



HAL
open science

Expanding the genetic and clinical spectrum of Tatton-Brown-Rahman syndrome in a series of 24 French patients

Hortense Thomas, Tom Alix, Émeline Renard, Mathilde Renaud, Justine Wourms, Stéphane Zuily, Bruno Leheup, David Geneviève, Natacha Dreumont, Emmanuelle Schmitt, et al.

► **To cite this version:**

Hortense Thomas, Tom Alix, Émeline Renard, Mathilde Renaud, Justine Wourms, et al.. Expanding the genetic and clinical spectrum of Tatton-Brown-Rahman syndrome in a series of 24 French patients. *Journal of Medical Genetics*, 2024, 61 (9), pp.878-885. 10.1136/jmg-2024-110031 . hal-04811418

HAL Id: hal-04811418

<https://univ-rennes.hal.science/hal-04811418v1>

Submitted on 19 Dec 2024

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution - NonCommercial 4.0 International License

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25

Expanding the Genetic and Clinical Spectrum of Tatton-Brown-Rahman Syndrome in a Series of 24 French Patients

Authors

Hortense Thomas* (1), Tom Alix* (1, 2), Émeline Renard (2, 3), Mathilde Renaud (2, 4), Justine Wourms (4), Stéphane Zuily (5, 6), Bruno Leheup (2), David Geneviève (7, 8), Natacha Dreumont (2), Emmanuelle Schmitt (9), Myriam Bronner (1), Marc Muller (1), Marion Divoux (1), Marion Wandzel (1), Jean-Marie Ravel (1, 2, 4), Mylène Dexheimer (1), Aurélie Becker (1), Virginie Roth (1), Marjolaine Willems (7), Christine Coubes (7), Gaëlle Vieville (10), Françoise Devillard (10), Elise Schaefer (11), Sarah Baer (11), Amélie Piton (11), Bénédicte Gérard (11), Marie Vincent (12), Mathilde Nizon (12), Benjamin Cogné (12), Lyse Ruaud (13), Nathalie Couque (13), Audrey Putoux (14), Patrick Edery (14), Gaëtan Lesca (14), Nicolas Chatron (14), Marianne Till (14), Laurence Faivre (15, 16), Frédéric Tran Mau-Them (16, 17), Jean-Luc Alessandri (18), Marine Lebrun (19), Chloé Quelin (20), Sylvie Odent (20), Christèle Dubourg (20), Véronique David (20), Marie Faucher (20), Cyril Mignot (21), Boris Keren (21), Elise Pisan (21), Alexandra Afenjar (21), Sophie Julia (22), Eric Bieth (22), Guillaume Banneau (22), Alice Goldenberg (23), Thomas Husson (24,25), Dominique Champion (24,25), François Lecoquierre (23), Gaël Nicolas (23), Camille Charbonnier (26), Anne de Saint Martin (27), Sophie Naudion (28), Manon Degoutin (28), Sophie Rondeau (29), Caroline Michot (29), Valérie Cormier-Daire (29), Abderrahim Oussalah (2, 30), Carine Bossenmeyer-Pourié (2), Laëticia Lambert† (2, 4), Céline Bonnet† (1, 2)

1 **Affiliations**

2 1. Laboratoire de génétique, CHRU de Nancy, Vandœuvre-lès-Nancy, France; 2.
3 INSERM NGERE U1256, Université de Lorraine, Vandœuvre-lès-Nancy, France; 3.
4 Endocrinologie pédiatrique, CHRU de Nancy, Vandœuvre-lès-Nancy; 4. Service de
5 Génétique Clinique, CHRU de Nancy, Vandœuvre-lès-Nancy, France; 5. Médecine
6 vasculaire, CHRU de Nancy, Vandœuvre-lès-Nancy, France; 6. INSERM UMR_S
7 916 DCAC, Université de Lorraine, Vandœuvre-lès-Nancy, France; 7. Centre de
8 référence anomalies du développement et syndromes malformatifs, Département de
9 Génétique Médicale, Maladies Rares et Médecine Personnalisée, Hôpital Arnaud de
10 Villeneuve, CHU de Montpellier, France; 8. Université Montpellier, Inserm U1183,
11 Montpellier, France; 9. Neuroradiologie, CHRU de Nancy, Vandœuvre-lès-Nancy,
12 France; 10. Département de Génétique et Procréation, Hôpital Couple Enfant, CHU
13 Grenoble, 38043 Grenoble Cedex, France; 11. Service de Génétique médicale,
14 Institut de Génétique Médicale d'Alsace, CHU Strasbourg, Strasbourg, France; 12.
15 CHU Nantes, Service de Génétique Médicale, Nantes Université, CNRS, INSERM,
16 l'institut du thorax, Nantes, France; 13. Département de Génétique, Hôpital Robert
17 Debré, APHP Nord, Paris, France; 14. Hospices Civils de Lyon, Groupe Hospitalier
18 Est, Service de Génétique, Bron, France; Institut NeuroMyoGene PNMG, CNRS
19 UMR5310, INSERM U1217, Université Claude Bernard Lyon 1, Lyon, France; 15.
20 Centre de référence anomalies du développement et syndromes malformatifs et
21 Centre de référence Déficiences Intellectuelles de causes rares, FHU TRANSLAD,
22 CHU Dijon Bourgogne, Dijon, France; 16. UMR1231 GAD, Inserm - Université
23 Bourgogne-Franche Comté, Dijon, France; 17. Unité Fonctionnelle Innovation en
24 Diagnostic génomique des maladies rares, FHU-TRANSLAD, CHU Dijon Bourgogne,
25 Dijon, France; 18. Service de génétique médicale, CHU de la Réunion - Hôpital Félix

