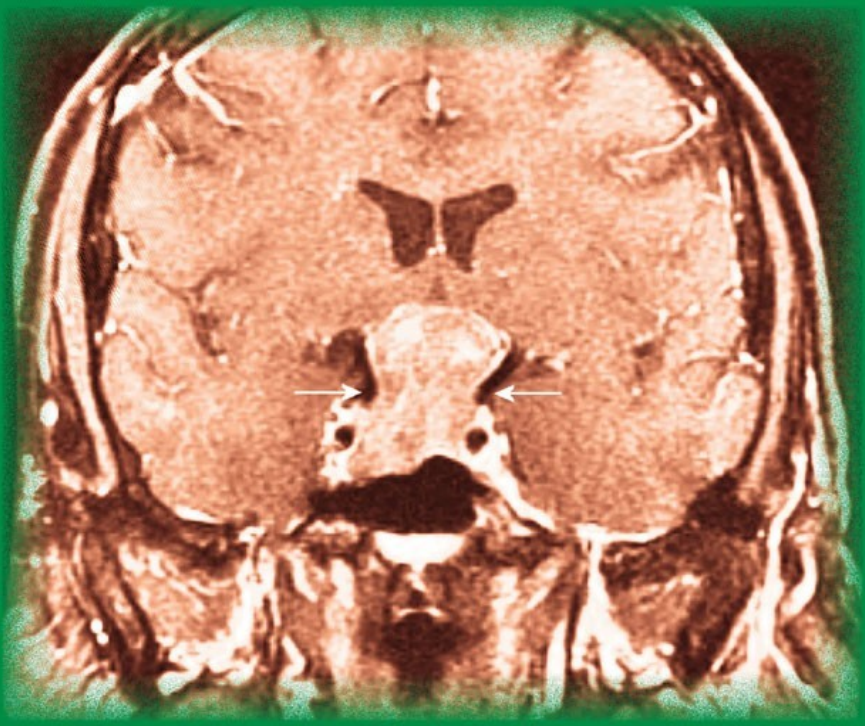
CONTEMPORARY ENDOCRINOLOGY™

Diagnosis and Management of Pituitary Disorders

Edited by

Brooke Swearingen, MD

Beverly M. K. Biller, MD



Humana Press

Diagnosis and Management of Pituitary Disorders

Contemporary Endocrinology

P. Michael Conn, *Series Editor*

Diagnosis and Management of Pituitary Disorders, edited by *Brooke Swearingen and Beverly M. K. Biller*, 2008

A Case-Based Guide to Clinical Endocrinology, edited by *Terry F. Davies*, 2008

Type 2 Diabetes Mellitus: An Evidence-Based Approach to Practical Management, edited by *Mark N. Feinglos and Mary Angelyn Bethel*, 2008

Autoimmune Diseases in Endocrinology, edited by *Anthony P. Weetman*, 2008

Energy Metabolism and Obesity: Research and Clinical Applications, edited by *Patricia A. Donohoue*, 2008

Polycystic Ovary Syndrome: Current Controversies, from the Ovary to the Pancreas, edited by *Andrea Dunaif, Jeffrey R. Chang, Stephen Franks, and Richard S. Legro*, 2008

The Metabolic Syndrome: Epidemiology, Clinical Treatment, and Underlying Mechanisms, edited by *Barbara Caleen Hansen and George A. Bray*, 2008

Genomics in Endocrinology: DNA Microarray Analysis in Endocrine Health and Disease, edited by *Stuart Handwerger and Bruce Aronow*, 2008

Controversies in Treating Diabetes: Clinical and Research Aspects, edited by *Derek LeRoith and Aaron I. Vinik*, 2008

Endocrine-Disrupting Chemicals: From Basic Research to Clinical Practice, edited by *Andrea C. Gore*, 2007

When Puberty is Precocious: Scientific and Clinical Aspects, edited by *Ora H. Pescovitz and Emily C. Walvoord*, 2007

Insulin Resistance and Polycystic Ovarian Syndrome: Pathogenesis, Evaluation and Treatment, edited by *Evanthia Diamanti-Kandarakis, John E. Nestler, D. Panidis, and Renato Pasquali,* 2007

Hypertension and Hormone Mechanisms, edited by *Robert M. Carey*, 2007

The Leydig Cell in Health and Disease, edited by *Anita H. Payne and Matthew Phillip Hardy*, 2007

Treatment of the Obese Patient, edited by *Robert F. Kushner and Daniel H. Bessesen*, 2007

Androgen Excess Disorders in Women: Polycystic Ovary Syndrome and Other Disorders, Second Edition, edited by *Ricardo Azzis, John E. Nestler, and Didier Dewailly*, 2006

Evidence-Based Endocrinology, edited by *Victor M. Montori*, 2006

Stem Cells in Endocrinology, edited by *Linda B. Lester*, 2005

Office Andrology, edited by *Phillip E. Patton and David E. Battaglia*, 2005

Male Hypogonadism: Basic, Clinical, and Therapeutic Principles, edited by *Stephen J. Winters*, 2004

Androgens in Health and Disease, edited by *Carrie Bagatell and William J. Bremner*, 2003

Endocrine Replacement Therapy in Clinical Practice, edited by *A. Wayne Meikle*, 2003

Early Diagnosis of Endocrine Diseases, edited by *Robert S. Bar*, 2003

Type I Diabetes: Etiology and Treatment, edited by *Mark A. Sperling*, 2003

Handbook of Diagnostic Endocrinology, edited by *Janet E. Hall and Lynnette K. Nieman*, 2003

Pediatric Endocrinology: A Practical Clinical Guide, edited by *Sally Radovick and Margaret H. Macgillivray*, 2003

Diseases of the Thyroid, Second Edition, edited by *Lewis E. Braverman*, 2003

Developmental Endocrinology: From Research to Clinical Practice, edited by *Erica A. Eugster and Ora Hirsch Pescovitz*, 2002

Osteoporosis: Pathophysiology and Clinical Management, edited by *Eric S. Orwoll and Michael Bliziotes*, 2002

Diagnosis and Management of Pituitary Disorders

Edited by

Brooke Swearingen, MD
Department of Neurosurgery, Massachusetts General Hospital, Harvard Medical School, Boston, MA

and

Beverly M. K. Biller, MD
Neuroendocrine Unit, Department of Medicine, Massachusetts General Hospital, Harvard Medical School, Boston, MA

Humana Press

Editors
Brooke Swearingen
Department of Neurosurgery
Massachusetts General Hospital
Harvard Medical School
Boston, MA
USA

Beverly M. K. Biller
Neuroendocrine Unit
Department of Medicine
Massachusetts General Hospital
Harvard Medical School
Boston, MA
USA

Series Editor
P. Michael Conn, PhD
Associate Director and Senior Scientist
Oregon National Primate Research Center
Professor
Oregon Health and Science University
505 NW 185th Ave.
Beaverton, OR 97006

ISBN: 978-1-58829-922-2 e-ISBN: 978-1-59745-264-9

Library of Congress Control Number: 2007942035

Cover illustration: Chapter 4, Fig. 3 by John T. Lysack et al.

