GH producing pituitary adenoma

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GH-secreting adenomas account for approximately 20% of PAs.

Patients have signs and symptoms of gigantism or acromegaly, as well as high serum levels of GH and IGF-I. Somatotroph adenomas (SA) occur in the anterior pituitary, arising from growth hormone-producing cells, often in the lateral wings of the gland. Prolactin co-secretion by the tumor is found in approximately 30%—50% of patients and results in signs and symptoms of hyperprolactinemia.
<table>
<thead>
<tr>
<th>Adenoma type</th>
<th>Transcription Factors</th>
<th>Hormones</th>
<th>Cytokeratin</th>
</tr>
</thead>
<tbody>
<tr>
<td>GH-producing adenomas</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Densely granulated somatotroph adenoma</td>
<td>Pit-1</td>
<td>GH, a-SU</td>
<td>diffuse</td>
</tr>
<tr>
<td>Sparsely granulated somatotroph adenoma</td>
<td>Pit-1</td>
<td>GH</td>
<td>dot-like</td>
</tr>
<tr>
<td>Mammosomatotroph adenoma</td>
<td>Pit-1, ER</td>
<td>GH, PRL, a-SU</td>
<td>diffuse</td>
</tr>
<tr>
<td>Mixed somatotroph and lactotroph adenoma</td>
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<td>diffuse</td>
</tr>
</tbody>
</table>
Pit-1 cell lineage.

It is well known that GH-producing pituitary adenomas may coexpress prolactin (PRL) and, less frequently, thyroid stimulating hormone (TSH) . Production of anterior pituitary hormones in non neoplastic as well as neoplastic cells is controlled by several transcription factors and cofactors. Accumulating evidence supports the hypothesis that normal and adenoma cells expressing GH, PRL, and TSH are regulated by the pituitary-specific transcription factor-1 (Pit-1) and, therefore, belong to the Pit-1 cell lineage.
Subtypes

Histologically, monohormonal GH-producing adenomas are classified in two subtypes, densely granulated (DG) and sparsely granulated (SG), based on the density of secretory granules in the cytoplasm of the adenoma cells.
DG- and SG-type cells have different cytoskeletal features, with or without the formation of fibrous bodies, the hallmark of SG type cells, which are globular aggregations of intermediate filaments as seen on electron micrographs.

In DG type cells, CK (CAM5.2) immunostaining is Perinuclear.
Distribution of CK also varies in **plurihormonal** GH adenomas. These adenomas are histologically subcategorized by **EM** analysis. However, the subtypes are difficult to predict on the basis of the CK pattern alone.
Perinuclear predominant group (P-pre), if more than 70% of the cells had perinuclear CK immunoreactivity.

Perinuclear intermediate group (P-inter), when 30% to 70% of the cells had a dot-like CK pattern.

Adenomas in which more than 70% of the cells had a dot-like CK pattern were classified (DP).

E-cadherin is another antibody that may be helpful in differentiating between them, because there is a loss of expression in sparsely granulated GH-secreting adenomas, but not in those densely granulated.
Densely granulated adenomas were the major subtype in all of the monohormonal and plurihormonal GH adenomas:

- 70% of GH
- 89% of GH-PRL
- 85% of GH-TSH
- all of GH-PRL-TSH adenomas.
Clinical difference

There is a growing body of evidence that DGSAs and SGSAs behave differently with SGSAs being larger, more common in younger, female patients, more proliferative (higher MIB1 indices) and with a greater capacity to invade surrounding structures. Some studies have found that SGSAs are more poorly responsive to somatostatin treatment than DGSAs although the extent of the impact of tumor subtype on behaviour is unclear.
Densely granulated
Perinuclear pattern

- Densely granulated adenoma have monotonous cells with moderate acidophilic cytoplasm and PAS positive
- <30% dot like pattern
- <5% PRL positive
Sparsely granulated
dot-like pattern

Sparsely granulated adenomas consisting of smaller tumor cells with chromophobic cytoplasms and eccentric nuclei and occasionally pleomorphism.

GH is heterogeneous and less intense, and cytoplasm may show paranuclear eosinophilic structures called “fibrous bodies” (an accumulation of intermediate filaments and endoplasmic reticulum), better visualized with IHC for cytokeratins 8/18 (CAM5.2)
Mixed densely and sparsely granulated somatotroph adenoma.
Intermediate (P-inter)

Both cell types 30% to 70% of the cells had a dot-like CK pattern. Behaviour is more like densely granulated
Negative pattern
Somatotroph Adenoma With Neuronal Differentiation

A rare but pathologically intriguing subtype of SA, always associated with acromegaly and usually presenting as macroadenoma with or without hypothalamic involvement, shows sparsely granulated GH-producing cells admixed with large atypical ganglion cells. These resemble tumor cells seen in gangliocytomas and represent truly metaplastic tumor cells, as they express a mixture of lineage markers that otherwise are virtually never co-expressed (synaptophysin, neurofilament, cytokeratin and GH).

