Expression of additional transcription factors is of prognostic value for aggressive behavior of pituitary adenomas

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OBJECTIVE According to the latest WHO classification of tumors of endocrine organs in 2017, plurihormonal adenomas are subclassified by their transcription factor (TF) expression. In the group of plurihormonal adenomas with unusual immunohistochemical combinations (PAWUC), the authors identified a large fraction of adenomas expressing TFs for gonadotroph adenoma (TFGA) cells in addition to other TFs. The aim of this study was to compare clinicopathological parameters of PAWUC with TFGA expression to gonadotroph adenomas that only express TFGA.

METHODS This retrospective single-center series comprises 73 patients with TFGA-positive pituitary adenomas (SF1, GATA3, estrogen receptor α): 22 PAWUC with TFGA (TFGA-plus group) and 51 with TFGA expression only (TFGA-only group). Patient characteristics, outcome parameters, rate of invasiveness (assessed by direct endoscopic inspection), and MIB1 and MGMT status were compared between groups.

RESULTS Patients in the TFGA-plus group were significantly younger than patients in the TFGA-only group (age 46 vs 56 years, respectively; $p = 0.007$). In the TFGA-only group, pituitary adenomas were significantly larger (diameter 25 vs 18.3 mm, $p = 0.002$). Intraoperatively, signs of invasiveness were significantly more common in the TFGA-plus group than in the TFGA-only group (50% vs 16%, $p = 0.002$). Gross-total resection was significantly lower in the nonfunctioning TFGA-plus group than in the TFGA-only group (44% vs 86%, $p = 0.004$). MIB1 and MGMT status showed no significant difference between groups.

CONCLUSIONS These data suggest a more aggressive behavior of TFGA-positive adenomas if an additional TF is expressed within the tumor cells. Shorter radiographic surveillance and earlier consideration for retreatment should be recommended in these adenoma types.

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KEYWORDS plurihormonal adenoma; WHO classification; aggressive pituitary adenoma; transcription factor; pituitary surgery

WITH the aim of differentiating pituitary adenomas according to their cell lineages,^{1,2} the latest WHO classification of tumors of endocrine organs in 2017 has included the transcription factors (TFs) PIT1 and TPIT and the TFs for gonadotroph adenoma (TFGA) cells SF1, GATA2/3, and estrogen receptor α in the classification of pituitary adenomas. In this context, plurihormonal adenomas are now defined by the immunohistochemical (IHC) expression of multiple hormones

and further subclassified according to their TF expression pattern. Exceptions are mammotroph (growth hormone [GH]–prolactin [PRL]) and gonadotroph adenomas (β follicle-stimulating hormone [β -FSH]– β luteinizing hormone [β -LH] $\pm \alpha$ subunit).

According to the WHO 2017 classification, plurihormonal adenomas have been subclassified on the basis of their TF expression into two groups: 1) plurihormonal with one TF, termed “PIT1-positive adenomas” (previ-

ABBREVIATIONS ACTH = adrenocorticotrophic hormone; ER = endocrine remission; FSH = follicle-stimulating hormone; GH = growth hormone; GKRS = Gamma Knife radiosurgery; GTR = gross-total resection; IHC = immunohistochemical; IQR = interquartile range; LH = luteinizing hormone; MEN1 = multiple endocrine neoplasia type 1; oGTT = oral glucose tolerance test; PAWUC = plurihormonal adenomas with unusual immunohistochemical combinations; PRL = prolactin; SRL = somatostatin receptor ligand; TF = transcription factor; TFGA = TF gonadotroph adenoma; TSH = thyroid-stimulating hormone.

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ously known as silent subtype III pituitary adenoma); and 2) plurihormonal with more than one TF, termed “plurihormonal adenomas with unusual immunohistochemical combinations” (PAWUC).

