Reial Acadèmia de Medicina i Ciències Afins de la Comunitat Valenciana

Excelentisimo Señor
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Prof. Dr. Enrique Merino
Valladolid

ASPECTOS MOLECULARES DEL CANCER DE RIÑON

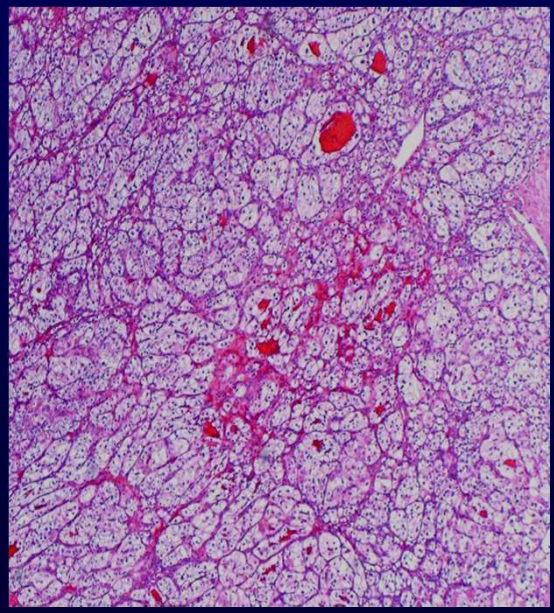
Maria J. Merino M.D

NCI

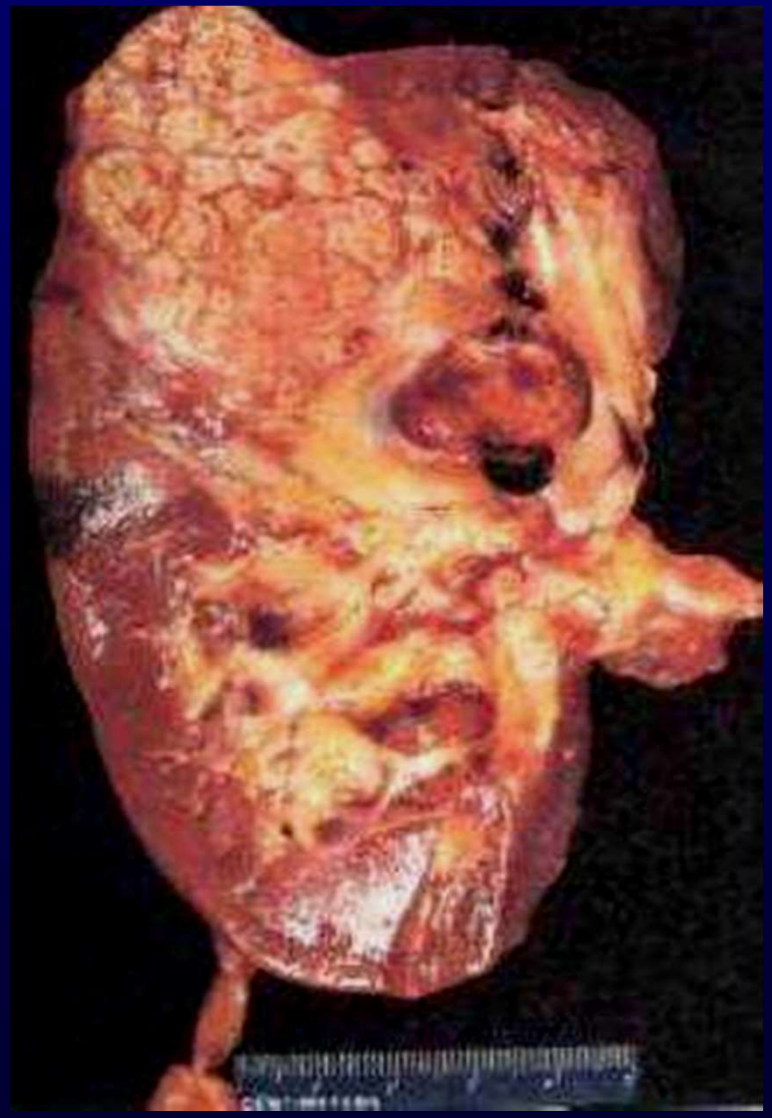
Paul Albert Grawitz (1850-1932)

Grawitz' tumour

(Hypernephroma)



Renal Cell Carcinoma



Problems:

****Disease diagnosed some times in advanced stages**

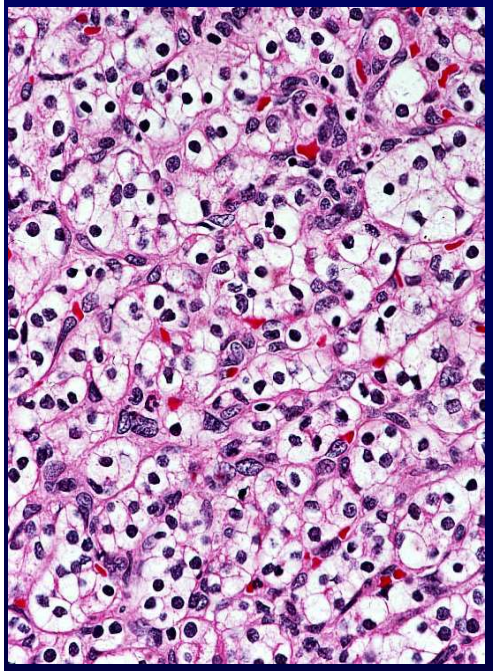
***Absence of markers for early detection or monitoring disease progression.**

***No good therapy. Surgery**

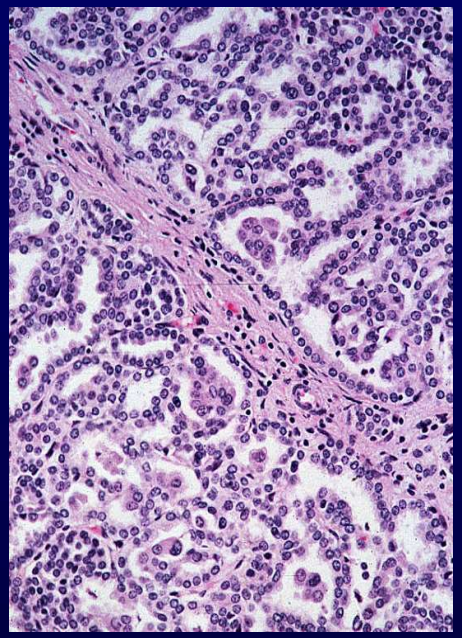
Kidney Cancer Statistic

- Kidney cancer is among the 10 most common cancers in both men and women.
- About 62,700 new cases of kidney cancer (39,650 in men and 23,050 in women) will occur.
- About 14,240 people (9,240 men and 5,000 women) will die from this disease.
- The rate of new kidney cancers has been rising since the 1990s world wide. Early diagnosis CT scans

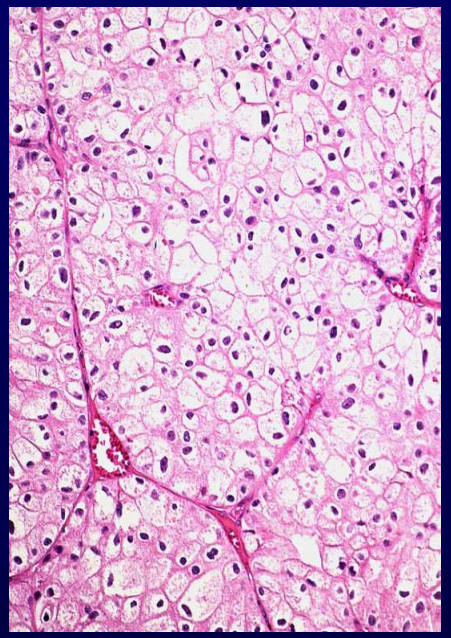
In the beginning….Kidney Tumors were simple…..



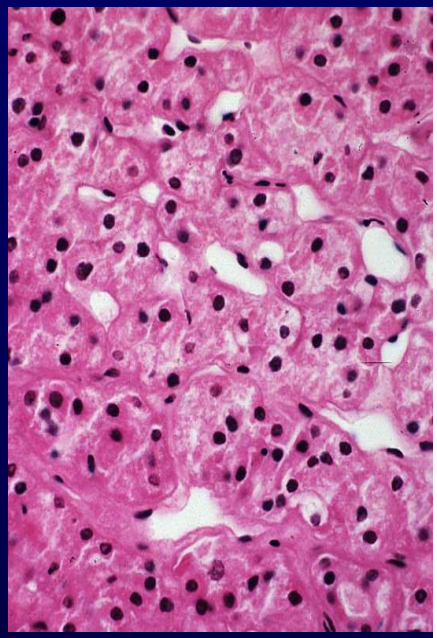
Clear cell



Pap 1

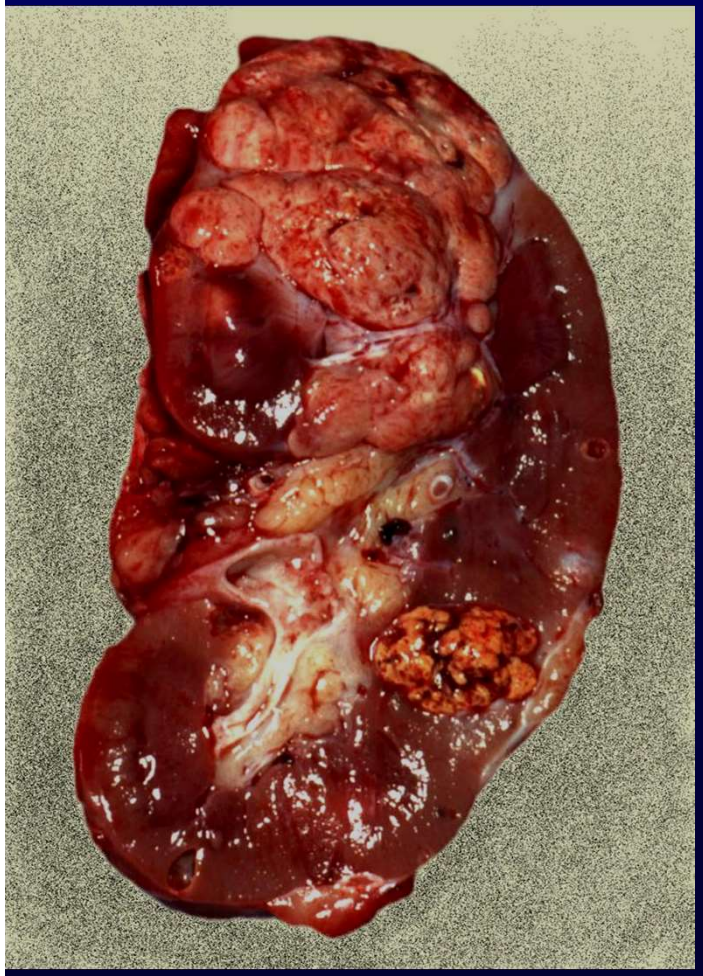


Chromo



Onco

RCC Clear Cell, Lack of response to treatment

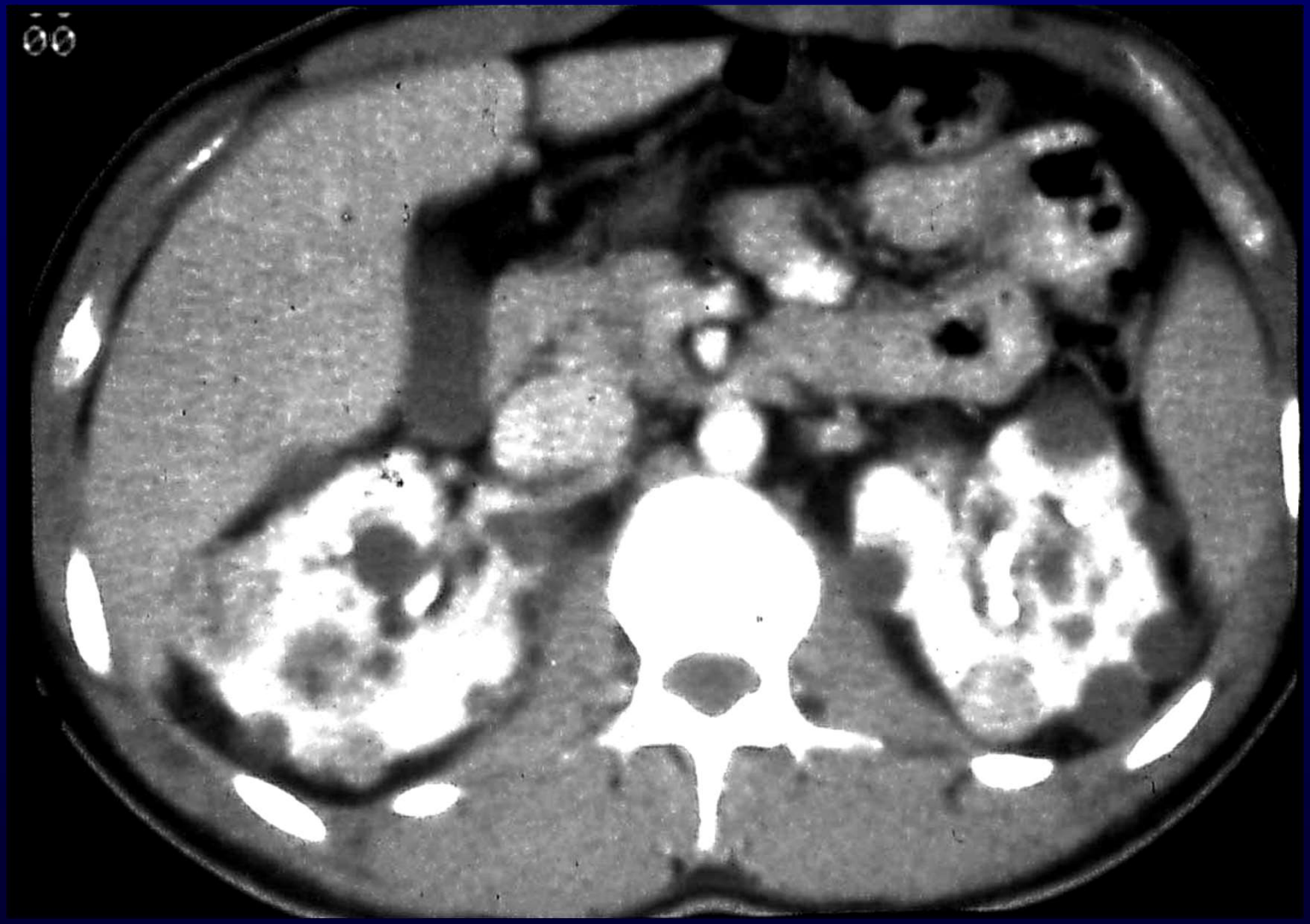


New experimental protocols in search of better therapies

Immunotherapy..
Dr. Steven Rosenberg

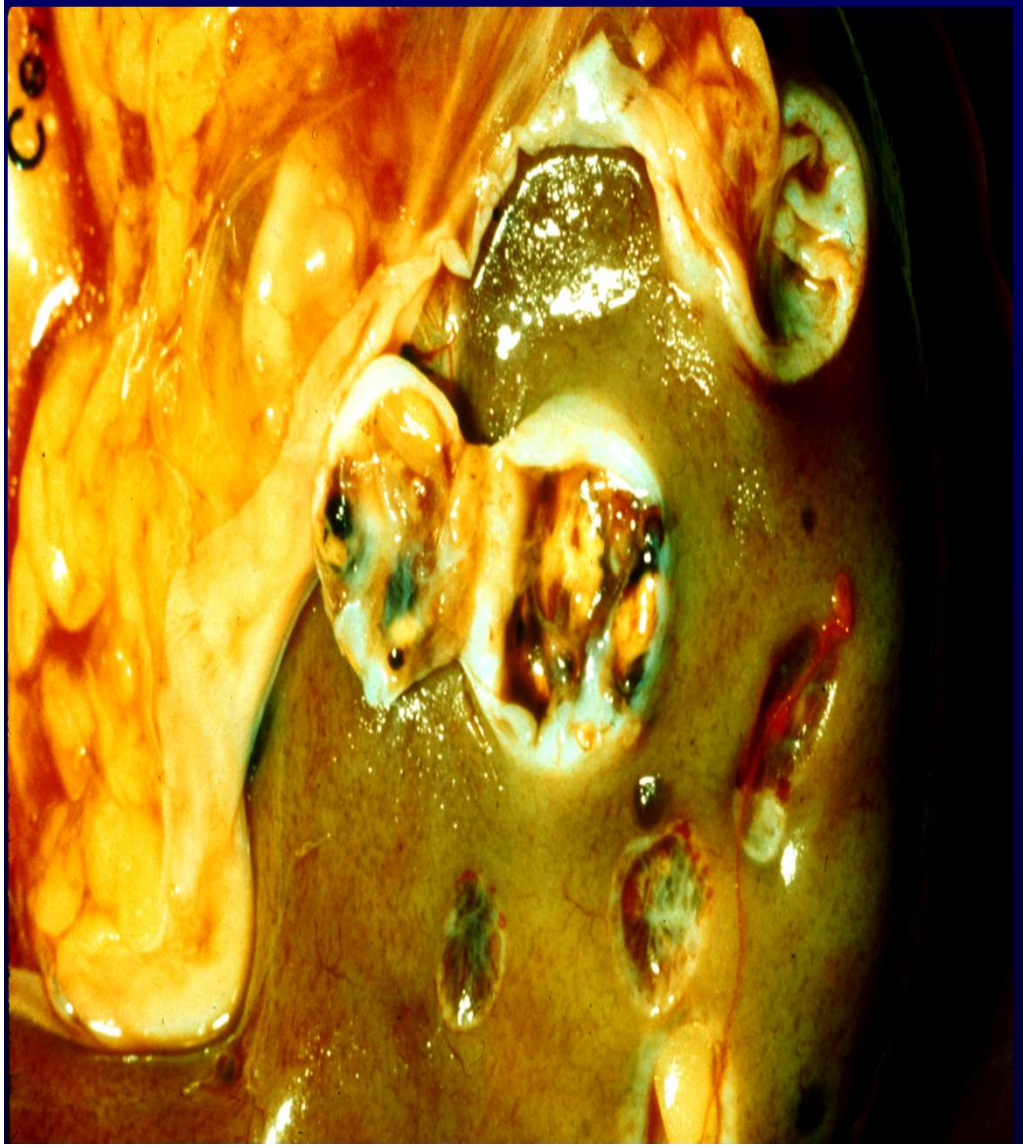
New Modalities of Surgical approach, genetics
Dr. Marston Linehan

