Kallmann's Syndrome: A Comparison of the Reproductive Phenotypes in Men Carrying *KAL1* and *FGFR1/KAL2* Mutations

Sylvie Salenave, Philippe Chanson, Hélène Bry, Michel Pugeat, Sylvie Cabrol, Jean Claude Carel, Arnaud Murat, Pierre Lecomte, Sylvie Brailly, Jean-Pierre Hardelin, Catherine Dodé, and Jacques Young

Assistance Publique-Hôpitaux de Paris, Hôpital de Bicêtre, Service d'Endocrinologie et des Maladies de la Reproduction and Centre de Référence des Maladies Endocriniennes Rares de la Croissance (S.S., P.C., H.B., J.Y.) and Laboratoire de Génétique moléculaire, Pharmacogénétique et Hormonologie (S.B.); Univ Paris-Sud 11 (P.C., S.B., J.Y.); and INSERM U693 (P.C., S.B., J.Y.), F94275 Le Kremlin-Bicêtre, France; Hôpital Neurologique, Fédération d'Endocrinologie (M.P.), F69500 Bron, France; Assistance Publique-Hôpitaux de Paris, Hôpital Trousseau, Laboratoire d'Exploration Fonctionnelles Endocriniennes (S.C.), F75012 Paris, France; Assistance Publique-Hôpitaux de Paris, Hôpital Robert Debré, Service d'Endocrinologie Pédiatrique, and Centre de Référence des Maladies Endocriniennes Rares de la Croissance (J.C.C.), F75935 Paris, France; Centre Hospitalier Universitaire, Service d'Endocrinologie (A.M.), F44000 Nantes, France; Centre Hospitalier Universitaire, Service d'Endocrinologie (P.L.), F37044 Tours, France; Institut Pasteur, Unité de Génétique des Déficits Sensoriels (J.-P.H.), F75724 Paris, France; and Assistance Publique-Hôpitaux de Paris, Hôpital Cochin, Laboratoire de, Biochimie et de Génétique Moléculaire (C.D.), F75014 Paris, France

Context: Kallmann's syndrome (KS) is a genetically heterogeneous disorder consisting of congenital hypogonadotropic hypogonadism (CHH) with anosmia or hyposmia.

Objective: Our objective was to compare the reproductive phenotypes of men harboring *KAL1* and *FGFR1/KAL2* mutations.

Design and Patients: We studied the endocrine features reflecting gonadotropic-testicular axis function in 39 men; 21 had mutations in *KAL1* and 18 in *FGFR1 /KAL2*, but none had additional mutations in *PROK-2* or *PROKR-2* genes.

Results: Puberty failed to occur in the patients with *KAL1* mutations, all of whom had complete CHH. Three patients with *FGFR1/KAL2* mutations had normal puberty, were eugonadal, and had normal testosterone and gonadotropin levels. Cryptorchidism was more frequent (14 of 21 *vs.* 3 of 15; $P < 00.1$) and testicular volume (2.4 ± 1.1 *vs.* 5.4 ± 2.4 ml; $P < 0.001$) was smaller in CHH subjects with *KAL1* mutations than in subjects with *FGFR1/KAL2* mutations. The mean basal plasma FSH level (0.72 ± 0.47 *vs.* 1.48 ± 0.62 IU/liter; $P < 0.05$), serum inhibin B level (19.3 ± 10.6 *vs.* 39.5 ± 19.3 pg/ml; $P < 0.005$), basal LH plasma level (0.57 ± 0.54 *vs.* 1.0 ± 0.6 IU/liter; $P < 0.01$), and GnRH-stimulated LH plasma level (1.2 ± 1.0 *vs.* 4.1 ± 3.5 IU/liter; $P < 0.01$) were significantly lower in the subjects with *KAL1* mutations. LH pulsatility was studied in 13 CHH subjects with KAL1 mutations and seven subjects with *FGFR1/KAL2* mutations; LH secretion was nonpulsatile in all the subjects, but mean LH levels were lower in those with *KAL1* mutations.

