ABSTRACT

The pituitary is an organ of dual origin. The anterior lobe (adenohypophysis) is epithelial in origin, whereas the posterior lobe (neurohypophysis) derives from the neural ectoderm. Precise spatial and temporal co-ordination of transcription factor expression in both structures is critical for pituitary formation and the differentiation of hormone-producing cells. Disruption of this regulation, for instance by mutation, can lead to numerous developmental disorders. We provide an overview of the molecular drivers of pituitary organogenesis and illustrate the anatomy and histology of the mature pituitary, comprising adenohypophysis (anterior lobe), neurohypophysis (posterior lobe), pars intermedia and infundibulum (pituitary stalk). For complete coverage of this and related areas of Endocrinology, please visit our free web—book, www.endotext.org.

PITUITARY ORGANOGENESIS

The pituitary is an organ of dual origin. The anterior lobe (adenohypophysis) is derived from oral ectoderm and is epithelial in origin, whereas the posterior lobe (neurohypophysis) derives from the neural ectoderm. The composite nature of the pituitary requires that the neural and oral ectoderm interact physically and developmentally. Precise spatial and temporal co-ordination and regulation of signals from both structures is critical for pituitary formation and the differentiation of the various hormone-producing cell types in the anterior lobe. The expression of transcription factors that control cell lineage commitment in the developing anterior lobe must be precisely regulated to ensure correct differentiation of hormone-producing cell types; the iterative and cumulative nature of this regulation renders it extremely sensitive to perturbation. Disruption of this process, for instance by mutation, can lead to numerous developmental disorders from congenital forms of hypopituitarism to pituitary tumours. Pituitary organogenesis is covered briefly below. For further description, please see Chapter 3c (Functional anatomy of the hypothalamus and pituitary Ronald M. Lechan, and Roberto Toni).

Pituitary organogenesis begins during week 4 of fetal development. A thickening of cells in the oral ectoderm form the hypophyseal placode, which gives rise to Rathke’s pouch, an upward evagination that extends towards the neural ectoderm. At the same time, a downward extension of the ventral diencephalon forms the posterior
lobe and the two nascent lobes connect to form the composite structure of the adult pituitary. Rathke’s pouch constricts at its base and eventually separates altogether from the oral epithelium during week 6-8. The cells of the anterior wall of Rathke’s pouch undergo extensive proliferation to form the anterior lobe while the posterior wall proliferates more slowly to form the vestigial (in humans) intermediate lobe. Cell patterning and terminal differentiation occurs within the anterior lobe to form the five principal specialised endocrine cell types of the pituitary gland.

**Transcriptional Control of Pituitary Organogenesis**

Development of the pituitary occurs broadly in three stages:

1. Initiation of pituitary organogenesis and formation of Rathke’s pouch
2. Evagination of Rathke’s pouch and cell proliferation
3. Lineage determination and cellular differentiation

Much of our understanding of the process of pituitary organogenesis comes from mouse studies, but the phenotypes associated with human disorders often share aspects with mouse models of defective pituitary development. Various transcription factors involved are presented in Table 3a-1. Reviewed extensively in (1-3).
Table 3a-1: Signalling molecules controlling pituitary organogenesis and associated dysfunction

