



Novel AIP mutation in exon 6 causing acromegaly in a German family

M. Detomas¹ · B. Altieri¹ · J. Flitsch² · W. Saeger³ · M. Korbonits⁴ · T. Deutschbein^{1,5}

Received: 28 November 2022 / Accepted: 1 February 2023 / Published online: 9 February 2023
© The Author(s) 2023

Abstract

The most frequent genetic alteration of familial isolated growth hormone producing pituitary neuroendocrine tumors is a germline mutation of the aryl hydrocarbon receptor-interacting protein (AIP) gene. Various AIP mutations are already known; however, an AIP mutation in exon 6 (c.811_812del; p.Arg271Glyfs*16) has not been reported yet. Here, we report a German family with two identical twins who were both affected by acromegaly and carried the above-mentioned novel AIP mutation. The father was found to be an unaffected carrier, while the paternal aunt most likely suffered from acromegaly as well and died from metastatic colorectal cancer. Apart from reporting a novel AIP mutation, this study does not only highlight the different clinical and histological features of the AIP mutated growth hormone producing pituitary neuroendocrine tumors but also confirms the poor responsiveness of dopamine agonists in AIP mutated acromegaly. Furthermore, it highlights the increased mortality risk of comorbidities typically associated with acromegaly.

Keywords AIP · Acromegaly · Colorectal cancer · Familial · Gene · GH · Growth hormone · Mutation · Pituitary adenoma · Twins

Rapid communication

Acromegaly is a rare disorder usually due to growth hormone (GH) excess caused by a pituitary adenoma. The most frequent genetic alteration, being responsible for about 22% of familial isolated GH-producing pituitary adenoma and around 4% of sporadic acromegaly, is a germline mutation of the aryl hydrocarbon receptor-interacting protein (AIP) gene [1, 2]. A heterozygous germline AIP mutation (c.646G > T; p.E216*) has previously been reported in non-identical female twins, each with a silent somatotroph pituitary

macroadenoma [3]. Identical twins with a de novo AIP mutation (c.910C < T; p.R304*) have also been reported, with one twin suffering from gigantism, while the other being an unaffected carrier [4]. Furthermore, acromegaly occurring in one of two uniovular twins (who did not undergo genetic workup) [5, 6] or in male twins (to our knowledge both affected by an AIP mutation) have sporadically been reported [7, 8]. Here, we report the case of identical male twins with active acromegaly and a novel AIP mutation in exon 6 (c.811_812del; p.Arg271Glyfs*16).

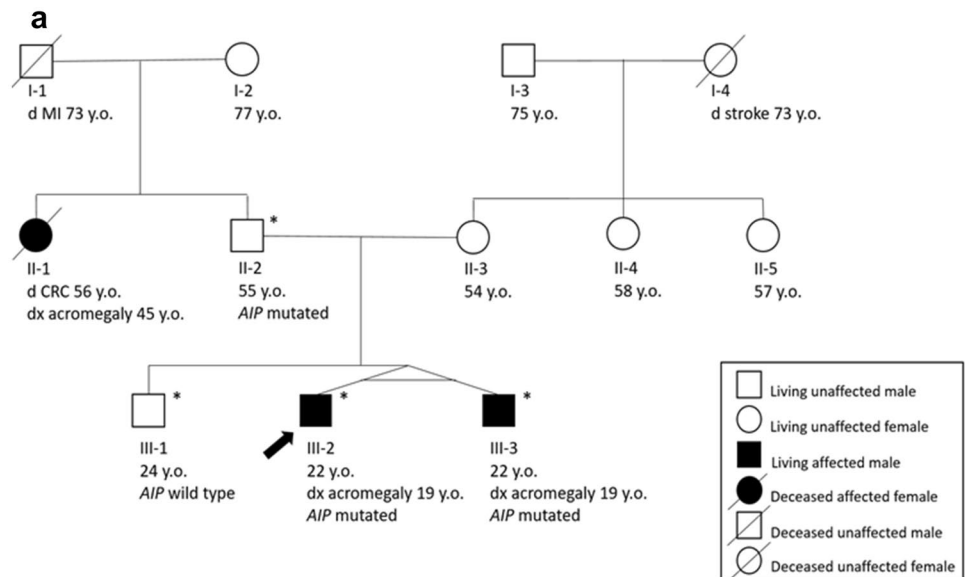
In April 2019, a 19-year-old male patient (patient III-2 in Fig. 1a) underwent a brain magnetic resonance imaging (MRI) because of recurrent headaches. Radiological workup revealed a pituitary macroadenoma of 2.5 × 2.1 × 2.0 cm (Fig. 1b). At initial presentation at our endocrine outpatient clinic, the patient was 200 cm tall and had a body weight of 114 kg. Apart from headaches, no complaints were reported. The patient was not on medication at that time. While the patient's own medical history only contained a wrist fracture during childhood, the paternal aunt was treated with a dopamine agonist (bromocriptine) because of a suspected prolactinoma. She was at that time 54 years old and also under chemotherapy because of metastatic colorectal cancer.