This is of no known clinical relevance and the mechanisms of trans differentiation remain unexplored.
Somatotroph Adenoma With Neuronal Differentiation
Somatostatin Analogue Effect On Somatotroph Adenomas

Densely granulated somatotroph adenomas tend to respond better to somatostatin analogue treatment than sparsely granulated tumors. This results in a distinct perivascular hyaline / fibrous reaction.

The reaction of somatotroph adenomas to somatostatin analogues is morphologically distinct to that of prolactinomas to dopamine agonists.
Somatostatin Analogue Effect On Somatotroph Adenomas
A large number of GH-secreting adenomas may show secondary immunoreactivity for other pituitary hormones (PRL, FSH, LH or TSH).
Mixed GH- and PRL-secreting adenomas

Mixed GH- and PRL-secreting adenomas account for approximately 6.5% of all Pas. Prolactin co-secretion by the tumor is found in approximately 30%--50% of GH adenoma and results in signs and symptoms of hyperprolactinemia.

Patients with these tumors show signs and symptoms of both acromegaly and hyperprolactinemia.
Mixed GH- and PRL-secreting adenomas

The diagnosis of this group of adenomas requires more complex IHC and ultrastructural analysis, and their differentiation is essential because it has clinical and prognostic implications.

Morphologically, three subtypes may be identified:

1. Mixed adenomas of cells secreting GH and cells secreting PRL.
2. Mammosomatotroph cell adenomas.
3. Acidophilic stem cell adenomas.
Mixed adenomas of cells secreting GH and cells secreting PRL

IHC shows labeling for GH and PRL with various degrees of intensity and distribution. (5% of all Pas)
At the ultrastructural level (EM), two separate cell populations are seen.
Mammosomatotroph cell adenomas

Are rare tumors (1% of all Pas)

Ultrastructural analysis shows a well-differentiated adenoma consisting of a population of monomorphic cells having characteristics of GH- and PRL secreting cells.
Acidophilic stem cell adenomas

Acidophilic stem cell adenomas are very rare and their diagnosis has great clinical importance because they may be confused with prolactinomas once most patients show characteristics of hyperprolactinemia. Histologically they are chromophobic tumors, with focal oncocytic changes in cytoplasms. IHC shows labeling for PRL and, to a lesser extent, for GH in the cytoplasm of the same tumor cells. Electron microscopy is required for the accurate identification of these adenomas, and may reveal megamitochondria responsible for the oncocytic appearance in light microscopy.
Molecular Genetics

- G protein α-subunit
- Somatostatin receptor
- Ghrelin receptor
- Aryl hydrocarbon interacting protein
- Gpr101 Mutations And X-Lag
- Signal Transducer and Activator of Transcription 3
- Micro RNA regulation
- Epigenetic regulation
Tumorigenic mechanisms in somatotroph cells. Several mechanisms increase cAMP production, which is key for somatotroph tumorigenesis. Hormones bind to receptors, including GHRH-R, SSTR, GPR101, and GIPR, on the somatotroph cell membrane and increase the activation of adenylyl cyclase through Gsα. The consequent increase in cAMP production leads to the dissociation of the regulatory subunits of PKA from the catalytic subunits, which then translocate to phosphorylate CREB in the nucleus and other targets, leading to increased GH expression and cell proliferation. Gsα activation induced by GNAS mutations also leads to upregulation of the cAMP pathway. In addition, ectopic expression of GIPR may lead to an activated cAMP pathway, and GPR101 is a Gsα-coupled constitutively active receptor that leads to increased cAMP signaling.

AIP, aryl hydrocarbon receptor-interacting protein; ATP, adenosine triphosphate; C, catalytic subunit; cAMP, cyclic adenosine monophosphate; CREB, cAMP response element; GHRH, growth hormone-releasing hormone; GHRH-R, GHRH receptor; GIPR, gastric inhibitory polypeptide receptor; GPR101, G protein-coupled receptor 101; Gsα, G protein stimulatory alpha subunit; GTP, guanine triphosphate; PKA, protein kinase A; R, regulatory subunit; SSTR, somatostatin receptor; ZAC1, zinc finger protein PLAGL1.
GNAS

- Guanine Nucleotide-Binding Protein G(s) Subunit Alpha is one of the earliest mutations to be associated with sporadic somatotroph adenomas was at the GNAS complex locus (15-58% of somatotroph adenomas)

- Gsα mutation leads inhibition of G protein and to constitutive activation of adenyl cyclase (termed the gsp oncogene) resulting in increased cAMP synthesis.