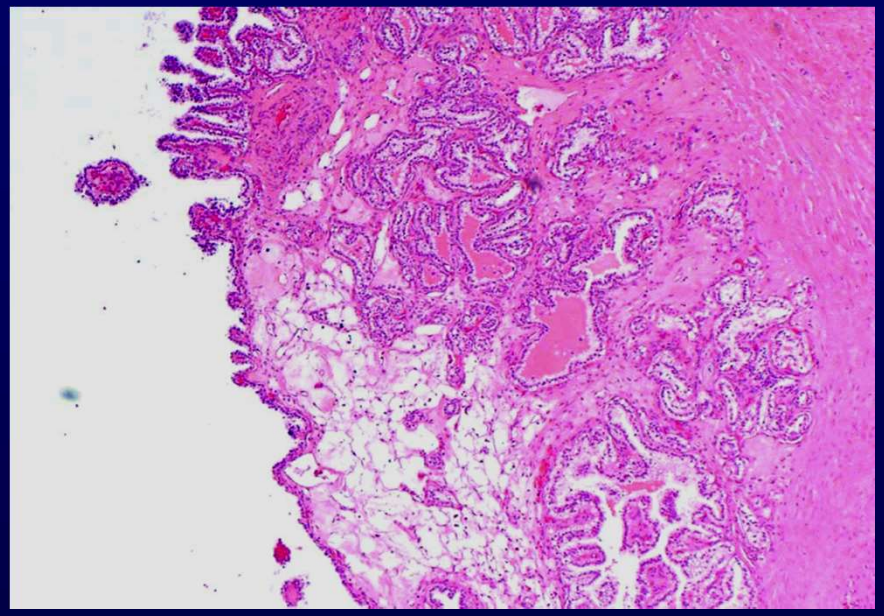
Renal Cell Carcinoma

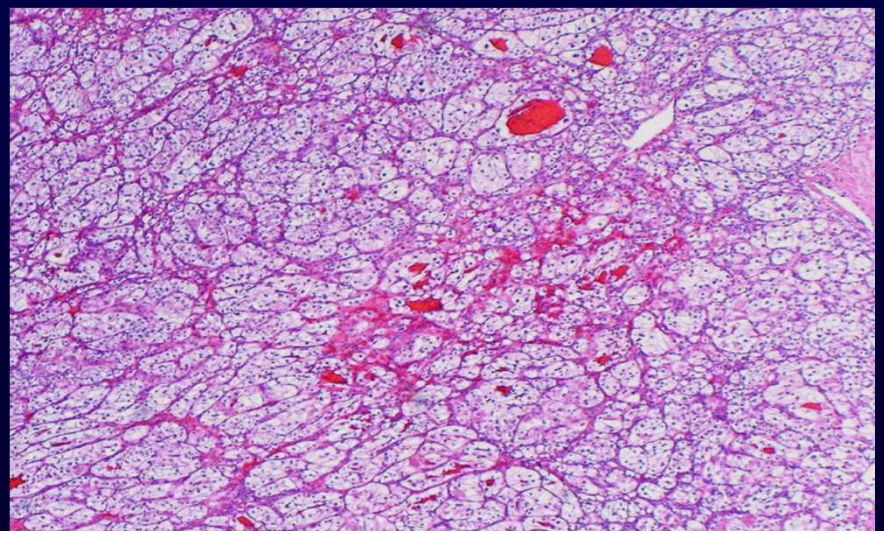


CT Scan: Bilateral, Multifocal lesions

von Hippel-Lindau (VHL)
Multiple Lesions



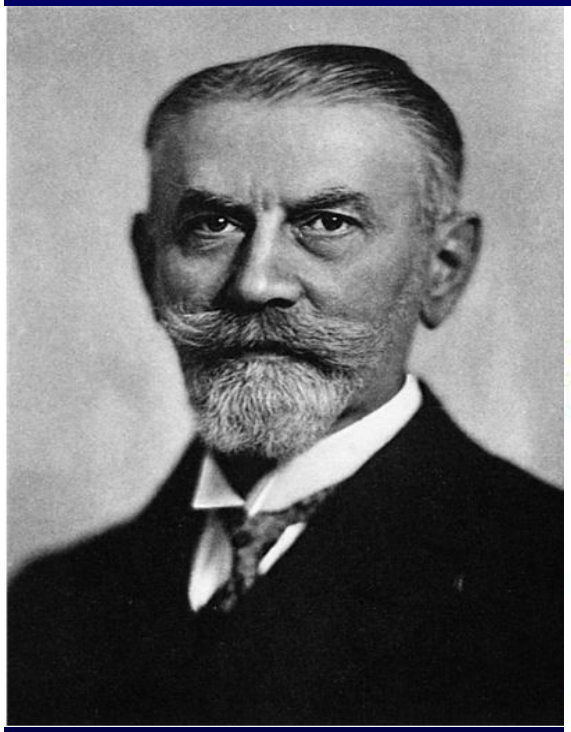




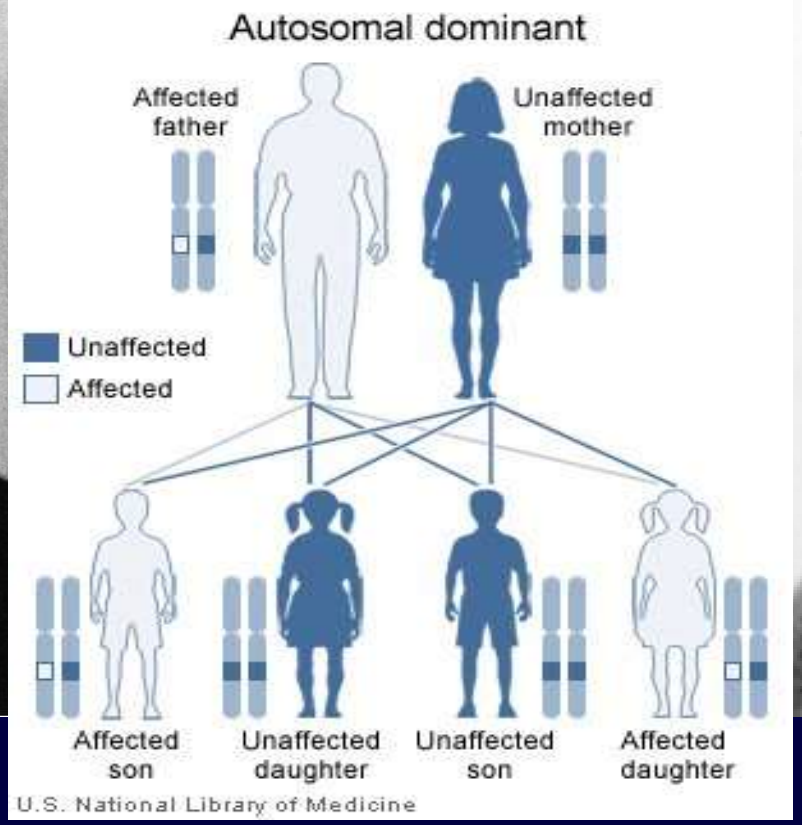
Hereditary syndromes

- Von Hippel-Lindau disease
- Tuberous Sclerosis
- Syndromes associated with Wilm's

Von Hippel Lindau Syndrome



In 1904 VH wrote "about a very rare disease of the retina. He studied one extended family, with several generations

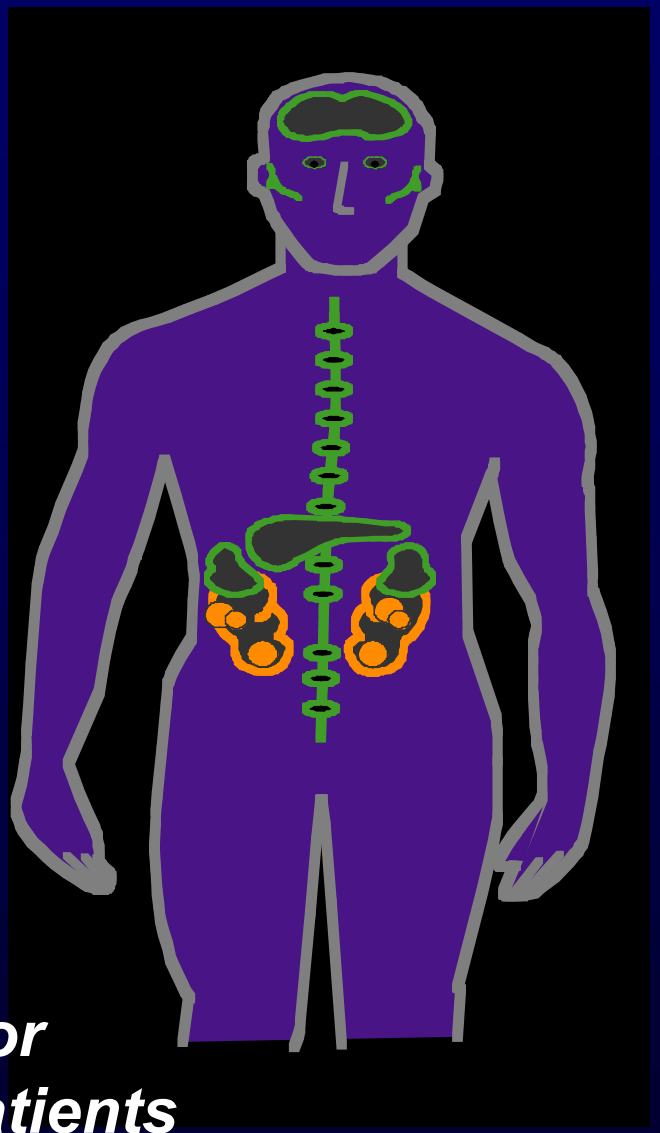


In 1964 Melmon and Rosen, described a large VHL family and codified the term "von Hippel-Lindau“.

hemangioblastomas of cerebellum and retina The condition was inheritable. He observe the visceral manifestations of renal and pancreatic involvement

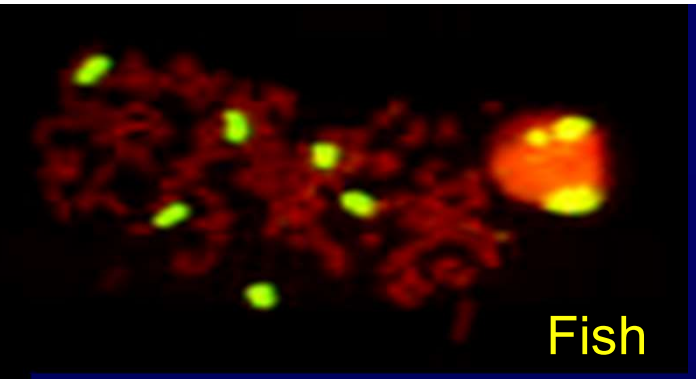
VHL Clinical Features

- **Autosomal dominant disease**
- **Tumors develop in:**
  - **-Both Kidneys (35-45%)**
  - -Pheochromocytomas
  - -Pancreas
  - -Hemangioblastomas
  - -Retinal angiomas
  - -Endolymphatic sac tumors
  - -Epididymal cysts
  - -Broad ligament cysts

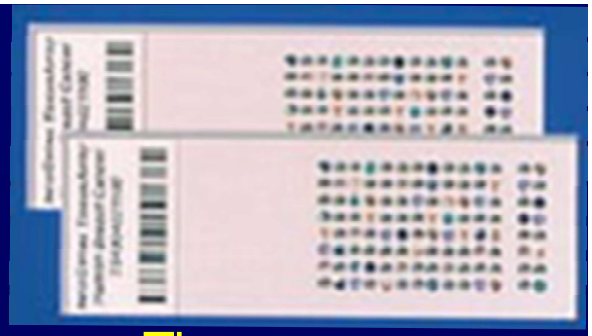


Diagnosis: ***Two separate tumors and/or family history of VHL. 20% occur in patients with no history***

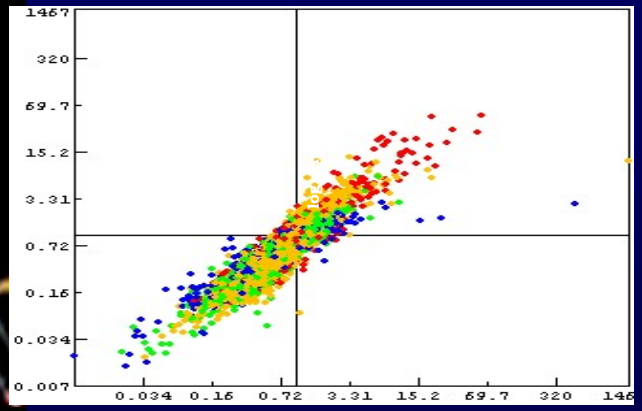
genome

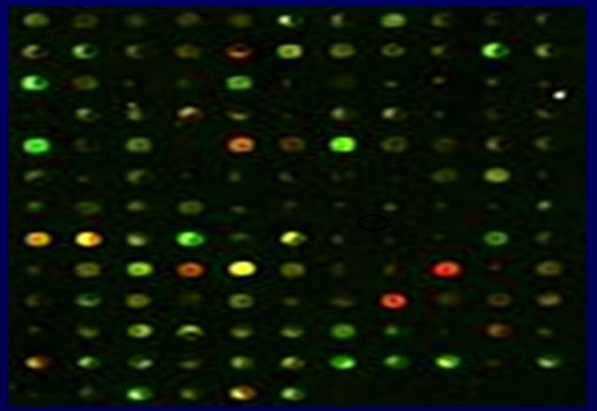


Fish

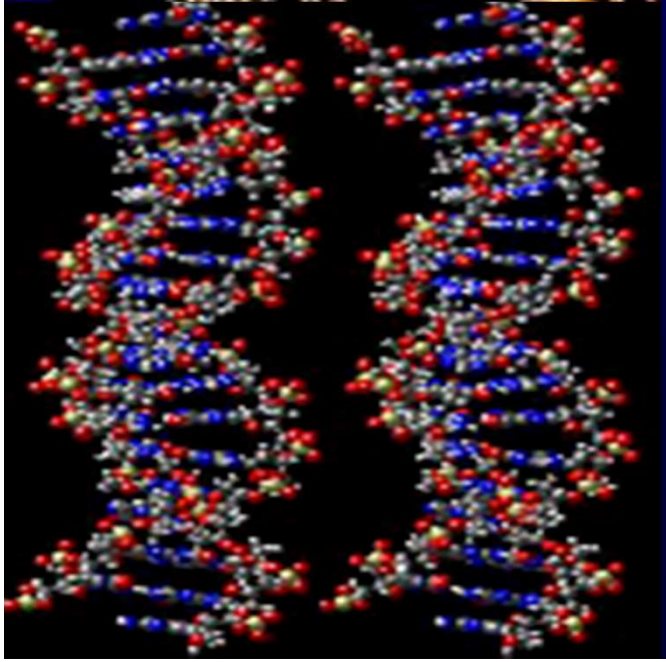


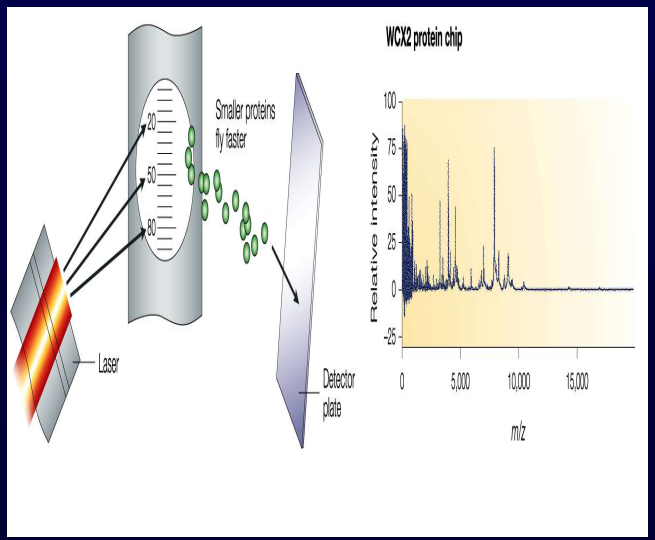
Tissue arrays

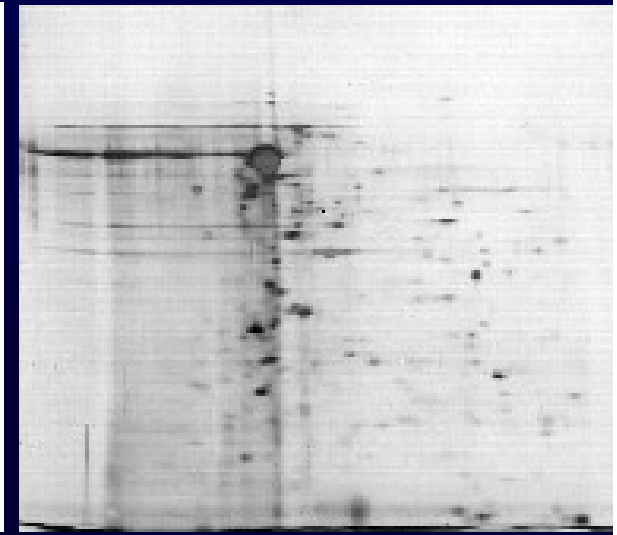




Microarrays







Proteomics

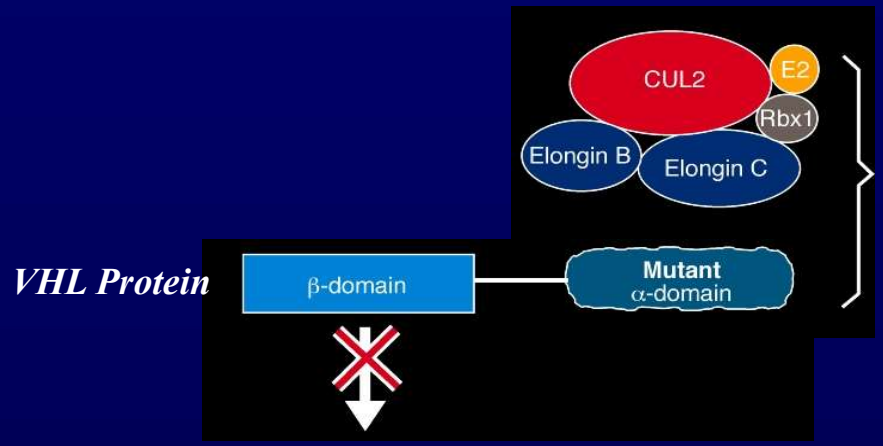
VHL

- DNA was microdissected from normal and tumor. 51/58 cases showed LOH on chromosome 3. Later studies identified the distal region of 3p as the region of the disease gene (3p21-26). (1991)

-An international consortium was formed and Glenn, Latiff discovered the gene in 1993.

-The VHL gene is a Tumor supressor gene

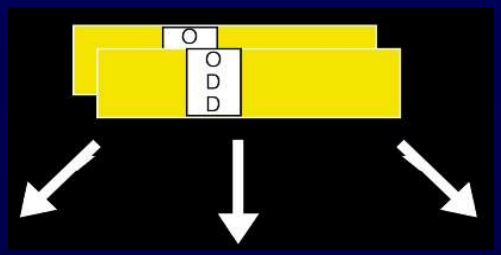
Downstream Effects of VHL Gene Mutation



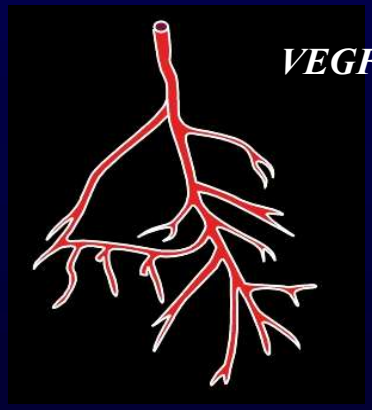
VHL Protein

VHL Complex Disrupted

HIF-α Accumulation

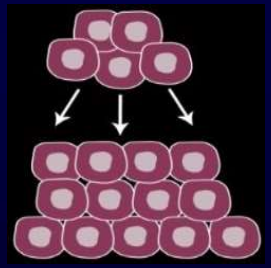


VEGF



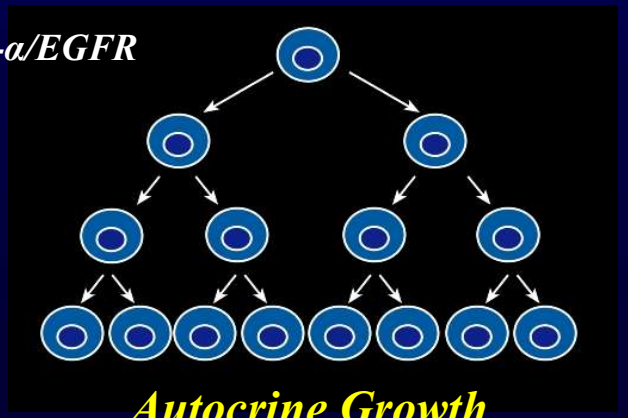
Angiogenesis

PDGF



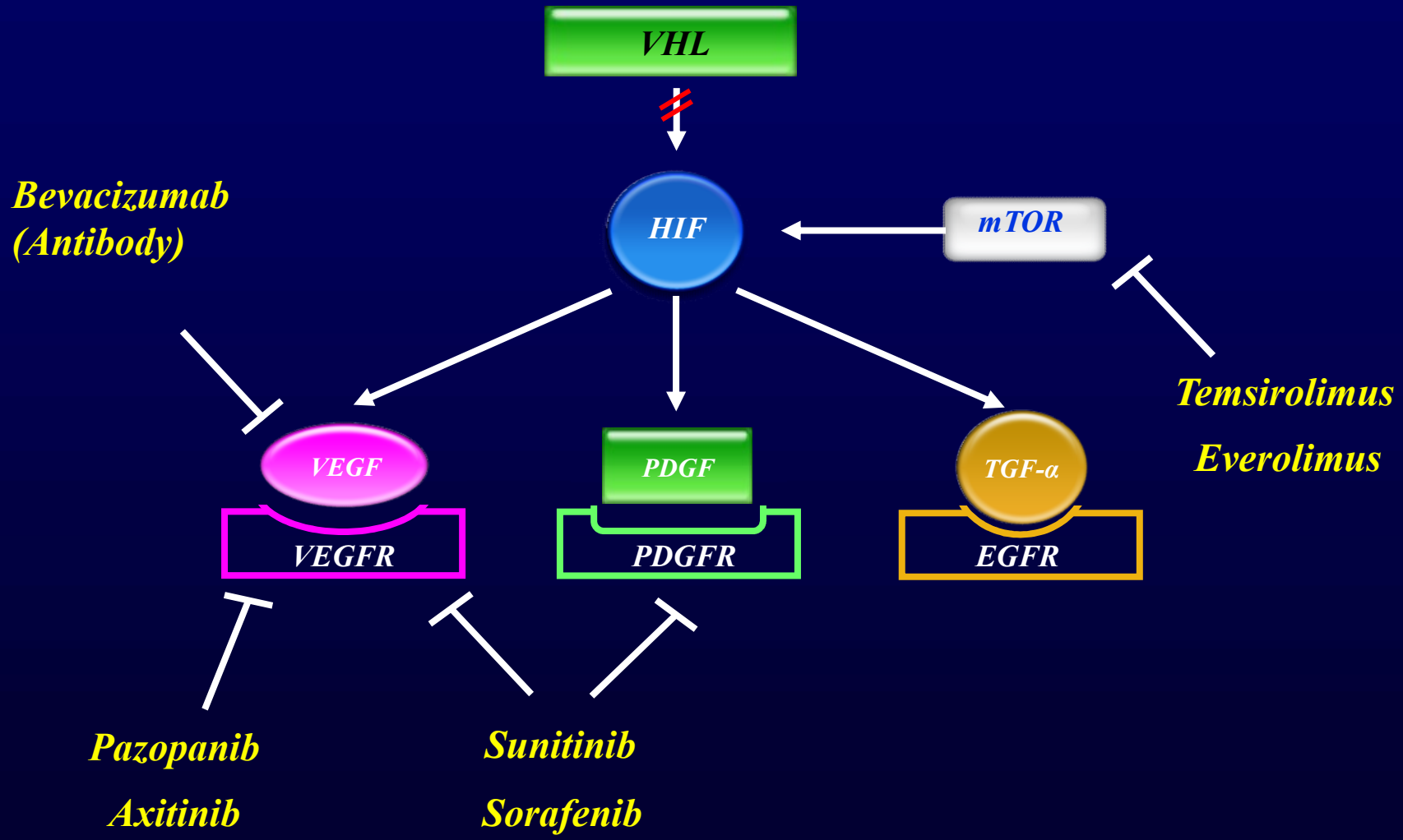
Paracrine Growth Stimulation

TGF-α/EGFR



Autocrine Growth Stimulation

Targeting VHL/HIF in Clear Cell RCC



VHL Gene

Two types of Clear Cell RCC

Sporadic: *Single masses, usually Higher grade. Alterations of the VHL gene occur in up to 60% -80% of sporadic tumors.*

Hereditary: *Multiple and bilateral, cysts, lower grade. VHL alterations 100%. Characteristic morphology*

Therapy (Sporadic)

- Surgery
- Cytokine therapy with interleukin 2 (IL-2). Response rate 15-20%.
- New therapies targeting specific molecules (protein kinase inhibitors, inhibitors of apoptosis binding proteins, HIF1-a, -anti-angiogenic factors…..)