During physical examination, our index patient presented with facial and acral dysmorphism. Hormonal analysis

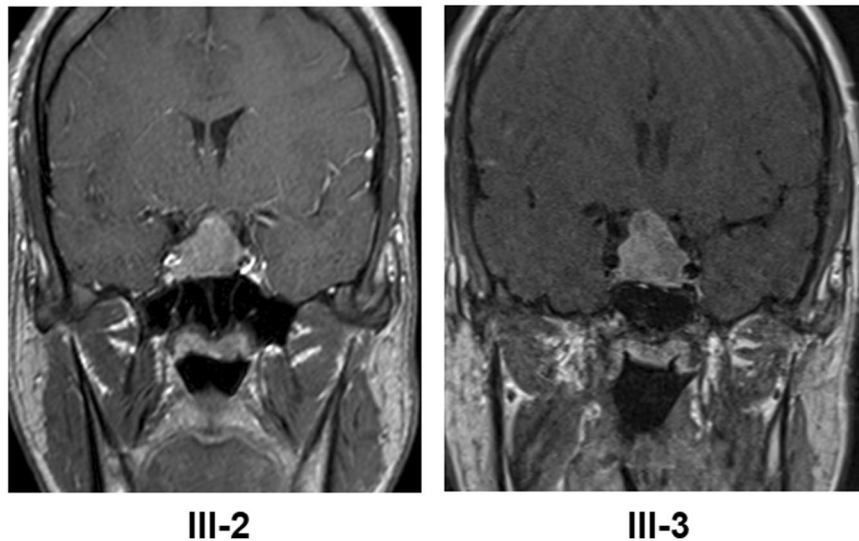
✉ M. Detomas
detomas_m@ukw.de

¹ Department of Internal Medicine I, Division of Endocrinology and Diabetes, University Hospital Würzburg, University of Würzburg, Würzburg, Germany
² Department of Neurosurgery, University Medical Center Hamburg-Eppendorf, Hamburg, Germany
³ Institute of Neuropathology, University of Hamburg, UKE, Hamburg, Germany
⁴ Centre for Endocrinology, Barts and the London School of Medicine and Dentistry, William Harvey Research Institute, Queen Mary University of London, London, UK
⁵ Medicovert Oldenburg MVZ, Oldenburg, Germany

Fig. 1 a Pedigree of the family. The arrow indicates the proband (subject III-2), while the asterisk indicate the patients undergoing genetic workup for *AIP* mutational status. The identified *AIP* mutation is c.811_812del; p.Arg271Glyfs*16. The provided ages are those at death, at the initial diagnosis of acromegaly, or at last follow-up. **b** Coronal T1-weighted magnetic resonance image with contrast medium of the sellar region of patient III-2 (left) and patient III-3 (right) *CRC* colorectal cancer, *d* age at patient's death, *dx* age at diagnosis of acromegaly, *MI* myocardial infarction, *y.o.* years old



b



revealed an insulin-like growth factor 1 (IGF-1) of 728 $\mu\text{g/l}$ ($1.8 \times$ upper limit of normal (ULN)), a random GH of 16.0 $\mu\text{g/l}$ (normal range 0–5), and a prolactin of 26.9 $\mu\text{g/l}$ (normal range 7.1–15.7). A subsequent oral glucose tolerance test confirmed acromegaly (insufficient GH decrease from 15.8 $\mu\text{g/l}$ to 13.40 $\mu\text{g/l}$ after 2 h, normal range ≤ 0.4 $\mu\text{g/l}$). Of note, the other pituitary hormones were normal (Table 1).