Conclusion: *KAL1* mutations result in a more severe reproductive phenotype than *FGFR1/KAL2* mutations. The latter are associated with a broader spectrum of pubertal development and with less severe impairment of gonadotropin secretion. **(*J Clin Endocrinol Metab* 93: 758–763, 2008)**

Kallmann's syndrome (KS) is a developmental disorder combining congenital hypogonadotropic hypogonadism (CHH) with anosmia or hyposmia. The anosmia is related to hypoplasia or aplasia of the olfactory bulbs. CHH is due to GnRH deficiency, which likely results from failed embryonic migration of GnRH-synthesizing neurons. The gene *KAL1*, re-

0021-972X/08/$15.00/0

Printed in U.S.A.

Copyright © 2008 by The Endocrine Society

doi: 10.1210/jc.2007-1168 Received May 29, 2007. Accepted December 19, 2007.

First Published Online December 26, 2007

Abbreviations: CHH, Congenital hypogonadotropic hypogonadism; FGFR1, fibroblast growth factor receptor 1; KS, Kallmann's syndrome.

Downloaded from https://academic.oup.com/jcem/article/93/3/758/2598286 by guest on 21 August 2022

sponsible for the X-chromosome-linked form of KS was identified 15 yr ago (1, 2). More recently, loss-of-function mutations in the fibroblast growth factor receptor 1 (FGFR1) gene (*FGFR1/KAL2*) were shown to cause one of the autosomal dominant forms of KS (3–7). *KAL1* encodes anosmin-1, a locally restricted glycoprotein of embryonic extracellular matrices, which is likely to be involved in FGFR1 signaling and in normal embryonic migration of GnRH neurons (4).

Previous studies performed before the discovery of *FGFR1/KAL2* as a gene underlying one autosomal form of KS suggested that CHH secondary to KS could be more severe than normosmic CHH (8, 9). In fact, KS is clinically heterogeneous, and patients display a broad spectrum of reproductive phenotypes ranging from CHH associated with micropenis and cryptorchidism to normal puberty (3–7, 10). The purpose of this study was to determine whether KS patients carrying *KAL1* mutations have a more severe reproductive phenotype than patients harboring *FGFR1/KAL2* mutations. We therefore compared parameters reflecting gonadotropic and testicular functions in 21 men with *KAL1* and 18 men with *FGFR1/KAL2* mutations and autosomal inheritance.

Patients and Methods

We included 39 men aged 18–55 yr and carrying either *KAL1* (n = 21) or *FGFR1/KAL2* (n = 18) mutations, all of the latter having an autosomal form of KS (Table 1). A detailed history was taken and a thorough physical examination was undertaken to evaluate spontaneous sexual maturation. Thirty-six men had KS and CHH (Table 1). The remaining three men were apparently normal (cases 23, 25, and 31; Table 1) and were identified during familial studies of patients with KS phenotype. The diagnostic criteria for CHH were the following: failure of spontaneous puberty at 17, small testicular volume, a normal hypothalamic-pituitary region on magnetic resonance imaging (MRI), a low plasma testosterone level, and a low or inappropriately normal plasma gonadotropin level. In the patients with CHH, all other pituitary functions were normal. All the patients with CHH were considered to have KS because olfactometry, used to achieve the measure of detection and identification threshold for five odorants (12) showed anosmia or hyposmia and MRI showed olfactory bulb aplasia or hypoplasia in the majority (Table 1). Stretched penile length was measured at diagnosis, before any androgen therapy; microphallus was defined as a length less than 2.5 cm (13). Testicular volume was measured at the same time by using both a Prader orchidometer and by sonography when intrascrotal and by sonography in patients with cryptorchidism. None of the patients had received gonadotropin replacement therapy before this study. Patients who were receiving androgen replacement therapy were taken off testosterone for at least 2 months before the hormonal investigations. On the day of admission, a blood sample was drawn between 0800 and 1000 h and stored until assay to determine baseline serum FSH, LH, testosterone, and inhibin B levels.

The study was approved by the local ethics committee, and the patients gave their written informed consent.

Assays

All the hormone measurements were performed in a single assay. Plasma LH, FSH, and inhibin B were measured with an immunoradiometric assay or with an ELISA as reported (14). The intra- and interassay coefficients of variation were, respectively, 1.5 and 5.2% for LH, 2.7 and 5.5% for FSH, and 6 and 15% for inhibin B. The detection limits were 0.15 IU/liter, 0.2 IU/liter, and 10 pg/ml for LH, FSH, and inhibin B, respectively. Plasma testosterone was measured with a commercial RIA method with a detection limit of 0.19 nmol/liter and intra- and interassay coefficients of variation of 5.8 and 8.0%, respectively.