Therapy (Hereditary)

- Morphology …dx of VHL …. genetic testing……
- Partial nephrectomy for tumors less than 3 cm with removal of cysts and smaller lesions.
- Close follow up of small lesions.
- Evaluation of family members, genetic testing and counseling
- Patients with Tumors smaller than 3-4 cm have excellent prognosis with prolonged survivals. (10-19)

Remember these patients used to be treated by nephrectomy..and dialysis…

VHL Clinical Features

-Cerebelar and Spinal Hemangioblastomas

-Retinal angiomas

-Endolymphatic sac tumors

-Pheochromocytomas

-Renal cell carcinomas and renal cysts

-Pancreatic cysts and neuroendocrine tumors

-Epididymal cysts

-Broad ligament cysts

-LUNG

-PLEURA

-TESTIS

The Cancer Genome Atlas

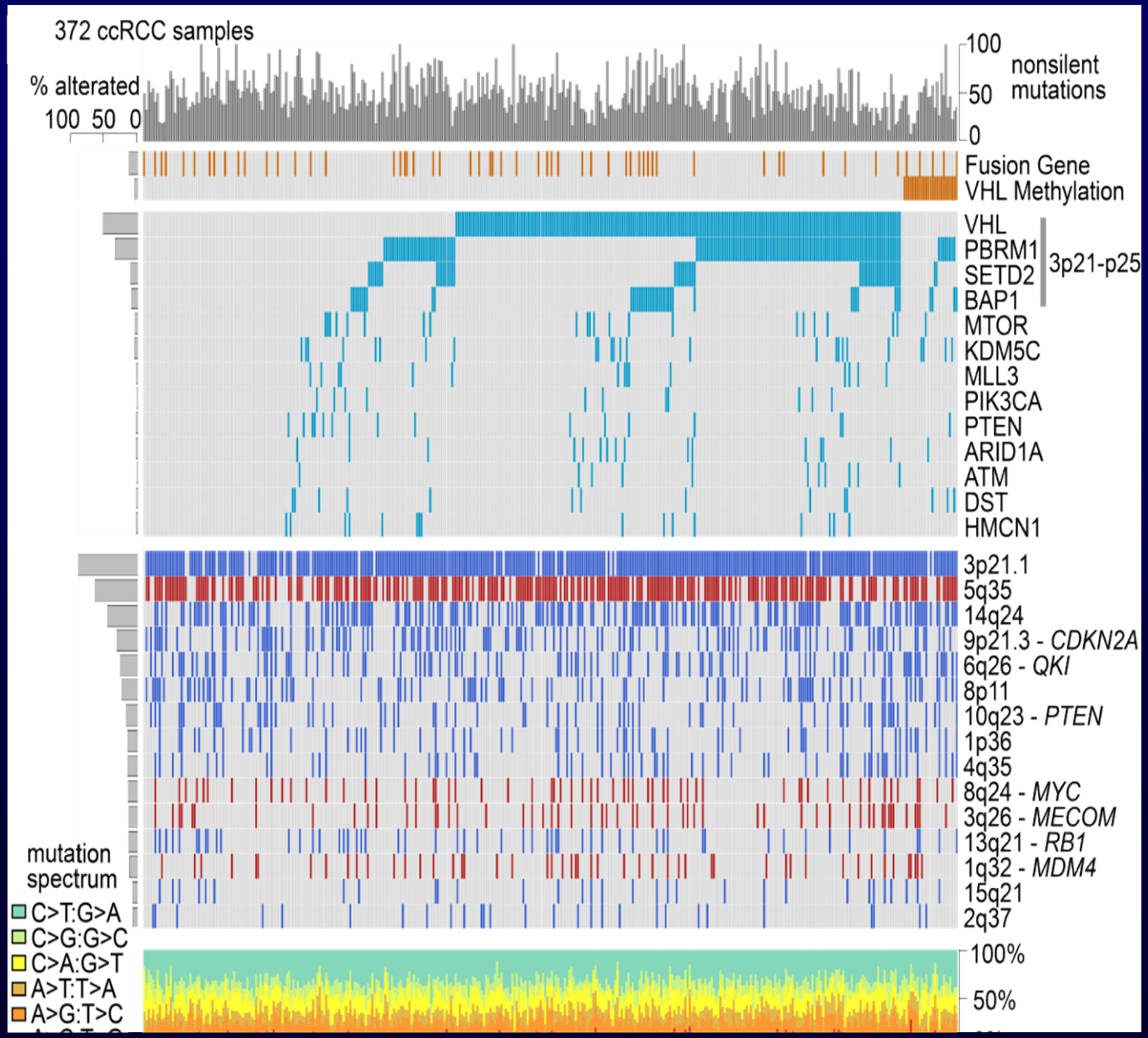
Genetic analysis of many tumors

In kidney:

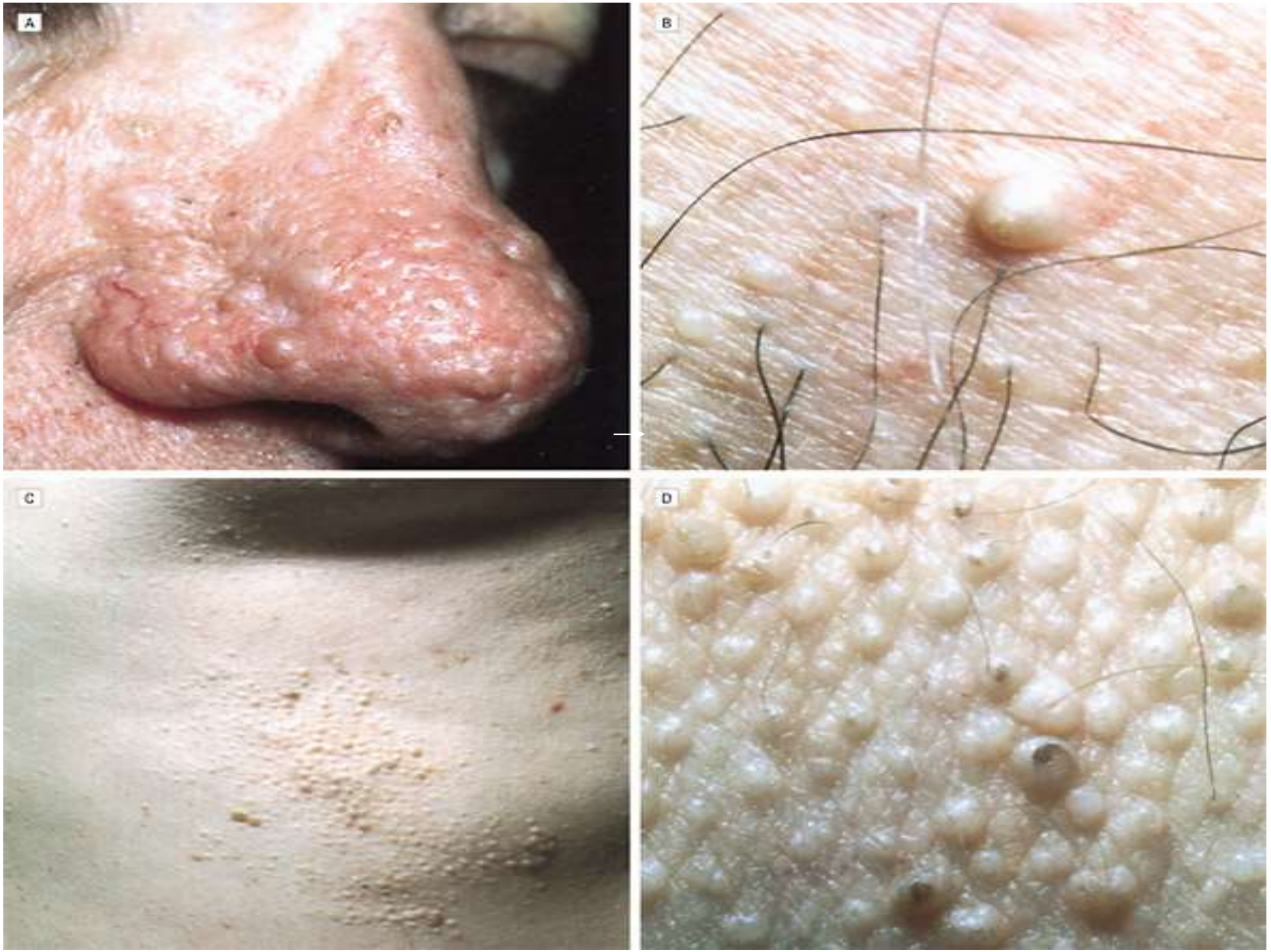
Clear Cell: : 3p (90%)–PBRM1, SETD2, BAP1, JARID1A, mTOR, PI3K, 14q, 8p, and 9p and gains at 5q and 12q

Chromophobe

TCGA Clear Cell Kidney Cancer



Nature 499:2013


A
B
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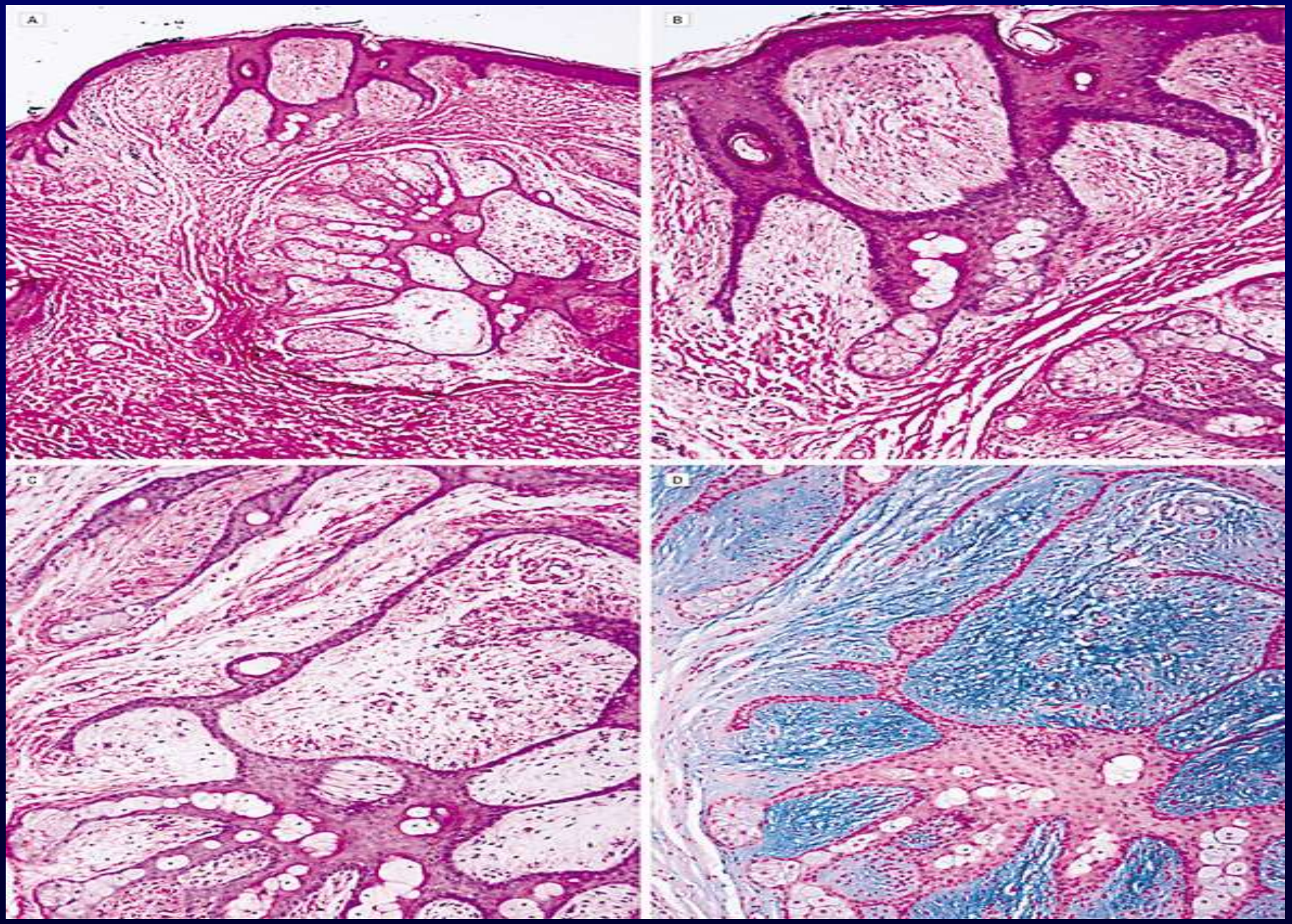


Fibrofolliculoma, achrocordon

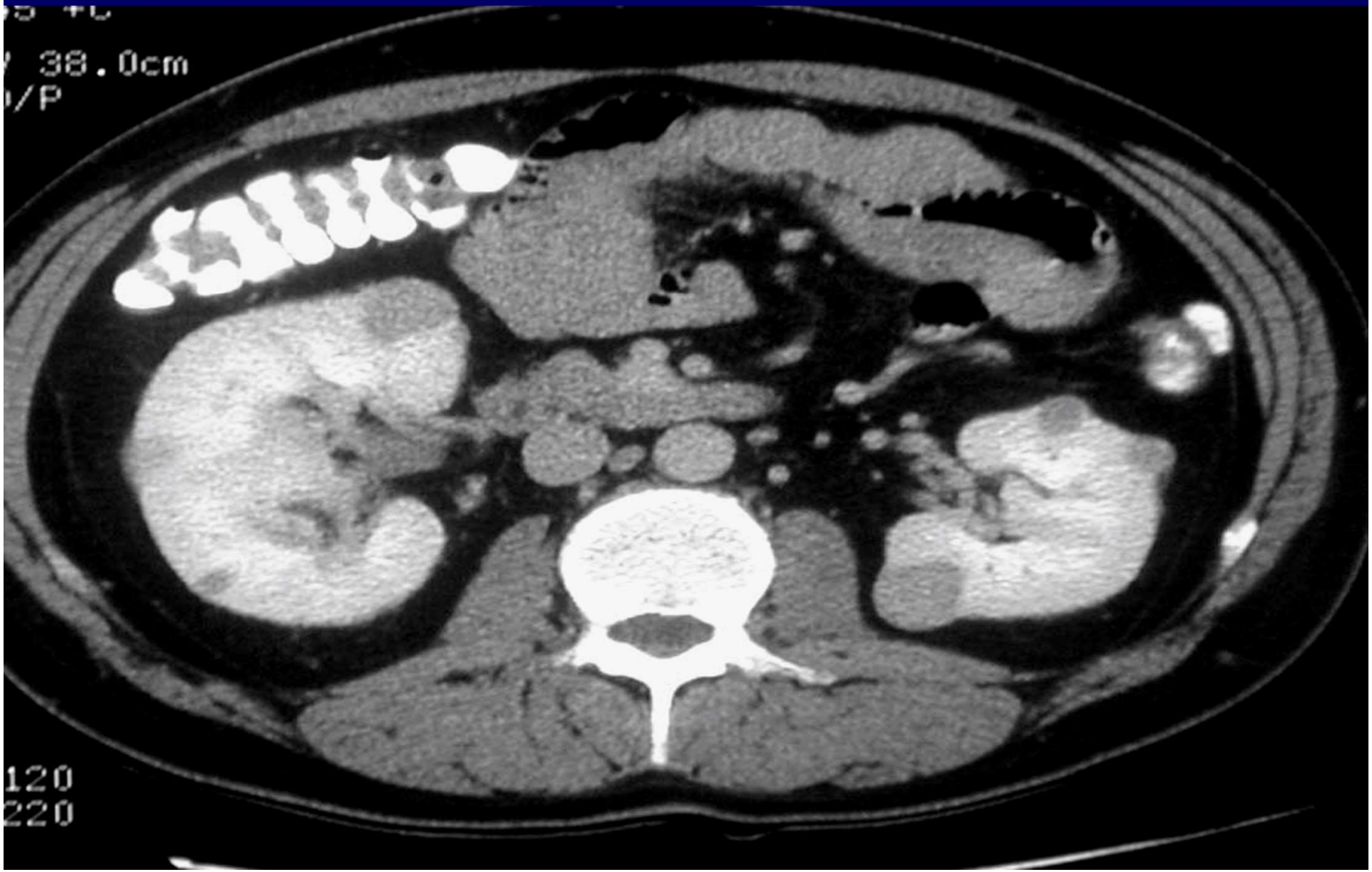
Hereditary Multiple Fibrofolliculomas With Trichodiscomas and Acrochordons