<table>
<thead>
<tr>
<th>Developmental Stage</th>
<th>Factor</th>
<th>Function</th>
<th>Dysfunction</th>
<th>Comments</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initiation of pituitary organogenesis and formation of Rathke’s pouch</td>
<td>SIX homeodomain proteins</td>
<td>Six1- Six6</td>
<td>Family of six transcriptional activators/ inhibitors Functional role difficult to determine due to redundancy and severity of mutations</td>
<td>Expression persists in adult pituitary; may mediate plasticity</td>
<td>(4,5)</td>
</tr>
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<td>Paired-like homeobox proteins</td>
<td>Hesx1</td>
<td>Transcriptional repressor Early marker of Rathke’s pouch Downregulation essential for endocrine cell differentiation</td>
<td>Mutations in patients with hypopituitarism including septo-optic dysplasia, combined pituitary hormone deficiency (CPHD) and isolated growth hormone deficiency (IGHD)</td>
<td>Expression activated by LIM homeodomain proteins</td>
<td>(6)</td>
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<td></td>
<td>Otx2</td>
<td>Transcription factor that regulates Hesx1</td>
<td>Mutations found in patients with ocular disorders (e.g. anophthalmia, microphthalmia) with or without hypopituitarism. In mice, deficiency results in craniofacial defects and pituitary gland dysmorphology, but normal pituitary cell specification</td>
<td></td>
<td>(7,8)</td>
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<td></td>
<td>Pitx 1/2/3</td>
<td>Interacts with various other factors to determine cell lineage</td>
<td>Mutations in Pitx2 (R91P) found in patients with Axenfeld-Rieger syndrome. Blocks expression of LH β</td>
<td>Expressed throughout oral ectoderm and Rathke’s pouch.</td>
<td>(9)</td>
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<tr>
<td>LIM homeodomain transcription factors</td>
<td>Isl1</td>
<td>Involved in cell lineage specification</td>
<td>No human mutations identified. Null mice do not develop Rathke’s pouch</td>
<td>First LIM protein to be expressed</td>
<td>Reviewed in (1)</td>
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<tr>
<td>Lhx3</td>
<td></td>
<td>Expression gradient required for differentiation of endocrine cell types</td>
<td>Heritable mutation in patients with CPHD with short, stiff neck and sensorineural hearing loss</td>
<td>Broad temporal and spatial expression pattern with many target genes</td>
<td>(10)</td>
</tr>
<tr>
<td>Lhx4</td>
<td></td>
<td>Expression gradient required for differentiation of endocrine cell types</td>
<td>Heterozygous mutations in patients with CPHD. Associated with pituitary hypoplasia, small sella and Arnold-Chiari malformation</td>
<td>Not critical for endocrine cell differentiation</td>
<td>(11, 12)</td>
</tr>
<tr>
<td>SOX2</td>
<td></td>
<td>Expressed throughout developing Rathke’s pouch</td>
<td>Downregulation essential for endocrine cell differentiation</td>
<td>Mutations found in patients with an- or microphthalmia, hypogonadotrophic hypogonadism, and growth hormone deficiency (GHD)</td>
<td>(13, 14)</td>
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<td></td>
<td></td>
<td></td>
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<td>Both duplications and loss of function mutations associated with hypopituitarism</td>
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<tr>
<td><strong>β-catenin</strong></td>
<td>Signalling activates Pitx2 expression promoting pituitary precursor proliferation. Required for Pit1 lineage determination and anterior pituitary formation</td>
<td>Premature activation of β-catenin results in Hesx1 repression and pituitary gland agenesis in mouse. Activating mutation of β-catenin leads to pituitary progenitor proliferation, loss of Pit1 lineage cells and adamantinomatous craniopharyngioma</td>
<td>High degree of interaction with other signalling pathways e.g. Notch. Not required for cell lineage determination</td>