Analysis of gonadotropin secretion

The GnRH challenge test (100 μg iv) was performed as reported (15). Endogenous LH secretion, analyzed according to the algorithm of Thomas *et al.* (15, 16) was evaluated overnight at 10-min intervals for 6 or 8 h, as reported (15), in 12 KS patients with *KAL1* mutations (cases 1, 2, 5, 9–13, 15, 16, 20, and 21) and in seven KS patients with *FGFR1/KAL2* mutations (cases 22, 26, 27, 29, 30, 35, and 37) four and two of whom had previous androgen therapy, respectively (Table 1).

Molecular studies

KAL1 and *FGFR1/KAL2* mutations were identified as previously reported (3, 5, 7, 11). Five of the *KAL1* mutations are novel mutations, whereas all the *FGFR1/KAL2* mutations have previously been reported (Table 1) (3, 5, 7). No mutations in the *Prokineticin-2* and *Prokineticin receptor-2* genes (10) were found in any patient (Table 1). No additional mutations in *FGFR1/KAL2* were found in the subjects with *KAL1* mutations (Table 1).

Results

Prevalence of CHH in patients with *KAL1* and *FGFR1/KAL2* mutations

Spontaneous pubertal development had failed in all the patients with *KAL1* mutations; physical examination showed a testicular volume of 4 ml or less, suggesting a complete lack of pubertal development. All these patients were therefore considered to have complete CHH. In contrast, familial investigations of the patients with *FGFR1/KAL2* mutations identified three ascendants (patients 23, 25, and 31; Table 1) who had mutations in this gene but who had undergone normal spontaneous puberty (virilization and growth spurt) between 13 and 14 yr of age; physical examination showed normal virilization and a postpubertal testicular volume (15–25 ml) compatible with normal gonadal function. These three men also had normal plasma testosterone and gonadotropin concentrations.

Clinical and hormonal evaluation of CHH patients with *KAL1* and *FGFR1/KAL2* mutations

Prevalence of microphallus and cryptorchidism

The three eugonadal subjects harboring *FGFR1/KAL2* mutations were excluded from this part of the analysis.

Four (19%) of the 21 CHH patients with *KAL1* mutations and two (13%) of the 15 CHH patients with *FGFR1/KAL2* mutations had micropenis at the time of diagnosis ($P = 0.09$). Cryptorchidism was significantly ($P < 0.01$, χ^2 test) more frequent in patients with *KAL1* mutations (14 of 21, 67%; bilateral in 11 patients) than in patients with *FGFR1/KAL2* mutations (three of 15, 20%; bilateral in two patients).

Testicular volume and secretions

Mean testicular volume was significantly lower in the CHH patients with *KAL1* mutations (2.5 ± 1.1 ml; mean ± SD) than in the CHH patients with *FGFR1/KAL2* mutations (5.4 ± 2.2 ml; $P < 0.0001$) (Fig. 1A). Figure 1B shows individual circulating testosterone levels, which were always below normal in both

Downloaded from https://academic.oup.com/jcem/article/93/3/758/2598286 by guest on 21 August 2022

TABLE 1. Characteristics and molecular analysis in patients with *KAL1* and *FGFR1/KAL2* mutations

