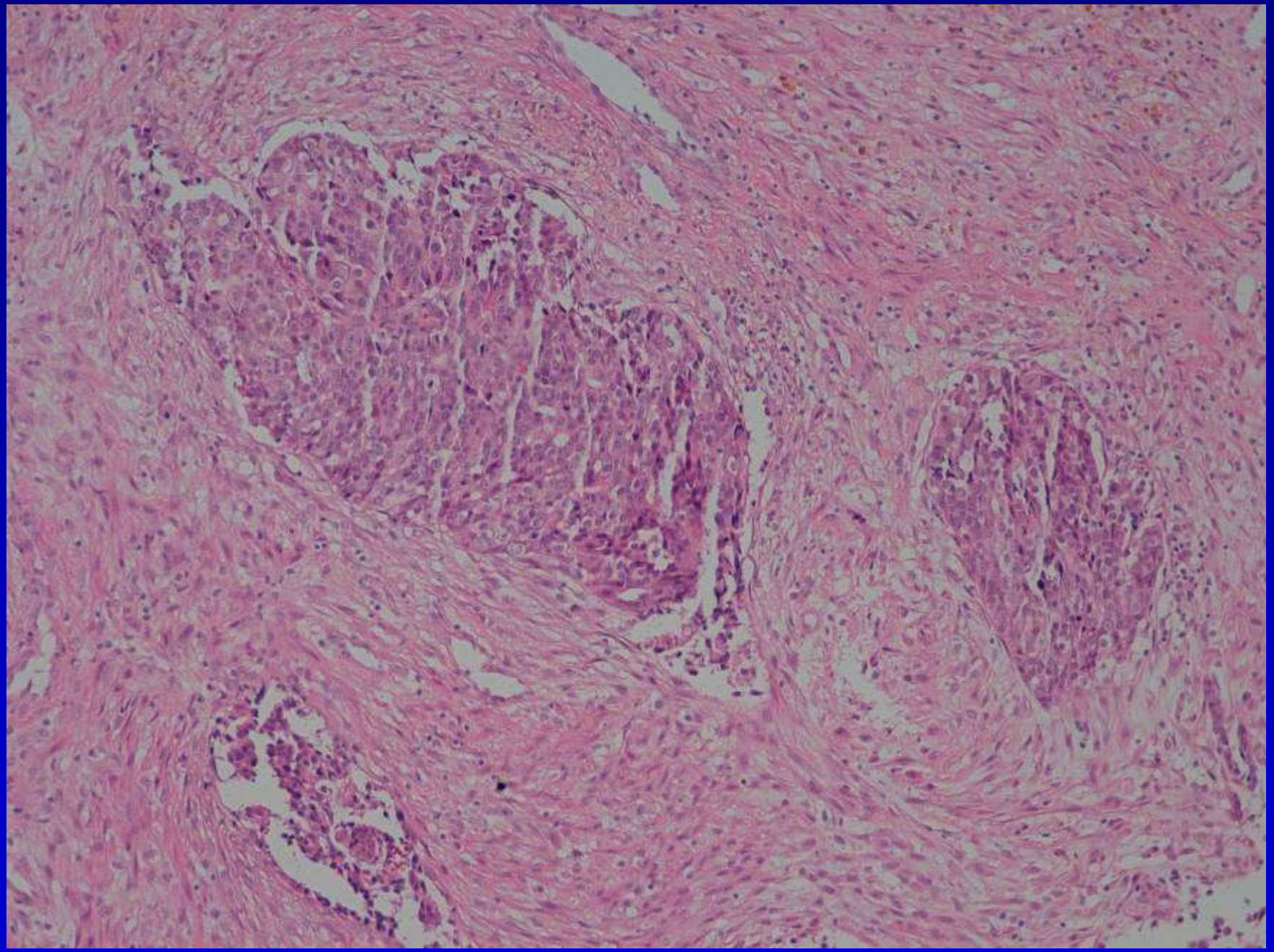
TISSUE	SIMILARITY SCORE	LOW 0 5 30 HIGH 100
Colorectal	85.5	
Ovarian	5.9	
Non-Small Cell Lung	2.3	
Gastric	1.9	
Breast	1.6	
Pancreas	0.7	
Testicular Germ Cell	0.6	
Kidney	0.4	
Hepatocellular	0.4	
Bladder	0.2	
Non-Hodgkin's Lymphoma	0.2	
Melanoma	0.1	
Prostate	0.1	
Thyroid	0.1	
Soft Tissue Sarcoma	0.1	

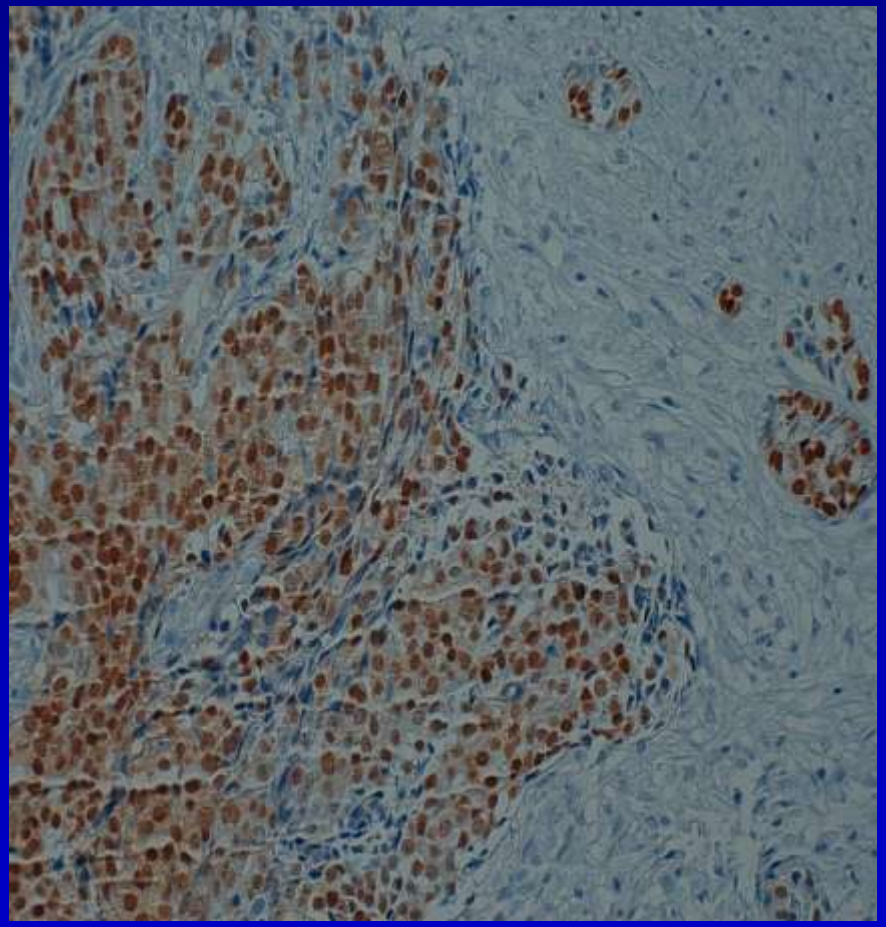
Diagnosis

Colon carcinoma metastatic to the ovary

Clinical History

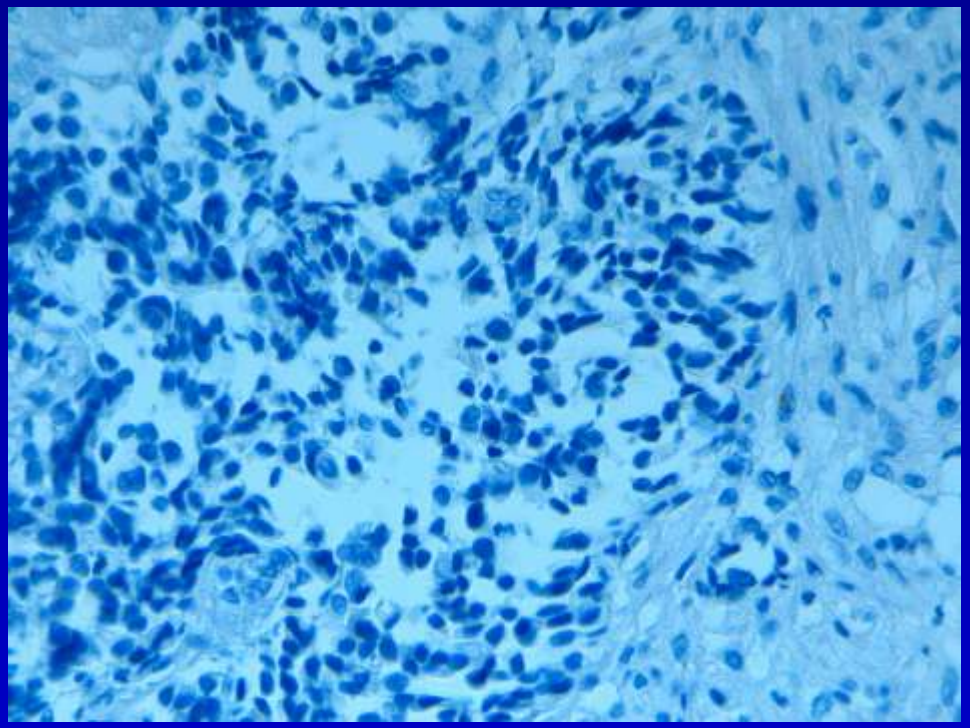
- 64 year old woman
- Breast Cancer, several years ago
- Peritoneal Carcinomatosis