1 Guyon, Bellepierre, 97405 Saint-Denis, France; 19. Département de Génétique, CHU
2 Saint Etienne, Saint Etienne, France; 20. Département de génétique moléculaire et
3 génomique, CHU Rennes, Rennes, France; 21. APHP Sorbonne Université,
4 Département de Génétique, Groupe Hospitalier Pitié-Salpêtrière, Paris, France; 22.
5 Département de Génétique médicale, CHU Toulouse, Toulouse, France; 23. Univ
6 Rouen Normandie, Inserm U1245 and CHU Rouen, Department of Genetics and
7 Reference Center for Developmental Disorders, F-76000 Rouen, France; 24. Univ
8 Rouen Normandie, Inserm U1245 and CHU Rouen, Department of Psychiatry, F-
9 76000, Rouen, France; 25. Department of Research, Centre hospitalier du Rouvray,
10 Sotteville-Lès-Rouen, France; 26. Univ Rouen Normandie, Inserm U1245 and CHU
11 Rouen, Department of Biotatistics, F-76000 Rouen, France; 27. Centre de Référence
12 des épilepsies Rares, CHRU de Strasbourg, Strasbourg, France; 28. Service de
13 Génétique Médicale, CHU Bordeaux, Bordeaux, France; 29. Université Paris Cité,
14 Centre de référence des maladies osseuses constitutionnelles, INSERM UMR 1163,
15 Institut Imagine, Hôpital Necker, APHP, Paris, France; 30. Department of Molecular
16 Medicine, Division of Biochemistry, Molecular Biology, and Nutrition, University
17 Hospital of Nancy, F-54000 Nancy, France.

18

19 * These authors contributed equally to this work.

20 † These authors contributed equally to this work and share senior authorship.

21

22 **Correspondence:** Céline BONNET, Laboratoire de génétique médicale, Centre
23 Hospitalier et Universitaire de Nancy, Rue du Morvan, 54511 Vandœuvre-lès-Nancy
24 Cedex, France. Email: ce.bonnet@chru-nancy.fr

1 **ABSTRACT**

2

3 Background: Tatton-Brown-Rahman syndrome (TBRS; OMIM 615879), also known
4 as *DNMT3A*-overgrowth syndrome (DOS), was first described by Tatton-Brown in
5 2014. This syndrome is characterized by overgrowth, intellectual disability, and
6 distinctive facial features and is the consequence of germline loss-of-function variants
7 in *DNMT3A*, which encodes a DNA methyltransferase involved in epigenetic
8 regulation. Somatic variants of *DNMT3A* are frequently observed in hematological
9 malignancies, including acute myeloid leukemia (AML). To date, one hundred TBRS
10 individuals with *de novo* germline variants have been described. We aimed to further
11 characterize this disorder clinically and at the molecular level in a nationwide series
12 of 24 French patients and to investigate the correlation between the severity of
13 intellectual disability and the type of variant. Methods: We collected genetic and
14 medical information from 24 individuals with TBRS using a questionnaire released
15 through the French National AnDDI-Rares Network. Results: Here, we describe the
16 first nationwide French cohort of 24 individuals with germline likely
17 pathogenic/pathogenic variants in *DNMT3A*, including 17 novel variants. We
18 confirmed that the main phenotypic features were intellectual disability (100% of
19 individuals), distinctive facial features (96%), and overgrowth (87%). We highlighted
20 novel clinical features, such as hypertrichosis, and further described the neurological
21 features and EEG results. Conclusion: This study of a nationwide cohort of
22 individuals with TBRS not only confirms previously published data but also provides
23 additional information and clarifies clinical features to facilitate diagnosis and improve
24 care. This study adds value to the growing body of knowledge on TBRS and
25 broadens its clinical and molecular spectrum.