Patient	Age (yr)	Puberty/sperm count	Sense of smell/ OB on MRI	Mutation (Ref.)/ additional mutation[a]
KAL1				
1[b]	17	No/az	A/ap (B)	Gene deletion (11)/No
2[b]	18	No/az	A/ap (B)	Gene deletion (11)/No
3	34	No/az	A/ap (B)	Gene deletion[c]/No
4	35	No/az	A/ap (B)	Gene deletion[c]/No
5	26	No/az	A/ap (B)	IVS1+1G→A (5)/No Truncated protein
6[b]	17	No/NE	A/ap (B)	p.R191fsX14 (5)/No
7[b]	20	No/az	A/ap (B)	p.R191fsX14 (5)/No
8	40	No/az	A/ap (B)	p.R262X (5)/No
9[b]	17	No/NE	A/ap (B)	p.N267K[c]/No
10[b]	19	No/az	A/ap (B)	p.N267K[c]/No
11	17	No/az	A/ap (B)	1016–1017insA (5) Truncated protein/No
12[b]	21	No/az	A/ap (B)	p.R423X (5)/No
13[b]	18	No/az	A/ap (B)	p.R423X (5)/No
14	21	No/az	A/hyp (B)	p.R424X [c]/No
15	18	No/NE	An/NE	p.R457X (5)/No
16[b]	22	No/az	An/ap (B)	p.P551-E552delinsSX (5)/No
17[b]	17	No/az	An/ap (B)	p.P551-E552delinsSX (5)/No
18[b]	25	No/az	An/hyp (B)	p.P551-E552delinsSX (5)/No
19	17	No/az	A/NE	p.W571R (5)/No
20[b]	19	No/az	A/ap (B)	p.Y617X[c]/No
21[b]	17	No/az	A/ap (B)	p.Y617X[c]/No
FGFR1/KAL2				
22[b]	17	No/az	A/ap (B)	p.Y99C (3)/No
23[b]	52	Yes/NE	N/N	p.Y99C (3)/No
24	22	No/az	A/ap (B)	p.C101F (7)/No
25	47	Yes/NE	H/hyp (B)	p.V102I (5)/No
26	18	No/az	A/ap (B)	p.D129A (5)/No
27	17	No/az	A/ap (B)	p.V273M (5)/No
28	27	No/az	A/ap (B)	p.C277Y (3)/No
29	20	No/az	A/ap (B)	p.R365fsX41 (5)/No
30[b]	19	No/az	A/ap (B)	p.V607M (3)/No
31[b]	55	Yes/NE	N/N	p.V607M (3)/No
32[b]	17	No/az	A/ap (B)	c.1970–1971delCA[d] (3)/No
33[b]	35	No/az	A/ap (B)	c.1970–1971delCA[d] (3)/No
34	24	No/az	A/ap (B)	p.R622X (3)/No
35	23	No/oligo[e]	A/ap (B)	p.R661X (7)/No
36	26	No/az	A/NE	p.S685F (7)/No
37	25	No/az	A/ap (B)	p.Y730X (5)/No
38	23	No/az	A/ap (B)	p.P772S (7)/No
39	21	No/az	H/hyp (U)	p.R822C (7)/No

In patients 23, 25, and 31, plasma testosterone levels were 21, 10.2, and 19.8 nmol/liter, respectively. A, Anosmia; ap, aplasia; az, azoospermia; B, bilateral; H, hyposmia; hyp, hypoplasia; NE, not examined; OB, olfactory bulbs; U, unilateral.

[a] In *Prokineticin-2*, *Prokineticin receptor-2*, and *FGFR1/KAL2* genes in patients 1–21 and in *Prokineticin-2* and *Prokineticin receptor-2* genes in patients 22–39.

[b] Patients with the same mutation are members of the same kindred.

[c] Present work.

[d] Frameshift mutation.

[e] Oligospermia: 3.9×10^6/ml.

groups of patients. Mean serum concentrations of inhibin B were below normal in the two groups of patients and significantly lower in the CHH patients with *KAL1* mutations than in those with *FGFR1/KAL2* mutations ($P < 0.005$) (Fig. 1C).

Basal and stimulated gonadotropin levels

Mean basal FSH (0.72 ± 0.47 and 1.48 ± 0.62 IU/liter, $P < 0.05$) (Fig. 1D) and LH (0.57 ± 0.54 and 1.0 ± 0.60 IU/liter, $P < 0.001$) (Fig. 1E) levels were much lower than normal in the two groups of patients and were significantly lower in the patients with *KAL1* mutations than in those with *FGFR1/KAL2* mutations. The mean poststimulation LH peak level (0.9 ± 0.9 *vs.* 4.0 ± 3.8 IU/liter; $P < 0.002$) was also significantly lower in CHH patients with *KAL1* mutations (Fig. 1F).