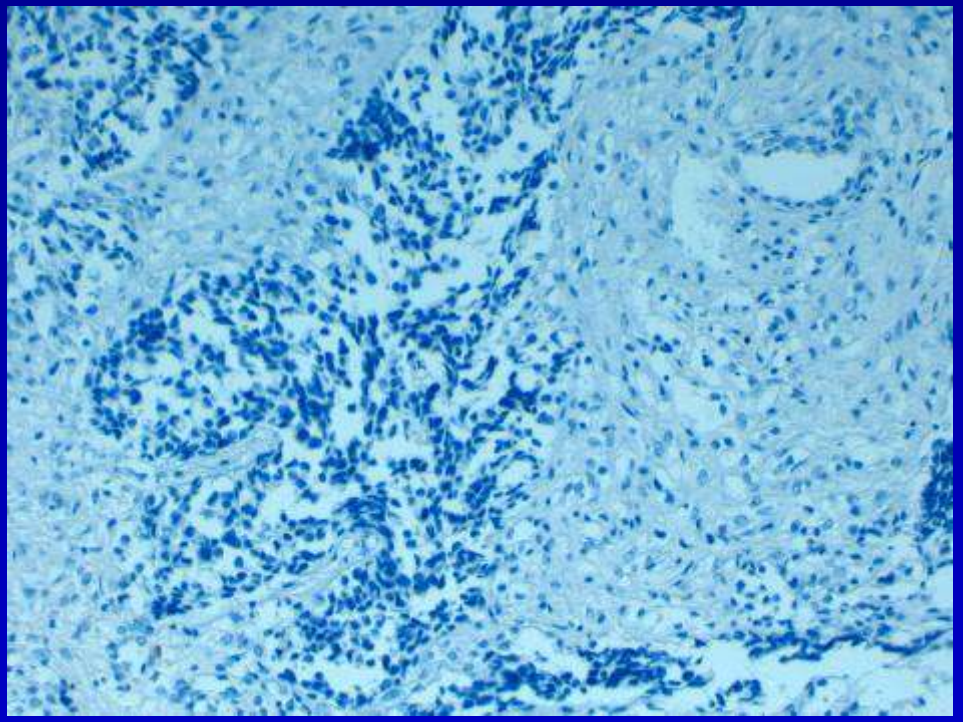


ER

WT-1



Mamoglobin



GCDF-15

Test Report Date: 23Oct2009 14:13PDT
Specimen ID: 2008B013420
Specimen Type: Frozen
File: 2008B013420.CEL

Pathwork Analysis ID: 30106
Pathchip Serial #: 80967
Version: TOOv3.1

Data Quality: Acceptable

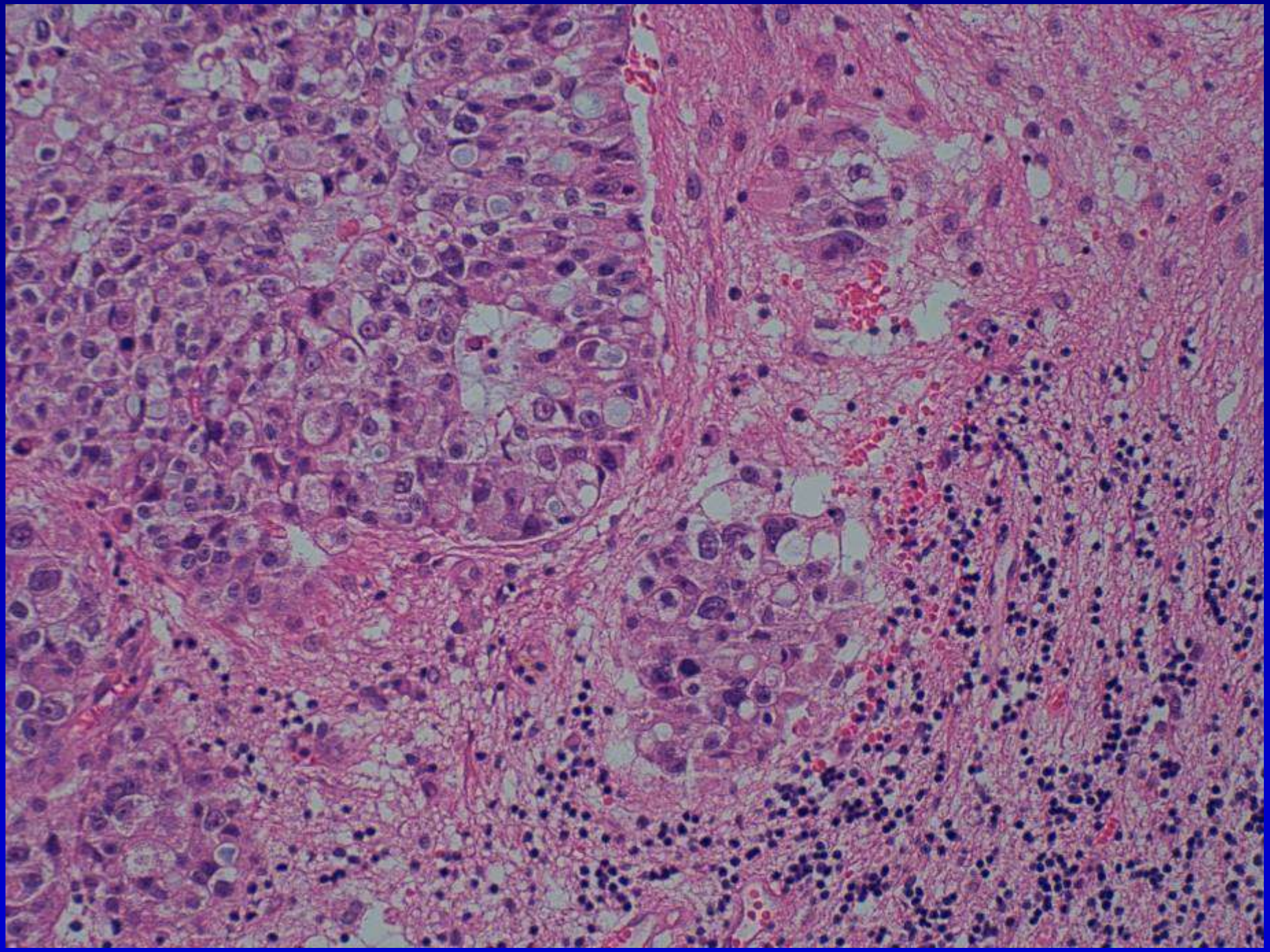
TISSUE	SIMILARITY SCORE	LOW 0 5 30 HIGH 100
Ovarian	97.2	
Non-Small Cell Lung	0.7	
Kidney	0.5	
Breast	0.3	
Soft Tissue Sarcoma	0.3	
Hepatocellular	0.3	
Thyroid	0.1	
Gastric	0.1	
Colorectal	0.1	
Pancreas	0.1	
Testicular Germ Cell	0.1	
Non-Hodgkin's Lymphoma	0.1	
Melanoma	0.1	
Bladder	0.1	
Prostate	0.0	

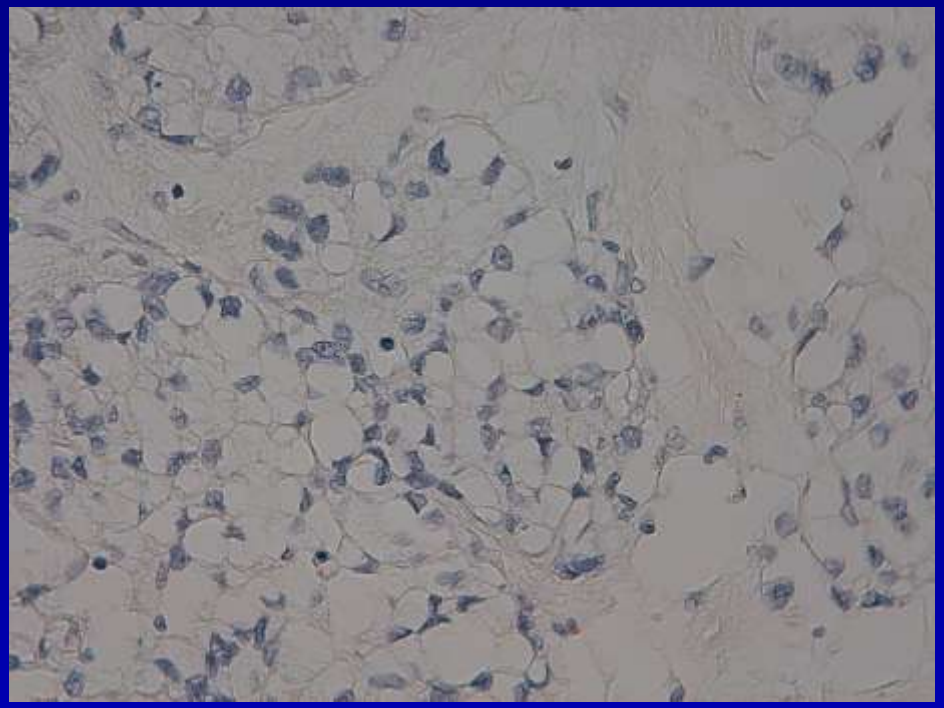
Diagnóstico

Metastatic ovarian carcinoma (serous type)