1

2 **Keywords:** *DNMT3A*, Tatton-Brown-Rahman syndrome, overgrowth, intellectual
3 disability

4

5 **Key messages**

6

7 **What is already known on this topic**

8 ⇒ Tatton-Brown-Rahman syndrome is a recently described overgrowth/intellectual
9 disability syndrome caused by loss-of-function variants in *DNMT3A*.

10 ⇒ Few large studies have delineated the phenotype–genotype spectrum of Tatton-
11 Brown-Rahman syndrome.

12

13 **What this study adds**

14 ⇒ Our study describes 24 additional affected individuals and 17 previously
15 unreported *DNMT3A* genetic variants.

16

17 **How this study might affect research, practice or policy**

18 ⇒ This study adds to the aggregate knowledge of Tatton-Brown-Rahman syndrome
19 and expands its clinical and molecular spectrum to facilitate the identification of
20 affected individuals and improve care.

1 INTRODUCTION

2

3 Tatton-Brown-Rahman syndrome (TBRS, OMIM #615879) was first described in
4 2014 by Katrina Tatton-Brown and Nazneen Rahman [1]. In a second publication, 55
5 individuals were clinically evaluated, including 13 previously reported individuals [2].
6 Numerous subsequent reports have confirmed the original findings [3–26]. TBRS is
7 characterized mainly by overgrowth (defined as height and/or head circumference at
8 least 2 standard deviations above the population mean); intellectual disability ranging
9 from mild to severe; and distinctive facial features, including coarse features, narrow
10 palpebral fissures, and heavy and horizontal eyebrows. In addition to these three
11 main characteristics, other features, such as hypotonia, behavioral disorders,
12 seizures, joint hypermobility, scoliosis, dental abnormalities, and predisposition to
13 malignant hemopathies, are more inconsistently described. The vast majority of
14 reported individuals are children. Most cases are sporadic and occur in individuals
15 with *de novo* heterozygous germline variants in *DNMT3A*. However, parent-to-child
16 transmission in some families has demonstrated autosomal dominant inheritance
17 with variable expressivity, and inheritance from unaffected mosaic parents has been
18 reported [5, 6, 16, 22]. An increasing number of case reports converge toward
19 phenotypic variability and multisystem involvement in this syndrome.

20 DNA methyltransferase 3 alpha (*DNMT3A*) is one of three active human
21 methyltransferases, with the others being *DNMT3B* and *DNMT1*. *DNMT3A* is
22 involved in *de novo* DNA methylation during development. This enzyme also plays a
23 role in the maintenance and remodeling of DNA methylation patterns, especially at
24 CpG sites and in histone proteins. In addition to this central role in epigenetic
25 regulation and chromatin configuration, a role in gene regulatory functions such as

1 transcriptional silencing and activation has been reported [27, 28]. DNMT3A is also
2 essential for parental imprinting [29] and XIST repression, which is involved in X
3 chromosome inactivation [30].

4 DNMT3A contains three functional domains: i) a proline-tryptophan-tryptophan-
5 proline (PWWP) domain present in transcription factors involved in the regulation of
6 developmental processes, ii) an ATRX-DNMT3-DNMT3L (ADD) domain implicated in
7 protein–protein and protein–DNA binding interactions, and iii) a methyltransferase
8 (MTase) domain essential for methyltransferase activity.

9 In individuals with TBRS, *DNMT3A* germline variants lead to protein loss of function.
10 Germline gain-of-function variants in the *DNMT3A* PWWP domain have been
11 implicated in Heyn-Sproul-Jackson syndrome (HESJAS, OMIM #618724), which is
12 characterized by a mirrored phenotype to TBRS, i.e., microcephalic dwarfism.

13 In addition to *DNMT3A* constitutional variants, somatic mutations have been reported
14 in acute myeloid leukemia (AML, OMIM #601626) and other hematopoietic
15 malignancies. More than half of *DNMT3A* somatic variants occur at amino acid
16 p.Arg882, which is located in the MTase domain [31, 32]. However, the spectrum of
17 germline variants overlaps with that observed in hematological malignancies [15].
18 Furthermore, TBRS is associated with perturbed hematopoiesis and the development
19 of hematopoietic malignancy in children and young adults [2, 10, 11, 13, 33].

20 Approximately one hundred individuals have been reported in the literature to date;
21 however, with the increasing availability of next-generation sequencing, the number
22 of individuals diagnosed with germline mutations in *DNMT3A* is growing, making it
23 necessary to improve our knowledge of the disease, its evolution and its individual
24 management.