One month later, the patient was operated in a tertiary neurosurgical referral center for sellar masses. The neuropathological examination confirmed presence of a somatotroph pituitary neuroendocrine tumor (along with strong hormonal expression, sparse granulation, globular expression of Cam5.2, fibrous bodies in more than 70% of tumor cells, Ki67 ranging from 5 to 10% in some hotspots, and strongly expressed p53 in 5% of the cells; Pit-1 was positive, whereas

T-pit and SF-1 were negative) (Fig. 2). According to the WHO classification from 2021, this tumor was described as an aggressive pituitary adenoma [9]. If a clinicopathological classification originally suggested by Trouillas et al. was used, the tumor was categorized as invasive and proliferative (grade 2b) [10].

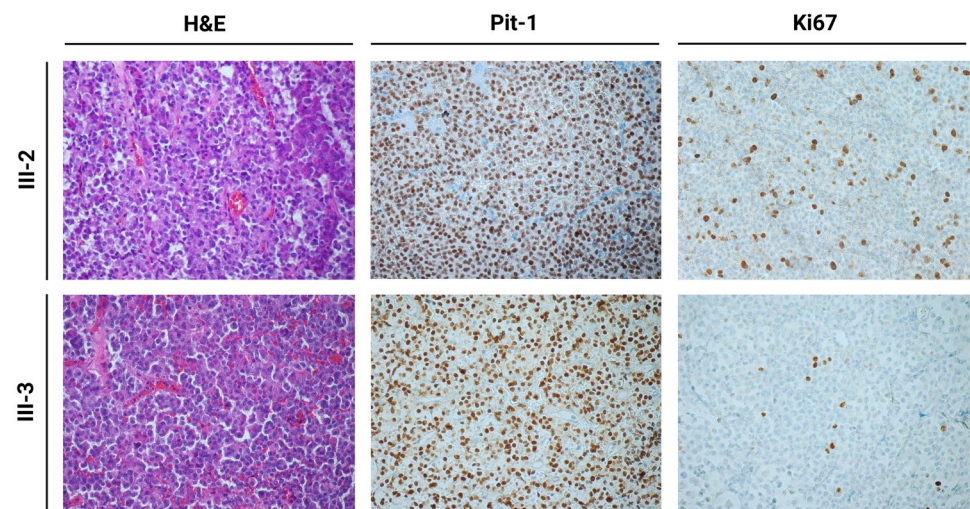
Because of the invasion of the right cavernous sinus (see Fig. 1b), a residual tumor persisted after surgery, and the patient remained biochemically uncontrolled (IGF-1: 424 $\mu\text{g/l}$, $1.3 \times$ ULN; random GH: 2.6 $\mu\text{g/l}$). Accordingly, a first-generation long-acting somatostatin analog was introduced (lanreotide 120 mg as i.m. injection every 4 weeks) but caused persistent nausea and fatigue. In January 2021, the patient therefore underwent a cyberknife stereotactic radiosurgery of the residual tumor. By June 2022, both IGF-1 (to 242 $\mu\text{g/l}$) and GH (to 0.7 $\mu\text{g/l}$) normalized without

Table 1 Clinical, hormonal, and genetic characteristics of the investigated family members at the time of their initial diagnostic workup at our endocrine outpatient clinic