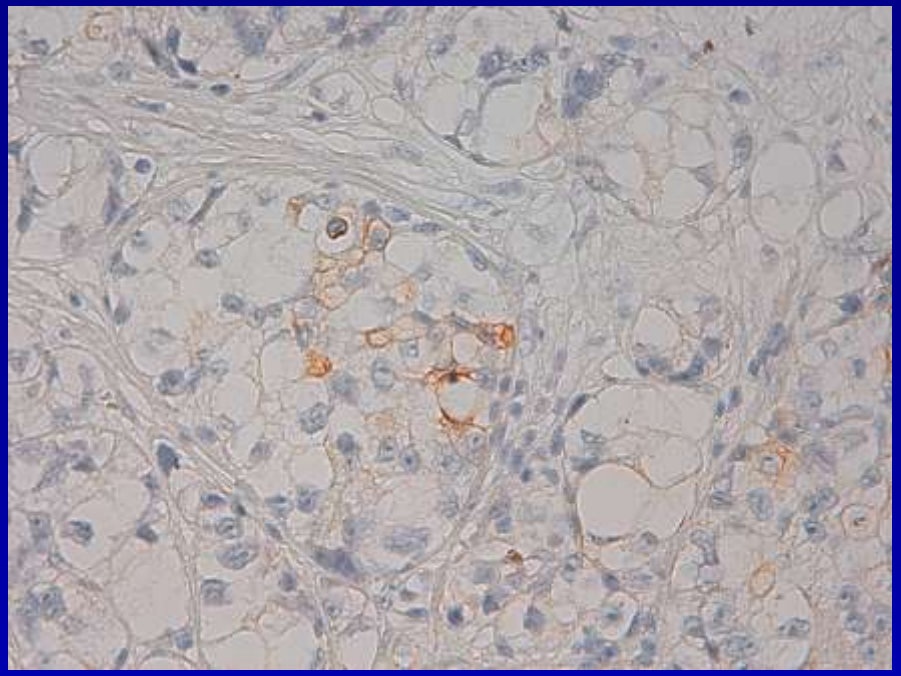
Clinical History

- **59 year old man.**
- **Brain tumor**

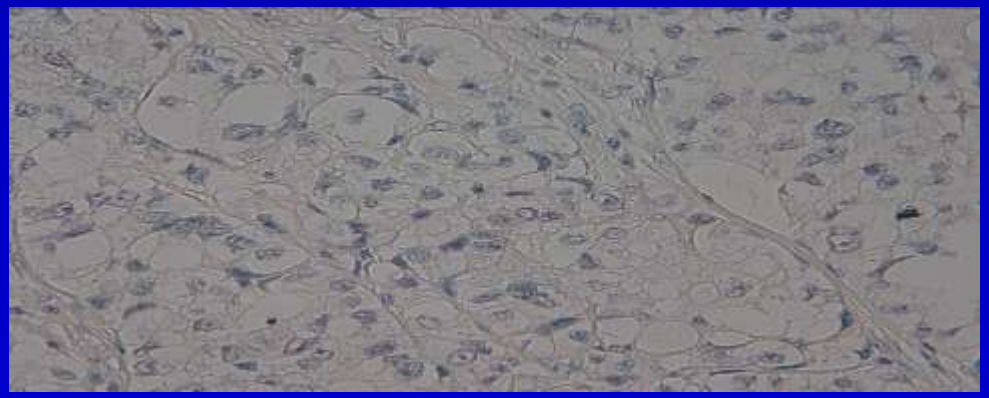




TTF-1



CK-7



CK-20

Test Report Date: 23Oct2009 14:09PDT
Specimen ID: 2004B008843
Specimen Type: Frozen
File: 2004B008843.CEL

Pathwork Analysis ID: 30102
Pathchip Serial #: 80967
Version: TOOv3.1

Data Quality: Acceptable

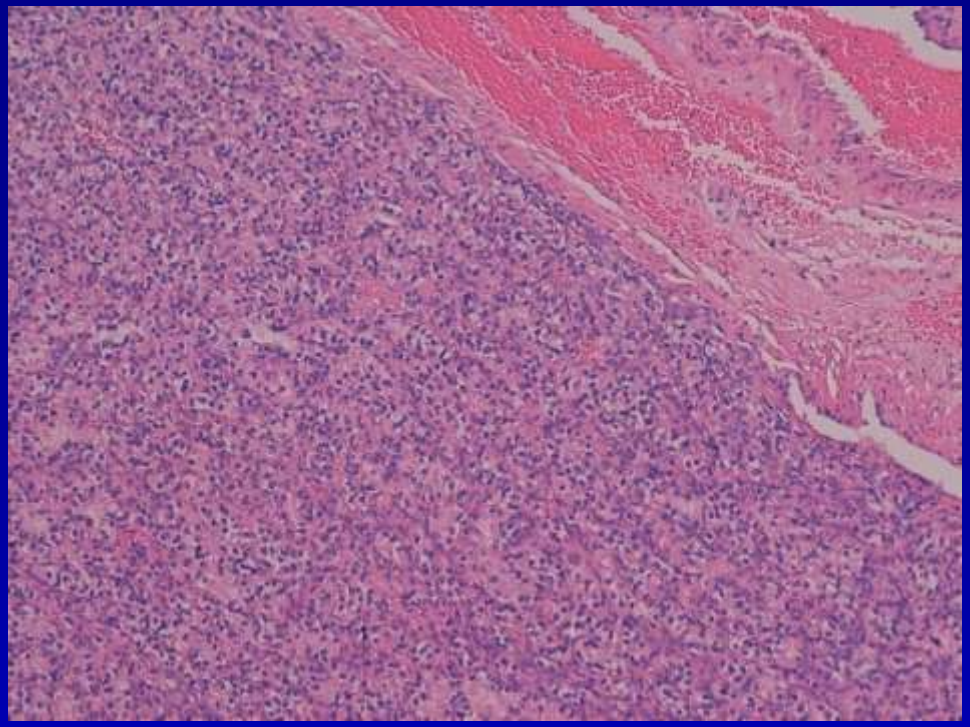
TISSUE	SIMILARITY SCORE	LOW 0 5 30 HIGH 100
Pancreas	19.6	
Non-Small Cell Lung	9.9	
Colorectal	5.0	
Gastric	3.8	
Kidney	1.9	
Breast	1.6	
Thyroid	1.2	
Hepatocellular	0.5	
Soft Tissue Sarcoma	0.5	
Non-Hodgkin's Lymphoma	0.5	
Testicular Germ Cell	0.3	
Bladder	0.3	
Prostate	0.1	
Melanoma	0.1	