Printed on acid-free paper

9 8 7 6 5 4 3 2 1

springer.com

Preface

This book presents a comprehensive update on the current diagnostic and treatment options for the management of disease of the sella, with an emphasis on pituitary adenomas. Over the past several decades, the techniques of molecular biology have been employed to investigate the pathogenesis of these tumors, as discussed by Drs. Lania, Mantovani, and Spada in Chapter 1. Their pathological analysis is discussed by Drs. Gejman and Hedley-Whyte in Chapter 2. The evaluation of patients presenting with sellar disease is based both on modern endocrine techniques, as discussed by Dr. Snyder in Chapter 3, as well as new imaging modalities, as discussed by Drs. Lysack and Schaefer in Chapter 4. Since Harvey Cushing first plotted visual fields, the intimate anatomic relationship between the sella and the optic structures has required careful neuro-ophthalmologic evaluation in these cases; this is discussed by Drs. Cestari and Rizzo in Chapter 5. The management of secretory adenomas remains challenging. Prolactinomas, since the introduction of medical treatment in the 1980s, have been primarily managed with dopamine agonists as discussed by Drs. Shibli-Rahhal and Schlechte in Chapter 6. The diagnosis of acromegaly, discussed by Dr. Clemmons in Chapter 7, is made by hormonal testing and depends on reliable GH and IGF-1 assays. The treatment of acromegaly, once primarily a surgical disease, is now increasingly amenable to new medical agents, including somatostatin analogs and growth hormone receptor antagonists. The relative advantages of these approaches are discussed by Dr. Freda, and Drs. Buchfelder and Nomikos, in Chapters 8 and 9, respectively. The patient with Cushing's disease requires an extensive and sophisticated endocrine evaluation before undergoing transsphenoidal surgery, as outlined by Drs. Findling and Raff in Chapter 10. The surgical approach is described by Dr. Kelly in Chapter 11, with options for medical treatment discussed by Drs. Lindsay and Nieman in Chapter 12. The diagnosis and treatment of the uncommon TSH adenomas is described by Drs. Zemskova and Skarulis in Chapter 13. Nonfunctioning tumors currently remain the province of the neurosurgeon, as discussed by Drs. Muh and Oyesiku in Chapter 14. Drs. Chandler and Barkan describe the surgical techniques used to remove sellar tumors in Chapter 15, while Drs. Barkan, Blank, and Chandler address

the perioperative management of patients with these lesions in Chapter 16. Although advances in medical treatment and surgical techniques have made its use less frequent, radiation therapy continues to have an important role in the management of these patients, as described by Drs. Shih and Loeffler in Chapter 17. Finally, a number of specialized and clinically important topics arise in caring for patients with pituitary disorders. The diagnosis and management of inflammatory disease of the pituitary is discussed by Drs. Ulmer and Byrne in Chapter 18, the management of apoplexy by Drs. Russell and Miller in Chapter 19, and the management of pituitary disease during pregnancy by Dr. Molitch in Chapter 20. Modern imaging techniques will sometimes demonstrate an incidental sellar abnormality when none was suspected; the evaluation of these patients is described by Dr. Frohman in Chapter 21. Although pituitary adenomas are relatively less common in children, other sellar pathologies, especially craniopharyngiomas, are more important and their endocrine management is critical in the developing child; these topics are discussed by Drs. Stanley, Prabhakaran, and Misra in Chapter 22. Finally, the management of cystic disease of the sella can be an especially thorny problem, and therapeutic options are described by Drs. Snyder, Naidich, and Post in Chapter 23.

It has been a pleasure to work with some of the leading authorities in the field of pituitary disease in the preparation of this volume and we would like to thank them both for their contributions to this volume and their commitment to the field of pituitary education. In addition, we would like to thank Dr. Michael Conn and Richard Lansing of Springer publishing for conceiving this project and asking us to participate in it, and the editorial staff at Springer for their expert assistance in preparing the volume.

Brooke Swearingen, ***MD***
Beverly M. K. Biller, ***MD***

Contents

Contributors

Ariel L. Barkan, MD • *Professor, Departments of Internal Medicine and Neurosurgery, University of Michigan Medical Center, Ann Arbor, MI*

Howard Blank, MD • *Fellow, Division of Metabolism, Endocrinology, and Diabetes, University of Michigan Medical Center, Ann Arbor, MI*

Michael Buchfelder, MD, PhD • *Professor and Chairman, Department of Neurosurgery, University of Erlangen-Nuremberg, Erlangen, Germany*

Thomas N. Byrne, MD • *Clinical Professor of Neurology and Health Sciences and Technology, Harvard Medical School, Massachusetts General Hospital, Boston, MA*

Dean M. Cestari, MD • *Assistant Professor of Ophthalmology, Massachusetts Eye & Ear Infirmary, Harvard Medical School, Boston, MA*

William F. Chandler, MD • *Professor, Departments of Neurosurgery and Internal Medicine, University of Michigan Medical Center, Ann Arbor, MI*

David R. Clemmons, MD • *Kenan Professor of Medicine, UNC School of Medicine, Chapel Hill, NC*

James W. Findling, MD • *Director, Endocrine-Diabetes Center, Aurora St. Luke's Medical Center, Clinical Professor of Medicine, Medical College of Wisconsin, Milwaukee, WI*

Pamela U. Freda, MD • *Associate Professor of Medicine, Columbia University, College of Physicians and Surgeons, New York, NY*

Lawrence A. Frohman, MD • *Professor Emeritus of Medicine, Section of Endocrinology, Metabolism, and Diabetes, University of Illinois at Chicago, Chicago, IL*

Roger Gejman, MD • *Research Fellow in Neuropathology, Massachusetts General Hospital, Research Fellow in Pathology, Harvard Medical School, Boston, MA*

E. Tessa Hedley-Whyte, MD • *Professor of Pathology, Harvard Medical School, Neuropathologist, Massachusetts General Hospital, Boston, MA*

Daniel F. Kelly, MD • *Director, Neuroendocrine Tumor Center, John Wayne Cancer Institute at Saint John's Health Center, Santa Monica, CA*

Andrea Lania, MD, PhD • *Endocrine Unit, Department of Medical Sciences, Fondazione Policlinico, IRCCS, University of Milan, Milan, Italy*

John R. Lindsay, MD • *Altnagelvin Hospital, Western Health and Social Care Trust, Londonderry, UK*
Jay S. Loeffler, MD • *Herman and Joan Suit Professor of Radiation Oncology, Harvard Medical School, Chair, Department of Radiation Oncology, Massachusetts General Hospital, Boston, MA*
John T. Lysack, MD, FRCPC • *Clinical Assistant in Neuroradiology, Massachusetts General Hospital, Harvard Medical School, Boston, MA*
Giovanna Mantovani, MD, PhD • *Endocrine Unit, Department of Medical Sciences, Fondazione Policlinico, IRCCS, University of Milan, Milan, Italy*
Karen Klahr Miller, MD • *Neuroendocrine Unit, Massachusetts General Hospital, Harvard Medical School, Boston, MA*
Madhusmita Misra, MD, MPH • *Assistant in Pediatrics, Pediatric Endocrinology Unit, MassGeneral Hospital for Children, Assistant in Biology, Neuroendocrine Unit, Massachusetts General Hospital, Assistant Professor of Pediatrics, Harvard Medical School, Boston, MA*
Mark E. Molitch, MD • *Professor of Medicine, Division of Endocrinology, Metabolism, and Molecular Medicine, Northwestern University Feinberg School of Medicine, Chicago, IL*
Carrie R. Muh, MD, MS • *Department of Neurological Surgery, Emory University School of Medicine, Atlanta, GA*
Thomas P. Naidich, MD • *Director of Neuroradiology, Professor of Radiology and Neurosurgery, Irving and Dorothy Regenstreif Research Professor of Neuroscience (Neuroimaging), Mount Sinai Medical Center, New York, NY*
Lynnette K. Nieman, MD • *Senior Investigator, RBMB, NICHD, Associate Director, IETP, NICHD-NIDDK, Reproductive Biology and Medicine Branch, NICHD, National Institutes of Health, Bethesda, MD*
Panagiotis Nomikos, MD • *Senior Neurosurgeon, Department of Neurosurgery, Hygeia Hospital, Marousi, Greece*
Nelson M. Oyesiku, MD, PhD, FACS • *Professor and Vice-Chairman, Department of Neurological Surgery, Emory University School of Medicine, Atlanta, GA*
Kalmon D. Post, MD • *Professor of Neurosurgery, Mount Sinai Medical Center, New York, NY*
Rajani Prabhakaran, MD • *Fellow, Pediatric Endocrinology, MassGeneral Hospital for Children, Harvard Medical School, Boston, MA*
Hershel Raff, PhD • *Director, Endocrine Research Laboratory, Aurora St. Luke's Medical Center, Professor of Medicine, Medical College of Wisconsin, Milwaukee, WI*
Joseph F. Rizzo III, MD • *Associate Professor of Ophthalmology, Massachusetts Eye & Ear Infirmary, Harvard Medical School, Boston, MA*