| | III-2 (Proband) | III-3 (Twin brother) | II-2 (Father) | II-1 (Paternal aunt) |
|--------------------------|--------------------|-------------------------|------------------|-------------------------|
| Clinical characteristics | | | | |
| Age | 19 | 19 | 53 | 45 |
| Size (cm) | 200 | 201 | 194 | 184 |
| Weight (kg) | 114 | 100 | 100 | 74 |
| BMI (kg/m ²) | 28.5 | 24.8 | 26.7 | 21.9 |
| Hormonal parameters | | | | |
| IGF-1 (µg/l) | 728 | 554 | 201 | 365 |
| Random GH (µg/l) | 16.0 | 17.4 | 0.7 | 6.5 |
| Prolactin (µg/l) | 26.9 | 30.8 | 12.6 | 26.4 |
| TSH (mIU/l) | 1.3 | 1.4 | 1.2 | 1.6 |
| FT4 (pmol/l) | 12.5 | 10.3 | 13.8 | 17.1 |
| ACTH (ng/l) | 45.1 | 27.5 | 14.9 | 22.4 |
| Cortisol (µg/dl) | 12.2 | 3.9 | 18.6 | 17.3 |
| LH (IU/l) | 3.0 | 5.2 | 6.7 | 6.6 |
| FSH (IU/l) | 5.3 | 3.8 | 10.0 | 5.4 |
| Testosterone (µg/dl) | 2.0 | 3.5 | 3.9 | 0.4 |
| Estradiol (ng/l) | n.a. | n.a. | n.a. | 42 |
| Genetic analysis | | | | |
| AIP gene status | Mutated | Mutated | Mutated | n.a. |

Values reported in bold are pathological

ACTH adrenocorticotropin, AIP aryl hydrocarbon receptor-interacting protein gene, BMI body mass index, FSH follicle-stimulating hormone, FT4 free thyroxine, GH growth hormone, IGF-1 insulin-like growth factor 1, LH luteinizing hormone, n.a. not available, TSH thyroid stimulating hormone

Fig. 2 Staining for hematoxylin and eosin, Pit-1, and Ki67 in both twins' growth hormone-secreting pituitary neuroendocrine tumors. The main difference was observed for Ki67 (up to 10% in patient III-2, facing 5% in patient III-3). H&E hematoxylin and eosin

drug therapy. In September 2022, an asymptomatic minimal increase of IGF-1 to 336 µg/l ($1.1 \times \text{ULN}$) was observed. Accordingly, a dopamine agonist (cabergoline) was initiated, and the dose was progressively increased from 0.25 mg to 0.5 mg twice weekly. After 6 weeks of therapy, however, the IGF-1 further increased to 409 µg/l ($1.3 \times \text{ULN}$), resulting in a therapeutic switch from cabergoline to a GH receptor antagonist (pegvisomant, introduced with an initial

80 mg s.c. injection which was followed by daily s.c. injections of each 10 mg).

Because of the patient's young age at initial presentation of the somatotroph pituitary tumor, a germline genetic analysis was performed. A novel AIP mutation in exon 6 (c.811_812del; p.Arg271Glyfs*16) was identified. As a consequence, first-degree family members were screened for this mutation as well. The identical twin brother and the father were both found to be carriers of the same mutation,

while the twins' older brother was having a wild-type *AIP* gene status.

The patient's twin brother (patient III-3 in Fig. 1a) was referred to our endocrine outpatient clinic for the first time in June 2019. He did not have any complaints, was not on medical treatment, and had an inconspicuous medical history. The patient was 201 cm tall and weighed 100 kg. Physical examination revealed modest facial dysmorphism. However, no other signs and symptoms of acromegaly were detectable. IGF-1 (554 µg/d, 1.3 × ULN), random GH (17.40 µg/l), and prolactin (30.8 µg/l) were all elevated, whereas fT4 and basal serum cortisol were slightly decreased. All other pituitary hormones were normal (Table 1). Secondary adrenal insufficiency was confirmed with a pathological ACTH stimulation test (serum cortisol at 60 min 12.0 µg/dl, normal range ≥ 20.0 µg/dl). Accordingly, hydrocortisone and l-thyroxin replacement therapy were initiated. A 2.4 × 2.3 × 1.7 cm pituitary macroadenoma was identified in a sellar MRI (Fig. 1b). Following transsphenoidal surgery, the hormonal profile normalized (IGF 256 µg/l; random GH 0.8 µg/l; prolactin 3.9 µg/l). The neuropathologist described a pituitary neuroendocrine tumor with low-to-medium expression of somatotroph cells and negative staining for prolactin, ACTH, TSH, and LH. Further neuropathological workup of the tumor revealed sparse granulation, globular expression of Cam.5.2 with fibrous bodies in more than 70% of tumor cells, Ki67 5%, strong expression of p53 in 5% of the cells; Pit-1 was positive, whereas T-pit and SF-1 were negative (Fig. 2). These data were also consistent with an aggressive pituitary adenoma [9], or an invasive and proliferative tumor (grade 2b) [10].