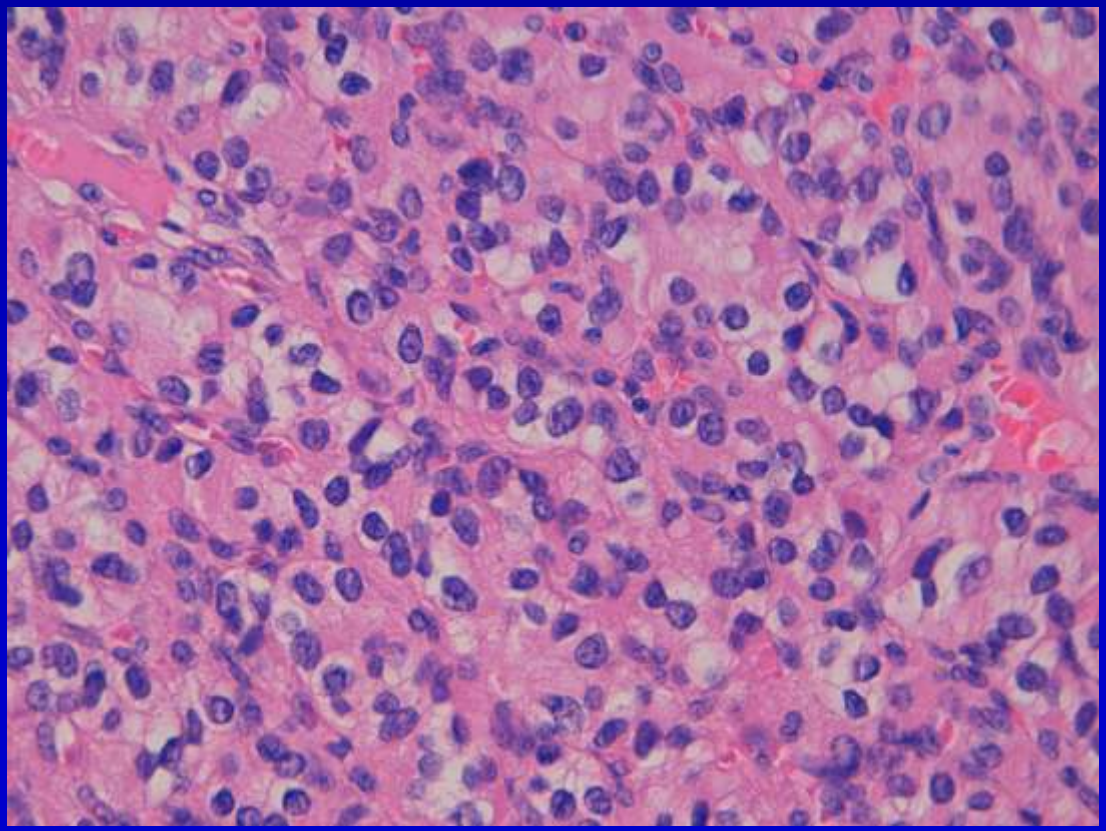
Diagnosis

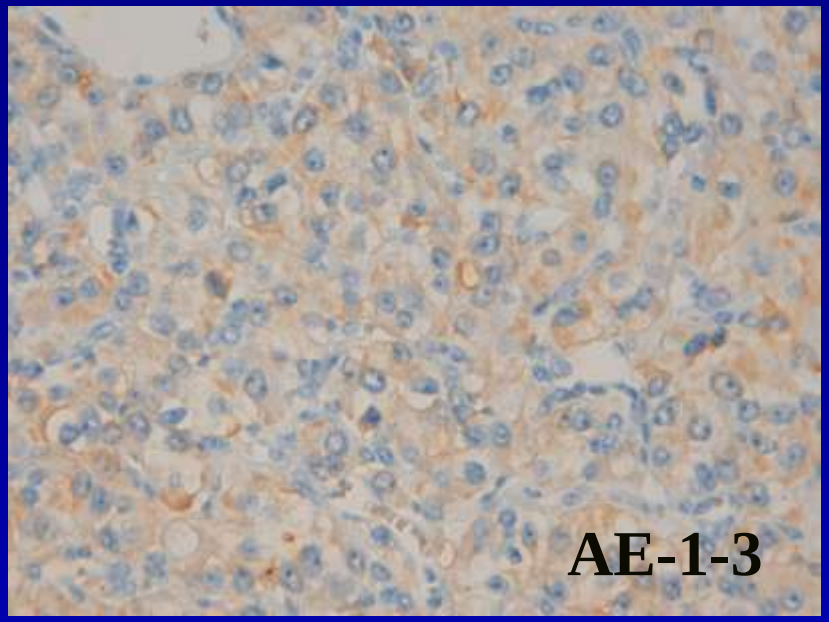
Metastatic carcinoma of pancreatic origin.

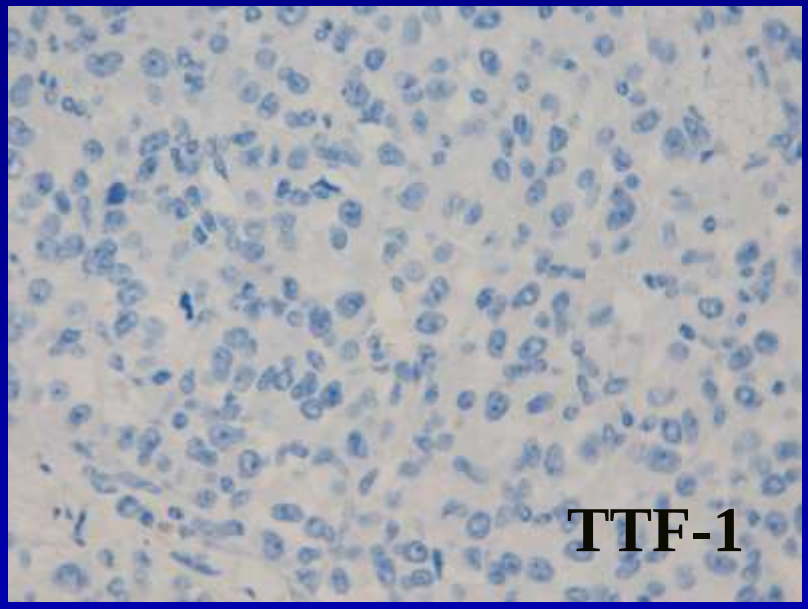
Clinical History

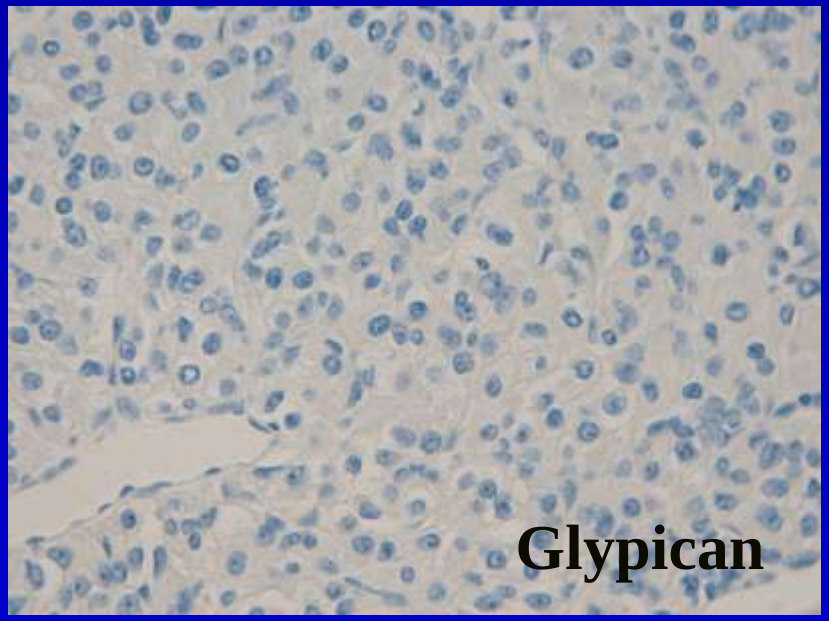
- A 92 year-old man
- No previous history of cancer
- Mass in the thoracic wall
- Biopsy