Steven J. Russell, MD, PhD • *Instructor in Medicine, Harvard Medical School, Assistant in Medicine, Massachusetts General Hospital, Boston, MA*

Pamela W. Schaefer, MD • *Associate Director of Neuroradiology, Clinical Director of MRI, Massachusetts General Hospital, Associate Professor of Radiology, Harvard Medical School, Boston, MA*

Janet A. Schlecte, MD • *Professor of Medicine, University of Iowa Hospitals and Clinics, Iowa City, IA*

Amal Shibli-Rahhal, MD • *Assistant Professor of Medicine, University of Iowa Hospitals and Clinics, Iowa City, IA*

Helen A. Shih, MD, MS, MPH • *Instructor in Radiation Oncology, Massachusetts General Hospital, Harvard Medical School, Boston, MA*

Monica C. Skarulis, MD • *Clinical Endocrinology Branch, National Institute of Diabetes, Digestive and Kidney Diseases, National Institutes of Health, Bethesda, MD*

Brian J. Snyder, MD • *Department of Neurosurgey, Mount Sinai Medical Center, New York, NY*

Peter J. Snyder, MD • *Professor of Medicine, University of Pennsylvania, Philadelphia, PA*

Anna Spada, MD • *Professor of Endocrinology, Endocrine Unit, Department of Medical Sciences, Fondazione Policlinico, IRCCS, University of Milan, Milan, Italy*

Takara Stanley, MD • *Fellow, Pediatric Endocrinology, MassGeneral Hospital for Children, Harvard Medical School, Boston, MA*

Stephan Ulmer, MD • *Institute of Neuroradiology, University Hospital of Schleswig-Holstein, Kiel, Germany*

Marina S. Zemskova, MD • *Associate Investigator, Clinical Endocrinology Branch, National Institute of Diabetes, Digestive and Kidney Diseases, National Institutes of Health, Bethesda, MD*

Color Plate

The following color illustrations are printed in the insert.

Chapter 2

Fig. 1: Prolactinoma composed of cells with chromophobic cytoplasm arranged in a diffuse architectural pattern (A). Prolactinoma with small, hyperchromatic cells after dopamine agonist therapy (B). Positive immunohistochemical reaction for PRL with diffuse (C) and paranuclear patterns (D) in two prolactinomas.

Fig. 2: Densely granulated somatotroph pituitary adenoma with acidophilic and densely granulated cytoplasm (A), strong positive immunoreaction for GH (B) and diffuse immunohistochemical staining pattern for CAM 5.2 (C). Sparsely granulated somatotroph pituitary adenoma with a chromophobic and less granular cytoplasm (D). The same tumor as in (D) with slightly positive reaction for GH (E) and the dot-like positive reaction with CAM 5.2 corresponding to fibrous bodies (F).

Fig. 3: ACTH-producing pituitary tumor composed of densely granular basophilic cells (A) with strong positive immunohistochemical reaction for ACTH (B).

Fig. 4: Gonadotropic pituitary adenoma with a papillary pattern (A); perivascular pseudorosettes (B); and focal and weak expression of beta-FSH (C). Ultrastructural appearance of a tumor cell with oncocytic changes, i.e., many mitochondria (D).

Fig. 5: Craniopharyngioma composed of cords and islands of squamoid epithelium limited by columnar cells (A). Some cavities contain keratin material (*) (B). A cystic area has a thin epithelial wall(C) and adjacent inflammatory reaction with many foamy macrophages (D) (hematoxylin and eosin stain).

Fig. 6: Germinoma with a dense lymphocytic population and scattered groups of bigger round tumor cells with clear cytoplasm (arrows) (hematoxylin and eosin stain).

Fig. 7: Granular cell tumor composed of closely apposed acidophilic cells with bland nuclei and granular cytoplasm (hematoxylin and eosin stain).

Fig. 9: Lymphocytic hypophysitis with a dense inflammatory infiltrate including lymphocytes and plasma cells. Scattered pituitary cells are seen between the inflammatory cells (arrows) (hematoxylin and eosin stain).

Chapter 5

Fig. 6: Horizontal section of the visual pathways. The visual fields demonstrate the correlation of lesion site and field defect. (Reproduced with permission, Yanoff M, Duker JS, editors. Ophthalmology, 2nd ed. St Louis, Mo: Mosby; 2004.)

Fig. 7: Localization and probable identification of masses by pattern of field loss. Junctional scotomas occur with compression of the anterior angle of the chiasm (sphenoid meningiomas). Bitemporal hemianopia results from compression of the body of the chiasm from below (e.g., pituitary adenoma, sellar meningiomas). Compression of the posterior chiasm and its decussating nasal fibers may cause central bitemporal scotomas (e.g., hydrocephalus, pinealoma, craniopharyngioma). (Reproduced with permission, Yanoff M, Duker JS, editors. Ophthalmology, 2nd ed. St Louis, Mo: Mosby; 2004.)

Fig. 9: Parasympathetic and sympathetic innervation of the iris muscles. (Reproduced with permission, Yanoff M, Duker JS, editors. Ophthalmology, 2nd ed. St Louis, Mo: Mosby; 2004.)

Fig. 10: (A) Early papilledema. The optic disk of an 18-year-old man 2 weeks after he had complained of diplopia arising from sixth cranial nerve palsies caused by increased intracranial pressure. Note the minimal evidence of edema. (Reproduced with permission, Yanoff M, Duker JS, editors. Ophthalmology, 2nd ed. St Louis, Mo: Mosby; 2004.) (B) Developed papilledema. The optic disk of a 36-year-old woman who suffered headache and blurred vision for 2 months. Fully developed disk edema present—note the engorged veins and peripapillary hemorrhages. (Reproduced with permission, Yanoff M, Duker JS, editors. Ophthalmology, 2nd ed. St Louis, Mo: Mosby; 2004.) (C) Chronic papilledema. Severe and chronic disk edema in a 27-year-old very obese woman who has pseudotumor cerebri. Note that the disk cup is obliterated and hard exudates are present. (Reproduced with permission, Yanoff M, Duker JS,

editors. Ophthalmology, 2nd ed. St Louis, Mo: Mosby; 2004.) (D) Secondary optic atrophy from chronic papilledema. The same 27-year-old obese female patient 5 months later. Note the secondary optic atrophy has developed fully. The disk margins appear hazy or "dirty." (Reproduced with permission, Yanoff M, Duker JS, editors. Ophthalmology, 2nd ed. St Louis, Mo: Mosby; 2004.)

Fig. 11: Optic disk tilting and the resulting visual field defects. (A, B) Visual fields demonstrate bilateral relative superotemporal defects not respecting the vertical midline. (C, D) Fundus photos show bilateral tilted disks, with flattening of the inferonasal disk margins. (Reproduced with permission from The American Academy of Ophthalmology, Basic and Clinical Science Course, Section 5: Neuro-ophthalmology 2005–2006.)

Fig. 18: Acute compressive optic neuropathy in pituitary apoplexy. (A, B) Fundus photographs in a patient with acute severe visual loss bilaterally. The optic disks appear relatively normal. (C, D) Axial (left) and sagittal (right) MRI scans show a large pituitary tumor with suprasellar extension. Inhomogeneity within the tumor represents hemorrhage and infarction. (Reproduced with permission from The American Academy of Ophthalmology, Basic and Clinical Science Course, Section 5: Neuro-ophthalmology 2005–2006.)

Chapter 13

Fig. 1: TSH-oma cells by light microscopy (40× magnification). H&E stain shows significant cytological and nuclear pleomorphism of tumor cells.