Plurihormonal adenomas have been found to occur with higher frequency in younger patients when compared to other types of pituitary adenomas. In approximately 70% of cases, plurihormonal adenomas are discovered through their mass effect and do not show signs of hormonal activity.³ In the remaining cases, clinical effects most often reflect the presence of GH (acromegaly) and to a lesser extent adrenocorticotrophic hormone (ACTH) overexpression (Cushing’s disease) and PRL cells (hyperprolactinemia). A clinically relevant overexpression of glycoprotein hormone (thyroid-stimulating hormone [TSH], FSH, LH) production is rare. Furthermore, in patients with multiple endocrine neoplasia type 1 (MEN1), plurihormonal adenomas have a higher incidence rate than in non-MEN1 patients.^{4–6} Plurihormonal adenomas are more often macroadenomas than microadenomas, and signs of invasiveness have been reported in as many as 30% in PIT1-positive tumors.⁴ However, apart from PIT1-positive plurihormonal adenomas (IHC expression of GH, PRL, TSH $\pm$ α subunit) that were identified to be more aggressive, little is known about the subgroup of PAWUC.

From our series of PAWUC, we identified a large fraction of adenomas expressing FSH/LH as well as the TFs for this cell lineage. Nonfunctioning adenomas in general, but also specifically the adenomas with immunocytochemical FSH/LH positivity, can exhibit very different biological behavior in terms of invasiveness and recurrence,^{6–8} and the expression of additional TFs to TFGA may be of prognostic value. The aim of this study was therefore to compare PAWUC with TFGA expression plus additional TFs to gonadotroph adenomas that only express the TFGA, in terms of biological behavior and clinical outcome.

Methods

Of all 458 pituitary adenomas surgically treated at our institution over 10 years (2008–2018), plurihormonal adenomas accounted for 5.7% (26 cases). This retrospective study comprises a single-center series of 73 patients with IHC-verified TFGA-positive pituitary adenomas: 22 of those with TFGA expression plus an additional TF (the TFGA-plus group), and 51 with TFGA expression only (the TFGA-only group). All patients were operated on using a purely endoscopic transsphenoidal approach.

Histopathological analysis was performed according to the criteria of the WHO 2017 classification of tumors of endocrine organs.² The TFGA-plus group was defined as a combination of the TFGA TF (LH, FSH $\pm$ α subunit) plus the PIT1 (GH, PRL, TSH) and/or TPIT (ACTH), corresponding to a subgroup of PAWUC. The TFGA-only group was defined as no other IHC TF expression additional to TFGA (LH, FSH $\pm$ α subunit) corresponding to gonadotroph adenomas.

Exclusion criteria were double adenomas (separate tumors with different cell types), combinations formerly classified as plurihormonal adenomas such as mammoso-

matotroph adenomas (GH-PRL), solely PIT1-positive plurihormonal adenomas, or patients lost to follow-up. Furthermore, patients with TFGA only without showing an IHC expression of LH/FSH ($\pm$ α subunit) were excluded.

Hormone expression was classified as negative if $< 1\%$ of tumor cells stained positive by IHC.⁹ Hormone production was classified as active if hormonal levels were found higher than sex-adjusted upper limits and if stigmata of acromegaly, Cushing’s disease, symptoms of hyperprolactinemia (which could not be explained by stalk effect), or secondary hyperthyroidism were present.

Histopathological examinations were performed (by T.R. and R.H.) without the knowledge of the extent of resection. Endocrine remission (ER) and gross-total resection (GTR) were assessed by endocrinologists and neurosurgeons (G.V., A.M., and S.W.) on the basis of follow-up hormone levels and follow-up MRI, blinded to the histopathological findings.

This study was approved by the Institutional Ethics Committee and was performed in accordance with the principles of the Declaration of Helsinki. The tumor samples were evaluated with the consent of the patients for further histopathological examination.