LH secretion

All the tested patients had apulsatile LH secretion. The mean plasma LH level (0.37 ± 0.24 IU/liter) was lower in the patients

Downloaded from https://academic.oup.com/jcem/article/93/3/758/2598286 by guest on 21 August 2022

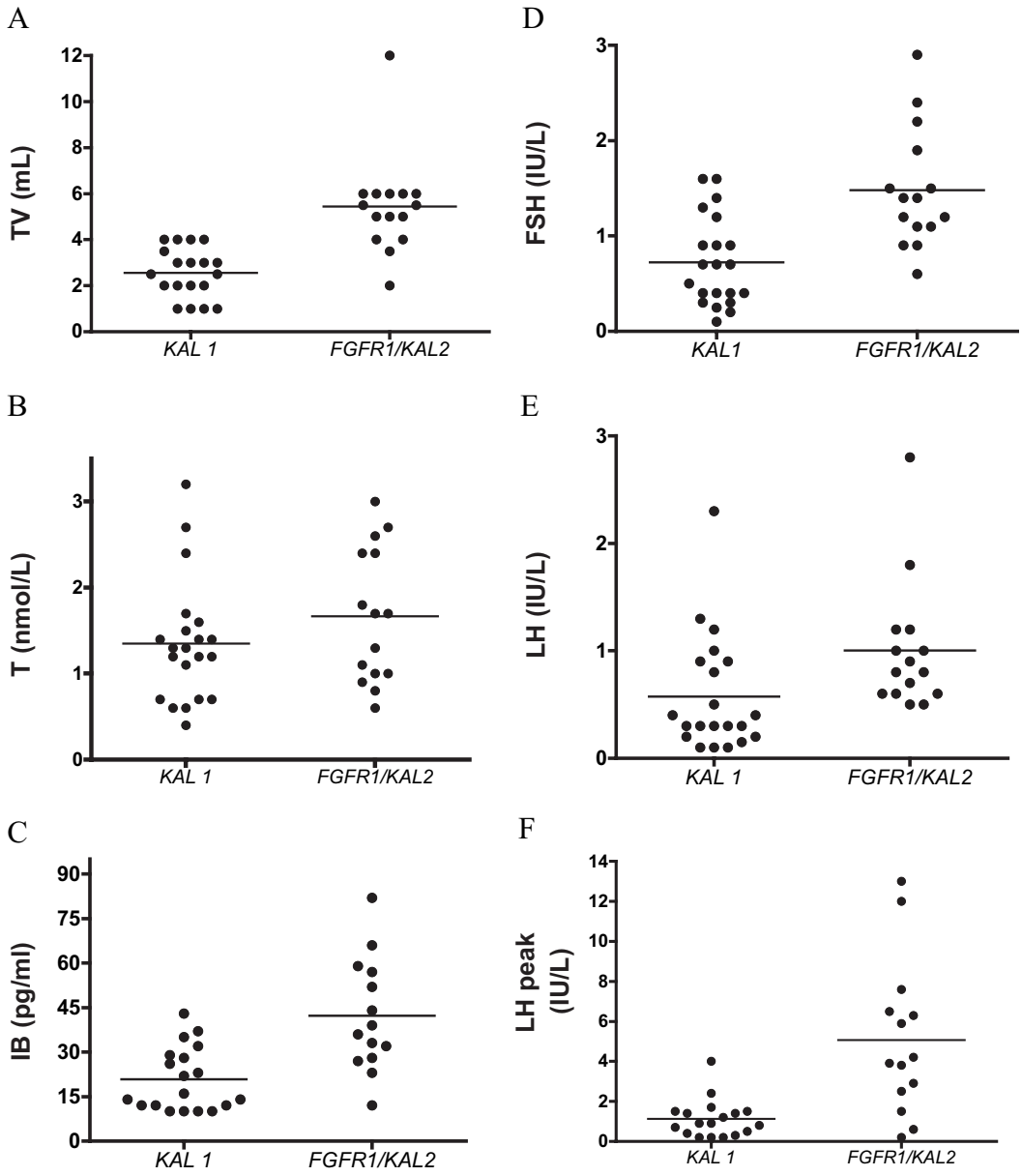

FIG. 1. Individual mean testicular volume (TV) (A), plasma total testosterone (T) (B), serum inhibin B (IB) (C), basal plasma FSH (D), basal plasma LH (E) levels, and individual LH response (peak) to GnRH (100 μg iv) (F) in CHH patients with *KAL1* and *FGFR1/KAL2* mutations. The normal ranges in adult men were 15–30 ml for testicular volume, 9.7–28.4 nmol/liter for testosterone, 96–360 pg/ml for inhibin B, 3.0–7.0 IU/liter for basal FSH and 2.9–8.0 IU/liter for basal LH. A significant positive correlation (Spearman's rank correlation procedure) was observed between inhibin B and testicular volume in CHH patients with *KAL1* ($r^2 = 0.52$; $P < 0.001$) and *FGFR1/KAL2* ($r^2 = 0.49$; $P < 0.01$) mutations. The Kolmogorov-Smirnov nonparametric test was used to compare above quantitative variables between CHH patients with *KAL1* and *FGFR1/KAL2* mutations. *P* values < 0.05 were considered to denote statistical significance (see text). In two patients with *KAL1* mutations and cryptorchidism, testicular volume measurements were not available.

with *KAL1* mutations than in those with *FGFR1/KAL2* mutations (1.13 ± 0.77 IU/liter; $P < 0.01$).

Discussion

We compared the prevalence and severity of CHH in 38 patients with characterized *KAL1* and *FGFR1/KAL2* mutations. Indeed, before *FGFR1/KAL2* mutations were shown to cause an autosomal dominant form of the syndrome, some studies were designed to compare the overall severity of CHH between patients with KS and patients with normosmic CHH (8, 9). The *FGFR1/KAL2* gene was not screened for mutations, and/or the *KAL1* gene was screened only in a minority of patients considered to have the X-linked mode of inheritance (8, 9, 17). However, we now know that some patients with a pedigree suggesting an X-linked mode of inheritance in fact have *FGFR1/KAL2* mutations, undermining comparisons based mainly on pedigree classification (3–5, 7). Overall, these former studies indicated a more severe gonadotrope phenotype in patients with KS, but the genetic and clinical heterogeneity of this syndrome was not always taken into account. Our results clearly show the more severe gonadal status of patients with *KAL1* mutations relative to those with *FGFR1/KAL2* mutations. The former patients also had a

Downloaded from https://academic.oup.com/jcem/article/93/3/758/2598286 by guest on 21 August 2022