Fig. 2: Immunohistochemical staining of TSH-oma (40× magnification). Tumor cells show positive reaction for TSH. The intensity of staining is variable from cell to cell.

1 Molecular Pathogenesis of Pituitary Adenomas

Andrea Lania, MD, PhD,
Giovanna Mantovani, MD, PhD,
and Anna Spada, MD

Contents

Summary

The genesis of pituitary tumors is still under debate. Although these neoplasia are monoclonal in origin, mutations of GNAS1, the gene encoding the α subunit of Gs is the only mutational change unequivocally associated with GH-secreting adenomas. In addition, multiple events, including the overexpression of cell cycle regulators, growth factors, and stimulatory hormones together with epigenetic disruption of genes with antioncogenic properties, frequently occur in pituitary tumors; their relative importance is still uncertain.

Key Words: Pituitary adenomas, Tumorigenesis, Oncogene, Oncosuppressor genes, *gsp*.

1. INTRODUCTION

The pathogenesis of pituitary tumors remains controversial. The respective role and importance of intrinsic alterations of the pituicytes themselves, dysregulation of hypothalamic hormones, and autocrine/paracrine action of locally produced growth factors are still under debate *(1–4)*. The demonstration by X-chromosome inactivation analysis that the majority of pituitary adenomas

From: *Contemporary Endocrinology: Diagnosis and Management of Pituitary Disorders*
Edited by: B. Swearingen and B. M. K. Biller © Humana Press, Totowa, NJ

are monoclonal in origin represents a milestone in this debate *(5,6)*. Indeed, these data unequivocally indicate that pituitary neoplasia arise from the replication of a single mutated cell, suggesting that growth advantage results from either activation of protooncogenes or inactivation of tumor-suppressor genes. Both in vivo and in vitro evidence suggest that, in addition to mutational changes, tumor formation requires a secondary event for clonal expansion and progression. The need for a "second hit" is indicated by the clinical observation that high-resolution neuroradiological imaging "incidentally" detects pituitary microadenomas in about 20% of subjects without signs or symptoms of pituitary disorders, a value that is about 1,000-fold higher than the clinical prevalence of the disease and approaches the incidence of pituitary adenomas found in unselected autopsies *(7,8)*. In this chapter we will summarize the molecular abnormalities that have been proposed to be responsible for pituitary tumor formation and progression.

2. ACTIVATION OF PROTOONCOGENES IN PITUITARY TUMORS

Pituitary tumors may originate from genetic abnormalities able to confer gain of function of either common or pituitary-specific protooncogenes. Moreover, in the absence of genetic abnormalities, dysregulation or overexpression of signal molecules that are components of proliferative pathways may promote cell growth (Table 1).

Table 1
Gain-of-function Events in Pituitary Tumors

Human pituitary tumor	*Gene*	*Defect*
ACTH-omas	Cyclin E	Increased expression
GH-omas	GNAS1	Somatic mutations
NFPA, GH-omas	Gi2α	Somatic mutations
PRL-omas	HMGA2	Increased expression
All types	PTTG	Increased expression
All types	FGFR4	Alternative transcription initiation
Pituitary carcinoma metastases; Aggressive PRL-omas	Ras	Somatic mutations
Aggressive adenomas	Cyclin D1	Increased expression
Invasive NFPA	PKC	Somatic mutations

FGFR4, fibroblast growth factor receptor 4; PKC, protein kinase C; PTTG, pituitary tumor transforming gene; NFPA, nonfunctioning pituitary adenoma.

2.1. Genetic Abnormalities of Protooncogenes

Common and pituitary specific protooncogenes have been extensively screened for genetic abnormalities in pituitary tumors during the past two decades. This extensive search has failed to identify the initial pathogenetic event in most tumors, and at present few genetic defects in protooncogenes are unequivocally associated with pituitary tumorigenesis.

2.1.1. Gain-of-Function Mutations of Monomeric and Heterotrimeric GTP-Binding Proteins

The family of RAS protooncogene encodes a 21-kD monomeric GDP/GTP-binding protein mainly involved in the activation of the mitogen activated protein kinase (MAPK) cascade and growth factor signaling. This protooncogene may acquire mitogenic properties by point mutations in codons 12 and 13 that increase the affinity for GTP, or mutations in codon 61 that prevent GTPase activity. RAS mutations are present with relatively high frequency in human malignancies, while they are uncommon in pituitary tumors. Indeed, a Gly12 to Val substitution has been observed in one single, unusually aggressive, and ultimately fatal prolactinoma resistant to dopaminergic inhibition *(9)*. Consistent with the view that this mutational change probably represents a late event associated with unusual malignant features, RAS mutations have been detected in metastases of pituitary carcinomas, but not in the primitive tumors *(10,11)*.

In contrast to the rare occurrence of RAS mutations, mutations in the gene encoding the α subunit of Gs (GNAS1) are frequent events, occurring in about 30–40% of GH-secreting adenomas *(12,13)*. Gs is a ubiquitously expressed protein that belongs to the family of heterotrimeric G proteins and is constituted by the specific α subunit and the common βγ subunits. Gs protein mediates the activation of adenylyl cyclase and generation of cAMP in pituitary target cells in response to several hormones. In particular, by interacting with specific G protein-coupled receptors, hypothalamic releasing hormones such as GH-releasing hormone (GHRH), corticotroph-releasing hormone (CRH), pituitary adenylate cyclase activating peptide (PACAP) and vasoactive intestinal peptide (VIP) activate the cAMP-dependent pathway. Although in vitro mutagenesis studies have documented a number of possible activating substitutions in the GNAS1 gene, the only amino acid changes so far reported replace either Arg 201 with Cys or His or Ser, or, less frequently, Gln 227 with Arg or Leu. These changes result in the constitutive activation of the subunit due to the reduction of GTPase activity *(12,13)*. Since somatotrophs belong to a set of cells that recognize cAMP as a mitogenic signal, Gsα may be considered the product of a protooncogene that is converted into an oncogene, designated *gsp* (for Gs protein) in selected cell types. Although this oncogene has been demonstrated to

confer growth advantage in vitro, patients carrying *gsp*-positive or *gsp*-negative tumors have the same clinical and biochemical phenotype, recurrence rate, and outcome *(14–17)*. The discrepancy between the mitogenic action of the mutant Gsα observed in vivo and in vitro strongly suggests the presence of events able to counteract in vivo the putative growth advantage conferred by the *gsp* oncogene. In this respect, some counteracting mechanisms, such as the instability of the mutant protein and the expression of cAMP-regulated genes with opposing actions, i.e., cAMP-specific phosphodiesterase isoforms and the inducible cAMP early repressor, have been identified in *gsp*-positive tumors *(18–22)*.

The phenotype of pituitary tumors is also related to the imprinting of GNAS1. The GNAS1 locus that maps on human chromosome 20q13 is under a complex imprinting control, with multiple maternally, paternally and bi-allelically alternatively spliced transcripts *(23,24)*. Recent reports demonstrated a predominant, though not exclusive, maternal origin of Gsα in adult human thyroid, gonad, and pituitary tissue *(25–27)*. Almost all *gsp*-positive tumors show mutations on the maternal allele *(25,27)*. Moreover, a partial loss of GNAS1 imprinting, resulting in Gsα overexpression, has been found in *gsp*-negative GH-secreting adenomas, although subsequent studies did not confirm this observation *(25,27)*.

Following the first identification in GH-secreting adenomas, *gsp* mutations have been infrequently detected also in other pituitary tumors, i.e., in about 10% of nonfunctioning pituitary adenomas and < 5% of ACTH-secreting adenomas *(28,29)*.