Arthur R. Birt, MD, FRCP (C); Georgina R. Hogg, MD, FRCP (C); W. James Dubé, MD

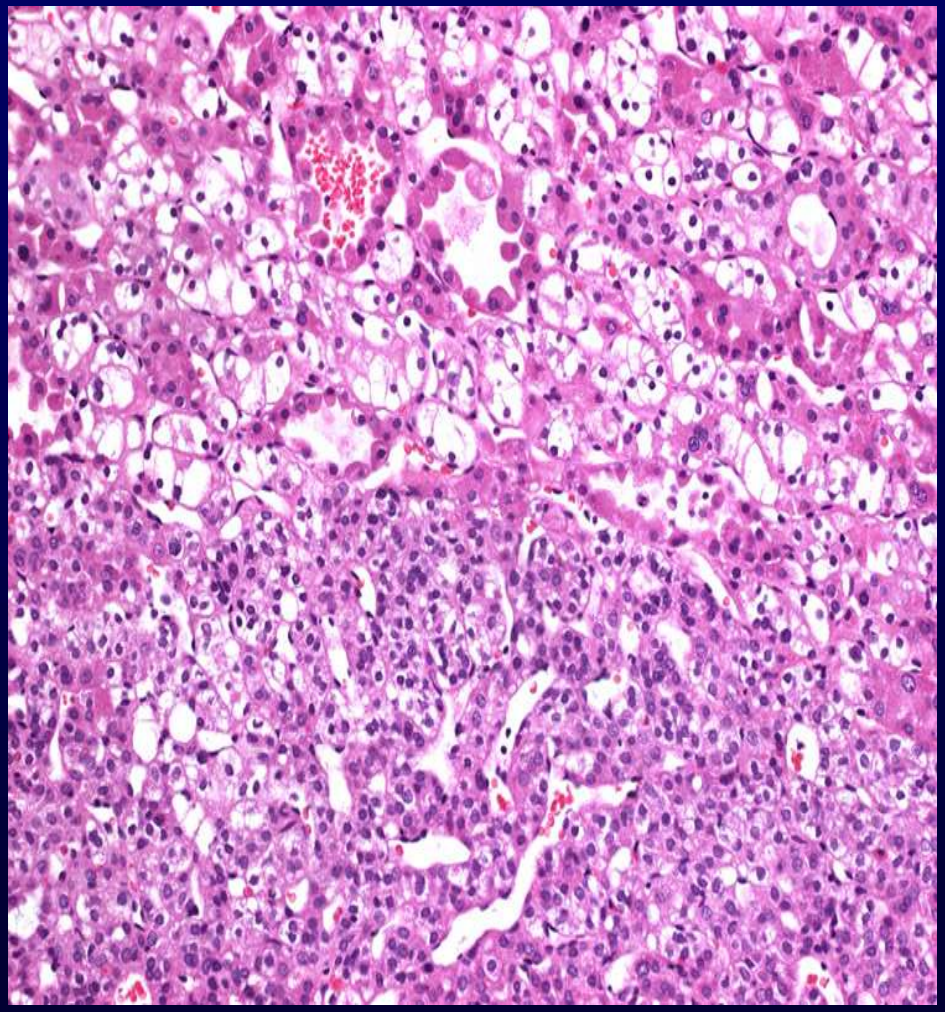
Arch Derm 113:1674-1677
Dec 1977

Arthur Birt

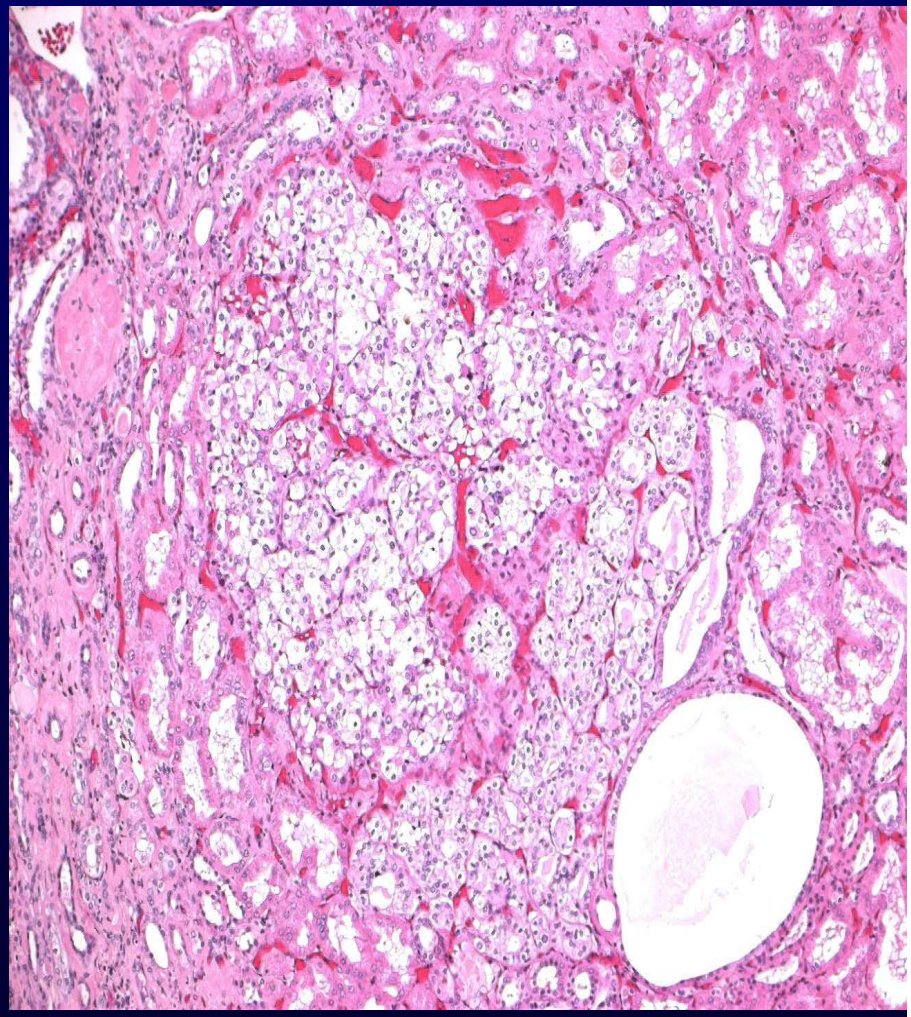
Kidney Tumors



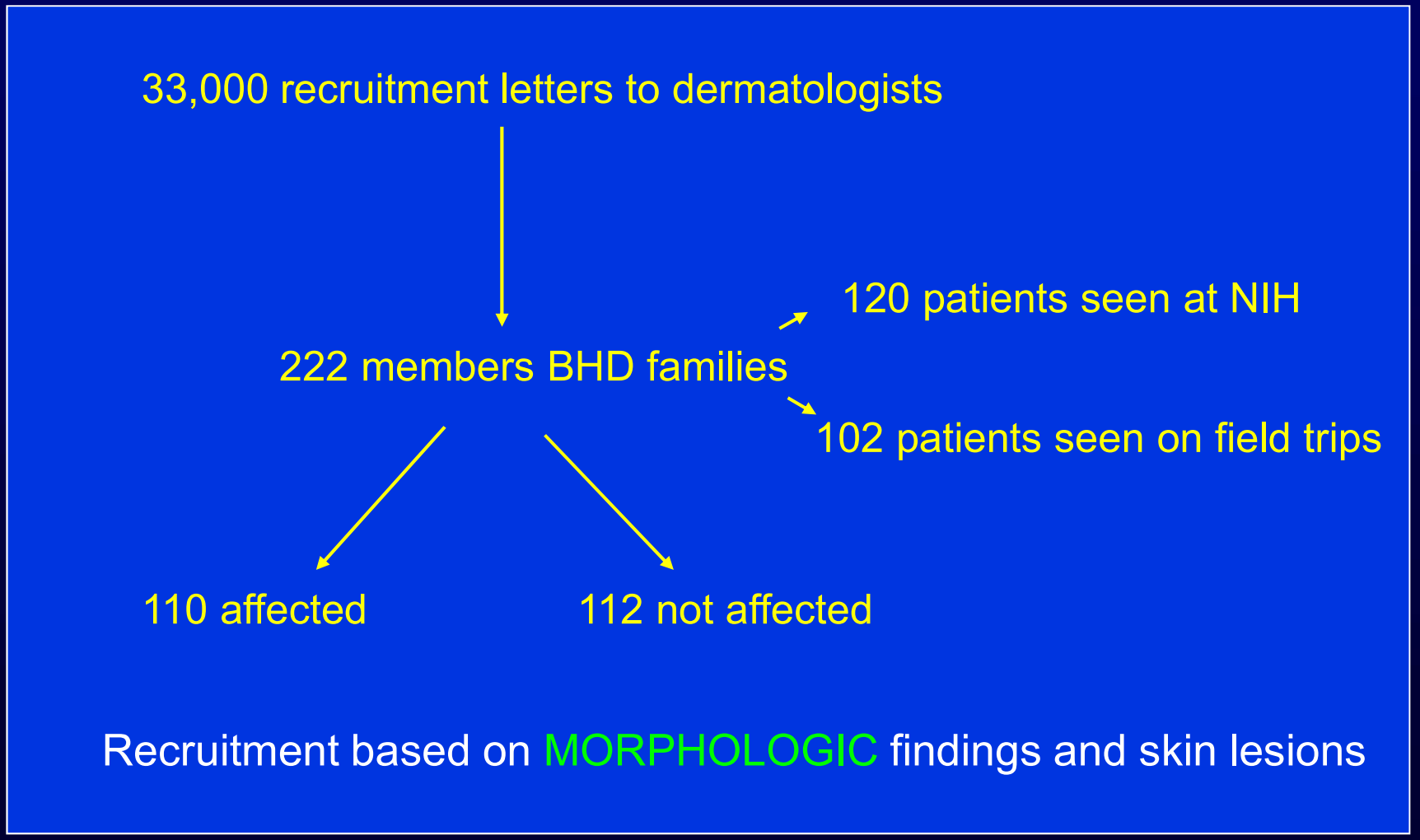
Twin 1



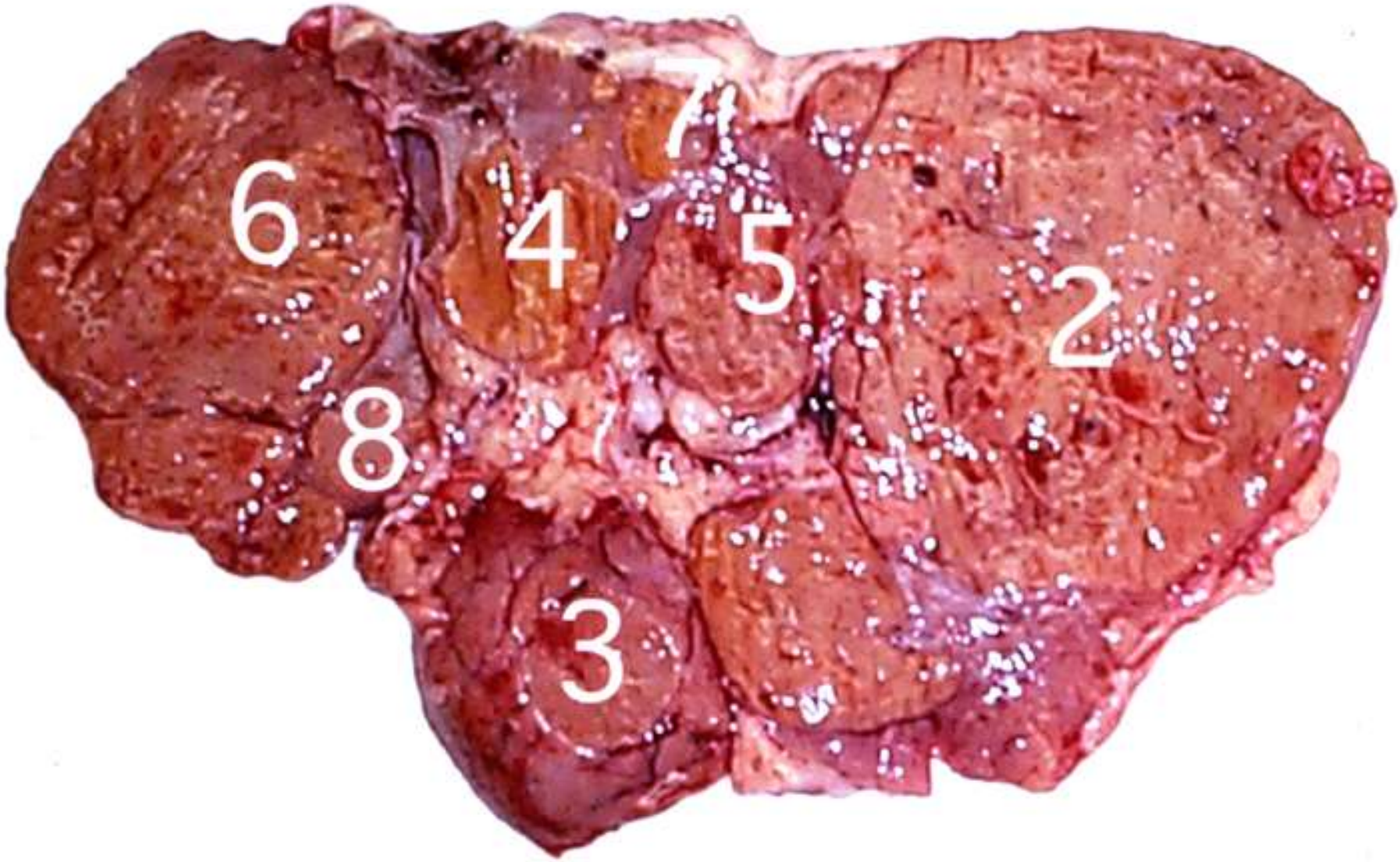
Twin 2



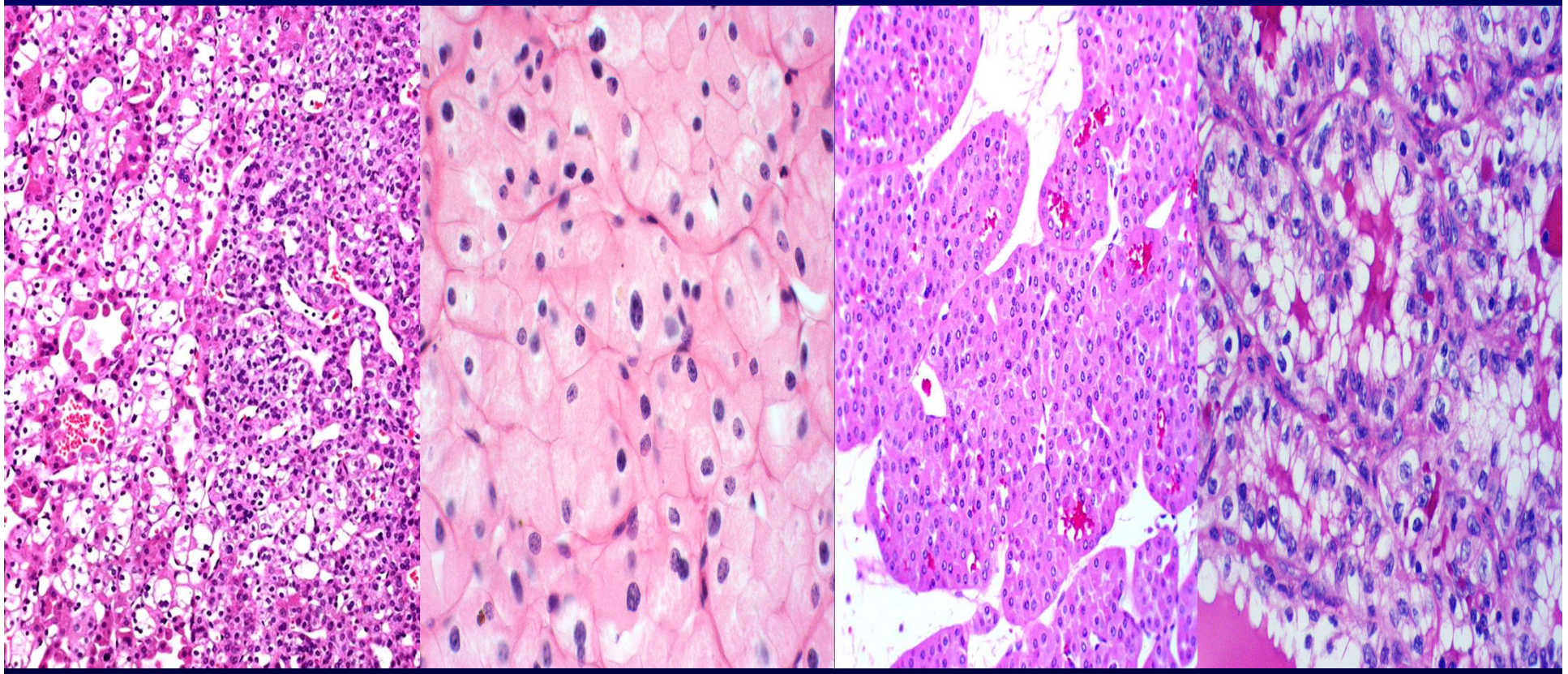
BHD Study Design



BHD



Birt Hogg Dubé Renal Tumors



Hybrid
LOH 17

Chromophobe
LOH 17

Oncocytoma
LOH 17

RCC
LOH 3p

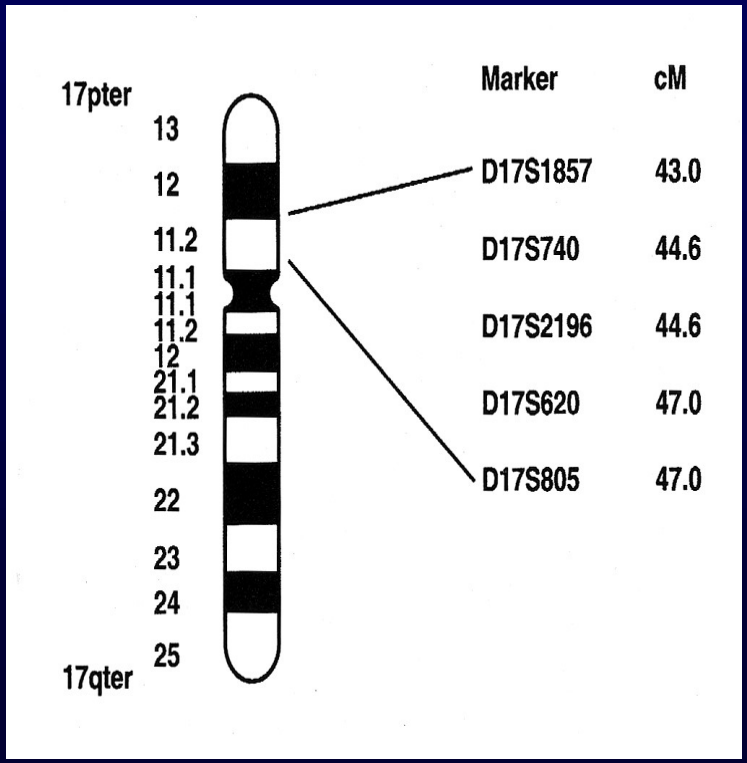
Microdissected DNA in 88 cases

Pavlovich, Merino

BHD1 Gene Locus Chromosome 17

Mutations in a novel gene lead to kidney tumors, lung wall defects, and benign tumors of the hair follicle in patients with the Birt-Hogg-Dubé syndrome

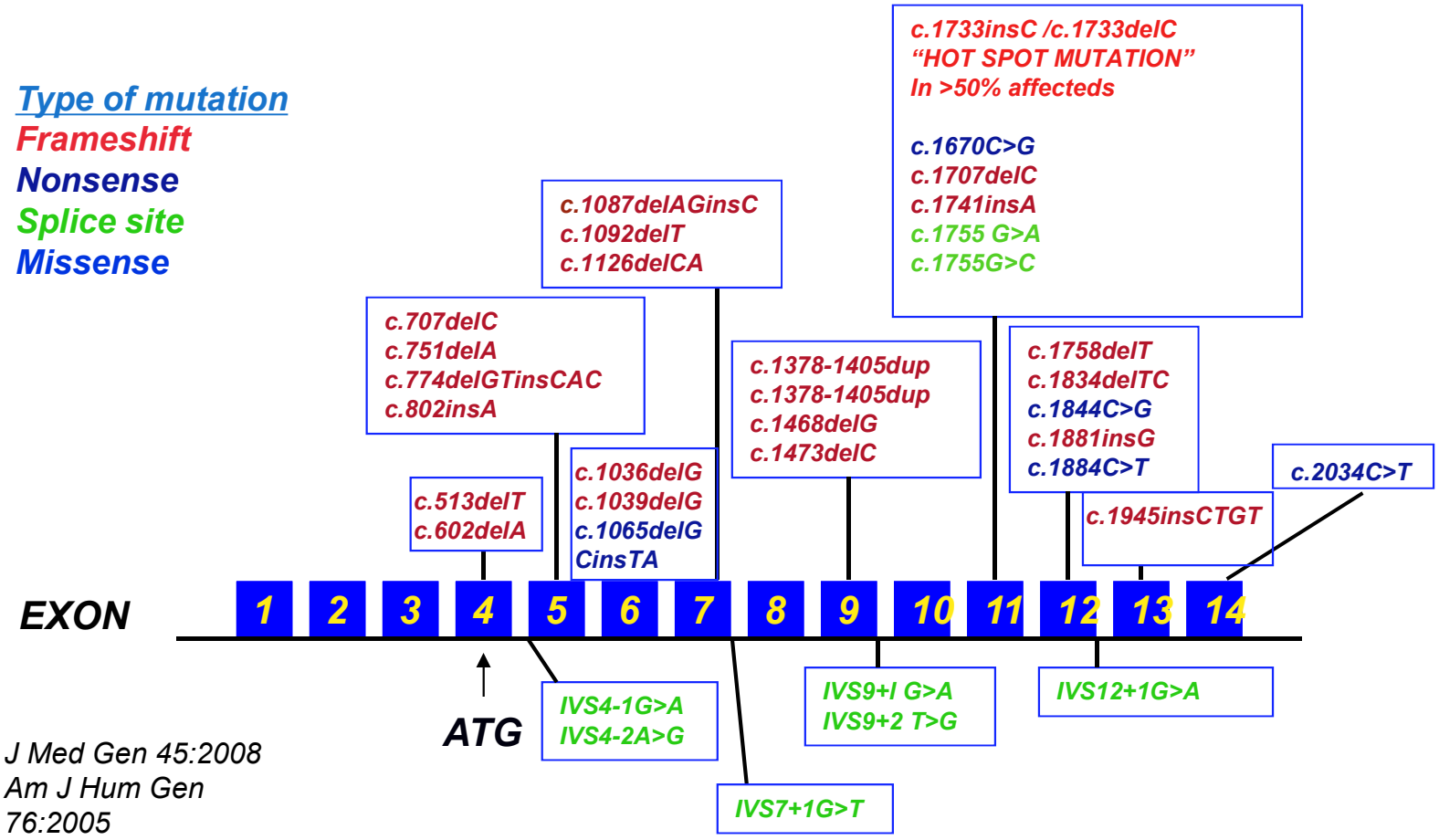
Michael L. Nickerson,[1] Michelle B. Warren,[1] Jorge R. Toro,[4] Vera Matrosova,[1] Gladys Glenn,[4] Maria L. Turner,[5] Paul Duray,[6] Maria Merino,[6] Peter Choyke,[8] Christian P. Pavlovich,[7] Nirmala Sharma,[1] McClellan Walther,[7] David Munroe,[3] Rob Hill,[3] Eamonn Maher,[9] Cheryl Greenberg,[10] Michael I. Lerman,[1] W. Marston Linehan,[7] Berton Zbar,[1] and Laura S. Schmidt[2,11]