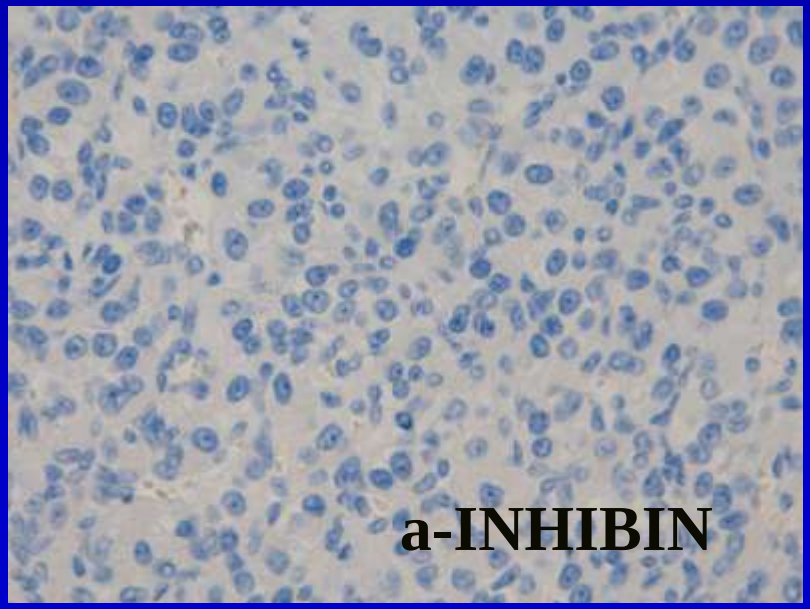





AE-1-3


TTF-1


Glypican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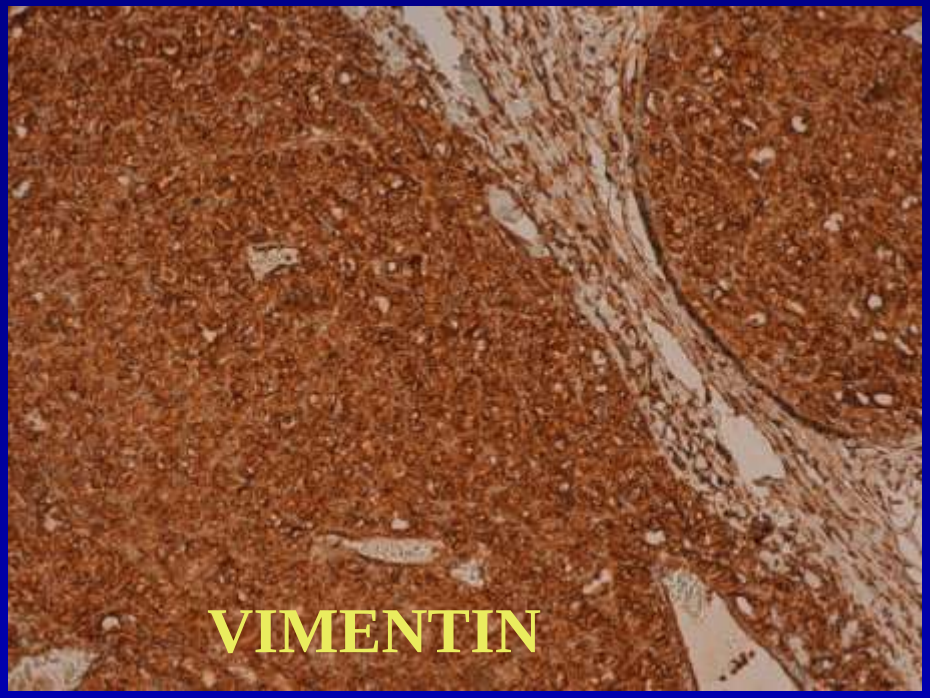
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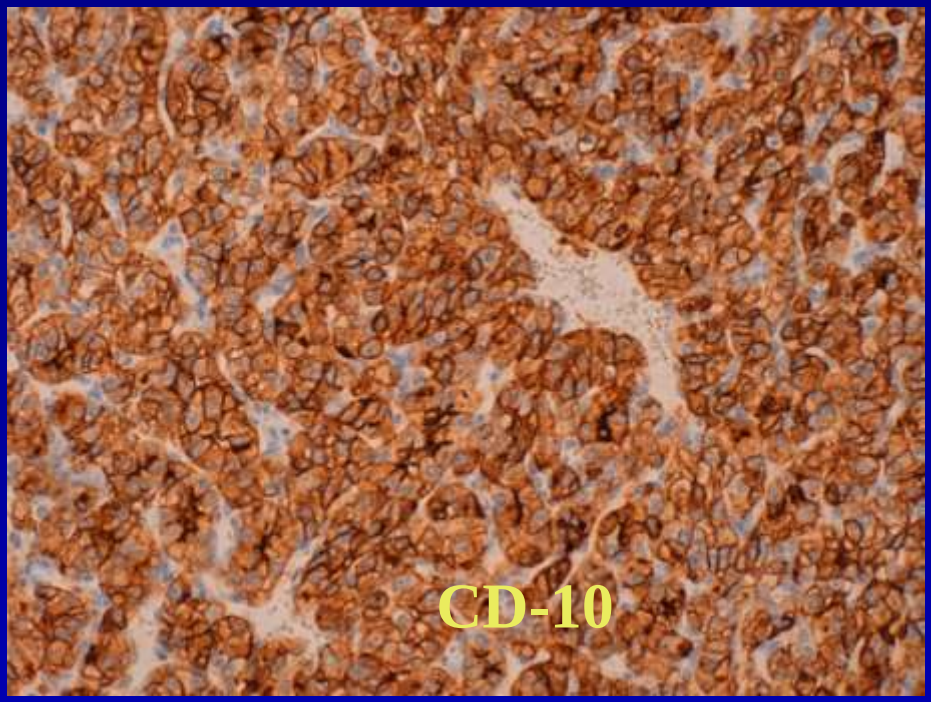
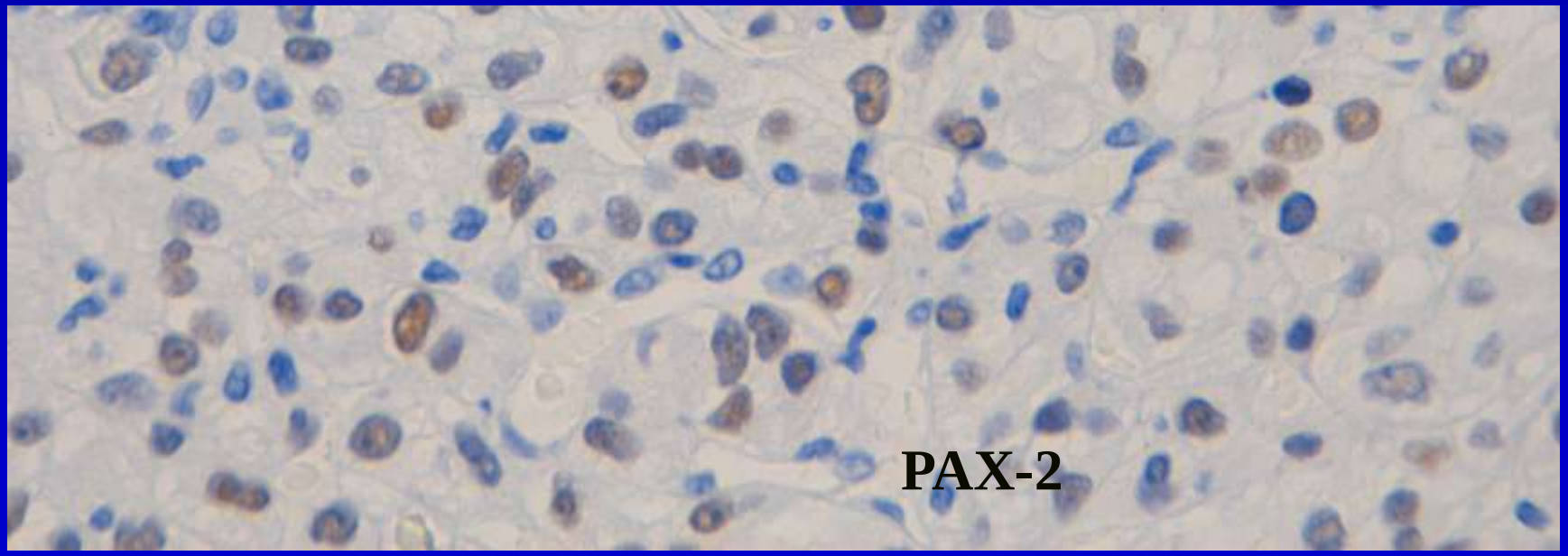