Tissue Processing

The surgical biopsy specimens were routinely processed and were immunohistochemically stained for pituitary hormones (ACTH, FSH, GH, LH, PRL, TSH, α subunit), TFs (PIT1, TPIT, SF1, GATA3, estrogen receptor α),^{10,11} the proliferation marker MIB1, and the DNA repair protein MGMT. GATA3 was used for tissue processing instead of GATA2 due to the high degree of homologous amino acids of GATA2 and GATA3 and the widespread use of GATA3 antibodies in pathological analysis.¹²

All biopsy specimens were fixed in 4% neutral buffered formalin, routinely processed, embedded in paraffin, cut in 5- μ m slices, and stained with H & E. Immunohistochemistry was performed with a streptavidin-biotin-peroxidase complex method. Two neuropathologists (T.R. and R.H.) separately reviewed specimens blinded to the intraoperative findings. Positive control tissues consisted of paraffin-embedded sections of appropriate positive controls for each antibody. Negative controls were assessed from nonrelevant antibodies of the same species and of the same immunoglobulin isotype (immunoglobulin G1).¹³

According to a study by Nishioka et al.,⁹ TFs were considered positive only if a significant fraction of relevant tumor cells showed nuclear reactivity. In cases of plurihormonal adenomas, multiple TF expression was graded positive if a significant fraction of tumor cells showed a positive expression of gonadotroph tumor cells (SF1, GATA3, or estrogen receptor α).

The MIB1 index, counted per 500 cells in the hotspot area, was used as a marker of the proliferation potential. MGMT expression status was assessed as the percentage of stained cells, i.e., $< 10\%$ (low), 10%–50% (moderate), and $> 50\%$ (high).^{13–16} Parasellar tumor extension was evaluated due to the revised Knosp classification.¹⁷ Tumor consistency was assessed based on the intraoperative status: soft tumors were defined as easy to remove with a suction device (easily suckable), whereas fibrous tumors showed

FIG. 1. Flowchart of patient characteristics. *Excluded: patients lost to follow-up, insufficient tumor tissue available for further IHC examinations.

a stiff and septated structure.¹⁸ The size (mean diameter) and tumor volume (cm³) of the tumor were assessed on preoperative isovoxel planning MRI (StealthStation S8; Medtronic). As part of our routine clinical practice, invasiveness was judged intraoperatively by the neurosurgical team through direct endoscopic visualization. Basal sellar dura samples of the anterior sellar wall were additionally obtained for histological examination.

Statistical Analysis

The data are presented as mean (range) for continuous variables and as frequencies for categorical variables, as well as median (interquartile range [IQR]) for preoperative hormonal levels. To assess the difference in GTR/ER rate, the rate of invasiveness (additional basal sellar dura), MGMT status, and tumor consistency between groups, the chi-square test with Pearson's correlation coefficient was performed. To analyze a difference in Knosp high grade between groups, a Mann-Whitney U-test was applied. Differences in age at first surgery, tumor size (diameter, volume), and MIB1 index between the two groups were assessed by an unpaired Student t-test. A p value < 0.05 was considered significant. For statistical analyses, SPSS software (version 25.0; IBM Corp.) was used.

Results

Study Cohort

The study group of PAWUC with TFGA and additional TF expression (TFGA-plus group) consisted of 22 consecutive patients (22/458 cases, 4.8%). The control group (TFGA-only group) consisted of 51 patients with gonado-

troph adenomas as confirmed by TFGA positivity on histopathology who were operated on and followed-up within the same time frame. Altogether, 73 consecutive patients were included in this study with a mean follow-up of 3.8 years (range 0.5–9 years).

IHC TF analysis revealed the following subclass rates of TFGA-plus adenomas: 1) 20 TFGA/PIT1 patients (FSH/LH plus GH, PRL n = 9; FSH/LH plus GH n = 4; FSH/LH plus PRL n = 3; FSH/LH plus PRL, TSH n = 3; FSH/LH, TSH n = 1); 2) 1 TFGA/TPIT patient (FSH/LH plus ACTH); and 3) 1 TFGA/PIT1/TPIT patient (FSH/LH, GH, PRL, ACTH). For detailed information about TFGA distribution within groups, see Fig. 1.

Comparison of Patient and Tumor Characteristics

Patients in the TFGA-plus group were significantly younger than patients in the TFGA-only group (46 vs 56 years of age, p = 0.007; Table 1). Sex distribution did not significantly differ within the groups (female/male ratio = 1:1.2 in the TFGA-plus group, 1:2.2 in the TFGA-only group; p = 0.249).