Cancer Cell. August 2002

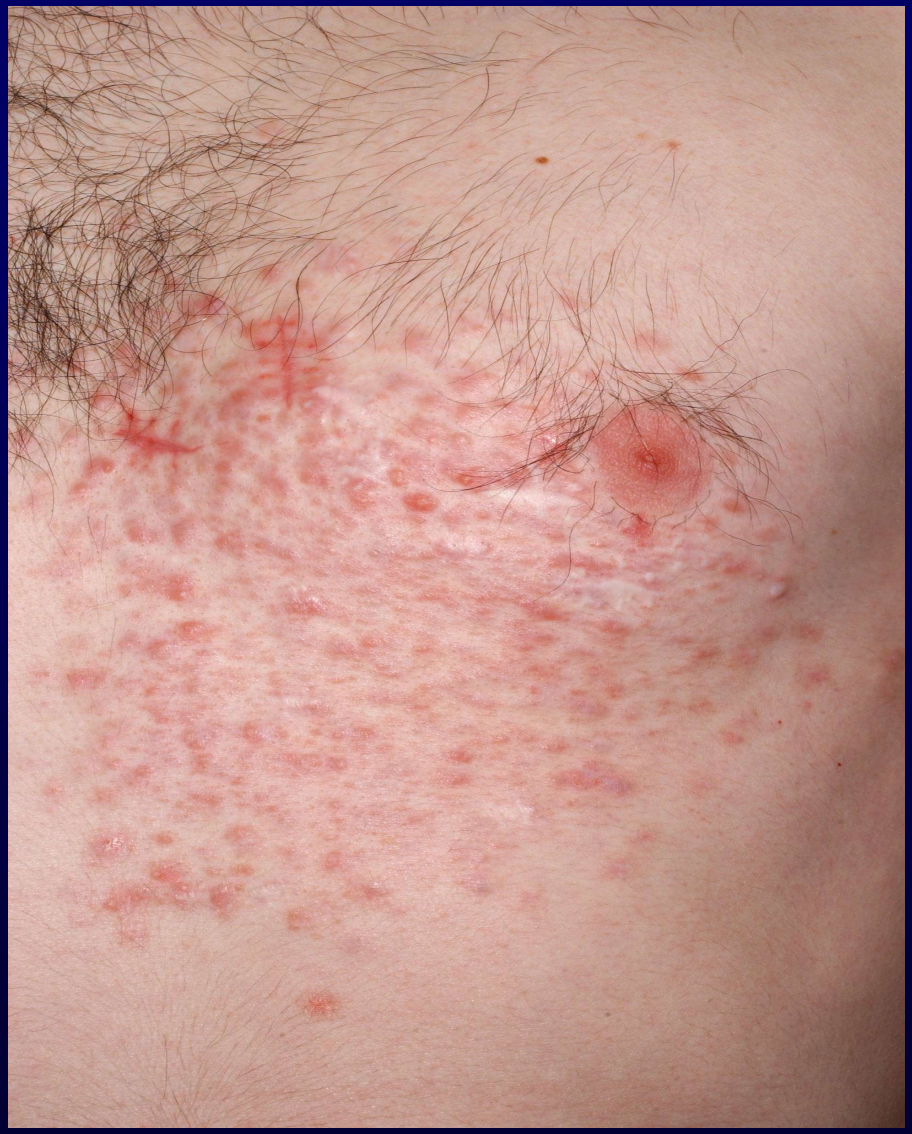
FLCN mutation spectrum

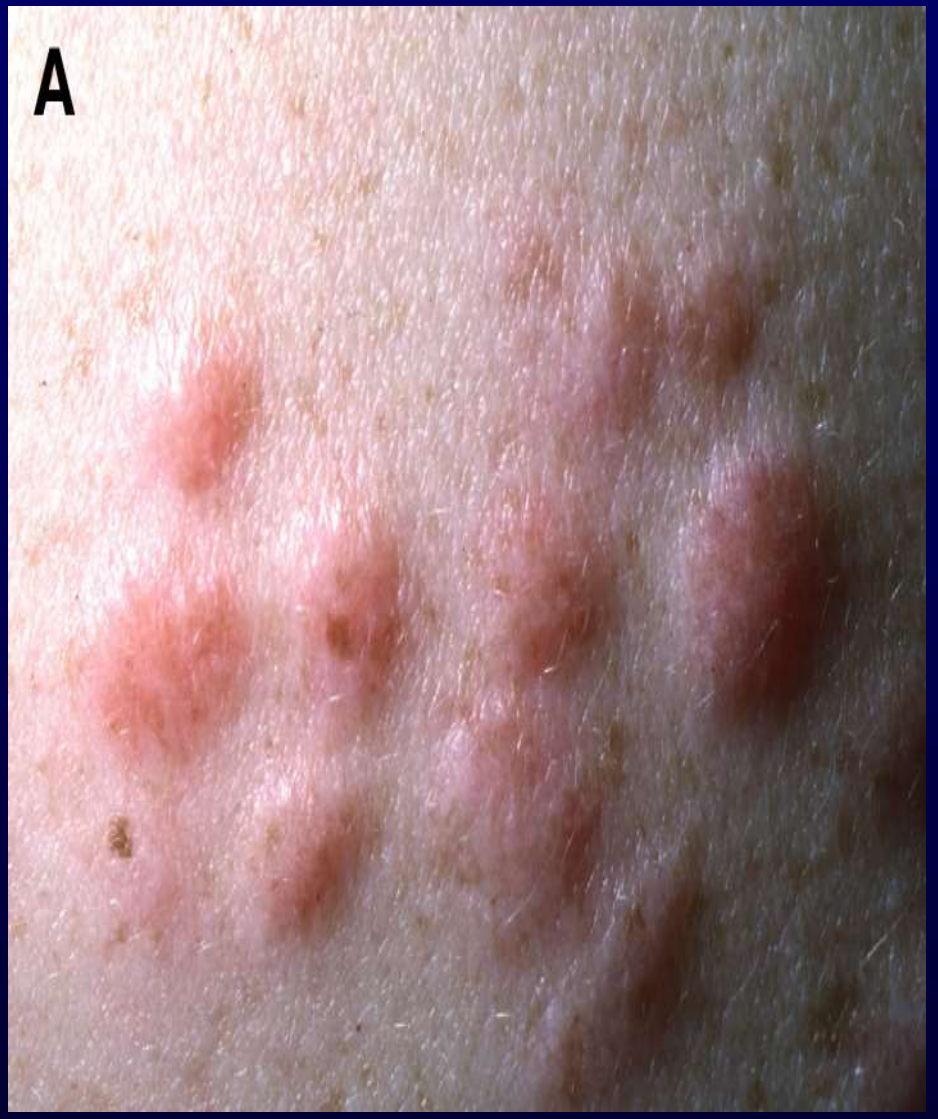
FLCN mutations across the entire gene
97% detection rate in 176 NCI BHD families

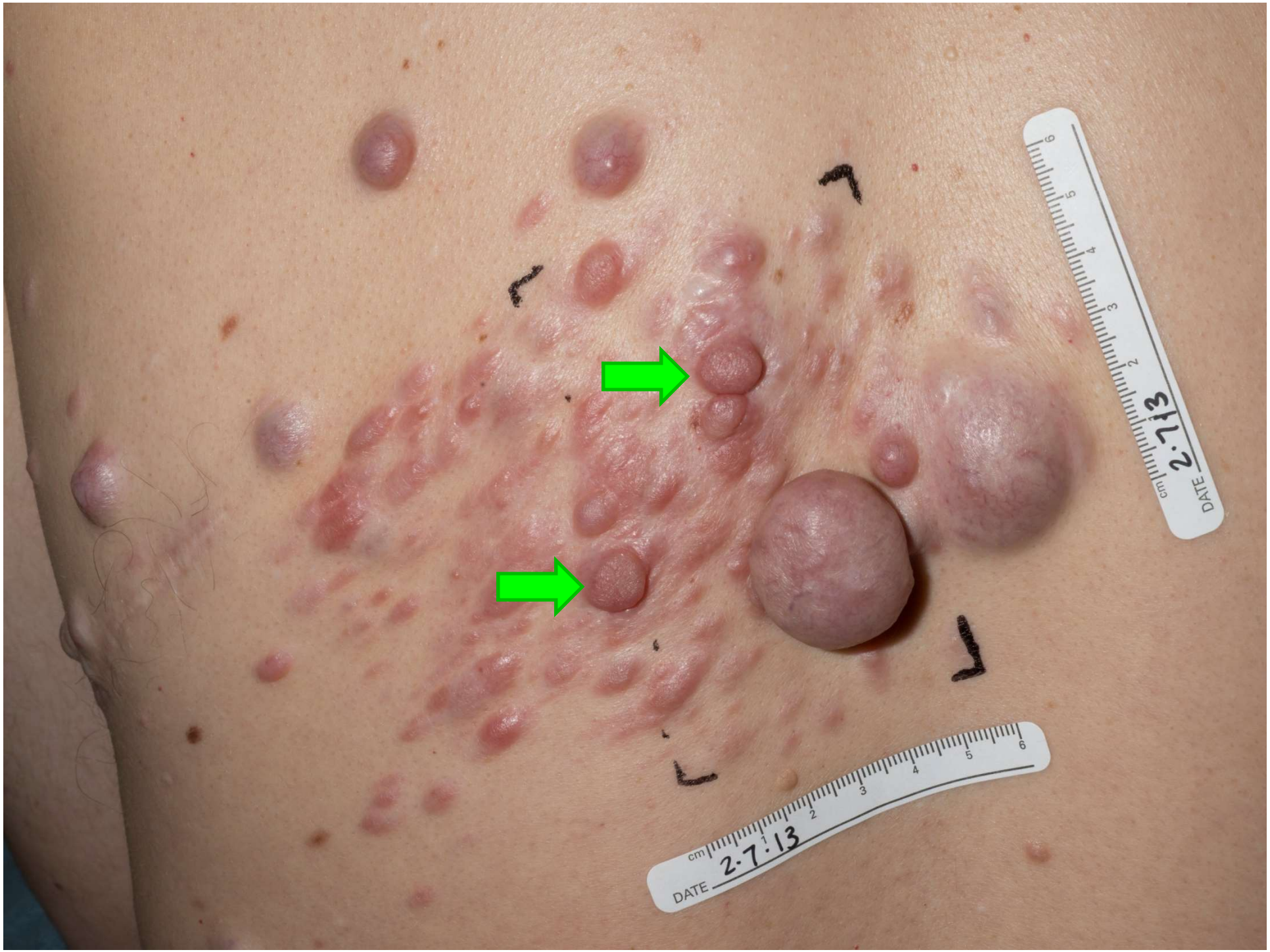


J Med Gen 45:2008
Am J Hum Gen 76:2005
Cancer Cell 2:2002

Brother



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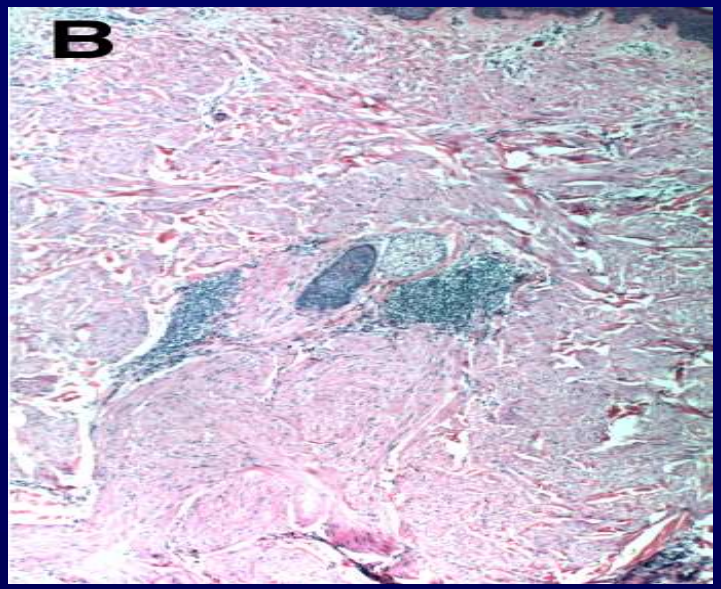
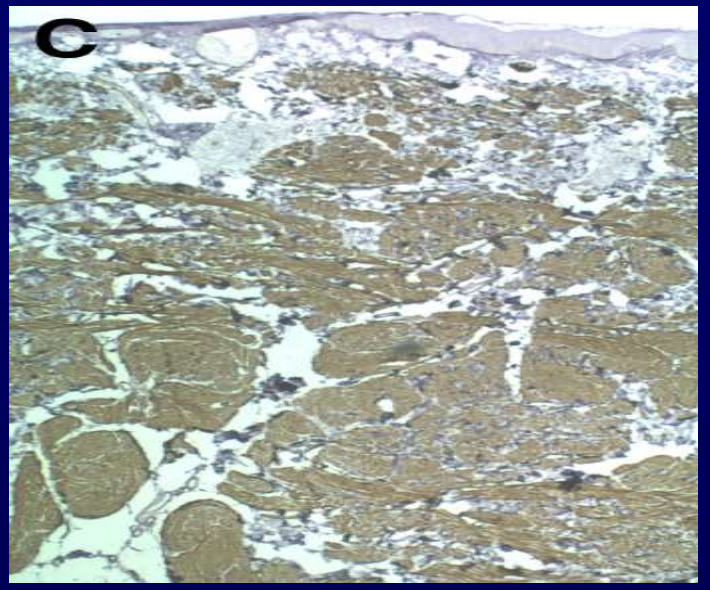




*Inherited susceptibility to uterine leiomyomas and renal cancer. (PNAS, 2001)

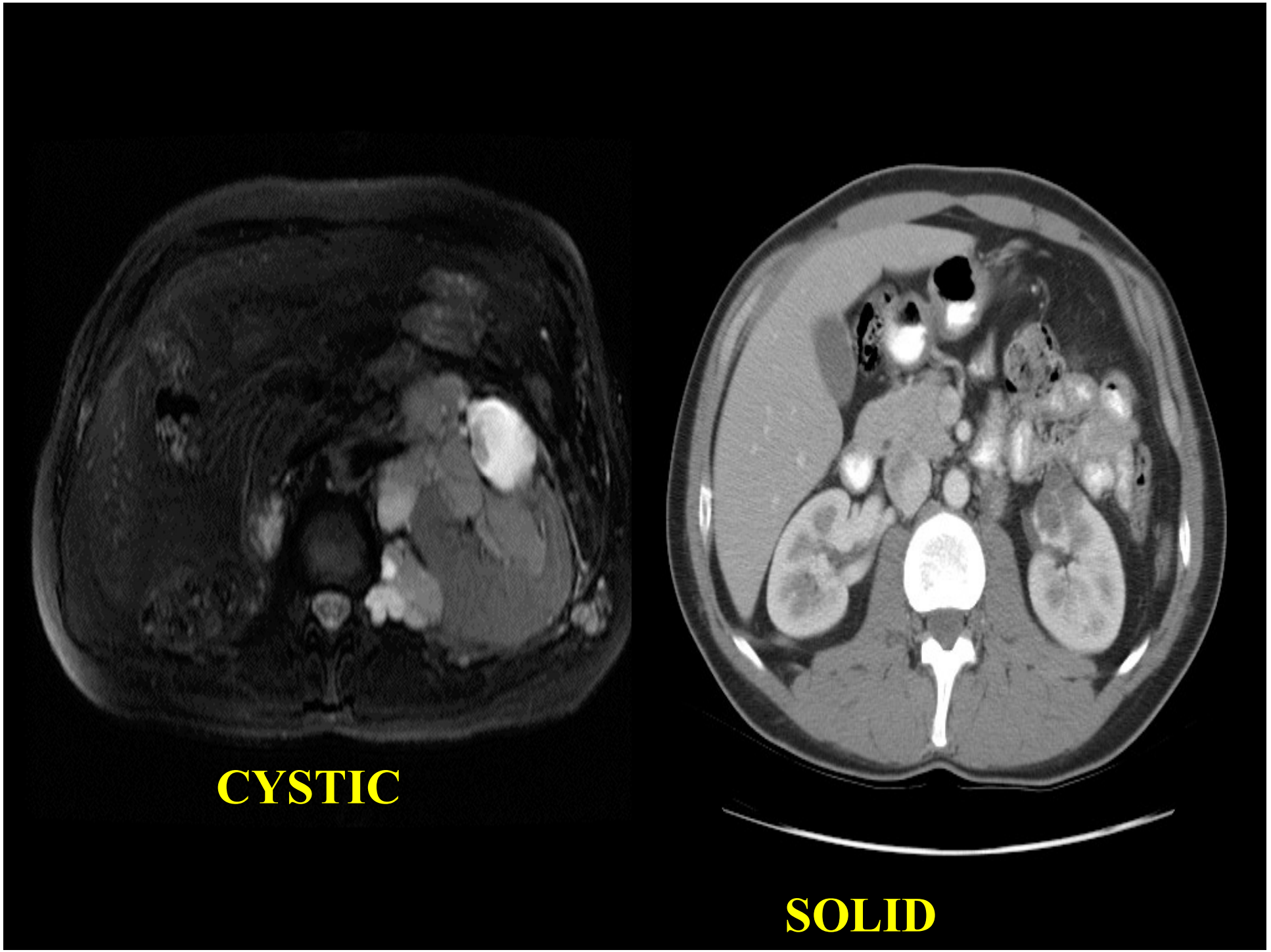
*Familial Cutaneous Leiomyomatosis Associated with RCC. (Am J Pathol 2001).

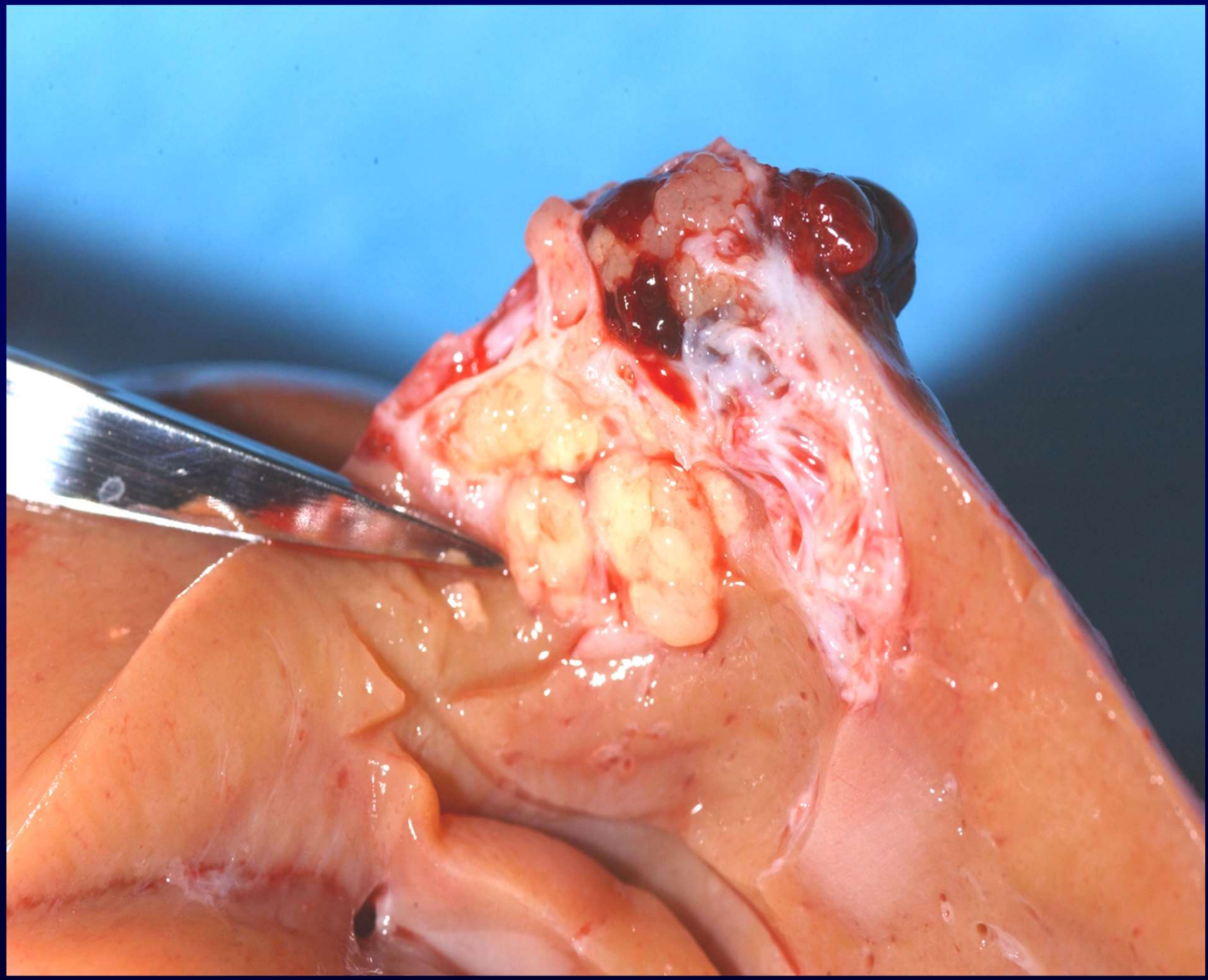
*Germ line mutations in the fumarate hydratase gene at 1q43. British J Cancer . (2002)

Hereditary Leiomyomatosis Renal Cell Ca

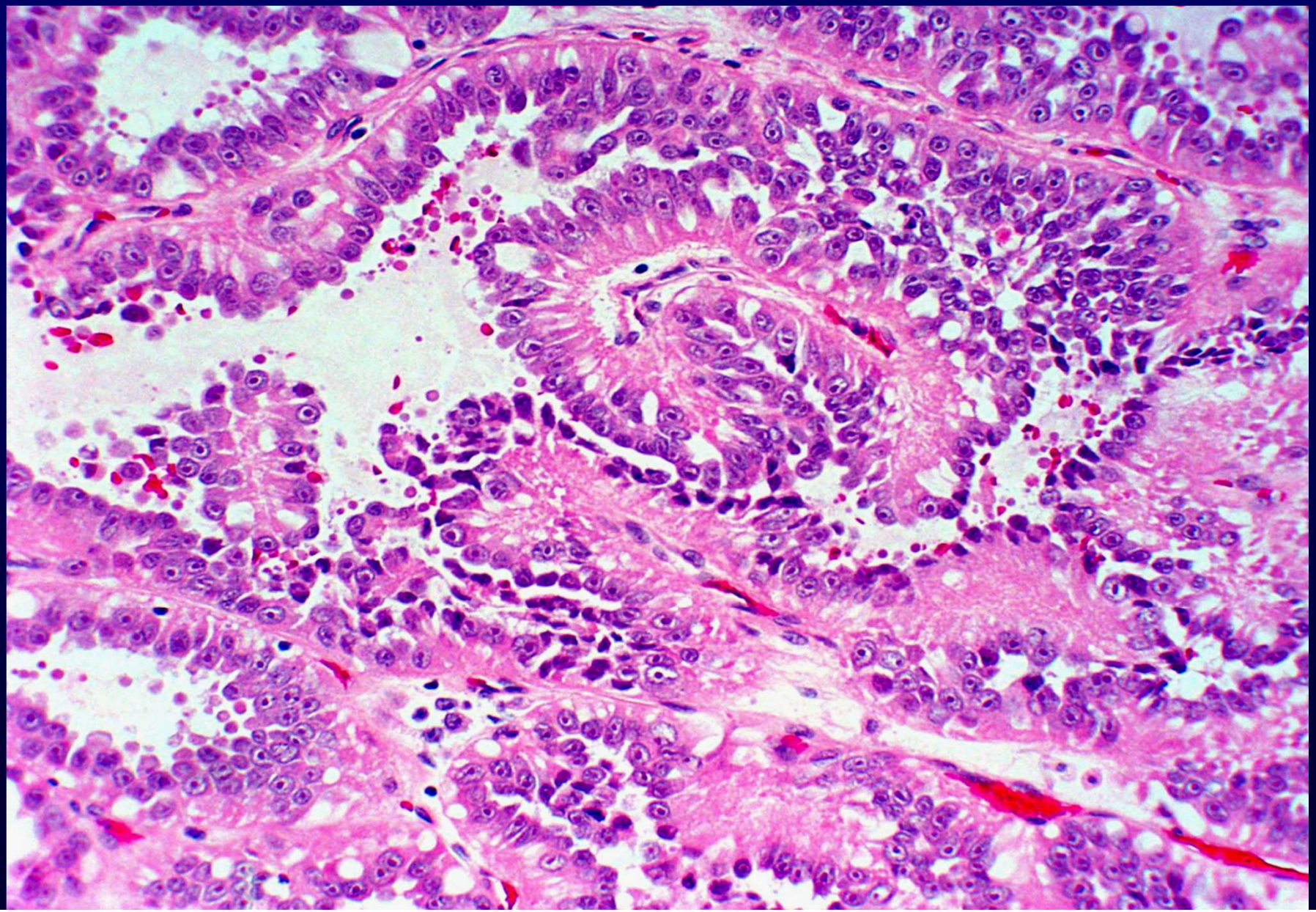
HLRCC

- Hereditary Leiomyomatosis and Renal Cell Carcinoma Syndrome (HLRCC) has been recently described (Launonen *et al*, 2001).
- HLRCC is inherited as an autosomal dominant condition with incomplete phenotype penetrance.
- It is characterized by predisposition to uterine and skin leiomyomas and renal cell carcinoma.
- Germline mutations in fumarate hydratase (FH, 1q42.3-q43) gene have been found to predispose this syndrome (The Multiple Leiomyoma Consortium, 2002).