In June 2020, we investigated the twins' father (subject II-2 in Fig. 1a), who was 194 cm tall (mid-parental height: 188 cm). The father (at that time 53 years old) had refused any endocrine workup before, since he did not have any complaints and biochemical workup (including IGF-1 and random GH) performed by the general practitioner had been normal. We also measured pituitary hormones in the normal range (e.g., IGF-1: 201 µg/dl; random GH: 0.7 µg/l; prolactin: 12.6 µg/l; testosterone: 3.9 µg/l, normal range 1.6–9.0) (Table 1). Because of his two affected sons and the results of the genetic workup, a pituitary MRI was performed (without any pituitary abnormalities). The paternal grandmother refused genetic testing.

The twins' paternal aunt (patient II-1 in Fig. 1a.) unfortunately died from metastatic colorectal cancer in 2021, at the age of 56. Interestingly, we noticed that she was referred to our endocrine outpatient clinic because of facial and acral dysmorphism already in March 2010. At that time, elevated levels of IGF-1 (365 µg/l, 1.4 × ULN) and random GH (6.47 µg/l) were observed, along with a slightly elevated prolactin of 26.4 µg/l and a body size of 184 cm. Of note, however, the biochemical evaluation was performed under

7.5 mg bromocriptine per day. This treatment had already been initiated elsewhere because of hyperprolactinemia. Unfortunately, the patient never came back to our institution despite further diagnostic measures were imperatively suggested.

In conclusion, we here report a novel *AIP* mutation in exon 6, which was responsible for acromegaly in two homozygous twins, and possibly in their paternal aunt. A mutation affecting the same nucleotide at c.811 (missense change c.811C > T; p.Arg271Trp) was already reported repeatedly in the context of familial isolated pituitary adenomas with GH-hypersecretion [11, 12], and indeed, it represents one of the genetic hotspots in this gene. This locus of the *AIP* gene can therefore be considered as important in the pathophysiological context of acromegaly.

The impact of the novel *AIP* mutation in exon 6 (c.811_812del; p.Arg271Glyfs*16) on the phenotype remains to be elucidated, since it caused a variable spectrum of clinical impairment and tumor-aggressiveness in the affected family members. Recently, it has been hypothesized that methylation processes may have a relevant influence on the variable behavior of pituitary neuroendocrine tumors [13], and this may have been true also for our case III-2. Considering that methylation of the *GSTP1* gene may be associated with an increased resistance to somatostatin analogues [14], resistance to dopamine agonists (as observed here and in other studies [15, 16]) may be explained as well. Finally, this report highlights the importance of early diagnosis and adequate treatment of GH excess, since some comorbidities related to acromegaly (e.g., colorectal cancer) may be lethal.

Acknowledgements We are grateful to the family members who agreed with providing their data to this manuscript. This work was supported by the Clinician Scientist programme RISE funded by the Else-Kröner-Fresenius Stiftung & the Eva-Luise-und-Horst-Köhler Stiftung (to MD), and by the European Reference Network on Rare Endocrine Conditions (Endo-ERN).

Funding Open Access funding enabled and organized by Projekt DEAL.

Data availability The authors declare that the data supporting the findings of this study are available within the article.

Declarations

Conflict of interest The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

Research involving human participants and/or animals This study was approved by the local ethics committee of the University Hospital of Würzburg (Approval number 85/12) and carried out in compliance with the declaration of Helsinki.