1 In this nationwide study, we report 24 novel individuals exhibiting TBRS associated
2 with 17 novel disease-causing variants in *DNMT3A*.

3

4 **SUBJECTS AND METHODS**

5

6 **Individual recruitment.** A national collaboration through the French National AnDDI-
7 Rares (*Developmental Abnormalities with or without Intellectual Deficiency of Rare*
8 *Causes*) health network made it possible to collect 24 individuals with TBRS between
9 January 2021 and January 2023. The inclusion criteria were as follows: 1) the
10 presence of a germline likely pathogenic or pathogenic variant in *DNMT3A* and 2)
11 available molecular and clinical data. A *DNMT3A*-specific clinical proforma was sent
12 to genetic specialists (including clinical elements present in overgrowth and
13 intellectual disability syndromes and clinical symptoms already reported in the
14 literature in individuals with TBRS). When available, we collected photographs,
15 growth curves, and imaging data. Brain imaging was reinterpreted by a local
16 specialist for seven individuals with available radiological data. *DNMT3A* variants
17 were detected via gene panel, exome, or genome sequencing. The reference
18 transcript used for variant designation was NM_022552.5. Variants were interpreted
19 according to the American College of Medical Genetics and Genomics (ACMG)
20 guidelines [34]. All individuals or legal guardians provided written informed consent
21 for genetic analyses. As required by French law, we informed all participants of their
22 right to oppose the use of their data. Individuals or legal guardians have given their
23 consent for the use of their photographs in the publication. This study was approved
24 by the ethics committee of Nancy University Hospital (referral n°429) and was
25 conducted in accordance with the Declaration of Helsinki.

1

2 **Literature review.** We performed a comprehensive PubMed search with the search
3 terms “DNMT3A” and “Tatton-Brown-Rahman syndrome” and manually assessed the
4 articles to identify those reporting TBRS cases. A total of 107 individuals with
5 germline likely pathogenic/pathogenic variants in *DNMT3A* and available clinical data
6 were collected.

7

8 **Statistical analyses.** We determined whether there was an association between the
9 severity of intellectual disability and the type of variant using a chi-square test in the
10 83 individuals with available data merged from our cohort and the literature.

11

12 **RESULTS** (Table 1, Supplementary Table 1)

13

14 We collected clinical and molecular data from 24 individuals (10 males and 14
15 females) from 23 families (Tables 1 and Supplementary Table 1). The mean age was
16 15.6 years (we excluded Patient 7’s mother from the calculation). Patient 5 has
17 already been reported in the literature [14]. All individuals benefited from molecular
18 diagnosis that identified pathogenic or likely pathogenic variants in *DNMT3A* with
19 predicted loss of function.

20

21 **Molecular characteristics** (Figure 1)

22 We identified 13 different missense variants (in 15 individuals), 5 different nonsense
23 variants, a frameshift variant, a splice-site variant, a deletion of several exons and an
24 in-frame duplication. The pathogenicity of this in-frame duplication relies on the high
25 interspecies conservation level of the 6 duplicated amino acids, proximity to the

1 PWWP domain and DNA methylation profile showing a concordant TBRS
2 epesignature. The results of a methylation study performed for this individual using
3 the Illumina Infinium EPIC array v1.0 have previously been reported [35]. Among the
4 variants, 17 have never been reported before (9 missense variants, 4 nonsense
5 variants, 1 frameshift variant, 1 splice-site variant, 1 multiexon deletion, and 1 in-
6 frame duplication). The remaining 5 variants (found in 6 individuals: 3, 5, 11, 15, 22
7 and 24) have previously been reported in individuals with TBRS [2, 5, 11, 14, 15, 18].
8 Nineteen individuals carried a *de novo* variant, and 2 individuals inherited the variant
9 from an affected parent; for the remaining 3 individuals, parental DNA was
10 unavailable. Only one of the two affected parents was included in this study due to
11 the lack of clinical information.

12

13 **Pregnancy and the perinatal period**

14 No toxic exposure or assisted reproductive technology was reported. The average
15 age at delivery was 28 years for the mothers and 35 years for the fathers.
16 Pregnancies were marked by various nonspecific complications in 33% (8/24) of the
17 patients, including increased nuchal translucency (n = 1), macrosomia without
18 gestational diabetes (n = 1), intrauterine growth retardation and further normalization
19 (n = 1), intrauterine growth restriction with preterm delivery and neonatal hypoxia (n =
20 1), and fetal distress (n = 1). In 17% (4/24) of the patients, various nonspecific
21 neonatal abnormalities, including hypoglycemia, hypotonia, sleeping disorders,
22 feeding difficulties, and bronchopulmonary dysplasia, were reported.