CYSTIC
SOLID

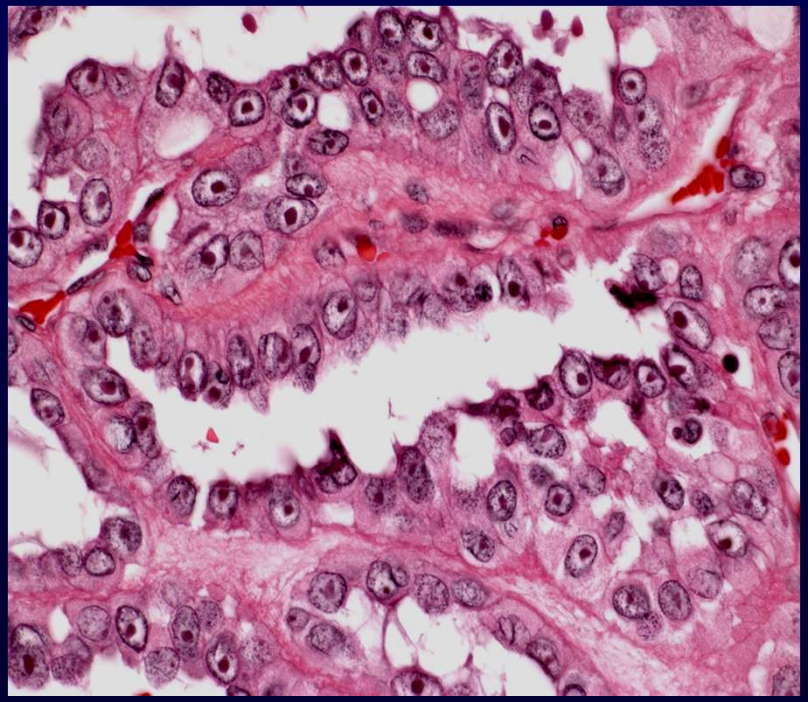


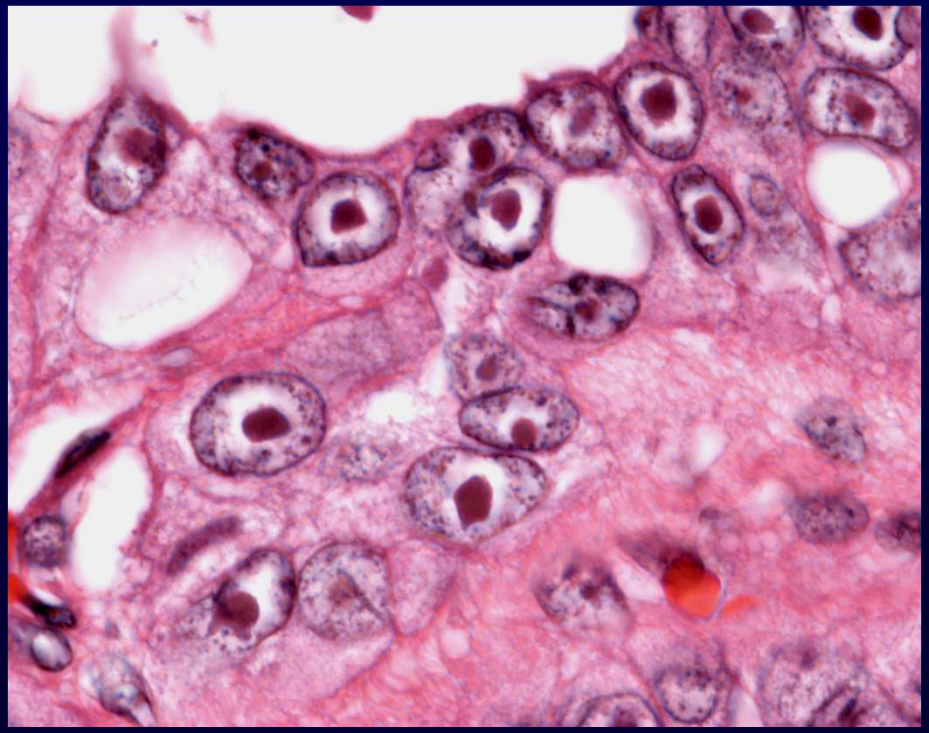
- 21 y.o. female, stage IV RCC



Kidney Tumors in HLRCC

- Predominantly Unilateral Tumors (2)
- Histology
  - Wide morphologic spectrum: papillary,solid, cystic
- Metastatic at time of diagnosis
- 11/16 patients died within 5 years of diagnosis





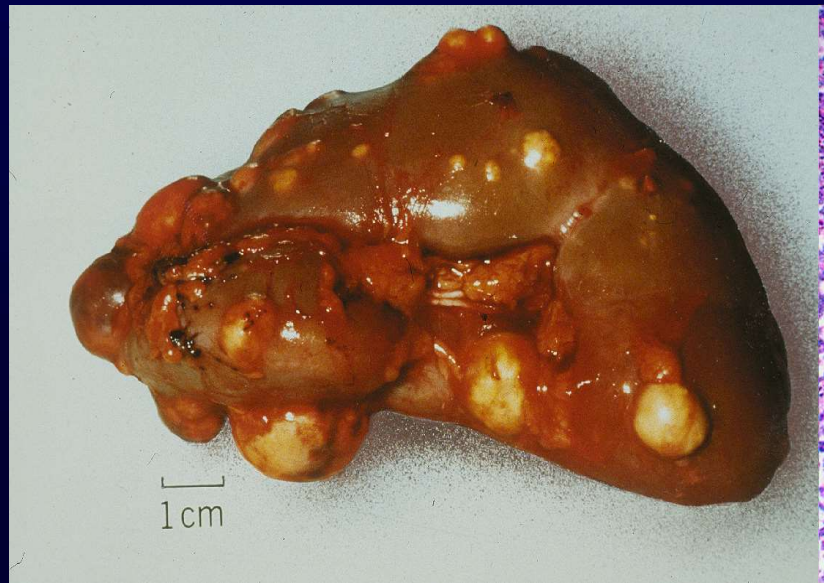
Papillary renal cell carcinoma: a clinicopathologic and immunohistochemical study of 105 tumors.

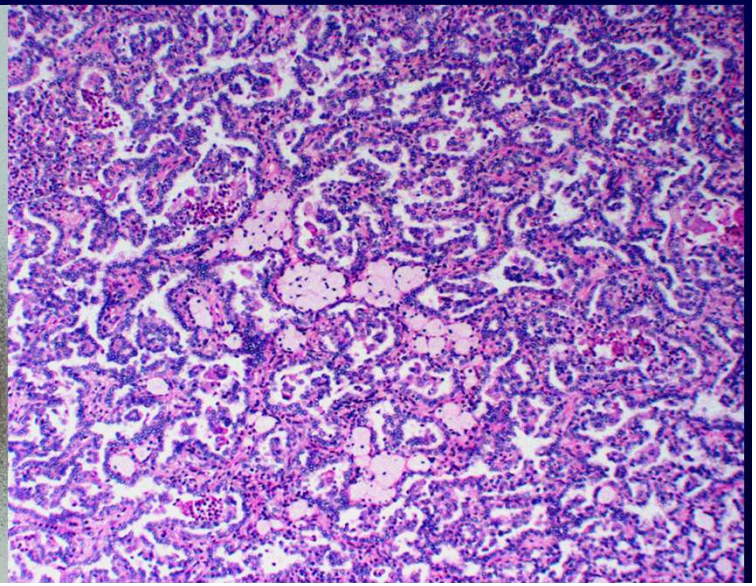
Delahunt B1, Eble JN. Mod Pathol. 1997 Jun;10(6):537-44.

Type 1 (67 tumors) consisted of papillae and tubular structures covered by small cells with pale cytoplasm and characterized by small oval nuclei with inconspicuous nucleoli, frequent glomeruloid papillae, papillary edema, foamy macrophages in papillary cores, and psammoma bodies. Type 2 (38 tumors) consisted of papillae covered by large cells with abundant eosinophilic cytoplasm and characterized by pseudostratification and large spherical nuclei with prominent nucleoli, glomeruloid papillae, psammoma bodies, edematous papillae, and foamy macrophages in papillary cores are uncommon. Type 2 tumors were larger, more common in patients younger than age 40, and more frequently Stages 3 or 4 than were Type 1 tumors. Pseudocapsules were common in both and often were infiltrated by carcinoma. Sarcomatoid foci were found in five tumors. Eleven were stage T1, 54 T2, 23 T3, and 12 T4. Reaction for cytokeratin 7 was strong or moderate in 48 of 61 Type 1 tumors, and reaction was null in 24 of 30 Type 2 tumors. No tumor reacted with U. europaeus lectin.

HPRC Pap RCC type I

- Autosomal dominant trait
- Usually late onset (50 years)
- Multiple bilateral lesions of variable sizes.
- Mutations of the MET oncogene (codes for a tyrosine kinase receptor)
- Hereditary and sporadic
- Trisomies in chromosomes 7, 17





Papillary type 2

HLRCC

Collecting Duct

Papillary RCC with eosinophilic features

Papillary Clear cell

Papillary, non type 1 or 2

Papillary, unclassified

TFE3

THE CANCER GENOME ATLAS

:

Comprehensive Molecular Characterization of Papillary Renal-Cell Carcinoma

The Cancer Genome Atlas Research Network

N Engl J Med 2016; 374:135-145 January 14, 2016DOI: 10.1056/

BACKGROUND

Papillary renal-cell carcinoma, which accounts for 15 to 20% of renal-cell carcinomas, is a heterogeneous disease that consists of various types of renal cancer, including tumors with indolent, multifocal presentation and solitary tumors with an aggressive, highly lethal phenotype. Little is known about the genetic basis of sporadic papillary renal-cell carcinoma, and no effective forms of therapy for advanced disease exist.

Results: Type 1 and type 2 papillary renal-cell carcinomas were shown to be clinically and biologically distinct. Alterations in the MET pathway were associated with type 1, and activation of the NRF2-ARE pathway was associated with type ognosis. Furthermore, type 2 papillary renal-cell carcinoma consisted of at least three subtypes based on molecular and phenotypic features.

CONCLUSIONS

Type 1 and type 2 papillary renal-cell carcinomas were shown to be clinically and biologically distinct. Alterations in the MET pathway were associated with type 1, and activation of the NRF2-ARE pathway was associated with type 2; *CDKN2A* loss and CIMP in type 2 conveyed a poor prognosis. Furthermore, type 2 papillary renal-cell carcinoma consisted of at least three subtypes based on molecular and phenotypic features.

PAPILLARY TUMORS MUST BE SUBCLASSIFIED

Hereditary syndromes

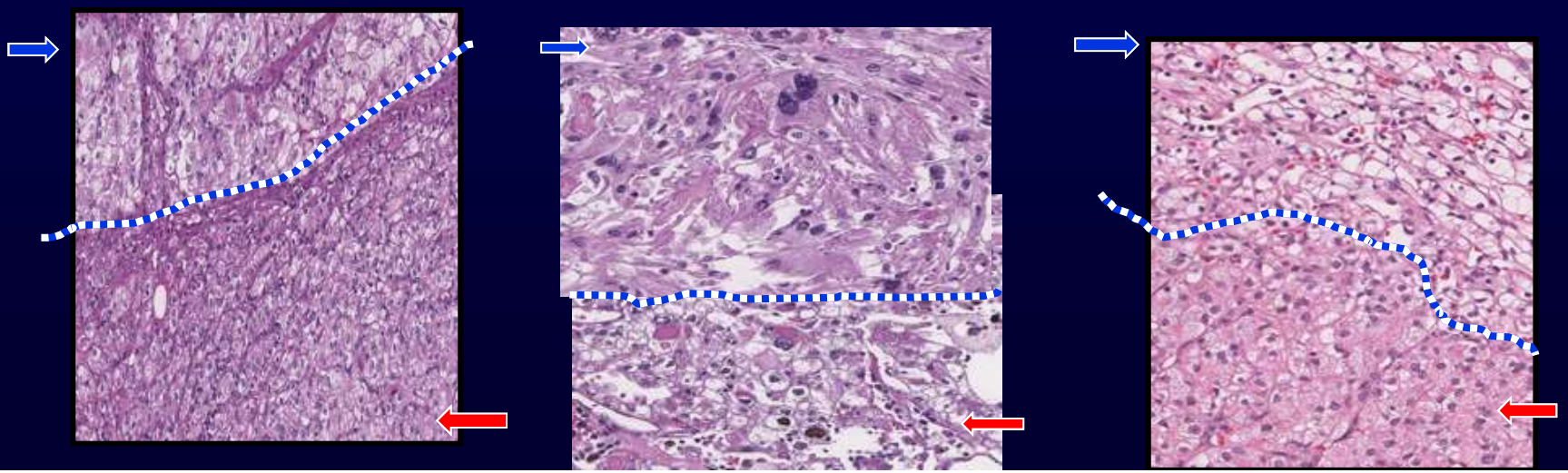
- Von Hippel-Lindau
- Tuberous sclerosis
- Syndromes associated to Wilm's

Kidney cancer is a complex disease

- Von Hippel-Lindau disease
- Hereditary Papillary type I
- Birt-Hubb-Dube
- HLRCC
- SDHB
- Tuberous Sclerosis
- Familial oncocytoma
- Mixed epithelial stromal
- Syndromes associated
- with Wilm's

Intratumoral Heterogeneity in Kidney Cancer

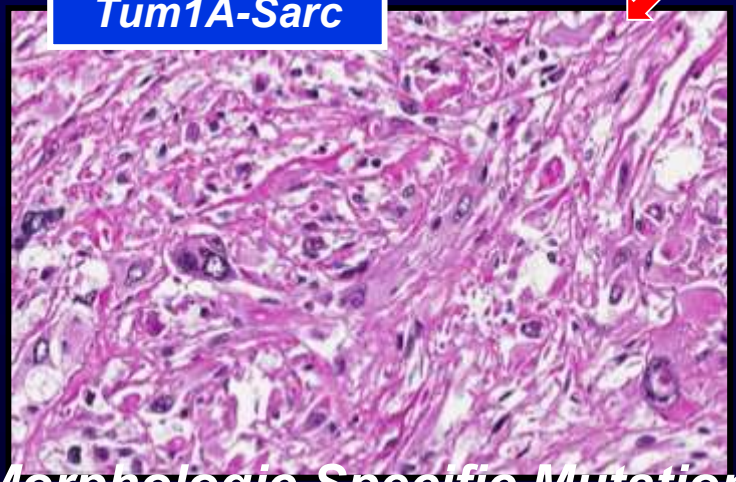
- *Intratumoral heterogeneity (ITH) in kidney cancer has been previously demonstrated at the genetic level from random sampling of tumors (Gerlinger et al., 2012).*
- ***The behavior of kidney cancer can be unpredictable, irrelevant of stage or grade; this may relate to ITH.***
- ***Single tumors can demonstrate different morphologic patterns that may reflect the presence of ITH (e.g. regions highlighted by white and red arrows in three examples).***
- ***Identification of ITH may have great implications for patient prognosis, the development of new molecular targets and effective treatment regimes.***



Genetic Intratumoral Heterogeneity Correlates with Differing Tumor Morphologies

Clear Cell RCC Tumor 1 demonstrates two distinct morphologies,) and).