<td>(15, 6)</td>
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<td><strong>Notch</strong></td>
<td>Mediates lateral inhibition and cell lineage determination. Activates Hes1 expression</td>
<td>Dysregulation of the pathway associated with premature corticotroph differentiation and pituitary hypoplasia in mice</td>
<td>Expression in the adult gland co-localises with SOX2</td>
<td>(17)</td>
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<td><strong>Bone morphogenic proteins</strong></td>
<td>BMP4 Expressed in ventral diencephalon. Required for hypophyseal placode formation</td>
<td>Downregulation results in arrested development of Rathke’s pouch in mice</td>
<td></td>
<td>Revied in (1)</td>
<td></td>
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<td><strong>BMP2</strong></td>
<td>Induces Isl1 expression. Downregulation required for cell differentiation</td>
<td>Prolonged expression results in hyperplastic pituitary and lack of terminal differentiation</td>
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<td>(18)</td>
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<td><strong>Fibroblast growth factors</strong></td>
<td>FGF8, 10, 18 Expressed in the posterior pituitary. Required for Lhx3 and Lhx4 expression and cell differentiation</td>
<td>In humans, mutations of FGF8 and its receptor are associated with Kallmann syndrome, resulting in isolated hypogonadotrophic</td>
<td></td>
<td>(19-21)</td>
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<tr>
<td>Gene</td>
<td>Function</td>
<td>Mutations</td>
<td>References</td>
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<td>Shh</td>
<td>Expressed in oral ectoderm and ventral diencephalon. Induces Lhx3 expression.</td>
<td>Antagonism in mouse oral ectoderm results in hypoplastic Rathke’s pouch.</td>
<td>(22)</td>
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<td>Prop1</td>
<td>Transcriptional activator and suppressor depending on context. Activates POU1F1 expression and switches developmental process from proliferation to differentiation.</td>
<td>Mutations are most common cause of CPHD in humans.</td>
<td>Revied in (1) and (23) (24-27)</td>
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<td>POU1F1 (Pit1)</td>
<td>Expressed in cells committed to somatotroph, lactotroph and thyrotroph lineage. Inhibits GATA2 and prevents gonadotroph cell fate.</td>
<td>Mutations in humans associated with GH PRL, TSH deficiency and small anterior pituitary. Mutations rarely present in sporadic CPHD and more common in familial CPHD.</td>
<td>Required for GH PRL, TSHβ expression. (26, 8-3)</td>
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<td>GATA2</td>
<td>Specifies gonadotroph and thyrotroph lineages. Induces expression of Nr5a1.</td>
<td>In mice, overexpression associated with gonadotroph and thyropro hypoplasia.</td>
<td>Expression persists in adult gland. (28)</td>
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<td>Nr5a1 (SF1)</td>
<td>Expressed throughout adrenal and reproductive axes. Regulates expression of GnRHR, LH, FSH and αGSU. Expression necessary for gonadotroph differentiation.</td>
<td>Mutations associated with 46XY sex reversal with adrenal failure, 46XY gonadal dysgenesis and 46XX ovarian insufficiency and premature ovarian failure in humans.</td>
<td>(31, 2)</td>
<td></td>
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<td>Tbx19 (TPIT)</td>
<td>Activates POMC expression in association with PITX1</td>
<td>Mutations are commonest cause of isolated ACTH deficiency in humans</td>
<td>Antagonists to Nr5a1 can prevent gonadotroph cell fate</td>
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(33, 4)
Figure 3a-1: Signalling molecules and transcription factors control pituitary development. Arrows represent regulation of expression in the direction indicated.
ANATOMY AND HISTOLOGY OF THE MATURE PITUITARY GLAND