In the TFGA-only group, pituitary adenomas were significantly larger in diameter (25 vs 18.3 mm, p = 0.002) at the time of diagnosis; however, tumor volume (9.8 vs 5.4 cm³, p = 0.077) only showed a trend to higher mean tumor volumes. However, this difference did not apply when comparing only nonfunctioning adenomas (TFGA-only vs TFGA-plus): diameter 25 vs 21.9 mm, p = 0.306; volume 9.8 vs 7.0 cm³, p = 0.457.

In the TFGA-plus group, 13 (59%) of 22 patients were found to harbor functioning active adenomas at the time of diagnosis: 11 cases of acromegaly (nadir oral glucose

TABLE 1. Patient and tumor characteristics

Characteristic	PAWUC, TFGA-Plus	TFGA-Only	p Value
No. of patients	22	51	
1st op	19 (86)	47 (92)	NS
Recurrent op	3 (14)	4 (8)	
Age (range), yrs	46 (20–72)	56 (27–75)	0.007
<40	8 (36)	10 (20)	NS
>40	14 (64)	41 (80)	
Sex (F/M ratio)	1:1.2	1:2.2	NS
Follow-up (range), yrs	3.9 (0.5–9)	3.8 (0.5–9)	NS
Functional classification			
Functional	13 (59)	—	<0.001
Nonfunctional	9 (41)	51 (100)	
Tumor size (range)			
Diameter, mm	18.3 (6–40)	25.0 (9–50)	0.002
Volume, cm ³	5.4 (0.1–21.3)	9.8 (0.8–63.8)	0.077
Tumor consistency			
Soft	19 (86)	47 (92)	NS
Fibrous	3 (14)	4 (8)	
Invasiveness	11 (50)	8 (16)	0.002
Knosp high grade	10 (45)	22 (43)	NS
3A	3 (30)	17 (77)	NS
3B	1 (10)	1 (5)	
4	6 (60)	4 (18)	
Degree of resection			NS
Complete	17 (77)	47 (92)	
Subtotal, >80%	3 (14)	1 (2)	
Partial, <80%	2 (9)	3 (6)	
GTR			
Functional	7/13 (54)	—	—
Nonfunctional	4/9 (44)	44 (86)	0.004
GKRS	5 (23)	2 (4)	0.012

NS = nonsignificant.

Data presented as mean (%) unless otherwise indicated.

tolerance test [oGTT]: GH median 8.5 ng/ml, IQR 6.3–22.2 ng/ml; IGF-1 median 938 ng/ml, IQR 542–1047 ng/ml, 1 case of acromegaly and hyperprolactinemia (nadir oGTT: GH 177 ng/ml, IGF-1 1493 ng/ml, PRL 464 ng/ml), and 1 case of hyperprolactinemia (PRL 350 ng/ml). In the patient with only hyperprolactinemia, the indication for operation was resistance to dopamine agonist treatment and mass effect on the chiasm.

In the TFGA-only group (n = 51), no patients exhibited any signs of hormone overexpression (functioning gonadotroph adenoma) and were operated on due to a mass effect of tumor. In recurrent cases, no tumor samples showed a different IHC hormonal or TF expression in comparison to the primary tumor.

When comparing parasellar extension on preoperative MRI between the groups, we more often found grade 3A in the TFGA-only group (77% vs 30%), and more often

grade 4 in the TFGA-plus group (60% vs 18%). Although not significant, a clear trend for a different distribution of Knosp high-grade adenomas (p = 0.057) between groups could be found.

Intraoperatively, signs of invasiveness as determined by intraoperative endoscopic direct visualization were significantly more common in the TFGA-plus group than in the TFGA-only group (50% vs 16%, p = 0.002). The histopathological examination of basal sellar dura invasiveness showed no statistical difference between the groups (21% vs 22%, p = 0.951). There was no difference in fibrous tumor consistency between the groups (14% in the TFGA-plus group vs 8% in the TFGA-only group, p = 0.440).

Overall, the GTR/ER rate was 75.3% (55/73); GTR/ER was significantly lower in the TFGA-plus group than in the TFGA-only group (50% vs 86%, p = 0.001). GTR was significantly lower in the nonfunctional TFGA-plus group than in the TFGA-only group (44% vs 86%, p = 0.004).