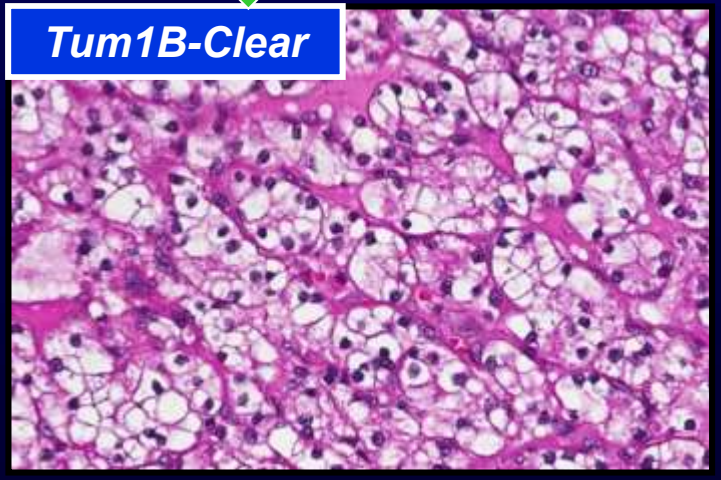
Tum1A-Sarc



Morphologic Specific Mutation TP53, CCND2, SYNE1

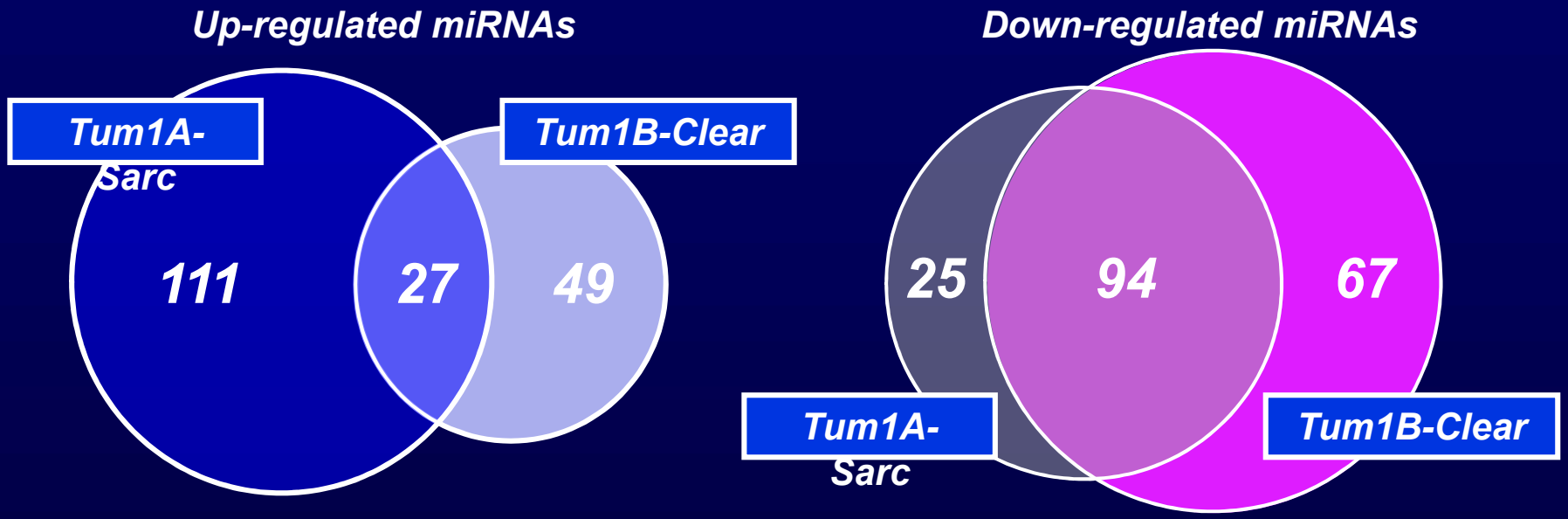
Shared Mutation NF1 SETD2 BTK

Tum1B-Clear



Morphologic Specific Mutation ARID1A

miRNA Expression Heterogeneity Observed with Different Tumor Morphologies

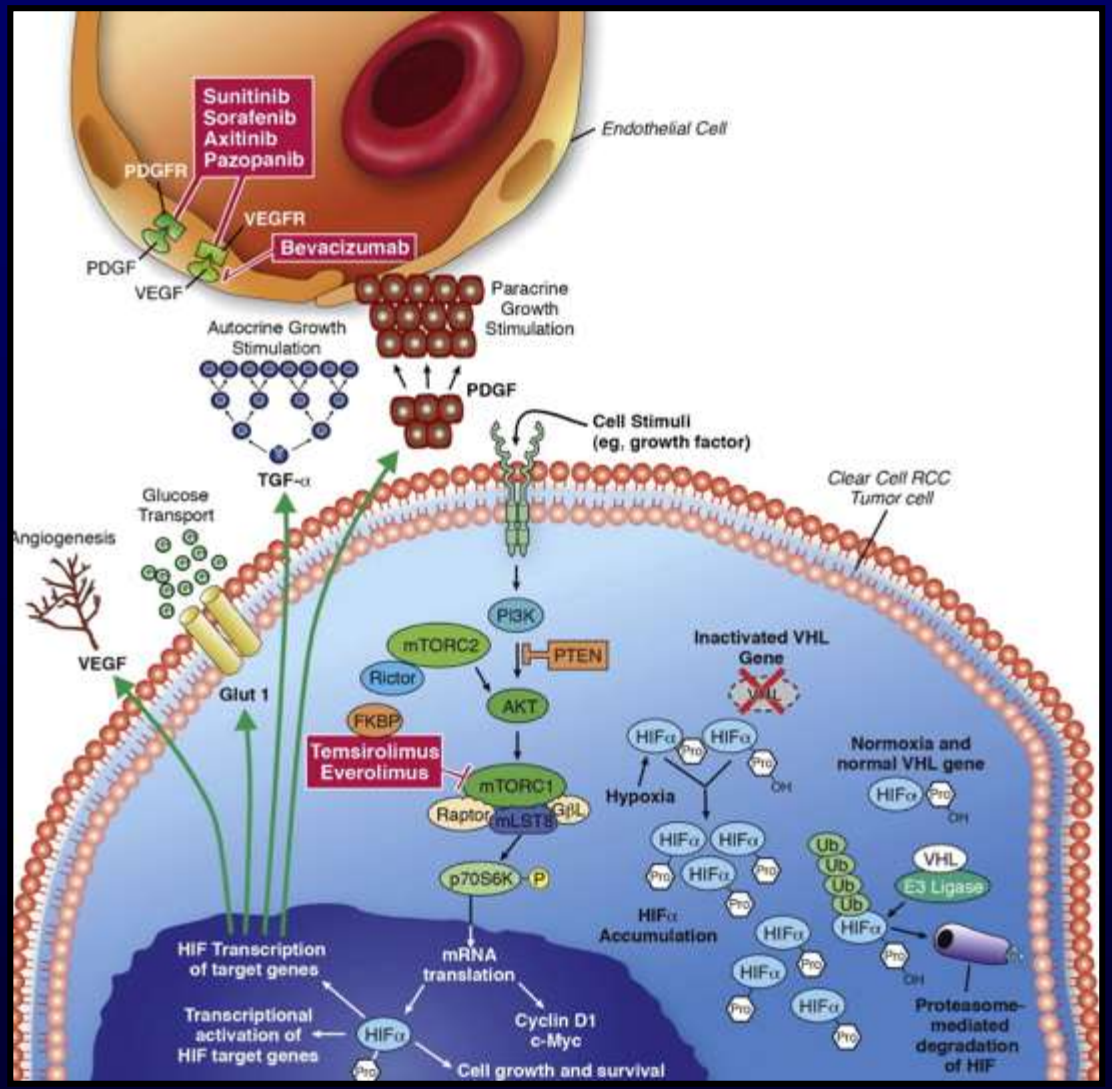


- *Tumor sections with differing morphologies demonstrated significant differences in their miRNA profile.*
- *Interestingly, miR-210 was one of the few miRNAs that was upregulated in all the specimens and represents part of the hypoxic phenotype in clear cell RCC.*
- *High levels of miR-210 are correlated with adverse prognostic factors such as high FNG and the presence of LN metastasis and it could be used as a biomarkers*

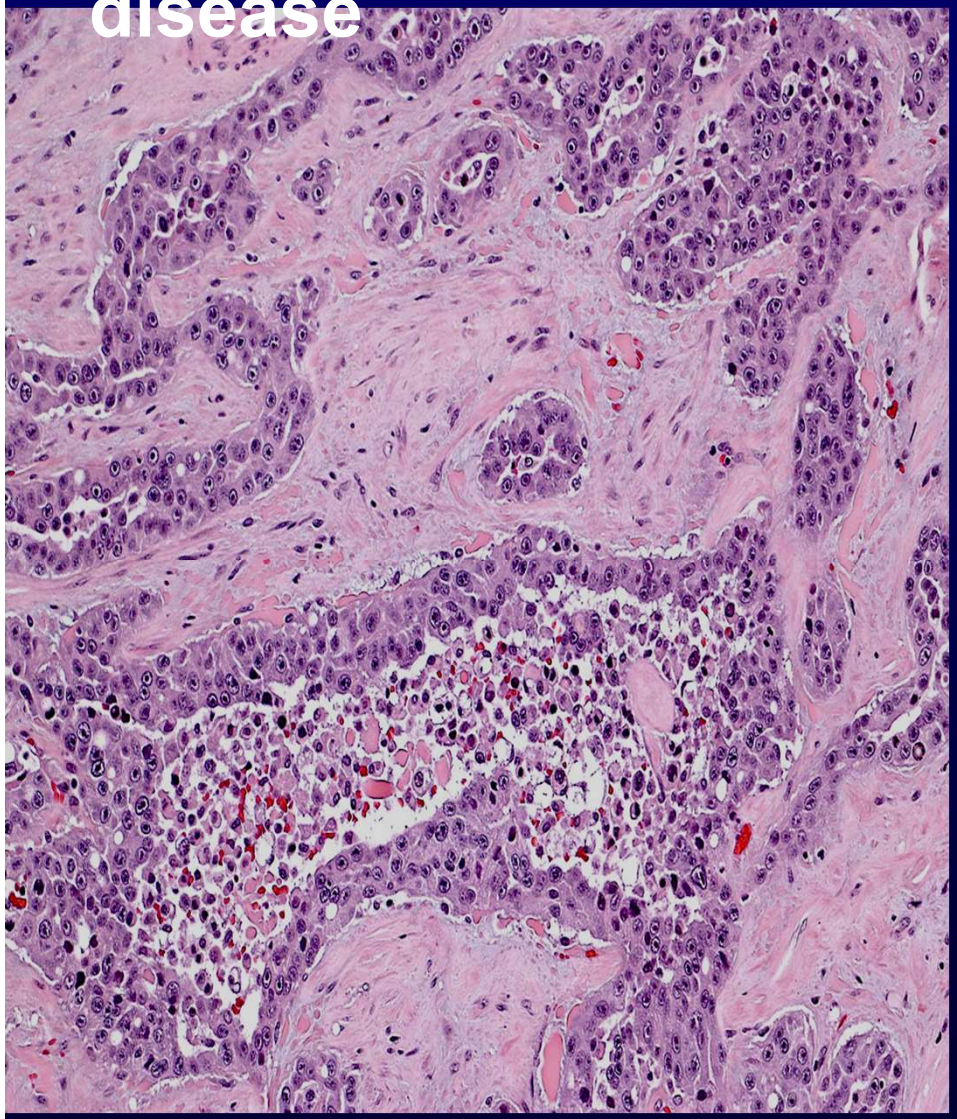
Valera, Walter, Linehan, Merino MJ. Cancer 2011

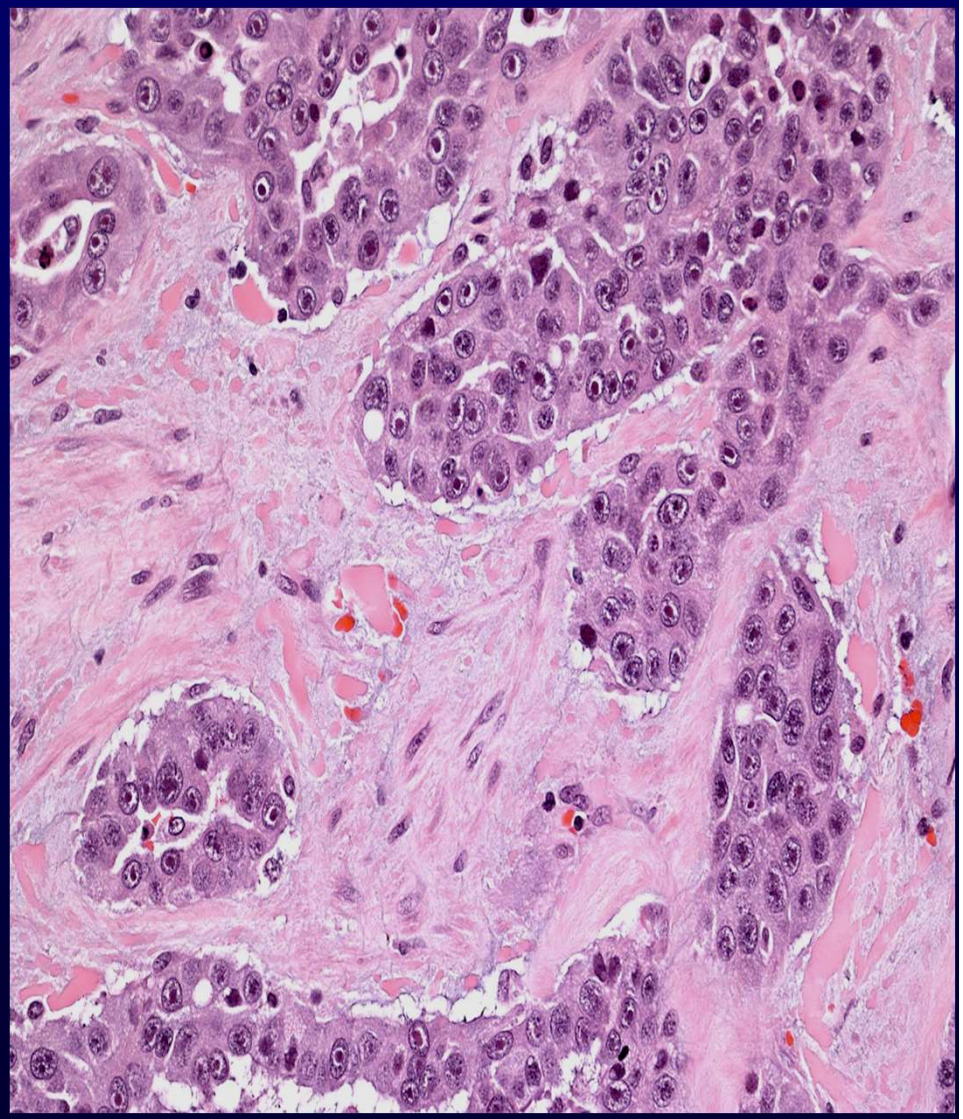
Clinical Relevance of Intratumoral Heterogeneity

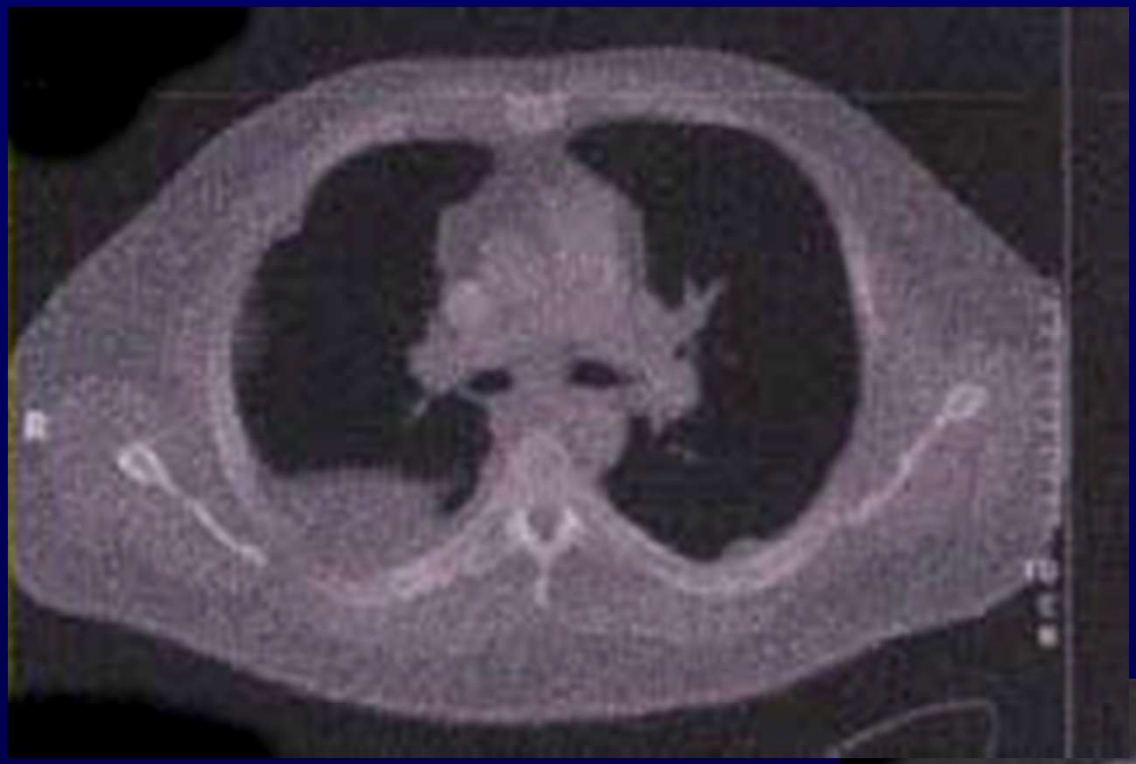
- ***Several targeted therapies exist for the treatment of kidney cancer, but these may only target part of a tumor with other regions immune to therapy.***
- ***Identification of biomarkers that are either based on the genetics of the tumor, some form of molecular biomarker (such as miRNA) or pathological morphology could aid in diagnosing multiple different subtypes of a tumor and suggest a series of therapeutic regimes to treat all subtypes of tumor present.***



38 year old man with disseminated disease



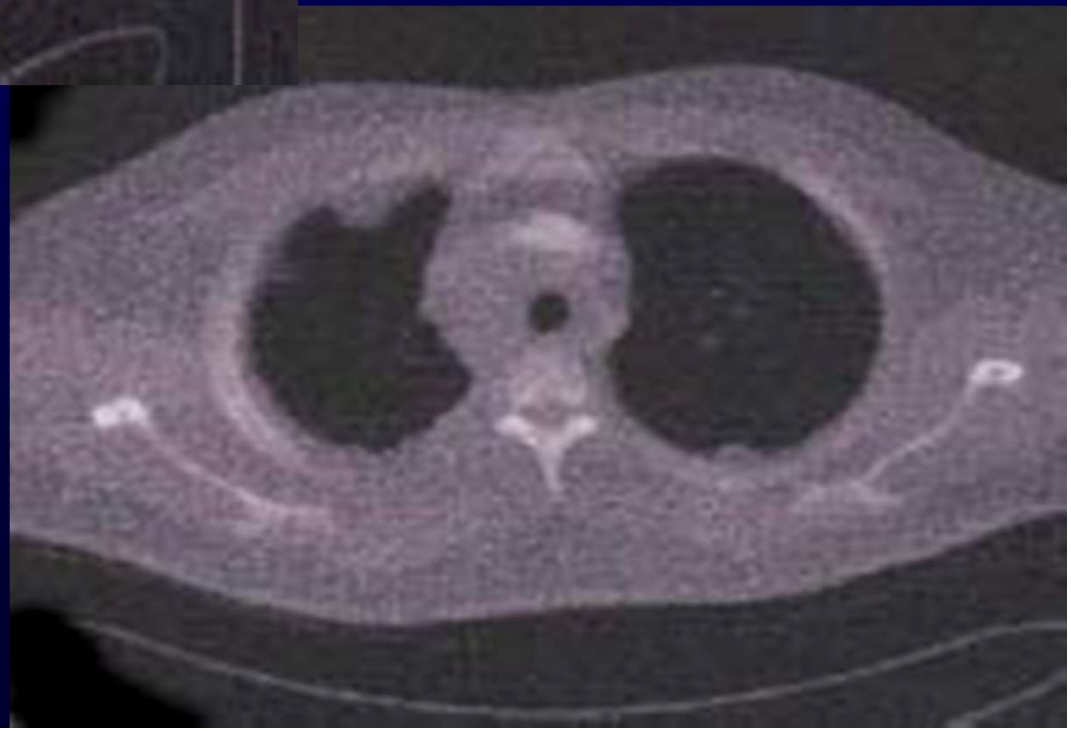




MAY

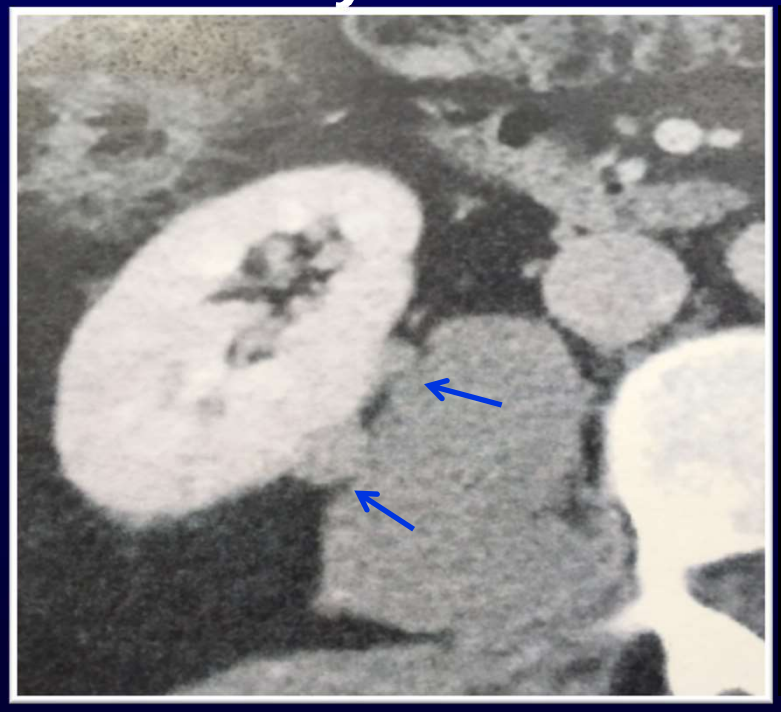
Received therapy with Iressa (targets the receptor of the tyrosine Kinase activity of the EGF receptor