- This mutation may promote tumorigenesis since cAMP can function as a mitogenic signal.
GNAS

GNAS-mutated tumors are often smaller and less invasive, respond better to SSAs, and are usually densely granulated somatotroph adenomas. In addition, GNAS-mutated tumors have relatively high expression of dopamine receptor (DRD), which suggests a good response to dopamine agonists.

Somatic mutations in GNAS can result in McCune–Albright syndrome. This syndrome is characterized by polyostotic fibrous dysplasia, skin hyperpigmentation, and autonomous endocrine hyperfunction.
Differing expression of the somatostatin receptor between adenoma subtypes has been observed and this pattern can be influenced by somatostatin analogue (SSA) treatment.

A positive correlation has been observed between SSTR2 expression and reduction in GH after SSA treatment.

Greater expression of SSTR2 has also been associated with densely granulated adenomas, while SSTR5 was associated with sparsely granulated tumors.
Ghrelin Receptor

Ghrelin, also called “the hunger hormone “is a growth hormone secretagogue that acts on the hypothalamus and pituitary and has been associated with increased cell migration and proliferation in certain cancers.
Aryl hydrocarbon interacting protein

- Mutations in AIP (aryl hydrocarbon interacting protein) are most frequently associated with somatotroph adenomas. They are generally truncations or nonsense mutations leading to loss of function, which has resulted in the classification of AIP as a tumor suppressor gene, although the mechanism by which it functions is not yet known.
Mutation in AIP in somatotroph adenomas is associated with larger tumors and more invasive behaviour and more recurrences.

Furthermore patients with AIP mutations are relatively resistant to treatment with somatostatin analogues although the mechanism of this resistance remains to be clarified.
Treatment with SSAs leads to and is associated with upregulation of AIP expression. The mechanism for this upregulation is not fully understood, but some authors have proposed that it is ZAC1- (zinc finger regulator of apoptosis and cell cycle arrest) mediated. ZAC1 induces G1 cell cycle arrest and apoptosis.

Low levels of AIP expression have been linked to tumor invasiveness suggesting that patients with AIP mutation require more stringent follow-up.
Gpr101 Mutations And X-Lag

- A study of early childhood onset gigantism with growth hormone hypersecretion found heritable microduplications on chromosome Xq26.3. The condition was termed X-LAG or x-linked acrogigantism.

- Analysis of the expression of the genes encoded in this region in a small number of patients showed that GPR101 mRNA was upregulated by up to 1000-fold.
In a screens of 263 patients with gigantism or acromegaly and 579 patients with acromegaly, the incidence of GPR101 mutation was shown to be 1.1% and 0.69% respectively.

GPR101 encodes an orphan G-protein-coupled receptor that is predicted to bind the stimulatory G protein and regulate activation of adenylyl cyclase, resulted in increased proliferation and growth hormone secretion, along with increased cAMP signalling.
Signal Transducer and Activator of Transcription 3 (STAT3)

is a member of the STAT family, and participates in cellular responses to cytokines and growth factors. Its expression is enhanced in somatotroph adenomas, leading to GH hypersecretion, which in turn promotes STAT3 expression.

In primary human somatotroph adenoma-derived cell cultures, the specific inhibitor S3I-201 can inhibit STAT3 expression, thus decreasing GH transcription and reducing GH secretion.
Micro RNA in Somatotroph Adenomas

- Microarray analysis of somatotroph adenomas and normal pituitary gland showed significant downregulation of miR-34b, miR-326, miR-432, miR-548c-3p, miR-570 and miR-603 in adenomas.

- Among the targets of these miRNAs are high-mobility group A1 (HMGA1), HMGA2 and E2F1, genes whose activation plays a role in pituitary tumorigenesis.

- Overexpression of these miRNAs resulted in reduced growth of pituitary adenoma cell lines.
A number of studies propose an epigenetic mechanism of pituitary somatotroph tumorigenesis.

The expression of the adherens junction component E-cadherin has been shown to be significantly lower in sparsely than densely granulated adenomas and lower levels of E-cadherin correlate with larger tumor size, invasiveness, GH and IGF-1 levels and poor acute response to SSAs.
Syndromic somatotroph adenoma

- MEN1
- MEN4
- Carney complex
- X-LAG syn
- NF1