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# How to Sustain Research Productivity and Funding

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# Disclosures

- None

# Overview

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- Surgeons as Scientists
- Align Clinical Interests with Research Interests
- Importance of Team Science
- The New Triple Threat…The Department
- Tissue as a Resource

# Surgeons as Scientists

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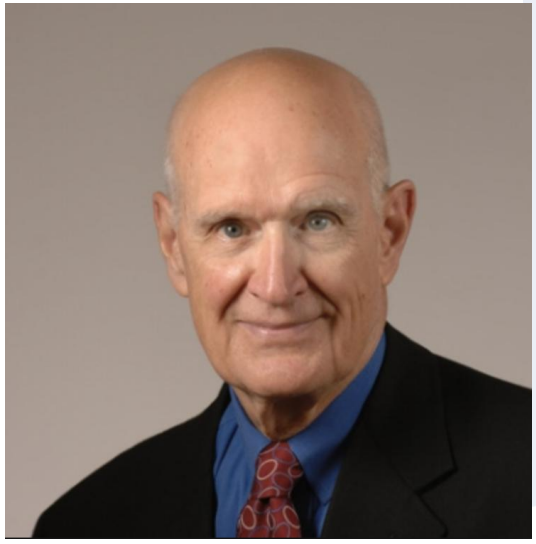
# Surgeons as Scientists

# Align Clinical Interests with Research Interests

# Alignment

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REVIEWS

## Evaluation and management of pancreatic lesions in patients with von Hippel–Lindau disease

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**Abstract** | von Hippel–Lindau (VHL) disease is a heritable cancer-predisposition syndrome with multiorgan involvement. Pancreatic lesions are detected in approximately two-thirds of patients with VHL disease at some point during their lifetime. In these patients, cystic pancreatic lesions are almost exclusively benign and, unless symptomatic, do not require surgical or endoscopic intervention; however, solid pancreatic lesions can often be recognized through imaging screens, and are commonly found to be nonfunctioning pancreatic neuroendocrine tumours (pNETs) with malignant potential. The natural history of these VHL-associated pNETs is variable, and lacks clinical or imaging features that predict disease progression or metastatic potential, and generally needs to be managed more conservatively than their sporadic counterparts. Treatment options for such lesions, which range from active surveillance to surgical intervention, can nevertheless be associated with considerable morbidity and even mortality. Of note, although several guidelines have been established for the management of tumours associated with VHL syndrome, none of these have specifically focused on pancreatic lesions. Thus, we aim to characterize the types of pancreatic lesions associated with VHL disease and their natural history, to identify particular lesions that necessitate treatment, and to define what forms of treatment should be undertaken.

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von Hippel-Lindau disease (VHL) is an autosomal dominant, heritable cancer-predisposition syndrome with a high penetrance of cancer[1,2]. The incidence of VHL disease is reported to be one in 36,000 live births in the USA, and the prevalence of the disease is around one in 100,000 inhabitants in the USA[1,2]. The disease is caused by germline mutations in the *VHL* tumour-suppressor gene, which is located on the short arm of chromosome 3 (3p25–3p26)[3,4]. Germline mutations associated with the disease are distributed widely across the coding sequence of *VHL*[5]. The phenotypic presentations of VHL disease include retinal and central nervous system (CNS) haemangioblastomas, endolymphatic sac tumours, cystadenomas of the epididymis, pheochromocytomas, renal cysts, and renal-cell carcinomas (RCCs), as well as pancreatic neuroendocrine tumours (pNETs) and pancreatic cysts[1,3]. Morbidity and mortality of patients with VHL disease is most commonly a result of CNS haemangioblastoma and/or RCC, which are observed in 60–80% and 24–45% of patients, respectively[1,6]. Nevertheless, pancreatic lesions associated with VHL disease can result in considerable morbidity, and unless patients are managed correctly, such lesions can result in lethal metastatic disease.

Several guidelines for the management of VHL disease have been disseminated; however, these have not focused specifically on interventions for pancreatic involvement[7–9]. Thus, the purpose of this Review is to provide an overview of state-of-the-art techniques for evaluating and treating patients with VHL-associated pancreatic lesions. This objective is particularly important because a broad spectrum of health-care providers — including internists, gastroenterologists, endocrinologists, medical oncologists, radiologists and surgeons — are involved in the management of such patients. We also propose what we believe, based on the currently available evidence (BOX 1), to be the optimal management strategies for pancreatic lesions that arise in patients with VHL disease.

### Epidemiology of pancreatic involvement

Pancreatic lesions in patients with VHL disease can manifest as solid tumours (most commonly pNETs), simple pancreatic cysts, or pancreatic serous cystadenomas[3].