At present, Gsα is the only G protein that has been identified as target for activating mutations unequivocally associated with pituitary tumors. In fact, data concerning mutations of Gi2α protein, a protein involved in the inhibition of adenylyl cyclase and calcium influx, are discordant. Previous screening studies reported amino acids substitutions of Gln 205 (corresponding to Gln 227 of the Gsα sequence) with Arg in a subset of pituitary tumors; these studies were not confirmed by subsequent reports *(30,31)*. Despite the absence of mutations in Gq and G11 genes that are involved in Ca^{2+}/calmodulin and phospholipid-dependent protein kinase C activation, some reports suggested an overactivity of this pathway due to mutations of protein kinase Cα gene in pituitary adenomas. In particular, point mutations replacing Gly 294, a domain containing the calcium-binding site, with Asp have been identified in four invasive pituitary tumors *(28)*, an observation not confirmed by subsequent studies *(32,33)*.

2.1.2. Genetic Abnormalities of Growth Factors

The normal pituitary and pituitary tumors produce a wide number of growth factors and express their specific receptors *(3,4)*. In contrast to other human

neoplasms, genetic abnormalities of these factors and receptors are a rare event in pituitary tumorigenesis, the only alteration occurring in fibroblast growth factor (FGF) signaling. Indeed, about 40% of pituitary adenomas show the aberrant expression of an *N*-terminally truncated variant of FGF receptor-4. This variant is constitutively phosphorylated in the absence of the ligand and causes transformation in vitro and in vivo *(34)*. Interestingly, in contrast to previous models of pituitary tumorigenesis, the expression of the truncated receptor in the pituitary of transgenic mice results in tumor formation in the absence of massive hyperplasia, a phenomenon similar to that observed in human pituitary adenomas *(34)*. Moreover, disruption of FGF receptor 4 signaling seems to be associated with tumor invasion, since this receptor is required, together with other molecules, such as *N*-cadherin, phospholipase C-γ, and tumor-suppressor neural cell-adhesion molecule, for normal cell contact *(34,35)*.

2.2. Overexpression of Protooncogenes and Proliferative Signals

In contrast to the few molecular changes detected in pituitary tumors, in these neoplasms amplification of proliferative signals frequently occurs by overexpression. While the resulting phenotypes and their clinical correlations have been extensively investigated, the molecular mechanisms responsible for this dysregulation remain largely undefined.

2.2.1. Overexpression of Cell Cycle Regulators

The expression of genes involved in cell progression to replication has been extensively investigated in pituitary tumors. Pituitary adenomas overexpress cyclins, particularly cyclin D1 and cyclin E. In particular, in a screening study reporting the expression of cyclins in about 100 pituitary tumors, cyclin D1 was overexpressed in aggressive functioning and nonfunctioning tumors, while cyclin E was preferentially present in corticotroph adenomas *(36)*. Moreover, using a frequent polymorphism in cyclin D1 gene (CCND1), allelic imbalance indicative of gene amplification has been found in about 25% of pituitary tumors, despite the absence of a clear increase of cyclin D1 protein *(37)*.

Almost all pituitary adenomas overexpress the pituitary tumor transforming gene (PTTG), an estrogen-inducible gene with high transforming properties originally isolated from the rat pituitary cell line and subsequently found to be expressed at high levels particularly in invasive hormone-secreting tumors *(38,39)*. Structural characterization has identified PTTG as a member of the securin family. PTTG is an anaphase inhibitor that prevents premature chromosome separation through inhibition of separase activity *(39)*. Therefore, its degradation is required to start anaphase and separation of sister chromatids during mitosis. Due to the critical role of PTTG in maintaining genomic

stability, it has been proposed that PTTG overexpression may be, at least in part, responsible for the aneuploidy frequently observed in pituitary tumors. Moreover, PTTG participates in cellular responses to DNA damage in humans, since it has been demonstrated that securin is a downstream target of the oncosuppressor p53 *(40)*. Finally, PTTG mediates the estrogen-induced upregulation of growth factors with potent mitogenic and angiogenic activity, such as FGF-2.

The high mobility group A nonhistone chromosomal protein 2 (HMGA2) is a nuclear architectural factor that plays a critical role in a wide range of biological processes including regulation of gene expression, embryogenesis, and neoplastic transformation. Overexpression of this protein is characteristic of rapidly dividing cells in embryonic tissues and in tumors and is probably related to interaction with the retinoblastoma gene (RB). Consistent with the observations that HMGA2 overexpression causes GH-secreting and PRL-secreting adenomas in transgenic animals and that high levels of HMGA2 protein are present in human prolactinomas, it has been suggested that this protein may be implicated in lactotroph proliferation *(41,42)*.

2.2.2. Overexpression of growth factors

Several growth factors are overexpressed in pituitary tumors. In particular, transforming growth factor-α, epidermal growth factor, and their common tyrosine kinase receptor are overexpressed in pituitary adenomas, particularly in those with high aggressiveness *(3,4)*. Although in the pituitary, unlike other tissues, vascularization is lower in adenomas compared to the normal gland, high levels of growth factors with angiogenic properties such as FGF and vascular endothelial growth factor (VEGF) are detected in pituitary tumors and particularly in aggressive prolactinomas *(3,4)*. In pituitary tumors derived from the gonadotroph lineage, activin/inhibin subunits appear highly expressed together with the specific type I and type II receptors, while follistatin, which prevents activin action by binding this subunit, is reduced *(43,44)*. Accordingly, it has been proposed that the imbalanced expression of these proteins, resulting in an enhanced activin signaling, may represent a pathogenetic mechanism in the development of this adenoma subtype.

2.2.3. Overexpression of Receptors for Hypothalamic Releasing Hormones

Pituitary function is under the strict control of hypothalamic neurohormones that are required for pituitary cell commitment and growth as well as hormone synthesis and release. It is a common clinical observation that ectopic overproduction of releasing hormones, such as GHRH or CRH, results in proliferation of the target cells. However, the vast majority of sporadic pituitary tumors do

not show hyperplasia in the surrounding tissue. Although these data suggest that hormonal stimulation is not a primary etiologic mechanism in pituitary tumorigenesis, it is worth noting that aggressive GH-secreting adenomas frequently express high intrapituitary amounts of GHRH *(45)*.

Receptors of hypothalamic neurohormones have been extensively investigated for either activating mutations or overexpression, both of which could mimic states of hormone excess. Studies carried out on a large series of functioning and nonfunctioning adenomas failed to identify mutational changes in the genes encoding TRH, GnRH, CRH and V3 receptors, while variants of the GHRH receptor devoid of any pathogenetic relevance have been found in about 20% of GH-secreting adenomas *(46)*. In contrast to the absence of mutational changes, these receptors are frequently overexpressed. Indeed, high levels of V3 and CRH receptor have been detected in ACTH-secreting adenomas, whereas most functioning and nonfunctioning adenomas possess TRH, GnRH, VIP, and PACAP receptors, normally coupled to intracellular effectors *(47,48)*.

3. LOSS OF ANTIPROLIFERATIVE SIGNALS

Proliferation may result from either inactivating mutations of common tumor suppressors or specific pituitary inhibitors, or epigenetic disruption of gene expression at mRNA or protein levels (Table 2).

3.1. Inactivating Mutations of Antiproliferative Signals

Few genetic defects have been so far identified in tumor-suppressor genes to confer constitutive activation of protooncogenes, while downregulation of inhibitory molecules at mRNA or protein levels is not a rare event.