Gamma Knife radiosurgery (GKRS) was performed in 2 of 5 patients in the TFGA-plus group and in 2 of 4 patients in the TFGA-only group with subtotal or partial tumor resection. The remaining 3 patients with subtotal or partial tumor resection in the TFGA-plus group had non-functioning adenomas and showed a stable tumor remnant after a second surgery. The same was found in 2 patients in the TFGA-only group.

In 8 (11%) of the 73 patients, including 5 (23%) of 22 in the TFGA-plus group and 3 (6%) of 51 in the TFGA-only group in whom no remaining tumor tissue on early postoperative MRI and normal postoperative hormone levels were found, relapse occurred postoperatively at a median of 2.9 years (IQR 1.8–3.8 years). In 5 (63%) of 8 patients with relapse in the TFGA-plus group, 4 patients had a functioning tumor (acromegaly in all cases) and were further treated with long-acting somatostatin receptor ligands (SRLs), and in 3 (75%) of 4 cases with GKRS. In 1 patient with nonfunctioning TFGA-plus adenoma and all patients in the TFGA-only group, a stable tumor remnant was noted during follow-up controls.

Evaluation of proliferation rate as assessed by the MIB1 index did not reveal any significant differences between the groups (3.8% vs 2.9% for the TFGA-plus and TFGA-only groups, respectively; p = 0.164). Low MGMT immunoreexpression (< 10%) was found in 7% (5/73) across all cases, including 80% (4/5) in the TFGA-plus group.

The frequency of low to moderate MGMT immunoreexpression (< 50%) was 23% (17/73) across all cases. Low to moderate MGMT immunoreexpression was not significantly differently distributed between groups (32% in the TFGA-plus group vs 20% in the TFGA-only group, p = 0.257; Figs. 2 and 3).

Discussion

To the best of our knowledge, the current series represents the first description of TFGA-positive PAWUC after the WHO classification revision in 2017. Although of significantly smaller size than TFGA-only (gonadotroph) adenomas, we found a higher rate of invasiveness and of Knosp grade 4 parasellar growth, indicating a more ag-

FIG. 2. Images of nonfunctioning PAWUC. **A:** Preoperative MR image depicting adenoma extension into the left cavernous sinus compartments. **B:** After a combination of 3 resections (2 transsphenoidal, 1 subfrontal) and 1 GKRS, 5-year postoperative MR image confirmed complete adenoma removal from the left cavernous sinus compartment. However, a recurrent tumor was noted on the right endo-/parasellar side that was treated by a second GKRS. **C–G:** IHC examination. PIT1 positive (C), TPIT negative (D), SF1 negative (E), MIB1 2.2% (F), and MGMT > 50% (G). Figure is available in color online only.

gressive behavior and, consequently, a lower rate of GTR/ER in TFGA-positive plurihormonal adenomas.

Plurihormonal Adenomas

Prior to the widespread use of immunohistochemistry for detecting hormonal expression in pituitary adenomas, classification of pituitary adenomas was based on H & E staining, which hampered the diagnosis of plurihormonal adenomas.¹⁹ With the revision of the 2004 WHO classification, immunohistochemistry has evolved to the gold standard in the diagnosis of pituitary adenomas as well as for defining pituitary adenoma subtypes. A possible benefit of using TFs to identify plurihormonal adenomas was presumed, but there were no sufficient available data for clarifying the cytological coherence. Furthermore, the group of subtype III adenomas was established (now termed “PIT1-positive adenomas”).

Within the 2017 WHO classification, TFs have now been defined as a mainstay for diagnosing pituitary adenomas, especially in the case of plurihormonal adenomas. Due to these improvements in diagnosis, true “null cell adenomas,” for example, are relatively rare. In an analysis by Nishioka et al., the authors accounted for only 0.6% of 1071 surgically treated adenomas.⁹ Many hormone-negative adenomas have been found to be TFGA-positive gonadotroph adenomas,²⁰ or in the case of plurihormonal

adenomas, show an additional expression of TFGA (FSH, LH $\pm$ α subunit).