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## Deciphering von Hippel-Lindau (VHL/Vhl)-Associated Pancreatic Manifestations by Inactivating *Vhl* in Specific Pancreatic Cell Populations

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**Abstract**

The von Hippel-Lindau (VHL) syndrome is a pleomorphic familial disease characterized by the development of highly vascularized tumors, such as hemangioblastomas of the central nervous system, pheochromocytomas, renal cell carcinomas, cysts and neuroendocrine tumors of the pancreas. Up to 75% of VHL patients are affected by VHL-associated pancreatic lesions; however, very few reports in the published literature have described the cellular origins and biological roles of VHL in the pancreas. Since homozygous loss of *Vhl* in mice resulted in embryonic lethality, this study aimed to characterize the functional significance of VHL in the pancreas by conditionally inactivating *Vhl* utilizing the Cre/LoxP system. Specifically, *Vhl* was inactivated in different pancreatic cell populations distinguished by their roles during embryonic organ development and their endocrine lineage commitment. With Cre recombinase expression directed by a glucagon promoter in α-cells or an insulin promoter in β-cells, we showed that deletion of *Vhl* is dispensable for normal functions of the endocrine pancreas. In addition, deficiency of VHL protein (pVHL) in terminally differentiated α-cells or β-cells is insufficient to induce pancreatic neuroendocrine tumorigenesis. Most significantly, we presented the first mouse model of VHL-associated pancreatic disease in mice lacking pVHL utilizing Pdx1-Cre transgenic mice to inactivate *Vhl* in pancreatic progenitor cells. The highly vascularized microcystic adenomas and hyperplastic islets that developed in Pdx1-Cre;Vhl f/f homozygous mice exhibited clinical features similar to VHL patients. Establishment of three different, cell-specific *Vhl* knockouts in the pancreas have allowed us to provide evidence suggesting that VHL is functionally important for postnatal ductal and exocrine pancreas, and that VHL-associated pancreatic lesions are likely to originate from progenitor cells, not mature endocrine cells. The novel model systems reported here will provide the basis for further functional and genetic studies to define molecular mechanisms involved in VHL-associated pancreatic diseases.

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**Competing Interests:** The authors have declared that no competing interests exist.

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### Introduction

The von Hippel-Lindau (VHL) syndrome is an autosomal, dominant inherited disorder caused by mutations in the *VHL* tumor suppressor gene. VHL patients are predisposed to develop highly vascular tumors in multiple organs, including hemangioblastomas of the retina and central nervous system (CNS), clear cell renal carcinomas, pheochromocytomas, cyst and neuroendocrine tumors in the pancreas [1]. This familial cancer syndrome is caused by germ-line mutations in the *VHL* gene, which was mapped to chromosome 3p25 by positional cloning [2]. Following Knudson's two-hit hypothesis, loss or inactivation of the remaining wildtype allele is associated with VHL tumorigenesis [3,4]. The spectrum of VHL tumors in affected families varies [5] and biochemical analysis of the *VHL* gene product has provided the molecular basis that explains the phenotype-genotype correlations evident in VHL disease [6,7,8,9].

At the molecular level, the von Hippel-Lindau protein (pVHL) is a critical factor in the oxygen sensing pathway. Under normoxic conditions, pVHL forms a multiprotein complex with E3 ubiquitin ligase that targets the α-subunits of hypoxia-inducible factor (HIFα) for degradation by the proteosome [10,11]. Under hypoxic conditions, HIFα subunits escape ubiquitin-mediated proteolysis, allowing HIFα to accumulate, translocate to the nucleus, and activate downstream targets. In subsets of VHL mutations, the lack of functional pVHL leads to accumulation of HIF, and results in the activation of HIFα target genes even in the presence of

# Alignment

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## The utility of routine transcervical thymectomy for multiple endocrine neoplasia 1-related hyperparathyroidism

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***Background.*** *Operation for multiple endocrine neoplasia (MEN)1-related hyperparathyroidism (HPT) includes a neck exploration with resection of 3.5 or 4 parathyroid glands and transcervical thymectomy (TCT). We reviewed our experience with initial operation for primary HPT to determine the outcome and utility of routine TCT.*

***Methods.*** *All patients with MEN1 who underwent initial neck exploration from 1993 to 2007 under an institutional review board-approved protocol were reviewed.*

***Results.*** *We identified 66 patients with initial operation for HPT in MEN1. In 34 patients, 4 glands were found; in 32 patients, <4 glands were found. In 2 of the 34 (6%) and 17 of the 32 (53%), intrathymic parathyroid tissue was found on permanent pathology. No thymic carcinoid tissue was found in any specimen.*

***Conclusion.*** *These data highlight the importance of performing TCT when <4 entopic parathyroid glands are found at first operation. (Surgery 2008;144:878-84.)*

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MULTIPLE ENDOCRINE NEOPLASIA TYPE 1 (MEN1) is an autosomal-dominant familial cancer syndrome with a prevalence of 1 in 10,000 to 1 in 100,000.[1] Endocrine manifestations include pituitary, parathyroid, and enteropancreatic endocrine neoplasms, as well as other rarer neoplasms.[1,2] The MEN1 gene and its protein product menin were identified in 1997; most of the mutations predicted a truncated protein product and, thus, suggest a loss of function role for MEN1 in tumorigenesis.[1] Primary hyperparathyroidism (HPT) due to parathyroid neoplasm(s) is usually the earliest and most frequent manifestation of MEN1 with a penetrance of almost 100% by age 50.[2] Parathyroid disease in MEN1 patients is multiglandular in nature secondary to its hereditary basis, and all parathyroid tissue is generally considered abnormal or at high risk for neoplasms.[3]

The recurrence rate for HPT in sporadic patients has been reported to be approximately 2%.[4] Recurrence rates in MEN1 patients are much greater, ranging from 14% to 69% depending on the operation performed, time after operation, and series.[5-8] The high recurrence rates and understanding of the underlying genetic mechanisms have led to efforts to clear as much abnormal parathyroid tissue as possible without causing permanent hypoparathyroidism.