CD-10


PAX-2

Data Quality: Acceptable

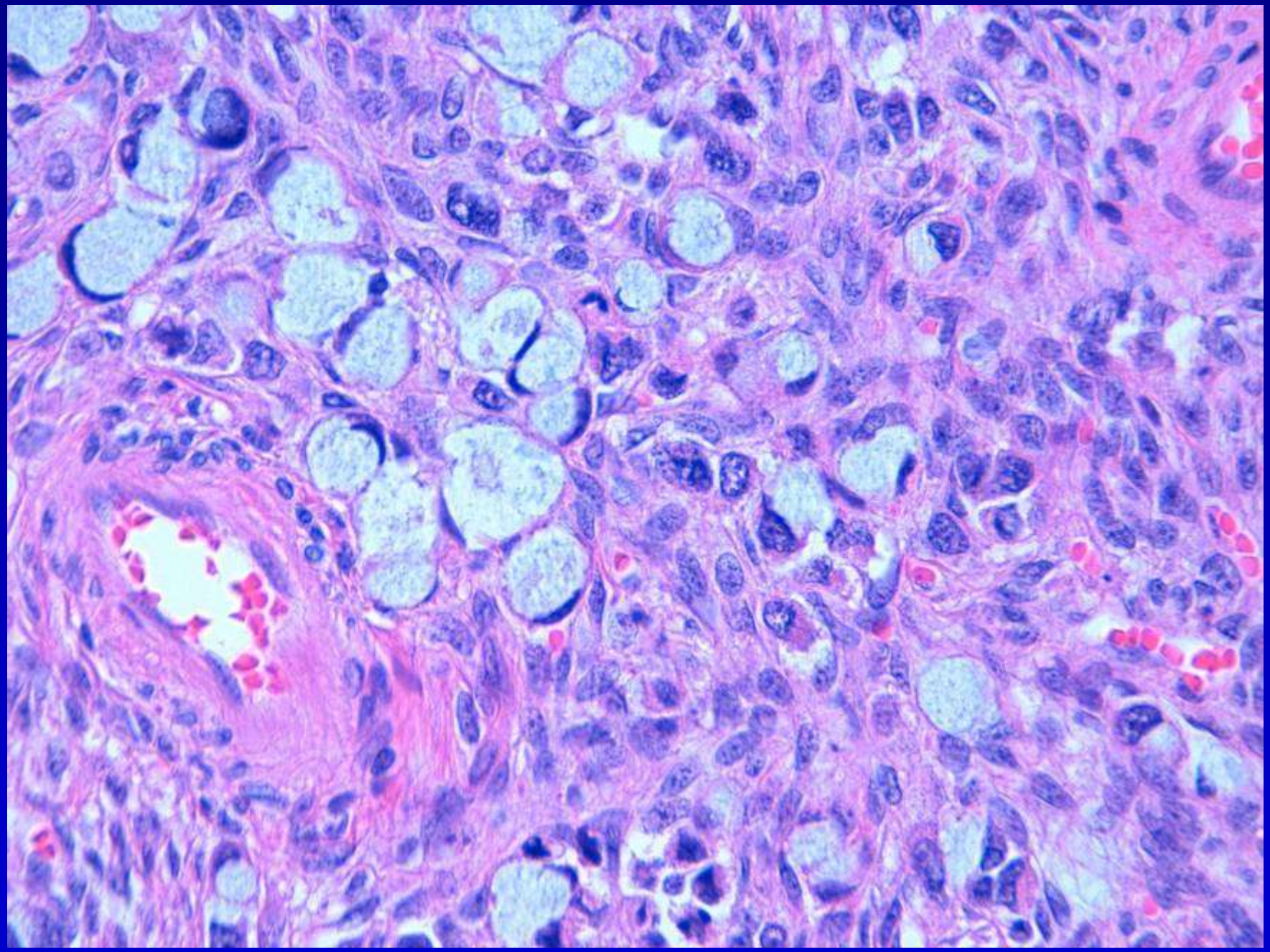
TISSUE	SIMILARITY SCORE	LOW 0 5 30 HIGH 100
Kidney	94.2	
Hepatocellular	2.0	
Ovarian	1.6	
Breast	0.7	
Thyroid	0.3	
Sarcoma	0.2	
Non-Hodgkin's Lymphoma	0.2	
Pancreatic	0.2	
Non-Small Cell Lung	0.1	
Germ Cell	0.1	
Colorectal	0.1	
Bladder	0.1	
Prostate	0.1	
Gastric	0.1	
Melanoma	0.0	

Diagnosis

Metastatic carcinoma of renal origin.

Clinical History

- **64 year old woman**
- **Bilateral ovarian tumors**



Follow-up

- **Gastroscopy: negative**
- **Gastric biopsies: negative**

Test Report Date: 27Jul2009 11:47PDT
Specimen ID: MM2
Specimen Type: Frozen
File: MM2_20090723_134958.CEL

Pathwork Analysis ID: 29148
Pathchip Serial #: 06968
Version: TOOv3.1

Data Quality: Acceptable

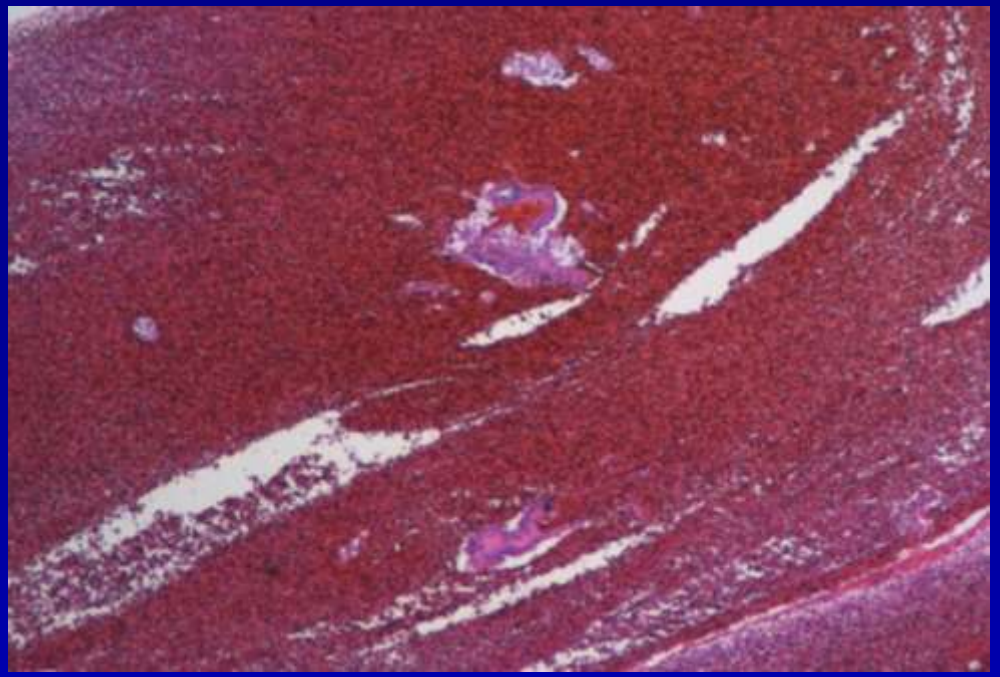
TISSUE	SIMILARITY SCORE	LOW 0 5 30 HIGH 100
Gastric	41.6	
Ovarian	33.3	
Colorectal	8.0	
Pancreas	4.6	
Testicular Germ Cell	2.9	
Non-Small Cell Lung	2.1	
Hepatocellular	1.5	
Soft Tissue Sarcoma	1.3	
Breast	1.3	
Kidney	0.9	
Non-Hodgkin's Lymphoma	0.7	
Bladder	0.6	
Thyroid	0.5	
Prostate	0.3	
Melanoma	0.3	

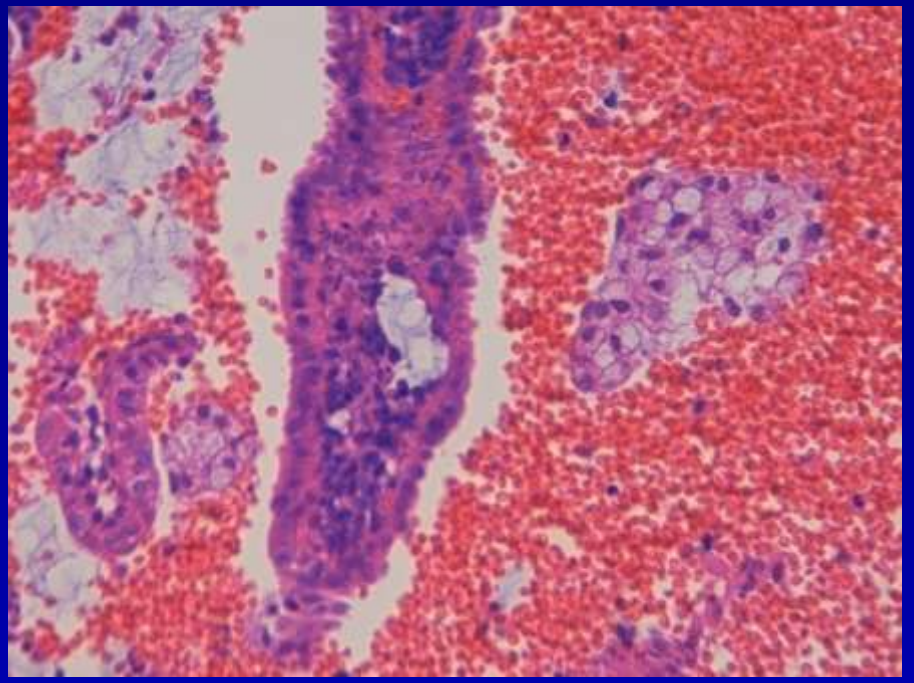
Diagnosis

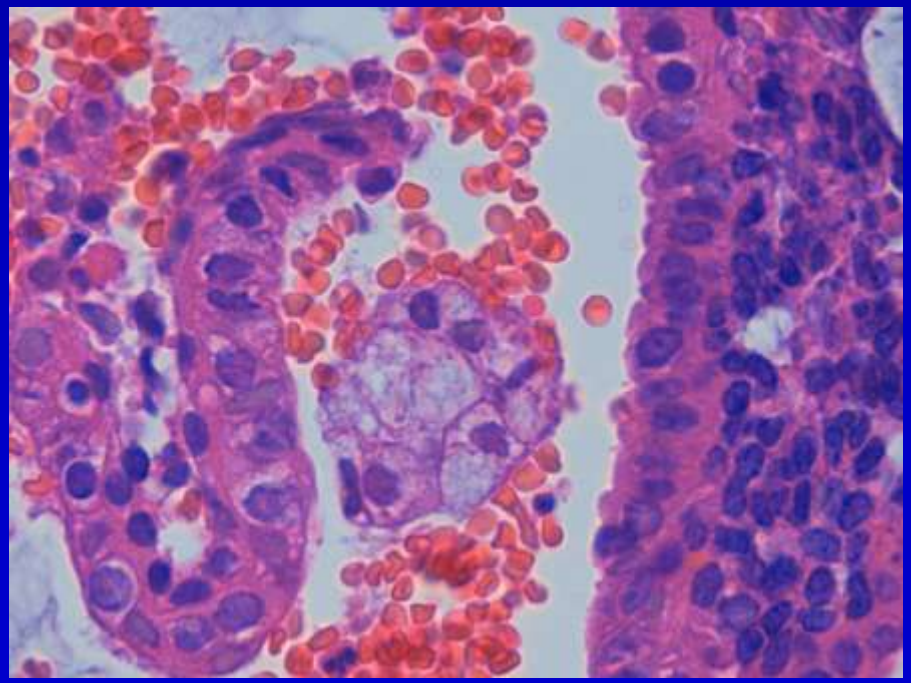
Gastric carcinoma metastatic to the ovaries