Several theories of how plurihormonal adenomas can possibly emerge have been published. On the one hand, the development may be based on a precursor cell that has undergone several transformation and/or mutation processes to advance to a plurihormonal adenoma.^{21–24} Alternatively, clone cells of an existing pituitary adenoma may undergo several multidirectional differentiations leading to the formation of this tumor entity.^{21,25,26} The difference of TF expression in plurihormonal adenomas has also been shown in our study cohort, as different types of TFGA (SF1, GATA3, estrogen receptor α) have been expressed by the tumor cells (Fig. 1).

PIT1-Positive Adenomas Versus PAWUC

The prototype of monomorphous plurihormonal adenomas is PIT1-only-positive adenomas (formerly termed “silent subtype III adenoma”). Initially considered as a third variant of silent corticotroph adenoma, silent subtype III pituitary adenomas have been recognized as biologically aggressive tumors.^{3,4,27} PIT1-positive plurihormonal adenomas have been found with a frequency of 0.9%–3.2% in large series of resected cases.^{3,4} The series from the Mayo Clinic, St. Michael’s Hospital, and the University Health Network showed that these neoplasms

FIG. 3. Images of gonadotroph adenoma. **A:** Preoperative MR image depicting adenoma extension into the left cavernous sinus, Knosp grade 2. **B:** Postoperative MR image obtained 3 years after endoscopic transsphenoidal resection confirmed complete adenoma removal. **C–G:** IHC examination. PIT1 negative (C), TPIT negative (D), SF1 positive (E), MIB1 2.6% (F), and MGMT 10%–50% (G). Original magnification $\times 40$. Figure is available in color online only.

were not always silent and may present with acromegaly, amenorrhea, and galactorrhea, as well as hyperthyroidism. Although a combination of clinical findings for different hormone oversecretions (e.g., acromegaly and hyperprolactinemia) is possible, it only occurs in rare cases.

The hallmark of these neoplasms is the expression of PIT1 and the presence of nuclear spheridia (nuclear inclusions).⁴ In contrast to differentiated PIT1 lineage adenomas (somatotroph, lactotroph, thyrotroph adenomas), these tumors often show a distinct IHC staining characterized by very focal or scattered positivity for one or more PIT1 hormones, including GH, PRL, and β -TSH.

In contrast to PIT1-only-positive plurihormonal adenomas, little is known about the subgroup of PAWUC. In a recently published retrospective cohort analysis, Aydin et al. reported PAWUC with a rate of 2.7% of all pituitary adenomas,²⁸ in contrast to 4.8% in our series. The most common IHC combinations in PAWUC include GH, PRL, and one or more glycoprotein hormone subunits (β -TSH, β -FSH, β -LH, and α subunit).²⁹ In concordance with our findings, Aydin et al.²⁸ found the combination of PIT1 and TFGA to be the most common subtype.

In our series of 22 patients with PAWUC, 13 were hormone secreting. Of those, 12 (92%) had acromegaly at the time of diagnosis, and 1 of these patients was diagnosed with additional hyperprolactinemia (464 ng/ml). In this patient, the postoperative PRL concentration was within the normal range without dopamine agonist treatment.

However, this patient needed SRLs and GKRS postoperatively.

Due to the inclusion of TFs and improvements in IHC hormonal antibodies, many nonfunctioning plurihormonal adenomas that were formerly classified as null cell adenomas or silent adenomas of a single hormonal type have now been discovered to be plurihormonal adenomas. In our series, the most common subtype of TFGA-plus plurihormonal adenomas presented with a combination of GH, PRL (PIT1), and FSH/LH (TFGA).