Transcervical thymectomy (TCT) has often been performed with neck exploration in MEN1 patients and mainly at initial parathyroidectomy.[7-10] The rationale for this has been concern that there may be ectopic or missing parathyroid tissues in the thymus.[11] An association between highly aggressive thymic carcinoid neoplasms and MEN1 has also been described.[12-14] However, no study has studied systematically the yield of these tissues in thymectomy specimens performed with parathyroid resection in MEN1 patients. We

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Research Article

## Recapitulation of Pancreatic Neuroendocrine Tumors in Human Multiple Endocrine Neoplasia Type I Syndrome via *Pdx1*-Directed Inactivation of *Men1*

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### Abstract

**Multiple endocrine neoplasia type 1 (MEN1) is an autosomal syndrome caused by mutations in the *MEN1* tumor suppressor gene. Whereas the protein product of MEN1, menin, is ubiquitously expressed, somatic loss of the remaining wild-type *MEN1* allele results in tumors primarily in parathyroid, pituitary, and endocrine pancreas. To understand the endocrine specificity of the MEN1 syndrome, we evaluated biallelic loss of *Men1* by inactivating *Men1* in pancreatic progenitor cells using the Cre-lox system. *Men1* deletion in progenitor cells that differentiate into exocrine and endocrine pancreas did not affect normal pancreas morphogenesis and development. However, mice having homozygous inactivation of the *Men1* in pancreas developed endocrine tumors with no exocrine tumor manifestation, recapitulating phenotypes seen in the MEN1 patients. In the absence of menin, the endocrine pancreas showed increase in cell proliferation, vascularity, and abnormal vascular structures; such changes were lacking in exocrine pancreas. Further analysis revealed that these endocrine manifestations were associated with up-regulation in vascular endothelial growth factor expression in both human and mouse MEN1 pancreatic endocrine tumors. Together, these data suggest the presence of cell-specific factors for menin and a permissive endocrine environment for MEN1 tumorigenesis in endocrine pancreas. Based on our analysis, we propose that menin's ability to maintain cellular and microenvironment integrity might explain the endocrine-restrictive nature of the MEN1 syndrome.** [Cancer Res 2009;69(5):1858–66]

### Introduction

Multiple endocrine neoplasia type 1 (MEN1; OMIM 131100) is a dominant inherited syndrome caused by mutations in the *MEN1* tumor suppressor gene (1, 2). Patients with a family history of the MEN1 syndrome are predisposed to develop multiple endocrine tumors, primarily affecting parathyroid, anterior pituitary, and pancreatic islets. More than 95% of MEN1 patients develop clinical manifestations of the disorder by the fifth decade (3, 4), whereas the earliest occurrence has been reported at 5 years old (5). Consistent with Knudson's two-hit hypothesis for tumor suppressor genes (6), MEN1 monoclonal expansion is initiated when loss of heterozygosity (LOH) at 11q13 occurs in patients with inherited germ line mutations of the *MEN1* gene (7–9). Additionally, somatic inactivation and LOH of the *MEN1* alleles have been reported in a variety of sporadic endocrine tumors, such as parathyroid adenomas and pancreatic insulinomas (10, 11). Mutations in the *MEN1* gene seem to be inactivating, and no clear genotype-phenotype correlations have been established for mutations detected along the coding sequence of *MEN1* in both familial and sporadic tumors (12).

The protein product of *MEN1*, menin, does not display significant homology to any known family of proteins, and it has been described predominantly as a transcriptional regulator by interacting with nuclear proteins, such as JunD, nuclear factor-κB, Smad3, and FANCD2 (13, 14). Further biochemical studies have shown that menin complexes with mixed-lineage leukemia protein to regulate gene expression via chromatin modification in mouse embryonic fibroblast cells (15), bone marrow cells (16), and human HeLa and leukemia cells (17, 18). Similar epigenetic regulation by menin has also been reported in endocrine tumors, wherein menin modulates histone methylation and expression of cyclin-dependent kinase inhibitors, p27 and p18 (19). More recently, menin has been implicated in the control of pancreatic β-cell growth during pregnancy (20). Together, these studies have broadened our knowledge of the biochemical, physiologic, and pathologic roles of menin in both endocrine and nonendocrine contexts.

Using mouse models to understand the human MEN1 syndrome has proved to be informative (21–23), perhaps due to the highly conserved genomic structures, nucleotide (89% identity), and amino acid (97% identity) sequences shared by mouse *Men1* and human *MEN1* genes (24, 25). Although mice deficient of both *Men1* alleles die *in utero* at E11.5-13.5 with developmental defects in multiple organs, mice heterozygous for *Men1* deletion develop endocrine tumors at maturity, similar to those found in human MEN1 patients (21, 22, 26). To circumvent the embryonic lethality, conditional inactivation of *Men1* has further confirmed that biallelic loss of menin in endocrine tissues can lead to the development of parathyroid adenoma (27), pancreatic insulinoma (28, 29), and pituitary prolactinoma (30). These observations are reminiscent of the tumor spectrum observed in mice with heterozygous germ line deletion of *Men1*, as well as human MEN1 patients. Whereas the generation of mouse models has effectively mimicked the human MEN1 syndrome, the mechanisms

**Note:** Supplementary data for this article are available at Cancer Research Online (http://cancerres.aacrjournals.org/).