Clinical History

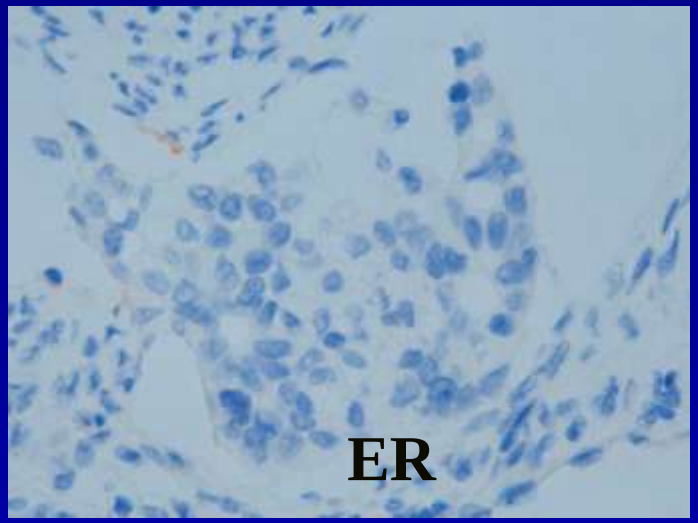
- **50 year old woman.**
- **Vaginal Bleeding**
- **Endometrial Biopsy**







AEI-AE3 +
CK7 –
CK 20 +
ER, PR –
Vimentin –
CDX-2 +
Villin +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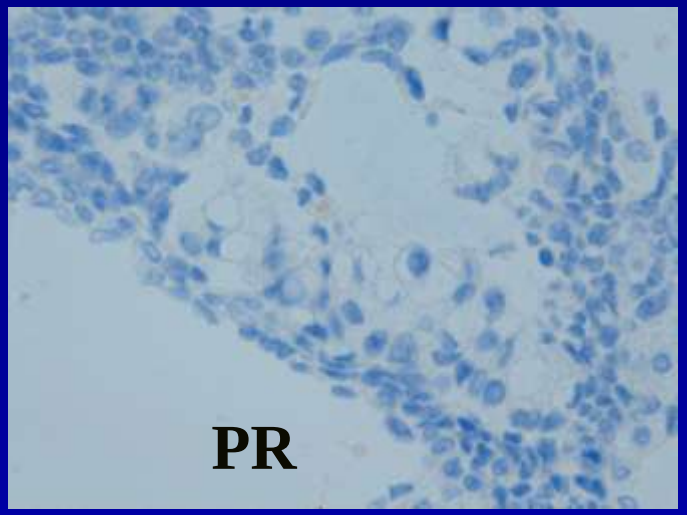
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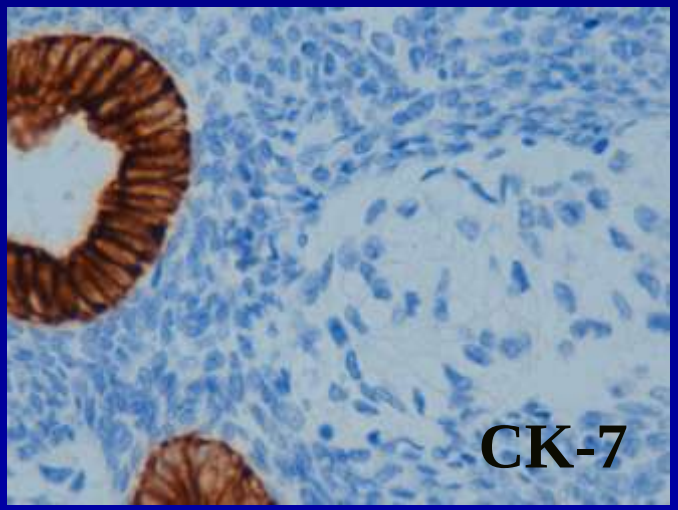
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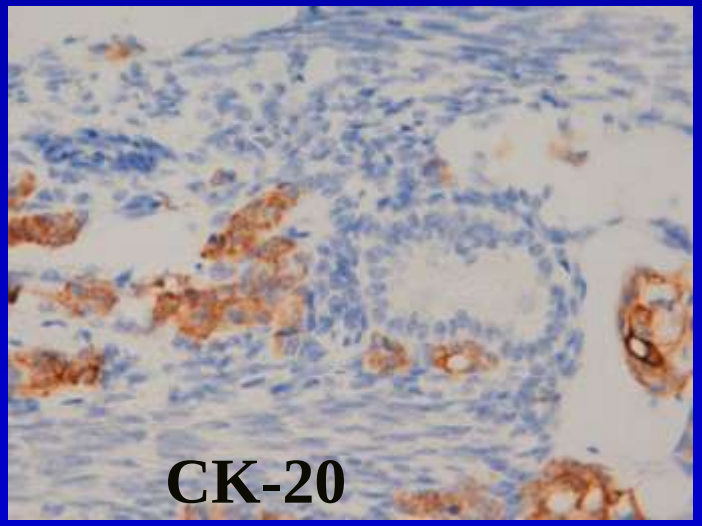
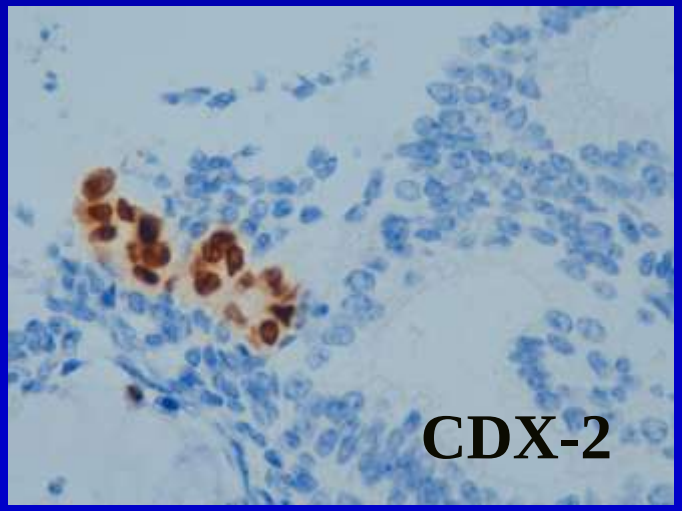
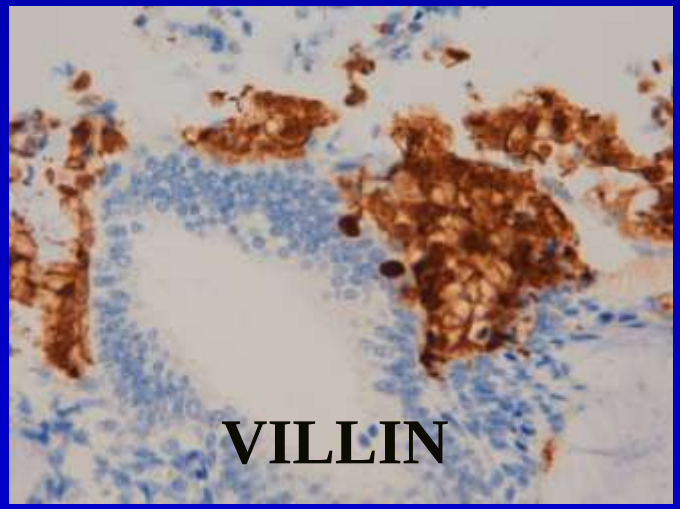
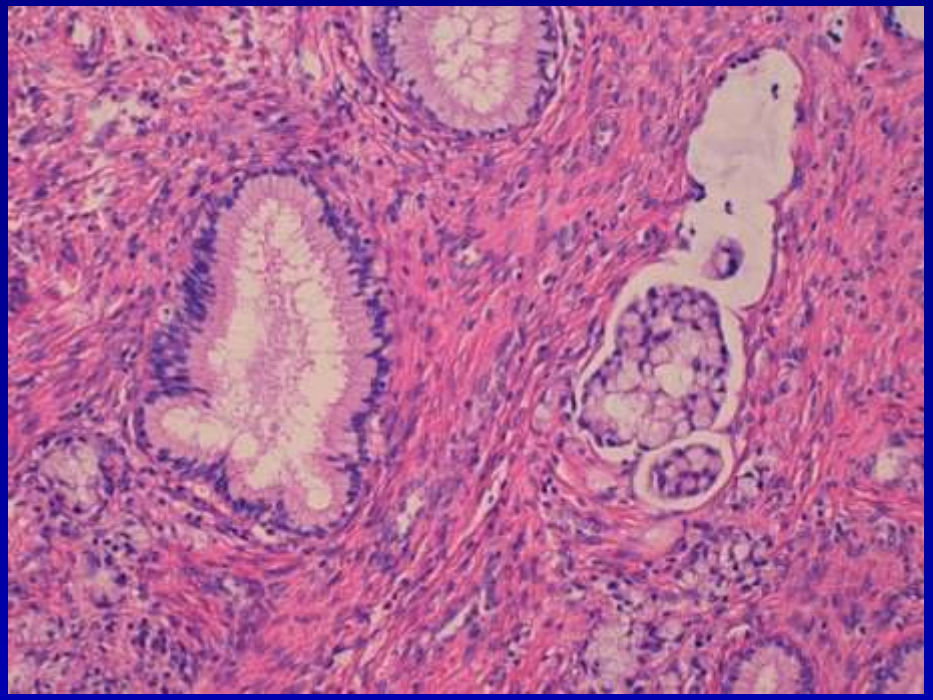