Informed consent The patients/participants provided written informed consent to provide their data in this manuscript.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

References

- Boguslawska A, Korbonits M (2021) Genetics of acromegaly and gigantism. *J Clin Med* 10(7):1377
- Gadelha MR, Kasuki L, Korbonits M (2017) The genetic background of acromegaly. *Pituitary* 20(1):10–21
- Villa C, Lagonigro MS, Magri F, Koziak M, Jaffrain-Rea ML, Brauner R, Bouligand J, Junier MP, Di Rocco F, Sainte-Rose C et al (2011) Hyperplasia-adenoma sequence in pituitary tumorigenesis related to aryl hydrocarbon receptor interacting protein gene mutation. *Endocr Relat Cancer* 18(3):347–356
- Ramirez-Renteria C, Hernandez-Ramirez LC, Portocarrero-Ortiz L, Vargas G, Melgar V, Espinosa E, Espinosa-de-Los-Monteros AL, Sosa E, Gonzalez B, Zuniga S et al (2016) AIP mutations in young patients with acromegaly and the tampico giant: the Mexican experience. *Endocrine* 53(2):402–411
- Briggs JH (1959) Acromegaly occurring in one of uniovular twins. *Proc R Soc Med* 52:1053–1054
- Lewis A (1934) Acromegaly in one of uniovular twins. *J Neurol Psychopathol* 15(57):1–11
- Izumi T, Kanazawa Y, Ishibashi M, Yamaji T, Kosaka K (1982) Pituitary gigantism and acromegaly in identical twin brothers. *Nippon Naika Gakkai Zasshi* 71(11):1586–1590
- Chahal HS, Stals K, Unterlander M, Balding DJ, Thomas MG, Kumar AV, Besser GM, Atkinson AB, Morrison PJ, Howlett TA et al (2011) AIP mutation in pituitary adenomas in the 18th century and today. *N Engl J Med* 364(1):43–50
- Asa SL, Mete O, Perry A, Osamura RY (2022) Overview of the 2022 WHO classification of pituitary tumors. *Endocr Pathol* 33(1):6–26
- Trouillas J, Roy P, Sturm N, Dantony E, Cortet-Rudelli C, Vignet G, Bonneville JF, Assaker R, Auger C, Brue T et al (2013) A new prognostic clinicopathological classification of pituitary adenomas: a multicentric case-control study of 410 patients with 8 years post-operative follow-up. *Acta Neuropathol* 126(1):123–135
- Daly AF, Vanbellinghen JF, Khoo SK, Jaffrain-Rea ML, Naves LA, Guitelman MA, Murat A, Emy P, Gimenez-Roqueplo AP, Tamburrano G et al (2007) Aryl hydrocarbon receptor-interacting protein gene mutations in familial isolated pituitary adenomas: analysis in 73 families. *J Clin Endocrinol Metab* 92(5):1891–1896
- Hernandez-Ramirez LC, Gabrovská P, Denes J, Stals K, Trivellin G, Tilley D, Ferrau F, Evanson J, Ellard S, Grossman AB et al (2015) Landscape of familial isolated and young-onset pituitary adenomas: prospective diagnosis in aip mutation carriers. *J Clin Endocrinol Metab* 100(9):E1242–E1254
- Giuffrida G, D'Argenio V, Ferrau F, Lasorsa VA, Polito F, Aliquo F, Ragonese M, Cotta OR, Alessi Y, Oteri R et al (2022) Methylation analysis in nonfunctioning and GH-secreting pituitary adenomas. *Front Endocrinol (Lausanne)* 13:841118
- Ferrau F, Romeo PD, Puglisi S, Ragonese M, Spagnolo F, Sapieto C, Ientile R, Curro M, Visalli G, Alibrandi A et al (2019) GSTP1 gene methylation and AHR rs2066853 variant predict resistance to first generation somatostatin analogs in patients with acromegaly. *J Endocrinol Invest* 42(7):825–831
- Beckers A, Aaltonen LA, Daly AF, Karhu A (2013) Familial isolated pituitary adenomas (FIPA) and the pituitary adenoma predisposition due to mutations in the aryl hydrocarbon receptor interacting protein (AIP) gene. *Endocr Rev* 34(2):239–277
- Marques P, Caimari F, Hernandez-Ramirez LC, Collier D, Iacovazzo D, Ronaldson A, Magid K, Lim CT, Stals K, Ellard S et al (2020) Significant Benefits of AIP testing and clinical screening in familial isolated and young-onset pituitary tumors. *J Clin Endocrinol Metab* 105(6):e2247–e2260

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.