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# Importance of Team Science

# Science is a Team Sport

## Aim 2: Novel Targets, Chemical Ligands and Mode of Action

Jin, Chen, Carpizo (CP/CIPT), LaVoie, Liu, Govdos (CIPT), Augeri, Mehnert (CIPT), Levine (GICG)

**Major Discoveries:**

- **Uncoupling mitochondria is a therapeutic strategy**
- **GRM1 is a new cancer drug target for melanoma**
- **Repurpose riluzole and develop a riluzole prodrug, (trigriluzole) as cancer drugs**
- **Pharmacological restoration of mutant p53 functions**

**Featured Publications**

Tao, Nature Med 2014
Yu, Cancer Cell, 2012
Blanden, Mol Pharm, 2015
Wen, Cancer Res, 2014
Teh, Pigm Cell Mel Res, 2014

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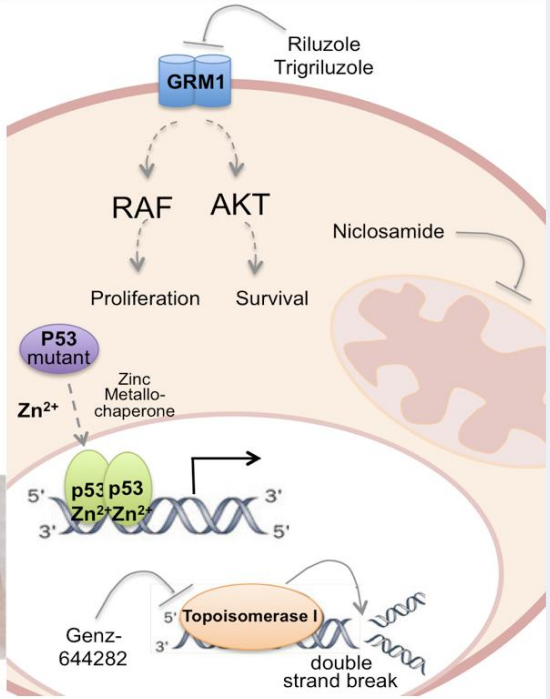
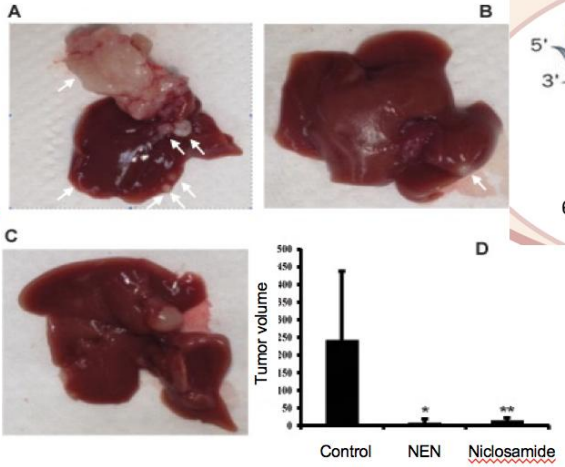
**Collaboration with Biopharma**

Biohaven Pharma (Chen)
Mito Biopharm (Jin)
Harrington Institute (Carpizo)
Sonafi/Genzyme (LaVoie/Liu)