3.1.1. Inactivating Mutations of Tumor-Suppressor Genes

According to the "two-hit" hypothesis, loss of tumor-suppressor genes requires a first "hit," represented by a germline or a somatic mutation, followed by a second "hit," that is usually a somatic deletion of the second allele in the involved tissue. This results in loss of heterozygosity (LOH), although evidence suggests other pathogenetic mechanisms beyond this hypothesis *(49)*. In pituitary tumors, LOH occurs with relatively high frequency (15–30%) and in several loci, such as 10q26, 11q13, 11p, 13q, and 22q13 *(2)*. However, the search for mutations of known antioncogenes in the retained allele has failed to reveal inactivating mutations in most cases. Indeed, in contrast to the pituitary tumor development observed in the knockout mice for RB and for p27Kip1, a cyclin-dependent kinase inhibitor that induces G1 arrest by RB hypophosphorylation, and the frequent LOH on chromosomes where these

Table 2
Loss-of-function Events In sporadic Pituitary Tumors

Human pituitary tumor	*Gene*	*Defect*
Aggressive adenomas	RB	Promoter methylation
All types	p16INK4a	Promoter methylation
ACTH-omas	p27Kip1	Reduced expression
TSH-omas	TRβ	Inactivating mutations
GH-omas	AIP	Inactivating mutations
NFPA	ZAC	LOH
ACTH-omas	GR	LOH
GH-omas	PRKAR1A	Reduced expression (in sporadic tumors) Inactivating mutation (in Carney Complex)
Resistant PRL-omas	D2R	Reduced expression
Resistant GH-omas	Sst2	Reduced expression

RB, retinoblastoma; LOH, loss of heterozygosity; D2R, dopamine receptor type 2; sst2, somatostatin receptor type 2; TRβ, thyroid hormone receptor β; GR, glucocorticoid receptor; AIP, aryl hydrocarbon receptor interacting protein; PRKAR1A, type 1 alpha regulatory subunit of protein kinase A.

genes are located *(50,51)*, no inactivating mutation of these genes has been reported so far *(50–52)*. Similarly, no mutation in the tumor-suppressor *p53* gene, the most frequently altered oncosuppressor gene in human neoplasia, has been ever found in human pituitary tumors *(53)*.

Since pituitary tumors are part of multiple endocrine neoplasia syndromes, such as MEN1 and Carney complex, the two genes responsible for the diseases, i.e., MEN1 and type 1α regulatory subunit of protein kinase A (PRKAR1A), have been screened for mutations in sporadic pituitary adenomas, yielding negative results *(54,55)*. However, consistent with the finding that LOH in the region 11q13, where MEN1 locus is located, is present in 10–20% of sporadic pituitary adenomas, genetic abnormalities in this region have been reported recently *(56)*. By combining chip-based technologies with genealogy data, germline loss-of-function mutations in the aryl hydrocarbon receptor (AHR) interacting protein (AIP) gene in individuals with pituitary adenoma predisposition have been reported recently. In particular, in a population-based series from Northern Finland, two AIP mutations accounted for 16% of all patients diagnosed with GH-secreting adenomas and for 40% of the affected patients younger than 35 years of age. AIP forms a complex with the AHR, a ligand-activated transcription factor that regulates a variety of xenobiotic metabolizing enzymes and mediates most of the toxic responses of dioxin-like chemicals.

However, the mechanisms by which AIP exerts its tumor-suppressive action in the pituitary remain to be determined. Recently, the occurrence of inactivating mutations of this tumor-suppressor gene was not confirmed in a series of US patients *(57)*.

3.1.2. Inactivating Mutations of Components of the Negative Feedback

It is well established that negative feedback is a potent inhibitory mechanism of both hormone secretion and cell growth. However, few genetic mutations have been identified to support the hypothesis that poor sensitivity to peripheral hormones is responsible for pituitary cell proliferation. Only one mutation of the glucocorticoid receptor (GR) has been far reported in one macroadenoma from a patient with Nelson's syndrome *(58)*. However, LOH at the GR gene locus is present in about a third of ACTH-secreting adenomas, suggesting a possible role of GR allelic deletion in glucocorticoid resistance and corticotroph tumorigenesis *(59)*. Similarly, the reduced inhibition of TSH secretion by T3 in TSH-secreting adenomas has been associated with mutations in the thyroid hormone receptor β isoform (TRβ), causing lack of T3 binding in two tumors *(60)*.

3.2. Downregulation of Antiproliferative Signals

The infrequent occurrence of mutations in genes encoding components of antiproliferative pathways strongly suggests that posttrascriptional events may cause antioncogene silencing by reducing mRNA/protein expression or stability. Indeed, epigenetic disruption and downregulation of common tumor-suppressor genes, probably due to gene promoter methylation as well as pituitary specific inhibitory signals, frequently occurs in pituitary tumors, although its relevancy in pituitary tumorigenesis remains uncertain.

3.2.1. Downregulation of Tumor-Suppressor Genes

Investigation of possible defects in RB occurring at the RNA or protein level in pituitary tumor tissues yielded contradictory results, with some immunohistochemical studies reporting low RB protein levels and other studies not confirming these data *(52,61)*. The low expression of p27Kip1 protein found in recurrent pituitary tumors and pituitary carcinomas by immunohistochemistry and not by mRNA analysis was consistent with protein degradation rather than reduced transcription *(62)*. A similar reduced expression, probably depending on methylation within the exon 1 CpG island, affects p16INK4a, another cyclin-dependent kinase inhibitor that prevents RB phosphorylation *(63)*. A widely expressed zinc finger protein named ZAC that shows transactivation

and DNA-binding activities and that, like p53, inhibits tumor cell proliferation has been found highly expressed in the normal anterior pituitary gland but downregulated in most pituitary adenomas *(64)*.

3.2.2. Downregulation of Inhibitory Signals

In addition to the component of the negative feedback, other hormones and receptors that physiologically inhibit pituitary hormone secretion may be considered as possible targets for inactivating mutations with pathogenetic impact. The best candidates among these are the dopaminergic D2 receptor (D2R) and the somatostatin receptor (sst) type 1–3 and 5.

Although the development of prolactinomas in D2R-deficient mice strongly suggests that inactivating mutations of this receptor might results in lactotroph proliferation *(65)*, studies carried out on prolactinomas, including those resistant to dopaminergic drugs, failed to find mutations in the D2R gene *(66)*. Conversely, resistant prolactinomas frequently show a reduction of D2R transcript, and particularly of the shortest isoform that is more efficiently coupled to phospholipase C *(67)*. In addition to the defect in D2R mRNA splicing and expression, the absence of D2R protein due to increased instability and degradation has been observed in metastases of a malignant prolactinoma resistant to different dopamine agonists *(68)*.

In analogy with the poor, if any, evidence of mutations in D2R, mutational changes of the sst genes seem to occur rarely. In fact, only one mutation in the sst5 gene has been identified so far in one octreotide-resistant acromegalic patient *(69)*. In the absence of mutations, several expression studies suggest that the different degree of responsiveness to somatostatin analog observed in acromegalic patients is probably related to the level of expression of somatostatin receptors. In particular, poor responsiveness to treatment seems to correlate with a low expression of sst2, while the role of sst5, the most highly expressed somatostatin receptor in normal and adenomatous somatotrophs, is still controversial *(70)*.

In addition to receptors, molecules that participate in the transduction of extracellular signals may have inhibitory functions. In particular, molecules that are involved in the negative control of the cAMP cascade may be considered as putative antioncogenes in tissues where cAMP is mitogenic, such as the pituitary. Accordingly, inactivating mutations of PRAKAR1A, the gene encoding the type 1A regulatory subunit of protein kinase A, that render the catalytic subunit more susceptible to activation by cAMP have been identified in patients with Carney complex, a multiple neoplasia syndrome that includes pituitary tumors. Although subsequent studies failed to identify mutations of PRKAR1A in sporadic pituitary adenomas *(54,55)*, the low expression of the

wild-type subunit due to proteasome-mediated degradation induces cAMP-dependent cell proliferation in GH-secreting adenomas *(71)*.

4. CONCLUSIONS

In the last years several candidate factors have been implicated in the genesis and progression of pituitary adenomas. To date, GNAS1 is the only gene that has been identified as a target for activating mutations that unequivocally cause cell proliferation in about 30–40% of GH-secreting adenomas. Abnormalities in the expression of cell cycle regulators, receptors, and growth factors and their signaling have been proposed to play a relevant role in cell transformation and/or clonal expansion. It is tempting to speculate that no single factor might effectively explain tumorigenesis in the pituitary.