Macroscopic Anatomy
The pituitary gland, or hypophysis cerebri, is an oval body approximately 12mm in transverse and 8mm in anterior-posterior diameter weighing approximately 500mg (Gray’s anatomy p380, 39th edition). The anterior lobe of the pituitary is generally smaller in men than women, and nullipara than multipara; during pregnancy the gland may increase by approximately 30% due to lactotroph hyperplasia. The hypophysis is connected to the brain via the infundibulum, a tubular structure arising from the tuber cinereum and median eminence of the hypothalamus. The gland rests in the sella turcica (pituitary fossa) of the sphenoid bone and is covered superiorly by the diaphragma sellae (dura), laterally by the wall of the cavernous sinus, and antero-inferiorly by the posterior wall of the sphenoid sinus, which is used as the standard route for pituitary surgery (transsphenoidal adenectomy (TSA)). Antero-superior the pituitary lies in close proximity to the optic chiasm; this explains why space-occupying lesions of the pituitary commonly present with bitemporal hemianopia.

Figure 3a-2: Coronal slice through the human brain at the level of the pituitary gland (left, MRI; right, post-mortem in situ appearance).
Normal Histology of the Pituitary

Adenohypophysis (Pars Anterior, Anterior Lobe)

The pituitary gland in adults has a distinct histological appearance, reflecting its divergent origin. The adenohypophysis (pars anterior, anterior lobe) is characterised by well-demarcated acini that usually contain a mixture of different hormone-producing cells. This nested pattern is best appreciated in reticulin preparations, which delineate the borders of the acini. The cellular heterogeneity within acini may be demonstrated histochemically in PAS-OG or preparations which were widely used before the adoption of antibody-based stains. Corticotroph cells are generally strongly basophilic (PAS-positive), somatotroph and lactotroph cells mostly acidophilic (orangeophilic), whilst gonadotrophs and thyrotrophs may be basophilic or chromophobe (reacting with neither acid nor basic stains). There is no perfect match of hormone-expression and type or degree of chromophilia; some chromophobe cells may represent degranulated chromophil cells or precursor cells (Gray’s anatomy p380, 39th edition). In a normal adult adenohypophysis approximately 10% of endocrine cells are basophils, 40% acidophils and 50% chromophobes. The PAS-OG stain is still a useful supplementary method in the differential diagnosis of some pituitary lesions (hyperplasia, corticotroph microadenoma, Crooke’s cell change or adenoma). Although most acini contain a mixture of different hormone-producing cells, there is evidence of zonation. The lateral wings of the gland mostly contain somatotrophs and lactotrophs, whilst corticotrophs are concentrated in the median mucoid wedge, which at its anterior border (the rostral tip) harbours clusters of thyrotrophs. Gonadotroph (LH/FSH) cells are diffusely scattered throughout the gland.
Figure 3a-4: Axial section of the anterior lobe of the human pituitary gland (adenohypophysis) at the level indicated by the dashed blue line in the diagram. Although there is a mixture of different hormone producing cells in most pituitary acini, the distribution of cells is not random: this is most pronounced in the ‘lateral wings’, which contain mostly somatotroph cells and the central ‘mucoid wedge’, which contains the majority of the corticotrophs. This is easily appreciated in periodic acid-Schiff / orange-G (PAS-OG) histochemistry, which stains somatotrophs yellow-orange (OG-positive) and corticotrophs purple (PAS-positive).

Follicular-stellate (FS) cells are (in the adult human pituitary) an agranular (non-hormone-producing) parenchymal component of the pars anterior. It has been postulated that they represent a stem cell capable of trans-differentiation into endocrine cells (35), but whether this is true in humans remains to be seen. FS cells are small, chromophobe, with slender processes that extend between the endocrine cells. They form small follicles at the centre of acini, comprised of apical tips of multiple FS cells. They may be visualised with S100 and GFAP antibodies, but their expression pattern is not always overlapping and may reflect different stages of maturation or function. In our hands, annexin-1 immunohistochemistry is a robust marker of FS cells. Annexin 1 (ANXA1) is a member of the annexin family of phospholipid- and calcium-binding proteins. ANXA1-positive FS cells may modulate glucocorticoid feedback loops in the anterior gland (36) or act as antigen-presenting cells.
**Figure 3a-5:** Non-endocrine cells of the anterior lobe include small folliculo-stellate (FS) cells with delicately branching processes that invest endocrine cells (left, annexin-1 staining) and (on the right) very rare cells that are postulated to represent adult pituitary stem cells (PSC, nestin staining).