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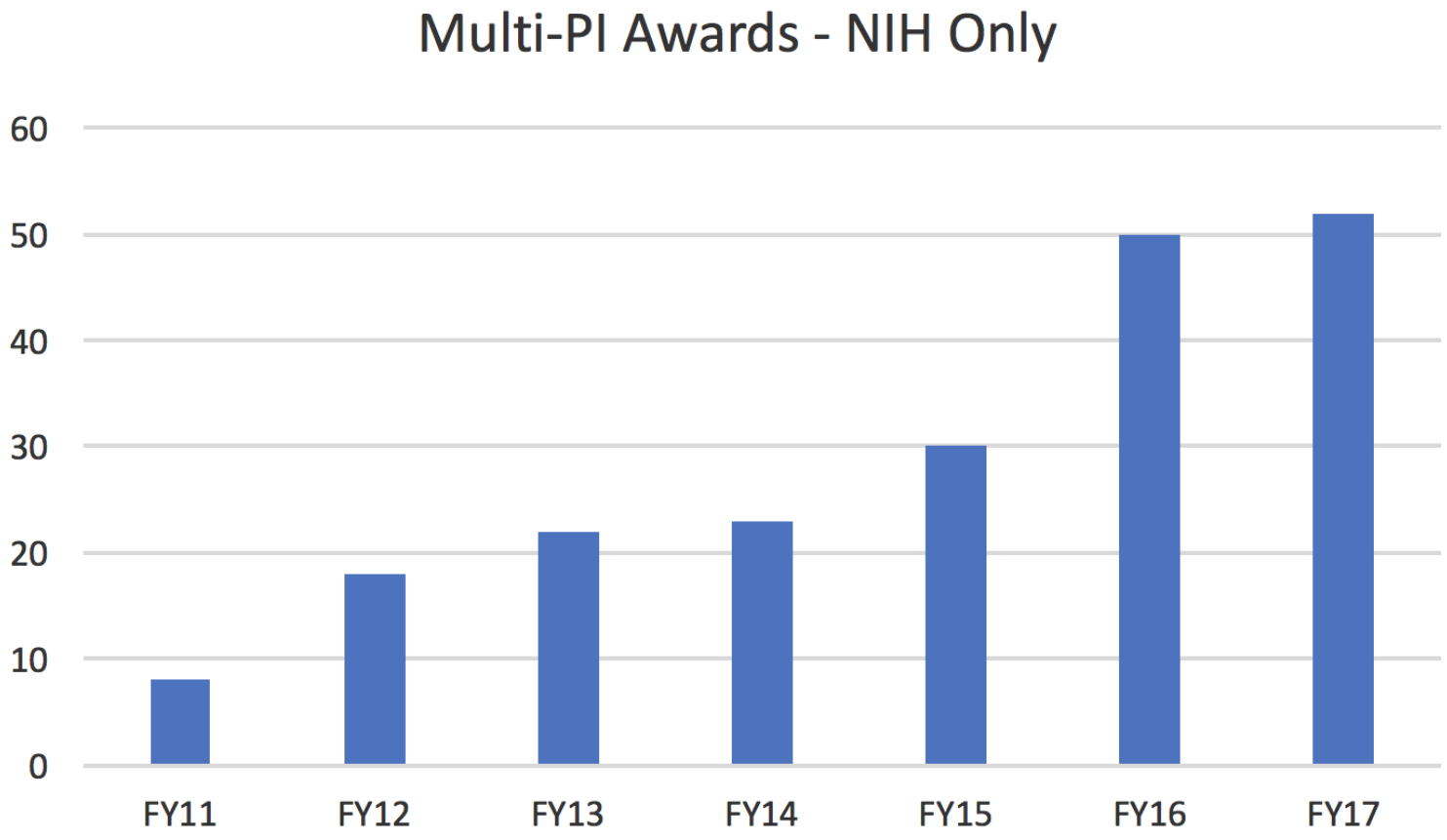
# Science is a Team Sport



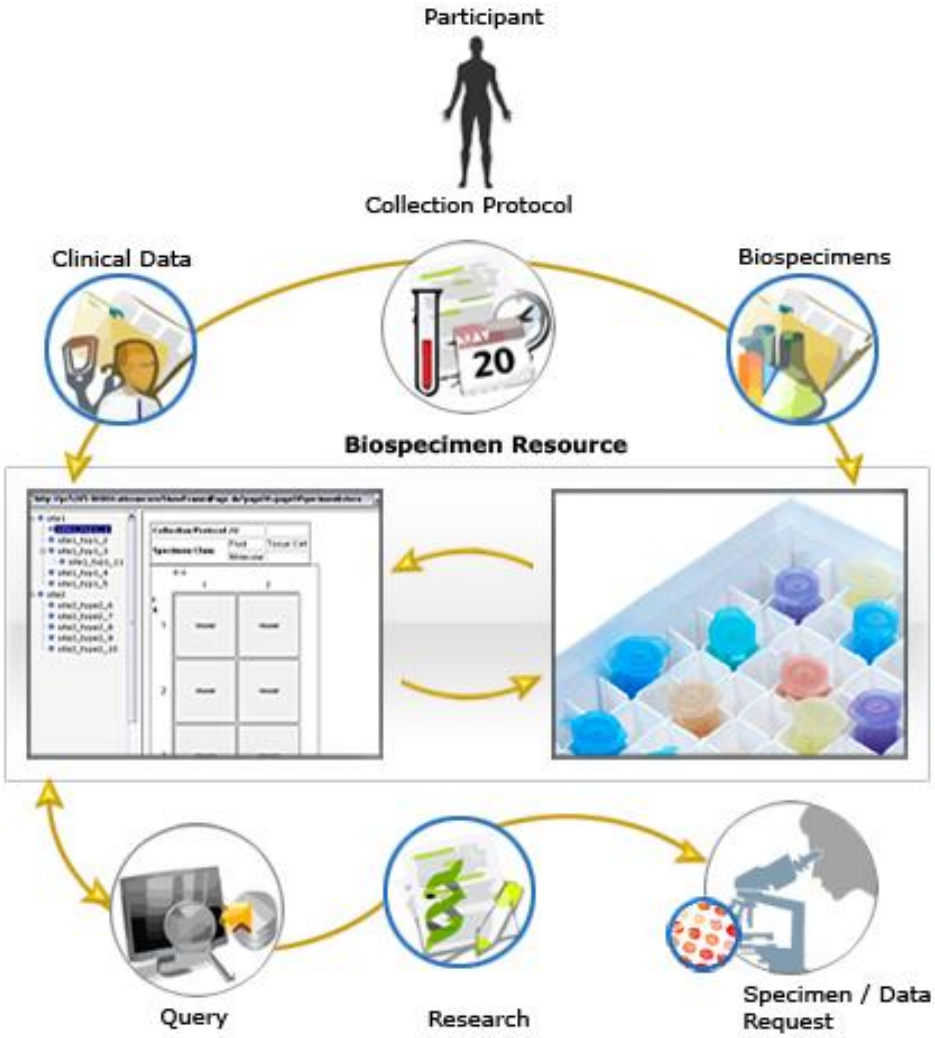
# The New Triple Threat…
# The Department