REFERENCES

1. Farrel WE, Clayton RN. Molecular pathogenesis of pituitary tumors. Front Neuroendocrinol 2000;21:174–98.
2. Lania A, Mantovani G, Spada A. G protein mutations in endocrine diseases. Eur J Endocrinol 2001145, 543–59.
3. Asa SL, Ezzat S. The pathogenesis of pituitary tumours. Nat Rev Cancer 2002;2:836–49.
4. Ezzat S, Asa SL. Mechanisms of disease: The pathogenesis of pituitary tumors. Nat Clin Pract 2006;2:220–30.
5. Alexander JM, Biller BMK, Bikkal H, Zervas NT, Arnold A, Klibanski A. Clinically non functioning pituitary adenomas are monoclonal in origin. J Clin Invest 1990; 86:336–40.
6. Herman V, Fagin J, Gonsky R, Kovacs K, Melmed S. Clonal origin of pituitary adenomas. J Clin Endocrinol Metab 1990;71:1427–33.
7. Burrow GN, Wortzman G, Rewcastle NB, Hodgate RC, Kovacs K. Microadenomas of the pituitary and abnormal sella tomograms in an unselected autopsy series. N Engl J Med 1981;304:156–8.
8. Elster AD. Modern imaging of the pituitary. Radiology 1993;187:1–14.
9. Karga HJ, Alexander JM, Hedley-Whyte ET, Klibanski A, Jameson JL. Ras mutations in human pituitary tumors. J Clin Endocrinol Metab 1992;74:914–9.
10. Cai WY, Alexander JM, Hedley-Whyte ET, et al. Ras mutations in human prolactinomas and pituitary carcinomas. J Clin Endocrinol Metab 1994;78:89–93.
11. Pei L, Melmed S, Scheithauer B, Kovacs K, Prager D. H-ras mutations in human pituitary carcinoma metastasis. J Clin Endocrinol Metab 1994;78:842–6.
12. Vallar L, Spada A, Giannattasio G. Altered Gs and adenylate cyclase activity in human GH-secreting pituitary adenomas. Nature 1987;330:566–7.
13. Landis C, Masters SB, Spada A, Pace AM, Bourne HR, Vallar L. GTPase inhibiting mutations activate the alpha chain of Gs and stimulate adenylyl cyclase in human pituitary tumours. Nature 1989;340:692–6.
14. Spada A, Arosio M, Bochicchio D, et al. Clinical, biochemical, and morphological correlates in patients bearing growth hormone-secreting pituitary tumors with or without constitutively active adenylyl cyclase. J Clin Endocrinol Metab 1990;71:1421–6.

15. Adams EF, Brockmeier S, Friedmann E, Roth M, Buchfelder M, Fahlbusch R. Clinical and biochemical characteristics of acromegalic patients harboring gsp-positive and gsp-negative pituitary tumors. Neurosurgery 1993;33:198–201.
16. Yang I, Park S, Ryu M, et al. Characteristics of gsp-positive growth hormone-secreting pituitary tumors in Korean acromegalic patients Eur J Endocrinol 1996;134:720–6.
17. Barlier A, Gunz G, Zamora AJ, et al. Prognostic and therapeutic consequences of Gs alpha mutations in somatotroph adenomas. J Clin Endocrinol Metab 1998;83:1604–10.
18. Lania A, Persani L, Ballaré E, Mantovani S, Losa M, Spada A. Constitutively active $G_{s\alpha}$ is associated with an increased phosphodiesterase activity in human growth hormone secreting adenomas. J Clin Endocrinol Metab 1998;83:1624–8.
19. Persani L, Borgato S, Lania A, et al. Relevant cAMP-specific phosphodiesterase isoforms in human pituitary: effect of Gs(alpha) mutations. J Clin Endocrinol Metab 2001;86: 3795–800.
20. Bertherat J, Chanson P, Montiminy M. The cyclic adenosine 3'-5'-monophosphate-responsive factor CREB is constitutively activated in human somatotrophs. Mol Endocrinol 1995;9:777–83.
21. Peri A, Conforti B, Baglioni-Peri S, et al. Expression of cyclic adenosine 3',5'-monophosphate (cAMP)-responsive element binding protein and inducible-cAMP early repressor genes in growth hormone-secreting pituitary adenomas with or without mutations of the Gs alpha gene. J Clin Endocrinol Metab 2001;86:2111–7.
22. Ballare E, Mantovani S, Lania A, Di Blasio AM, Vallar L, Spada A. Activating mutations of the Gs alpha gene are associated with low levels of Gs alpha protein in growth hormone-secreting tumors. J Clin Endocrinol Metab 1998;83:4386–90.
23. Hayward BE, Kamiya M, Strain L, et al. The human GNAS1 gene is imprinted and encodes distinct paternally and biallelically expressed G proteins. Proc Natl Acad Sci USA 1998;95:10038–43.
24. Hayward B, Bonthron DT. An imprinted antisense transcript at the human GNAS1 locus. Hum Mol Genet 2000;9:835–41.
25. Hayward BE, Barlier A, Korbonits M, et al. Imprinting of the G(s)alpha gene GNAS1 in the pathogenesis of acromegaly. J Clin Invest 2001;107:R31–6.
26. Mantovani G, Ballare E, Giammona E, Beck-Peccoz P, Spada A. The $G_{s\alpha}$ gene: predominant maternal origin of transcription in human thyroid gland and gonads. J Clin Endocrinol Metab 2002;87:4736–40.
27. Mantovani G, Bondioni S, Lania AG, et al. Parental origin of Gsalpha mutations in the McCune-Albright syndrome and in isolated endocrine tumors. J Clin Endocrinol Metab 2004;89:3007–9.
28. Tordjman K, Stern N, Ouaknine G, et al. Activating mutations of the Gs alpha gene in non functioning pituitary adenomas. J Clin Endocrinol Metab 1993;77:765–9.
29. Williamson EA, Ince PG, Harrison D, Kendall-Taylor P, Harris PE. G-protein mutations in human adrenocorticotrophic (ACTH) hormone-secreting adenomas. Eur J Clin Invest 1995;25:128–31.
30. Williamson EA, Daniels M, Foster S, Kelly WF, Kendall-Taylor P, Harris PE. Gs alpha and Gi alpha mutations in clinically non-functioning pituitary tumours. Clin Endocrinol 1994;41:815–20.
31. Petersenn S, Heyens M, Ludecke DK, Beil FU, Schulte HM. Absence of somatostatin receptor type 2 A mutations and gip oncogene in pituitary somatotroph adenomas Clin Endocrinol (Oxf) 2000;52, 35–42.
32. Alvaro V, Levy L, Dubray C, et al. Invasive human pituitary tumors express a point-mutated alpha-protein kinase C. J Clin Endocrinol Metab 1993;77:1125–9.