**Pars Intermedia**

In contrast to rodents, the pars intermedia is rudimentary in adult humans. It represents a narrow zone between the adeno- and neurohypophysis often containing microscopic remnants of Rathke’s cleft. This zone may also contain scattered intensely PAS-positive corticotrophs, which may extend from the mucoid wedge of the adenohypophysis into the neurohypophysis. This so-called “basophil invasion” must not be confused with corticotroph microadenomas; it is believed to increase with aging and it has been suggested that these basophil cells are functionally distinct from classical ACTH-producing cells of the adenohypophysis and do not respond with hyaline degeneration (“Crooke’s cell change”) in the setting of systemic hypercortisolaemia.
**Figure 3a-6:** Axial section of the human pituitary gland at the level of the vestigial intermediate lobe (approximately representing the boxed area in the diagram). Note the cluster of remnants of Rathke’s pouch / cleft (RC). The arcs indicate the posterior (centre) and postero-medial (left and right) edges of the mucoid wedge (with scattered basophils) and pituitary wings (with scattered somatotrophs), respectively. The asterisk indicates basophil corticotrophs ‘spilling’ into the neurohypophysis (‘basophil invasion’, see figure 3a-7).

**Figure 3a-7:** ‘Basophil invasion’ of corticotrophs from the vestigial pars intermedia into the neurohypophysis (pars posterior of the pituitary gland). The dashed line represents the border between pars intermedia and pars posterior.
Neurohypophysis (Pars Posterior, Posterior Lobe)
The neurohypophysis does not contain neuroendocrine epithelial cells. Instead, it is composed of the axons arising from groups of hypothalamic neurons, most prominently those originating from magnocellular neurons of the supraoptic and paraventricular nuclei. They form the hypothalamo-hypophyseal tract and their terminals end near the sinusoids of the posterior lobe. The neurosecretory granules mostly contain oxytocin or vasopressin and form axonal beads close to their termini. Whilst it is believed that normal astrocytes may populate (at least partially) the infundibulum, the axon terminals in the neurohypophysis are supported by so-called pituicytes, which are characterised by the expression of the TTF-1 transcription factor (absent in classic GFAP-positive astrocytes). These cells show elongated processes often running in parallel with axons, and demonstrate only patchy GFAP and S100 expression.

Figure 3a-8: The cytoarchitecture of the neurohypophysis (right) is strikingly different from the adenohypophysis, which contains the nested (inset, left) collection of endocrine cells (left). The neurohypophysis does not contain neurosecretory cell bodies; instead it is composed of specialised glial cells (pituicytes) that – unique in the human brain – express the TTF-1 (thyroid transcription factor 1) protein in their nuclei (inset, top right). The neurohypophysis contains the nerve endings of the oxytocin and vasopressin producing cells of the hypothalamus. Their large distended nerve endings can be identified on routine stains as so-called Herring-bodies (arrow), named after Percy Theodore Herring (University of Edinburgh) who described them in 1908 as the ‘physiologically active principle’ of the posterior gland.

Infundibulum (Pituitary Stalk)
The stalk is a tubular (funnel-shaped) structure divided into the anterior pars tuberalis and posterior pars infundibularis. The pars tuberalis is considered to be part of the
adenohypophysis and contains a few scattered gonadotroph or corticotroph cells. It 
surrounds anteriorly and superficially the pars infundibularis (infundibular stem), which 
contains the unmyelinated axons of the magnocellular supraoptic and paraventricular 
neurons. Large intraaxonal accumulations of oxytocin and vasopressin may be seen as eosinophilic ovoid granular swellings along the trajectory of these axons in the infundibular 
stem. These structures are called “Herring bodies”. Another notable feature of the rostral portion of the stalk are tortuous capillary loops surrounding a central capillary, termed gomitoli (see example page 327, Histology for pathologists, 3rd edition). These are part of the complex hypothalamo-hypophyseal portal system.

mutations of the LHX3 gene cause combined pituitary hormone deficiencies with or without limited neck rotation. J Clin Endocrinol Metab 2007; 92:1909-1919


27. Vieira TC, Boldarine VT, Abucham J. Molecular analysis of PROP1, PIT1, HESX1, LHX3, and LHX4 shows high frequency of PROP1 mutations in patients with familial forms of combined pituitary hormone deficiency. Arquivos brasileiros de endocrinologia e metabologia 2007; 51:1097-1103