# Position Players Make a Team

- Exceptional Clinicians
- Exceptional Educators
- Exceptional Investigators

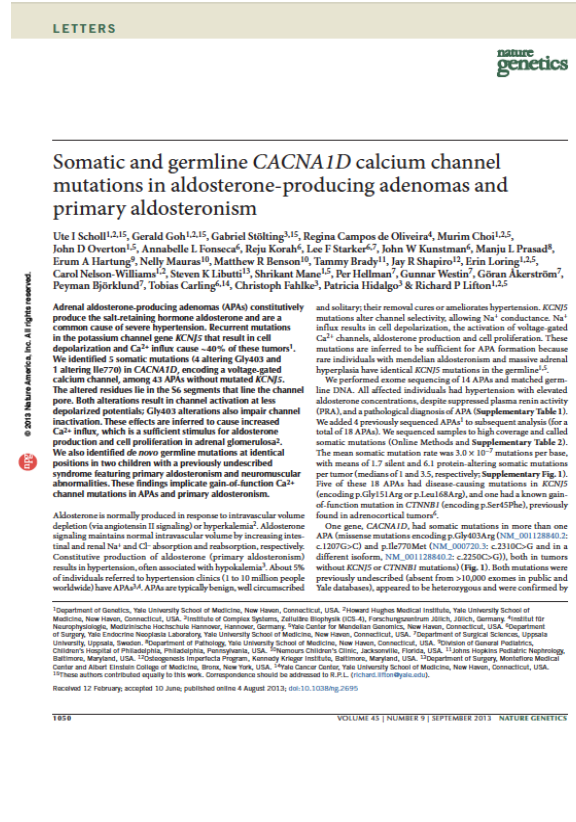
# Tissue as a Resource

# Biorepository



# Tissue as a Resource

Association for Academic Surgery



LETTERS

nature genetics

## Somatic and germline *CACNA1D* calcium channel mutations in aldosterone-producing adenomas and primary aldosteronism

Ute I Scholl[1,2,15], Gerald Goh[1,2,15], Gabriel Stölting[3,15], Regina Campos de Oliveira[4], Murim Choi[1,2,5], John D Overton[1,5], Annabelle L Fonseca[6], Reju Korah[6], Lee F Starker[6,7], John W Kunstman[6], Manju L Prasad[8], Erum A Hartung[9], Nelly Mauras[10], Matthew R Benson[10], Tammy Brady[11], Jay R Shapiro[12], Erin Loring[1,2,5], Carol Nelson-Williams[1,2], Steven K Libutti[13], Shrikant Mane[1,5], Per Hellman[7], Gunnar Westin[7], Göran Åkerström[7], Peyman Björklund[7], Tobias Carling[6,14], Christoph Fahlke[3], Patricia Hidalgo[3] & Richard P Lifton[1,2,5]

**Adrenal aldosterone-producing adenomas (APAs) constitutively produce the salt-retaining hormone aldosterone and are a common cause of severe hypertension. Recurrent mutations in the potassium channel gene *KCNJ5* that result in cell depolarization and $Ca^{2+}$ influx cause ~40% of these tumors[1]. We identified 5 somatic mutations (4 altering Gly403 and 1 altering Ile770) in *CACNA1D*, encoding a voltage-gated calcium channel, among 43 APAs without mutated *KCNJ5*. The altered residues lie in the S6 segments that line the channel pore. Both alterations result in channel activation at less depolarized potentials; Gly403 alterations also impair channel inactivation. These effects are inferred to cause increased $Ca^{2+}$ influx, which is a sufficient stimulus for aldosterone production and cell proliferation in adrenal glomerulosa[2]. We also identified *de novo* germline mutations at identical positions in two children with a previously undescribed syndrome featuring primary aldosteronism and neuromuscular abnormalities. These findings implicate gain-of-function $Ca^{2+}$ channel mutations in APAs and primary aldosteronism.**

Aldosterone is normally produced in response to intravascular volume depletion (via angiotensin II signaling) or hyperkalemia[2]. Aldosterone signaling maintains normal intravascular volume by increasing intestinal and renal $Na^+$ and $Cl^-$ absorption and reabsorption, respectively. Constitutive production of aldosterone (primary aldosteronism) results in hypertension, often associated with hypokalemia[3]. About 5% of individuals referred to hypertension clinics (1 to 10 million people worldwide) have APAs[3,4]. APAs are typically benign, well circumscribed and solitary; their removal cures or ameliorates hypertension. *KCNJ5* mutations alter channel selectivity, allowing $Na^+$ conductance. $Na^+$ influx results in cell depolarization, the activation of voltage-gated $Ca^{2+}$ channels, aldosterone production and cell proliferation. These mutations are inferred to be sufficient for APA formation because rare individuals with mendelian aldosteronism and massive adrenal hyperplasia have identical *KCNJ5* mutations in the germline[1,5].