In 40% (4/10) of nonfunctioning TFGA-plus PAWUC, a Knosp grade 4 parasellar invasion was found on preoperative MRI and therefore significantly more often than in the TFGA-only group (gonadotroph adenoma), with a frequency of 8%. In cases of nonfunctioning PAWUC, considerations of shorter surveillance of tumor remnants and GKRS are recommended. Other combinations of TF hormones occur more rarely. Especially in cases of combinations with TPIT (such as ACTH), only small case series have been published. To date, 20 cases have been reported to co-express PIT1 and TPIT (GH and ACTH).³⁰

It is common for GH- and ACTH-secreting pituitary tumors to show no clinical signs of Cushing's disease. In all reported cases of plurihormonal pituitary adenomas producing both GH and ACTH, the dominant clinical characteristics have been those of GH overproduction. Only 3 cases of TFGA and TPIT (LH/FSH and ACTH) have currently been presented in the literature. In these adeno-

mas, an increased risk of tumor recurrence was found to be associated with ACTH co-expression.^{31,32} Our series adds 2 more patients with TPIT and TFGA expression to the current literature (both nonfunctioning). One of these patients is currently in ER and the other showed relapse, with tumor regrowth.

Markers for Plurihormonal Adenomas

By analyzing the proliferation rate of plurihormonal adenomas, Pawlikowski et al. found significantly higher MIB1 values compared to monohormonal adenomas.³³ However, in our series no significant difference in the MIB1 index was found between TFGA-plus plurihormonal (mean MIB1 index 3.8) and gonadotroph (mean MIB1 index 2.9) adenomas ($p = 0.164$).

In cases of PIT1-positive plurihormonal adenomas, MGMT IHC negativity was found in 50% (5/10) of cases.³⁴ In our 3 PIT1-positive plurihormonal adenoma cases, 33% (1/3) had a negative MGMT IHC expression. In cases of TFGA-plus plurihormonal adenomas, low MGMT immunoreexpression (< 10%) was found in 80% (4/5) of cases in the TFGA-plus group. However, the frequency of low to moderate MGMT immunoreexpression (< 50%) was not significantly differently distributed between groups (32% in the TFGA-plus group vs 20% in the TFGA-only group, $p = 0.257$).

Richardson et al. found that plurihormonal adenomas had an upregulation of genes that encode for *ARG2* and *SEMA3A*, which regulate T-cell proliferation and function within the tumor microenvironment. This regulation of T-cell proliferation might be a possible target for future medical therapy in recurrent cases.³⁵

Limitations of the Study

The main limitations of the presented study are due to the inherent nature of a retrospective analysis and the relatively small number of plurihormonal adenomas. Although a trend for a smaller tumor volume in the TFGA-plus group was detected, no significant difference was found. However, tumor diameter is the common criteria for tumor size, which was found to be significantly different between the groups.

We were able to present the first comprehensive review of TFGA-positive plurihormonal adenomas with unusual hormonal combinations and report on the aggressive biological behavior of this subgroup of plurihormonal adenomas. Further multicenter studies are needed to investigate the entity of all subtypes of plurihormonal adenomas.

Conclusions

Our data suggest a more aggressive behavior of TFGA-positive adenomas if an additional TF is expressed within the tumor cells, especially in cases of nonfunctioning PAWUC. This is particularly important as tumor size has failed to act as a marker to predict GTR/ER in tumors expressing TFGA. Detailed IHC testing of TFs and hormones is therefore important to make an accurate statement about the further behavior of pituitary adenomas. Consideration of shorter surveillance of tumor remnants and early radio-surgery are recommended in these adenoma subtypes.

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Disclosures

Dr. Wolfsberger is currently an educational consultant for Medtronic Surgical Technologies.

Author Contributions

Conception and design: Wolfsberger, Micko. Acquisition of data: Micko, Rötzer, Vila, Oberndorfer. Analysis and interpretation of data: Micko, Rötzer, Vila. Drafting the article: Micko. Critically revising the article: Wolfsberger, Micko, Rötzer, Hoftberger, Vila, Frischer, Knosp. Reviewed submitted version of manuscript: all authors. Approved the final version of the manuscript on behalf of all authors: Wolfsberger. Statistical analysis: Micko. Administrative/technical/material support: Rötzer, Oberndorfer. Study supervision: Wolfsberger.

Supplemental Information

Online-Only Content

Supplemental material is available with the online version of the article.

Supplemental Table 1. <https://thejns.org/doi/suppl/10.3171/2020.2.JNS2018>.

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