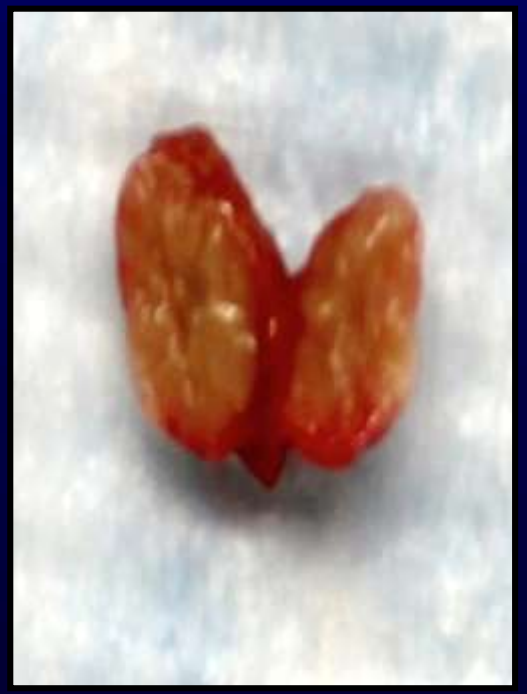
August

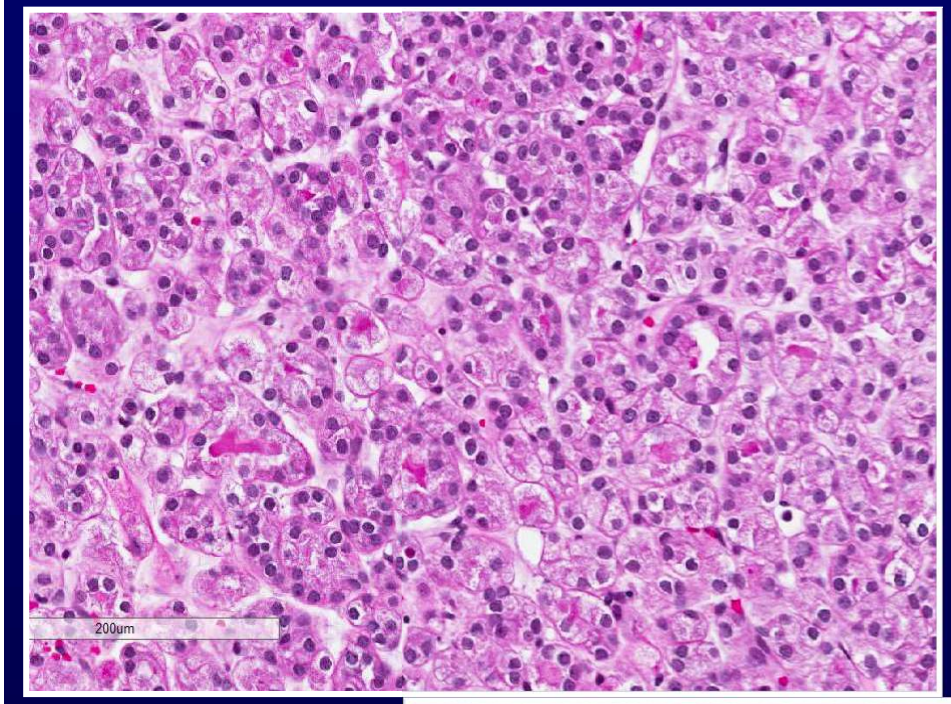


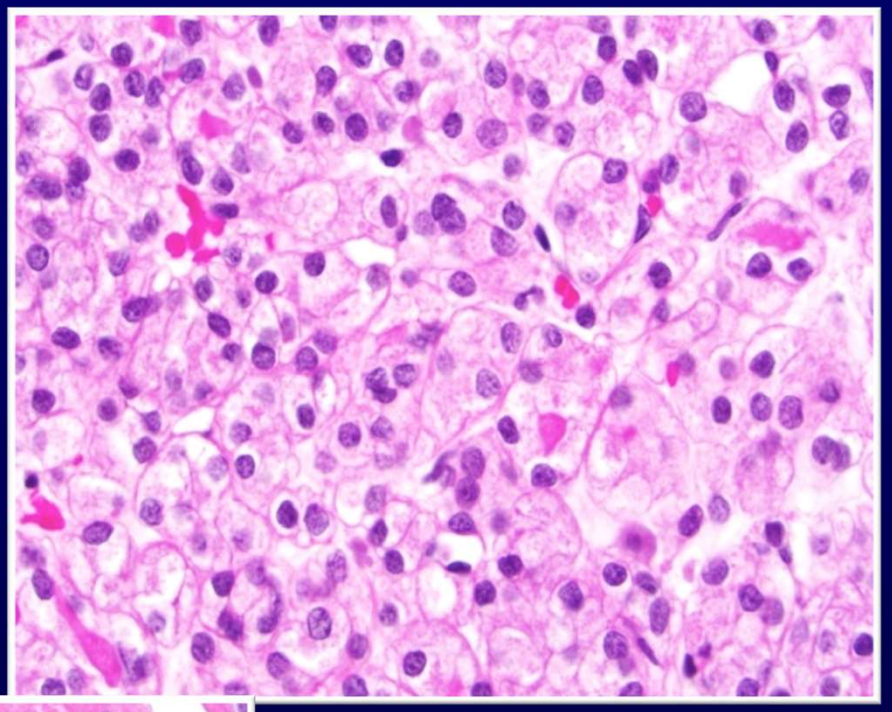
Identification of a New and Distinct Kidney Cancer

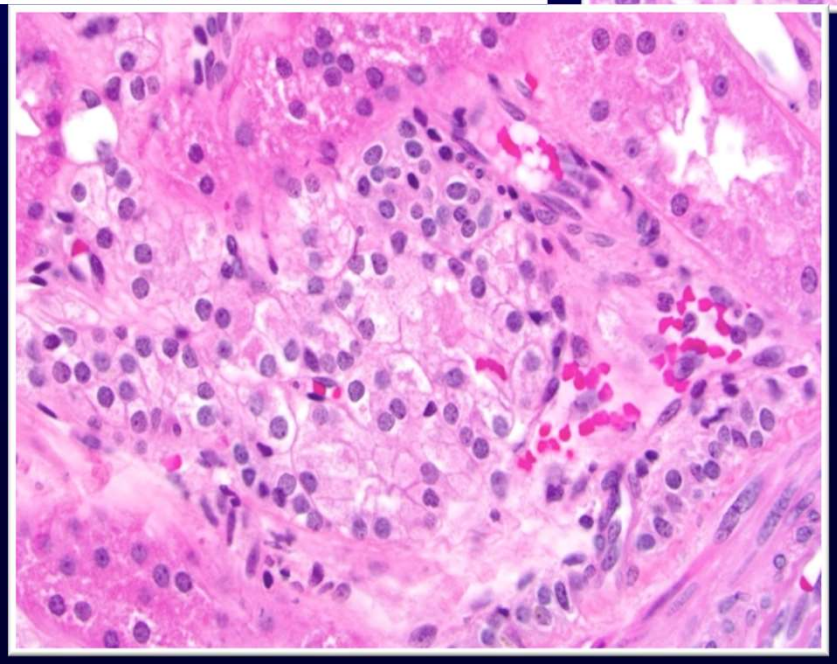
- *A 43 year old male patient was evaluated for hydronephrosis on the right kidney. CT scans revealed multiple tumors (6 in total) in his left kidney. Family history was non contributory*







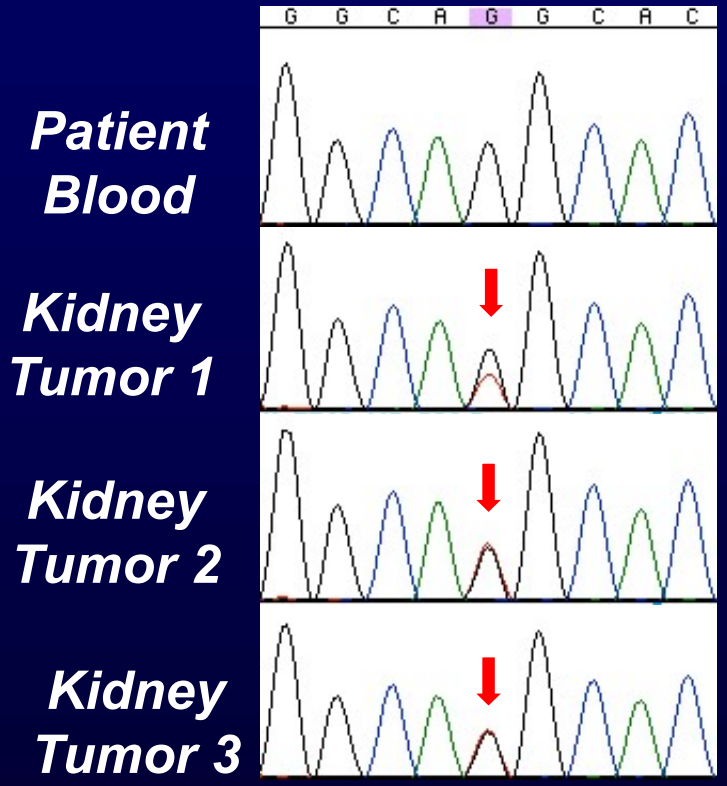




NEW Renal Cancer

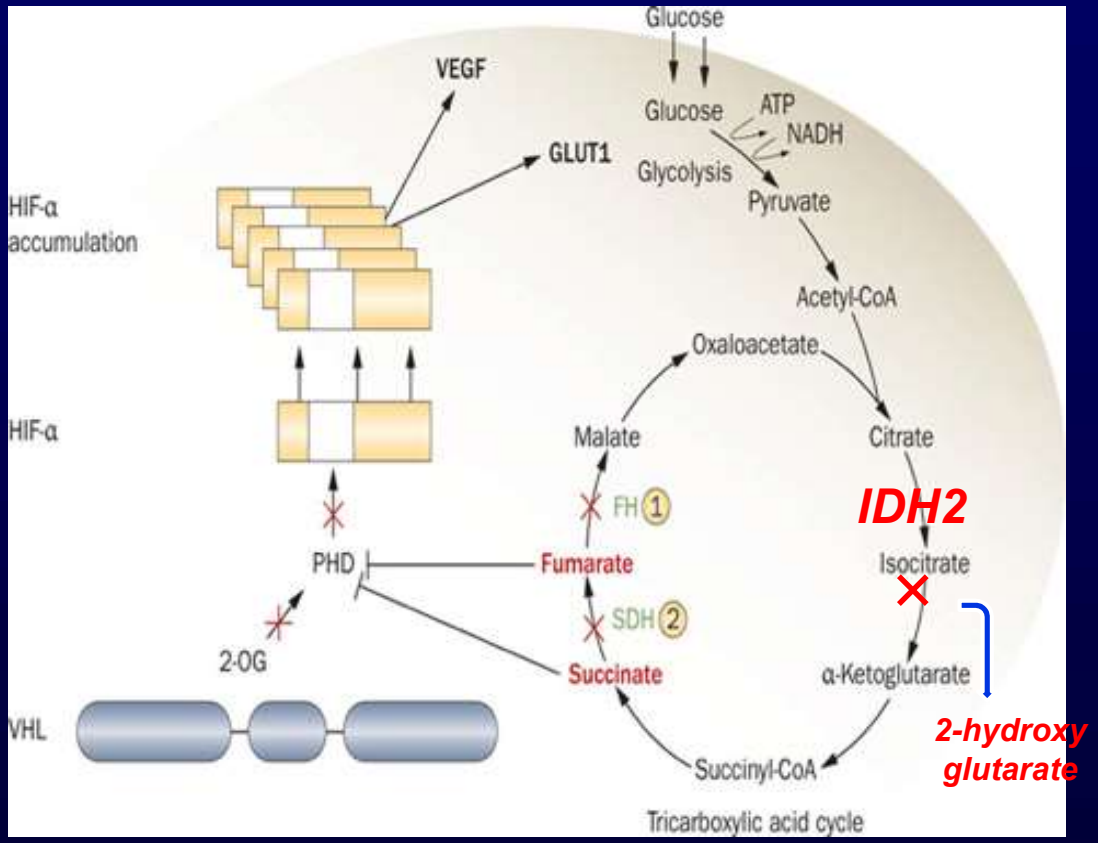
Identification of a Novel Histologically Distinct Kidney Cancer

- ***Mutation analysis of > 300 cancer related genes demonstrated no somatic mutations in the commonly mutated RCC genes.***
- ***<u>An IDH2 R172M</u> activating mutation was observed and was present in all three assessed tumors.***



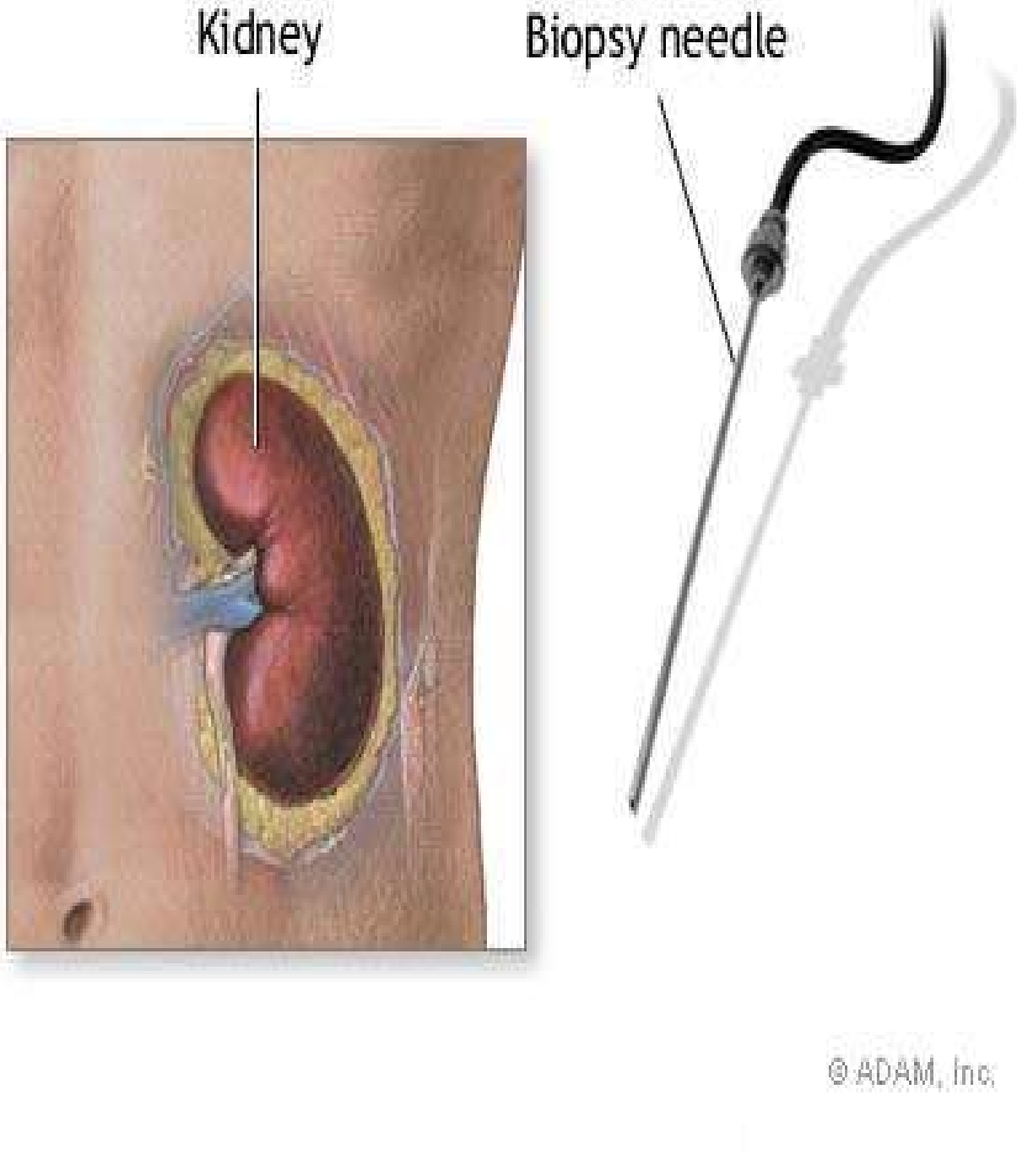
Potential Role of the IDH2 R172M Activating Mutation in Kidney Cancer

- ***Kidney cancer is associated with two other mutations of Krebs cycle genes, fumarate hydratase (1) and succinate dehydrogenase (2).***
- ***Mutation of these genes result in increased levels of the fumarate and succinate metabolites that have been shown to inhibit alpha-ketoglutarate dependent enzymes, such as the prolyl hydroxylases (PHDs).***
- ***Loss of IDH2 also increases the levels of a metabolite, 2-hydroxyglutarate, that acts in a similar manner. This is one of the areas of our future research.***



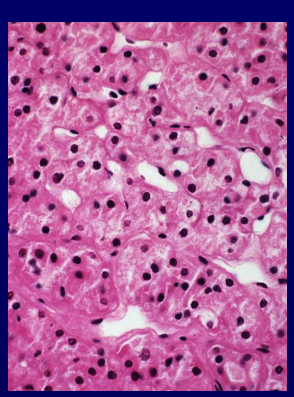
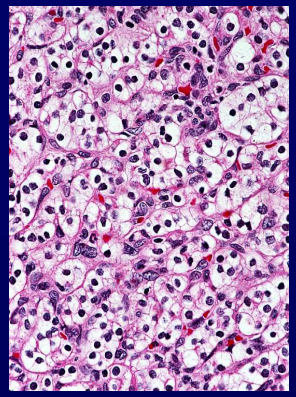
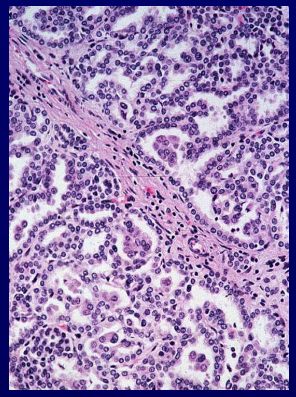
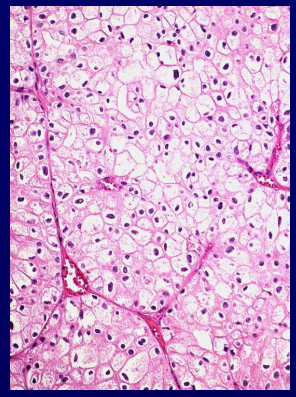
New and future approaches to treatment

- **Genetics:** For example, problems with the VHL tumor suppressor gene that allows genes such as HIF to be activated and drives the cell to become malignant.
- High-intensity focused ultrasound (HIFU) is a fairly new technique that is now being studied for use in kidney cancer. It involves pointing very focused ultrasound beams from outside the body to destroy the tumor
- ***Immunotherapy*.** Cancer cells use natural pathways in the body to help avoid being detected and destroyed by the immune system. For example, they often have a protein called PD-L1 on their surface that helps them evade the immune system. New drugs that block the PD-L1 protein, or the corresponding PD-1 protein on immune cells called T cells, can help the immune system recognize the cancer cells and attack them. (Nivolumab)
- Vaccines. To stimulate the immune system

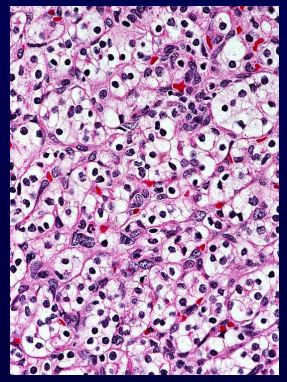
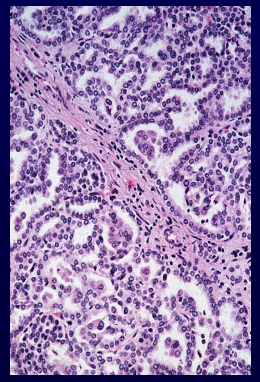
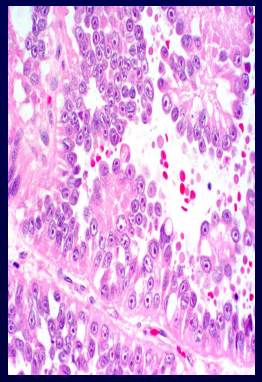
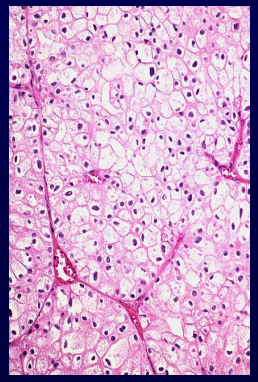
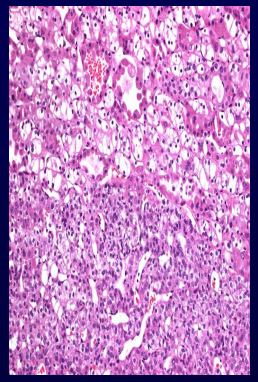
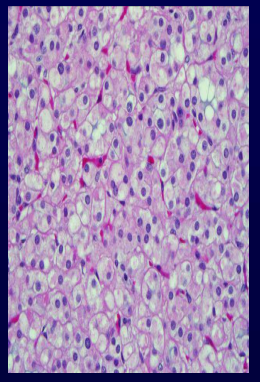
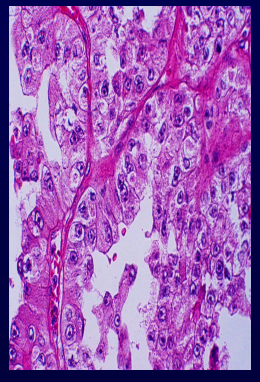

Kidney
Biopsy needle
A
B
C
© ADAM, Inc.

Partial Nephrectomy

In the beginning….Kidney Cancer was simple……

Now we need new classifications based in Genetic findings

Clear Cell	*Pap Type 1*	*HLRCC*	*Chromo*	*Hybrid*	*SDHB*	*TFE3*
H → VHL	*Met*	*FH*	*BHD*	*BHD*		
S → VHL	*Met*	*FH?*	*BHD?*			

This could be my family

SE HACE CAMINO AL ANDAR

Antonio Machado

Gracias amigos

- Dr. Carlos Monteagudo
- Dra. Josefina San Juan
- Dr. Miguel Perez
- Dr. Elias Campo
- Dr. Pedro Fernandez
- Dr. Julian Sanz
- Dr. Ernesto Moro
- Dr. Angel Panizo
- Dra. Ana Echegoyen
- Dr. Luis Vicioso

Dr. Jose Palacios
Dra. Carmen Garcia M
Dra. Elisa Muñoz
Dr. Pablo Canatta
Dr. Rafael Navas
Dr. Xavier San Juan
Dra. Roxana Sanchez
Dr. Daniel Val
Dra. Rosita Guarch
Dr. Alberto de La Cruz


Gracias Dr. Llombart

Why do we have to remember Hereditary Renal syndromes?

- Because Morphology is the only way to recognize these syndromes and diagnose them.
- Because once you make the diagnosis, you will be guiding the surgeon's and the oncologist's hand towards modalities of therapy that will benefit the patient.
- But more important……because you can save life's when patients are recognized and members of the family screened and early lesions identified.

Hereditary syndromes

- Von Hippel-Lindau
- Tuberous sclerosis
- Syndromes associated to Wilm's

50 families with KC

- Von Hippel-Lindau disease
- Hereditary Papillary type I
- Birt-Hubb-Dube
- HLRCC
- Tuberous Sclerosis
- SDHB
- Familial Oncocytomas
- MEST
- Syndromes associated with Wilm's