CK-20

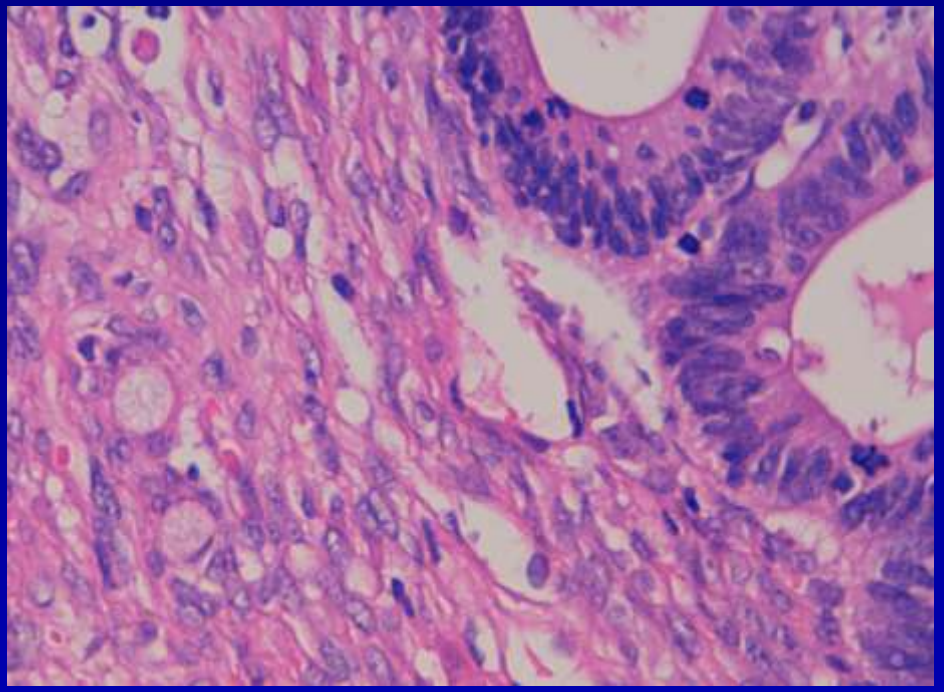

CDX-2

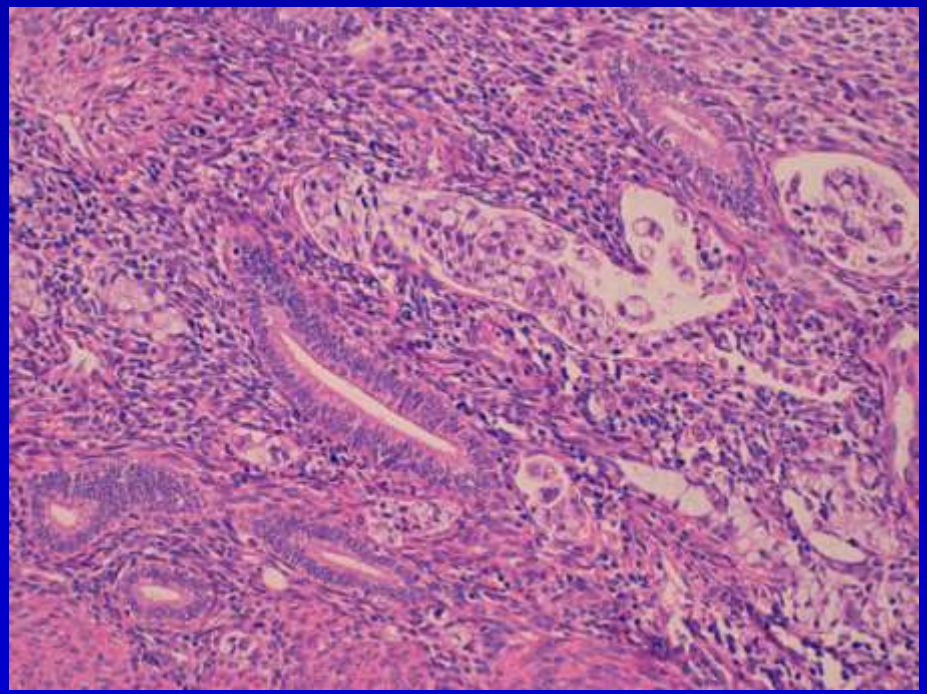

VILLIN

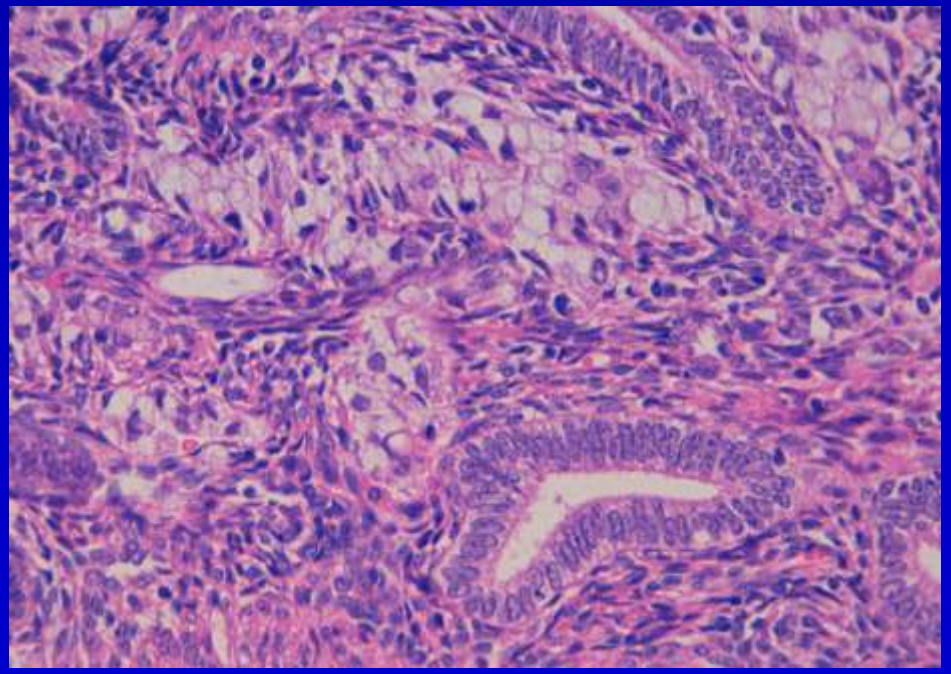
Clinical History

- **Endometrial bx: Adenocarcinoma (signet-ring features, consistent with gastrointestinal origin**
- **No evidence of gastrointestinal malignancy**
- **Hysterectomy and bilateral salpingooforectomy**









Data Quality: Acceptable

TISSUE	SIMILARITY SCORE	LOW 0 5 30 HIGH 100
Ovarian	41.9	
Colorectal	18.2	
Gastric	18.2	
Pancreatic	4.0	
Non-Small Cell Lung	3.6	
Germ Cell	3.2	
Kidney	2.1	
Breast	2.1	
Non-Hodgkin's Lymphoma	1.5	
Hepatocellular	1.4	
Thyroid	0.9	
Bladder	0.9	
Sarcoma	0.9	
Prostate	0.5	
Melanoma	0.4	

Diagnosis

Metastatic adenocarcinoma (signet-ring features) of unknown origin

Agradecimientos

- **Antonio Llombart-Cussac**
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- **Manuel Mata, J Antonio Lopez Guerrero**

Galacidalacidesoxyribonucleicacid, Salvador Dalí, 1963.