We performed exome sequencing of 14 APAs and matched germline DNA. All affected individuals had hypertension with elevated aldosterone concentrations, despite suppressed plasma renin activity (PRA), and a pathological diagnosis of APA (**Supplementary Table 1**). We added 4 previously sequenced APAs[1] to subsequent analysis (for a total of 18 APAs). We sequenced samples to high coverage and called somatic mutations (Online Methods and **Supplementary Table 2**). The mean somatic mutation rate was $3.0 \times 10^{-7}$ mutations per base, with means of 1.7 silent and 6.1 protein-altering somatic mutations per tumor (medians of 1 and 3.5, respectively; **Supplementary Fig. 1**). Five of these 18 APAs had disease-causing mutations in *KCNJ5* (encoding p.Gly151Arg or p.Leu168Arg), and one had a known gain-of-function mutation in *CTNNB1* (encoding p.Ser45Phe), previously found in adrenocortical tumors[6].

One gene, *CACNA1D*, had somatic mutations in more than one APA (missense mutations encoding p.Gly403Arg (NM_001128840.2: c.1207G>C) and p.Ile770Met (NM_000720.3: c.2310C>G and in a different isoform, NM_001128840.2: c.2250C>G)), both in tumors without *KCNJ5* or *CTNNB1* mutations) (**Fig. 1**). Both mutations were previously undescribed (absent from >10,000 exomes in public and Yale databases), appeared to be heterozygous and were confirmed by

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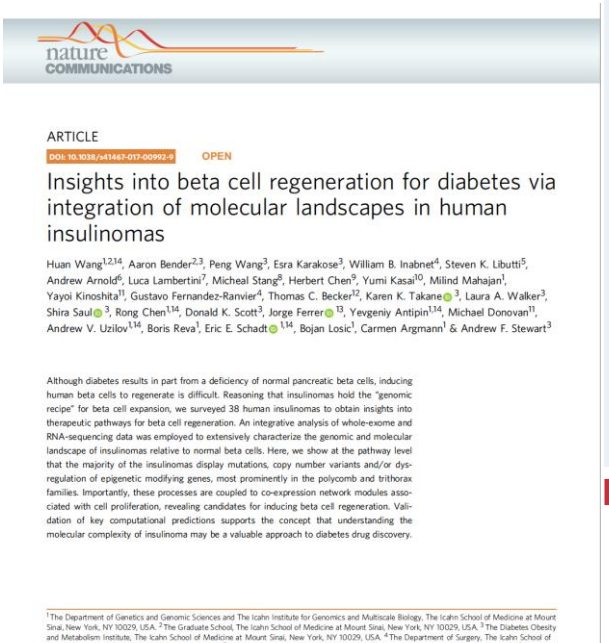

nature COMMUNICATIONS

ARTICLE

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## Insights into beta cell regeneration for diabetes via integration of molecular landscapes in human insulinomas

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Although diabetes results in part from a deficiency of normal pancreatic beta cells, inducing human beta cells to regenerate is difficult. Reasoning that insulinomas hold the "genomic recipe" for beta cell expansion, we surveyed 38 human insulinomas to obtain insights into therapeutic pathways for beta cell regeneration. An integrative analysis of whole-exome and RNA-sequencing data was employed to extensively characterize the genomic and molecular landscape of insulinomas relative to normal beta cells. Here, we show at the pathway level that the majority of the insulinomas display mutations, copy number variants and/or dysregulation of epigenetic modifying genes, most prominently in the polycomb and trithorax families. Importantly, these processes are coupled to co-expression network modules associated with cell proliferation, revealing candidates for inducing beta cell regeneration. Validation of key computational predictions supports the concept that understanding the molecular complexity of insulinoma may be a valuable approach to diabetes drug discovery.

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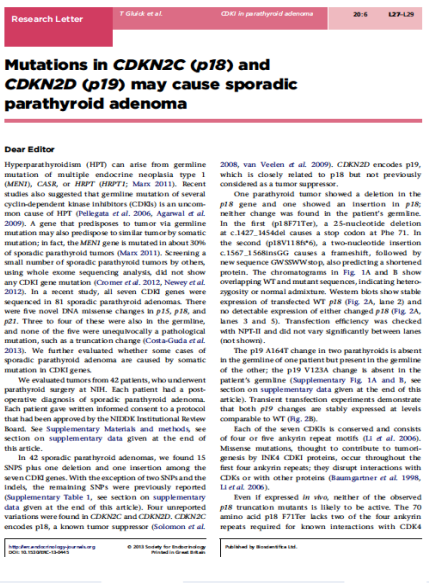



Diabetes-Related Cancer

## Mutations in *CDKN2C* (*p18*) and *CDKN2D* (*p19*) may cause sporadic parathyroid adenoma

**Dear Editor**

Hyperparathyroidism (HPT) can arise from germline mutation of multiple endocrine neoplasia type 1 (*MEN1*), *CASR*, or *HRPT* (*HRPT1*; Marx 2011). Recent studies also suggested that germline mutation of several cyclin-dependent kinase inhibitors (CDKIs) is an uncommon cause of HPT (Pellegata *et al.* 2006, Agarwal *et al.* 2009). A gene that predisposes to tumor via germline mutation may also predispose to similar tumor by somatic mutation; in fact, the *MEN1* gene is mutated in about 30% of sporadic parathyroid tumors (Marx 2011). Screening a small number of sporadic parathyroid tumors by others, using whole exome sequencing analysis, did not show any CDKI gene mutation (Cromer *et al.* 2012, Newey *et al.* 2012). In a recent study, all seven CDKI genes were sequenced in 81 sporadic parathyroid adenomas. There were five novel DNA missense changes in *p15*, *p18*, and *p21*. Three to four of these were also in the germline, and none of the five were unequivocally a pathological mutation, such as a truncation change (Costa-Guda *et al.* 2013). We further evaluated whether some cases of sporadic parathyroid adenoma are caused by somatic mutation in CDKI genes.