33. Dong Q, Brucker-Davis F, Weintraub BD, et al. Screening of candidate oncogenes in human thyrotroph tumors: absence of activating mutations of the G alpha q, G alpha 11, G alpha s, or thyrotropin-releasing hormone receptor genes. J Clin Endocrinol Metab 1996;81:1134–40.
34. Ezzat S, Zheng L, Zhu XF, Wu GE, Asa SL. Targeted expression of a human pituitary tumor-derived isoform of FGF receptor-4 recapitulates pituitary tumorigenesis. J Clin Invest 2002;109:69–78.
35. Cavallaro U, Niedermeyer J, Fuxa M, Christofori G. N-CAM modulates tumour-cell adhesion to matrix by inducing FGF-receptor signalling. Nat Cell Biol 2001;3:650–7.
36. Jordan S, Lidhar K, Karbonits M, Lowe DG, Grossman AB. Cyclin D and cyclin E expression in normal and adenomatous pituitary. Eur J Endocrinol 2000;143:R1–6.
37. Hibberts NA, Simpson DJ, Bicknell JE, et al. Analysis of cyclin DI (CCND1) allelic imbalance and overexpression in sporadic pituitary tumors. Clin Cancer Res 1999;5: 2133–9.
38. Pei L, Melmed S. Isolation and characterization of a pituitary tumor-transforming gene (PTTG). Mol Endocrinol 1997;11:433–41.
39. Chen LL, Puri R, Lefkowitz EJ, Kakar SS. Identification of the human pituitary tumor transforming gene (HPTTG) family: molecular structure, expression, and chromosomal localization. Gene 2000;246:41–50.
40. Zhou Y, Mehta KR, Choi AP, Scolavino S, Zhang X. DNA damage-induced inhibition of securin expression is mediated by p53. J Biol Chem 2003;278:462–70.
41. Fedele M, Battista S, Kenyon L, et al. Overexpression of the HMGA2 gene in transgenic mice leads to the onset of pituitary adenomas. Oncogene 2002;21:3190–8.
42. Finelli P, Pierantoni GM, Giardino D, et al. The High Mobility Group A2 gene is amplified and overexpressed in human prolactinomas. Cancer Res 2002;62, 2398–405.
43. Danila DC, Inder WJ, Zhang X, et al. Activin effects on neoplastic proliferation of human pituitary tumors. J Clin Endocrinol Metab 2000;85:1009–15.
44. Wessels HT, Hofland LJ, van der Wal R, et al. In vitro secretion of FSH by cultured clinically nonfunctioning and gonadotroph pituitary adenomas is directly correlated with locally produced levels of activin A. Clin Endocrinol (Oxf) 2001;54:485–92.
45. Thapar K, Kovacs K, Stefaneau L, et al. Overexpression of the growth-hormone-releasing hormone gene in acromegaly associated pituitary tumors. An event associated with neoplastic progression and aggressive behavior. Am J Pathol 1997;151:769–84.
46. Lee EJ, Kotlar TJ, Ciric I, et al. Absence of constitutively activating mutations in the GHRH receptor in GH-producing pituitary tumors. J Clin Endocrinol Metab 2001;86:3989–95.
47. de Keyzer Y, Rene P, Beldjord C, Lenne F, Bertagna X. Overexpression of vasopressin (V3) and corticotrophin-releasing hormone receptor genes in corticotroph tumours. Clin Endocrinol (Oxf) 1998;49:475–82.
48. Spada A, Reza Elahi F, Lania A, Gil del Alamo P, Bassetti M, Faglia G. Hypothalamic peptides modulate cytosolic free Ca^{2+} levels and adenylyl cyclase activity in human nonfunctioning pituitary adenomas. J Clin Endocrinol Metab 1991;71:913–8.
49. Tucker T, Friedman JM. Pathogenesis of hereditary tumors: beyond the "two-hit" hypothesis. Clin Genet 2002;62:345–57.
50. Jacks T, Fazeli A, Schmitt EM, Bronson RT, Goodell MA, Weinberg RA Effects of an Rb mutation in the mouse. Nature 1992;359:295–300.
51. Nakayama K, Ishida N, Shirane M, et al. Mice lacking p27(Kip1) display increased body size, multiple organ hyperplasia, retinal dysplasia, and pituitary tumors. Cell 1996;85: 707–72.

52. Simpson DJ, Magnay J, Bicknell JE, et al. Chromosome 13q deletion mapping in pituitary tumors: infrequent loss of the retinoblastoma susceptibility gene (RB1) despite loss of RB1 product in somatotropinomas. Cancer Res 1999;59:1562–6.
53. Levy A, Hall L, Yeudall WA, Lightman SL. p53 gene mutations in pituitary adenomas: rare events. Clin. Endocrinol 1994;41:809–14.
54. Tanaka C, Kimura T, Yang P, et al. Analysis of loss of heterozygosity on chromosome 11 and infrequent inactivation of the MEN-1 gene in sporadic pituitary adenomas. J Clin Endocrinol Metab 1998;83:2631–4.
55. Kaltsas GA, Kola B, Borboli N, et al. Sequence analysis of the PRKAR1A gene in sporadic somatotroph and other pituitary tumours. Clin Endocrinol (Oxf) 2002;57:443–8.
56. Vierimaa O, Georgitsi M, Lehtonen R, et al. Pituitary adenoma predisposition caused by germline mutations in the AIP gene. Science 2006;312:1228–30.
57. Yu R, Bonert V, Saporta I, Raffel LJ, Melmed S. Aryl hydrocarbon receptor protein variants in sporadic pituitary sporadic adenomas. J Clin Endocrinol Metab 2006;91:5126–9.
58. Karl M, Lamberts SW, Koper JW, et al. Cushing's disease preceded by generalized glucocorticoid resistance: clinical consequences of a novel, dominant-negative glucocorticoid receptor mutation. Proc Assoc Am Physicians 1996;108:296–307.
59. Huizenga NA, de Lange P, Koper JW, et al. Human adrenocorticotropin-secreting pituitary adenomas show frequent loss of heterozygosity at the glucocorticoid receptor gene locus. J Clin Endocrinol Metab 1998;83:917–21.
60. Ando S, Sarlis NJ, Oldfield EH, Yen PM. Somatic mutation of TRbeta can cause a defect in negative regulation of TSH in a TSH-secreting pituitary tumor. J Clin Endocrinol Metab 2001;86:5572–6.
61. Simpson DJ, Hibberts NA, McNicol AM, Clayton RN, Farrell WE. Loss of pRb expression in pituitary adenomas is associated with methylation of the RB1 CpG island. Cancer Res 2000;60:1211–6.
62. Bamberger CM, Fehn M, Bamberger AM, et al. Reduced expression levels of the cell-cycle inhibitor p27Kip1 in human pituitary adenomas. Eur J Endocrinol 1999;140:250–5.
63. Simpson DJ, Bicknell JE, McNicol AM, Clayton RN, Farrell WE. Hypermethylation of the p16/CDKN2A/MTSI gene and loss of protein expression is associated with nonfunctional pituitary adenomas but not somatotrophinomas. Genes Chromosomes Cancer 1999;24: 328–36.
64. Pagotto U, Arzberger T, Theodoropoulou M, et al. The expression of the antiproliferative gene ZAC is lost or highly reduced in nonfunctioning pituitary adenomas. Cancer Res 2000;60:6794–9.
65. Asa SL, Kelly MA, Grandy DK, Low MJ. Pituitary lactotroph adenomas develop after prolonged lactotroph hyperplasia in dopamine D2 receptor-deficient mice. Endocrinology 1999;140:5348–55.
66. Friedman E, Adams EF, Hoog A, et al. Normal structural dopamine type 2 receptor gene in prolactin-secreting and other pituitary tumors. J Clin Endocrinol Metab 1994;78:568–74.
67. Caccavelli L, Feron F, Morange I, et al. Decreased expression of the two D2 dopamine receptor isoforms in bromocriptine-resistant prolactinomas. Neuroendocrinology 1994;60:314–22.
68. Winkelmann J, Pagotto U, Theodoropoulou M, et al. Retention of dopamine 2 receptor mRNA and absence of the protein in craniospinal and extracranial metastasis of a malignant prolactinoma: a case report. Eur J Endocrinol 2002;146:81–8.
69. Ballare E, Persani L, Lania AG, et al. Mutation of somatostatin receptor type 5 in an acromegalic patient resistant to somatostatin analog treatment. J Clin Endocrinol Metab. 2001;86:3809–14.

70. Jaquet P, Saveanu A, Gunz G, et al. Human somatostatin receptor subtypes in acromegaly: distinct patterns of messenger ribonucleic acid expression and hormone suppression identify different tumoral phenotypes. J Clin Endocrinol Metab 2000;85:781–92.
71. Lania AG, Mantovani G, Ferrero S, et al. Proliferation of transformed somatotroph cells related to low or absent expression of protein kinase a regulatory subunit 1A protein. Cancer Res 2004;64:9193–8.