higher prevalence of cryptorchidism, which reflects severe perinatal gonadotropin deficiency (18).

Circulating levels of inhibin B were significantly lower in CHH patients with *KAL1* mutations. This finding also indicates that CHH is more severe in patients with *KAL1* mutations than in those with *FGFR1/KAL2* mutations. Given the known prognostic value of cryptorchidism, testicular volume, and inhibin B levels for fertility in patients with CHH (19), infertility could be more difficult to treat and the risk of testicular cancer could be increased in men with KS due to *KAL1* mutations.

Another finding is that LH secretion was apulsatile in both groups of patients. The mean LH level was significantly lower in *KAL1* patients than in *FGFR1/KAL2* patients, indicating that gonadotropin secretion is more severely affected in the former patients (17). This is further supported by the stronger decline in FSH and the smaller increase in LH after GnRH stimulation in *KAL1* patients.

The normal pubertal development and gonadal status in three of the men with *FGFR1/KAL2* mutations differs from the phenotype reported by Pitteloud *et al.* (6) in a man with CHH and an *FGFR1/KAL2* mutation who had late puberty but recovered normal gonadotropic function in adulthood. These three men were identified during familial investigations of their sons, who had CHH and anosmia, underlining the variable penetrance of this genetic form of KS. In contrast, CHH seems to show almost complete penetrance in men with documented *KAL1* mutations. Thus, men with *FGFR1/KAL2* mutations have a broad spectrum of pubertal development and less severe CHH than men with *KAL1* mutations. Our results are in line with a recent, noncomparative study by Pitteloud *et al.* (6) who reported 15 men with *FGFR1/KAL2* mutations.

In the absence of additional mutations in three others KS loci (Table 1) indicating a digenic mechanism (20), it is unclear why *KAL1* mutations should be associated with a more severe gonadotrope phenotype than *FGFR1/KAL2* mutations. The traits shared by patients with *KAL1* and *FGFR1/KAL2* mutations and *in vitro* studies suggest that anosmin and FGFR1 might interact (4, 21). We can speculate that in the absence of a *KAL1* healthy allele in men, loss-of-function mutations in *KAL1* could lead to more severe impairment of the common downstream pathway than heterozygous *FGFR1/KAL2* mutations; in the latter, residual expression of the normal FGFR1 protein encoded by the wild-type allele could attenuate the consequences of the lack of anosmin/FGFR1-mediated GnRH neuron migration during fetal life (4, 21). This could result in a milder reproductive phenotype in men with heterozygous *FGFR1/KAL2* mutations.