We evaluated tumors from 42 patients, who underwent parathyroid surgery at NIH. Each patient had a post-operative diagnosis of sporadic parathyroid adenoma. Each patient gave written informed consent to a protocol that had been approved by the NIDDK Institutional Review Board. See Supplementary Materials and methods, see section on supplementary data given at the end of this article.

In 42 sporadic parathyroid adenomas, we found 15 SNPs plus one deletion and one insertion among the seven CDKI genes. With the exception of two SNPs and the indels, the remaining SNPs were previously reported (Supplementary Table 1, see section on supplementary data given at the end of this article). Four unreported variations were found in *CDKN2C* and *CDKN2D*. *CDKN2C* encodes p18, a known tumor suppressor (Solomon *et al.* 2008, van Veelen *et al.* 2009). *CDKN2D* encodes p19, which is closely related to p18 but not previously considered as a tumor suppressor.

One parathyroid tumor showed a deletion in the *p18* gene and one showed an insertion in *p18*; neither change was found in the patient's germline. In the first (p18T71Ter), a 25-nucleotide deletion at c.1427_1454del causes a stop codon at Phe 71. In the second (p18V118fs*6), a two-nucleotide insertion c.1567_1568insGG causes a frameshift, followed by new sequence GWSWW stop, also predicting a shortened protein. The chromatograms in Fig. 1A and B show overlapping WT and mutant sequences, indicating heterozygosity or normal admixture. Western blots show stable expression of transfected WT *p18* (Fig. 2A, lane 2) and no detectable expression of either changed *p18* (Fig. 2A, lanes 3 and 5). Transfection efficiency was checked with NPT-II and did not vary significantly between lanes (not shown).

The p19 A164T change in two parathyroids is absent in the germline of one patient but present in the germline of the other; the p19 V123A change is absent in the patient's germline (Supplementary Fig. 1A and B, see section on supplementary data given at the end of this article). Transient transfection experiments demonstrate that both *p19* changes are stably expressed at levels comparable to WT (Fig. 2B).

Each of the seven CDKIs is conserved and consists of four or five ankyrin repeat motifs (Li *et al.* 2006). Missense mutations, thought to contribute to tumorigenesis by INK4 CDKI proteins, occur throughout the first four ankyrin repeats; they disrupt interactions with CDKs or with other proteins (Baumgartner *et al.* 1998, Li *et al.* 2006).

Even if expressed *in vivo*, neither of the observed *p18* truncation mutants is likely to be active. The 70 amino acid p18 F71Ter lacks two of the four ankyrin repeats required for known interactions with CDK4

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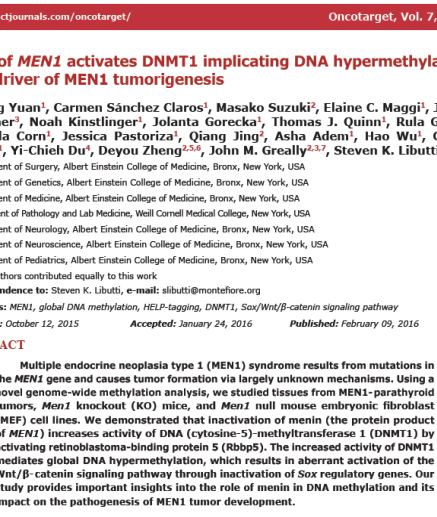

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## Loss of *MEN1* activates DNMT1 implicating DNA hypermethylation as a driver of MEN1 tumorigenesis

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ABSTRACT

**Multiple endocrine neoplasia type 1 (MEN1) syndrome results from mutations in the *MEN1* gene and causes tumor formation via largely unknown mechanisms. Using a novel genome-wide methylation analysis, we studied tissues from MEN1-parathyroid tumors, *Men1* knockout (KO) mice, and *Men1* null mouse embryonic fibroblast (MEF) cell lines. We demonstrated that inactivation of menin (the protein product of *MEN1*) increases activity of DNA (cytosine-5)-methyltransferase 1 (DNMT1) by activating retinoblastoma-binding protein 5 (Rbbp5). The increased activity of DNMT1 mediates global DNA hypermethylation, which results in aberrant activation of the Wnt/β-catenin signaling pathway through inactivation of *Sox* regulatory genes. Our study provides important insights into the role of menin in DNA methylation and its impact on the pathogenesis of MEN1 tumor development.**

# Final Thoughts

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- Inspiration, Passion, Focus and Follow Through
- Team Work and Mentorship
- Science and Discovery Must be Valued by Departments of Surgery and Health Systems