In conclusion, this study shows that men with KS and documented *KAL1* mutations have a more severe reproductive phenotype than men with *FGFR1/KAL2* mutations, which are associated with a broad spectrum of phenotypes, ranging from complete CHH to normality.

Acknowledgments

Address all correspondence and requests for reprints to: Jacques Young, Service d'Endocrinologie, Hôpital de Bicêtre, 94275 Le Kremlin-Bicêtre, France. E-mail: jacques.young@bct.aphp.fr.

Disclosure Statement: The authors have nothing to disclose.

References

1. **Legouis R, Hardelin JP, Levilliers J, Claverie JM, Compain S, Wunderle V, Millasseau P, Le Paslier D, Cohen D, Caterina D, Bougueleret L, Delemarre-Van de Waal H, Lutfalla G, Weissenbach J, Petit C** 1991 The candidate gene for the X-linked Kallmann syndrome encodes a protein related to adhesion molecules. Cell 67:423–435
2. **Franco B, Guioli S, Pragliola A, Incerti B, Bardoni B, Tonlorenzi R, Carrozzo R, Maestrini E, Pieretti M, Taillon-Miller P, Brown CJ, Willard HF, Lawrence C, Persico MG, Camerino G, Ballabio A** 1991 A gene deleted in Kallmann's syndrome shares homology with neural cell adhesion and axonal path-finding molecules. Nature 353:529–536
3. **Dodé C, Levilliers J, Dupont JM, De Paepe A, Le Du N, Soussi-Yanicostas N, Coimbra RS, Delmaghani S, Compain-Nouaille S, Baverel F, Pecheux C, Le Tessier D, Cruaud C, Delpech M, Speleman F, Vermeulen S, Amalfitano A, Bachelot Y, Bouchard P, Cabrol S, Carel JC, Delemarre-van de Waal H, Goulet-Salmon B, Kottler ML, Richard O, et al** 2003 Loss-of-function mutations in FGFR1 cause autosomal dominant Kallmann syndrome. Nat Genet 33:463–465
4. **Dodé C, Hardelin JP** 2004 Kallmann syndrome: fibroblast growth factor signaling insufficiency? J Mol Med 82:725–734
5. **Albuisson J, Pecheux C, Carel JC, Lacombe D, Leheup B, Lapuzina P, Bouchard P, Legius E, Matthijs G, Wasniewska M, Delpech M, Young J, Hardelin JP, Dode C** 2005 Kallmann syndrome: 14 novel mutations in KAL1 and FGFR1 (KAL2). Hum Mutat 25:98–99
6. **Pitteloud N, Meysing A, Quinton R, Acierno JS Jr., Dwyer AA, Plummer L, Fliers E, Boepple P, Hayes F, Seminara S, Hughes VA, Ma J, Bouloux P, Mohammadi M, Crowley Jr WF** 2006 Mutations in fibroblast growth factor receptor 1 cause Kallmann syndrome with a wide spectrum of reproductive phenotypes. Mol Cell Endocrinol 254–255:60–69
7. **Dodé C, Fouveaut C, Mortier G, Janssens S, Bertherat J, Mahoudeau J, Kottler ML, Chabrolle C, Gancel A, François I, Devriendt K, Wolczynski S, Pugeat M, Pineiro-Garcia A, Murat A, Bouchard P, Young J, Delpech M, Hardelin JP** 2007 Novel *FGFR1* sequence variants in Kallmann syndrome, and genetic evidence that the FGFR1c isoform is required in olfactory bulb and palate morphogenesis. Hum Mutat 28:97–98
8. **Pitteloud N, Hayes FJ, Boepple PA, DeCruz S, Seminara SB, MacLaughlin DT, Crowley Jr WF** 2002 The role of prior pubertal development, biochemical markers of testicular maturation, and genetics in elucidating the phenotypic heterogeneity of idiopathic hypogonadotropic hypogonadism. J Clin Endocrinol Metab 87:152–160
9. **Quinton R, Duke VM, Robertson A, Kirk JM, Matfin G, de Zoysa PA, Azcona C, MacColl GS, Jacobs HS, Conway GS, Besser M, Stanhope RG, Bouloux PM** 2001 Idiopathic gonadotrophin deficiency: genetic questions addressed through phenotypic characterization. Clin Endocrinol (Oxf) 55:163–174
10. **Dodé C, Teixeira L, Levilliers J, Fouveaut C, Bouchard P, Kottler ML, Lespinasse J, Lienhardt-Roussie A, Mathieu M, Moerman A, Morgan G, Murat A, Toublanc JE, Wolczynski S, Delpech M, Petit C, Young J, Hardelin JP** 2006 Kallmann syndrome: mutations in the genes encoding prokineticin-2 and prokineticin receptor-2. PLoS Genet 2:e175
11. **Hardelin JP, Levilliers J, Young J, Pholsena M, Legouis R, Kirk J, Bouloux P, Petit C, Schaison G** 1993 Xp22.3 deletions in isolated familial Kallmann's syndrome. J Clin Endocrinol Metab 76:827–831
12. **Eloit C, Trotier D** 1994 A new clinical olfactory test to quantify olfactory deficiencies. Rhinology 32:57–61
13. **Bin-Abbas B, Conte FA, Grumbach MM, Kaplan SL** 1999 Congenital hypogonadotropic hypogonadism and micropenis: effect of testosterone treatment on adult penile size why sex reversal is not indicated. J Pediatr 134:579–583
14. **Young J, Chanson P, Salenave S, Noel M, Brailly S, O'Flaherty M, Schaison G, Rey R** 2005 Testicular anti-Mullerian hormone secretion is stimulated by recombinant human FSH in patients with congenital hypogonadotropic hypogonadism. J Clin Endocrinol Metab 90:724–728
15. **de Roux N, Young J, Misrahi M, Genet R, Chanson P, Schaison G, Milgrom E** 1997 A family with hypogonadotropic hypogonadism and mutations in the gonadotropin-releasing hormone receptor. N Engl J Med 337:1597–1602
16. **Thomas G, Plu G, Thalabard JC** 1992 Identification of pulses in hormone time series using outlier detection methods. Stat Med 11:2133–2145
17. **Oliveira LM, Seminara SB, Beranova M, Hayes FJ, Valkenburgh SB, Schipani E, Costa EM, Latronico AC, Crowley Jr WF, Vallejo M** 2001 The

Downloaded from https://academic.oup.com/jcem/article/93/3/758/2598286 by guest on 21 August 2022

importance of autosomal genes in Kallmann syndrome: genotype-phenotype correlations and neuroendocrine characteristics. J Clin Endocrinol Metab 86:1532–1538
18. **Grumbach MM** 2005 A window of opportunity: the diagnosis of gonadotropin deficiency in the male infant. J Clin Endocrinol Metab 90:3122–3127
19. **Pitteloud N, Hayes FJ, Dwyer A, Boepple PA, Lee H, Crowley Jr WF** 2002 Predictors of outcome of long-term GnRH therapy in men with idiopathic hypogonadotropic hypogonadism. J Clin Endocrinol Metab 87:4128–4136
20. **Pitteloud N, Quinton R, Pearce S, Raivio T, Acierno J, Dwyer A, Plummer L, Hughes V, Seminara S, Cheng YZ, Li WP, Maccoll G, Eliseenkova AV, Olsen SK, Ibrahimi OA, Hayes FJ, Boepple P, Hall JE, Bouloux P, Mohammadi M, Crowley W** 2007 Digenic mutations account for variable phenotypes in idiopathic hypogonadotropic hypogonadism. J Clin Invest 117:457–463
21. **Gonzalez-Martinez D, Kim SH, Hu Y, Guimond S, Schofield J, Winyard P, Vannelli GB, Turnbull J, Bouloux PM** 2004 Anosmin-1 modulates fibroblast growth factor receptor 1 signaling in human gonadotropin-releasing hormone olfactory neuroblasts through a heparan sulfate-dependent mechanism. J Neurosci 24:10384–10392

Downloaded from https://academic.oup.com/jcem/article/93/3/758/2598286 by guest on 21 August 2022