23

24 **Growth and endocrinology**

1 Most individuals had generalized overgrowth since birth. At birth, 48% (10/21) had a
2 length above the 90th centile, 64% (14/22) had a weight above the 90th centile, and
3 53% (10/19) had a head circumference above the 90th centile. At the time of the last
4 clinical examination (mean age: 13 years, standard deviation: 8 years), overgrowth
5 (defined as height and/or head circumference at least 2 standard deviations above
6 the population mean) was observed in 87% (20/23) of the individuals, 65% (15/23)
7 had a tall stature, 46% (10/22) were overweight according to BMI, 76% (16/21) were
8 overweight according to weight, and 68% (15/22) had macrocephaly. The available
9 weight and height curves (for 4 male and 5 female individuals) showed that the
10 measurements remained approximately stable within the same range (data not
11 shown).

12 Thirty percent (3/10) of male patients had cryptorchidism. Twenty-nine percent (4/14)
13 of female patients had early onset of puberty. Hypertrichosis was observed in 46%
14 (11/24) of patients. Hair extended into the frontal regions associated with thick
15 eyebrows, and hypertrichosis sometimes involved the face, back, and arms (Figure
16 2C).

17

18 **Facial features**

19 Distinct facial features were observed in 96% of the individuals (23/24). Fifty percent
20 (12/24) had coarse features, and 38% (9/24) had a round face. Forty-six percent
21 (11/24) had narrow palpebral fissures, and 21% (5/24) had enophthalmia. Twenty-
22 nine percent (7/24) had a high palate. Seventy percent (17/24) had atypical eyebrows
23 (thick, horizontal or synophrys). Facial features were more apparent in early
24 adolescence and less specific in early childhood and adulthood. Sixty-seven percent
25 (16/24) had dental abnormalities: large upper incisors (n = 13), widely spaced upper

1 incisors (n = 6), abnormal dental implantation (n = 10), multiple caries lesions (n = 2),
2 and late (n = 1) or early loss (n = 1) of the milk teeth (Figure 2A).

3

4 **Neurodevelopment and behavior**

5 One hundred percent (24/24) of individuals had intellectual disability with varying
6 degrees of severity. Of the 14 individuals for whom impairment severity was
7 specified, 29% (4/14) had mild intellectual disability (ID), 57% (8/14) had moderate
8 ID, and 14% (2/14) had severe ID. For 21% (5/24) of the individuals, a regression in
9 psychomotor acquisitions was observed. Eight percent (2/24) of individuals had
10 stuttering disorders. Twenty-nine percent (7/24) had autism spectrum disorders
11 (ASDs). Sixty-three percent (15/24) had behavioral disorders, including attention-
12 deficit/hyperactivity disorder (ADHD) (n = 3), obsessive-compulsive disorder (n = 3),
13 aggressive behavior (n = 4), and anxiety disorders (n = 4). Twenty-five percent (6/24)
14 of the individuals had eating disorders, namely, compulsions (n = 2) and hyperphagia
15 (n = 4); all six were overweight.

16

17 **Neurology**

18 Forty-two percent (10/24) of individuals had at least one episode of clinical seizures.
19 6 individuals had isolated febrile or cluster of febrile seizures; and the remaining 4
20 individuals had non-febrile epilepsy (with or without previous febrile seizures): febrile
21 seizures and generalized epilepsy (n = 1); febrile seizures, focal seizures and later
22 continuous spikes and waves during sleep (CSWS) (n = 1); febrile seizures, focal
23 seizures and absence seizures (n = 1); and focal seizures and absence seizures (n =
24 1). Patient with generalized epilepsy was successfully treated with valproate. Patient
25 with CSWS was treated with clobazam, levetiracetam, ethosuximide and

1 corticosteroids. Patients with focal and absence seizures were successfully treated
2 by lacosamide and levetiracetam for one or lamotrigine and zonisamide for the other.
3 The two individuals with severe ID had at least one episode of clinical seizures.
4 Electroencephalography (EEG) information was available for 50% (12/24) of the
5 individuals, including 6 individuals with clinical seizures; focal abnormalities were
6 observed in 3 individuals, and generalized abnormalities were observed in one
7 individual. Among individuals without clinical seizures (n = 6), only one presented
8 with EEG abnormalities, i.e., focal left centrotemporal spikes.

9
10 Other neurological manifestations were observed in 58% (14/24) of the individuals,
11 including cerebellar syndrome (n = 7), hypotonia (n = 6), a decrease in fine motor
12 coordination (n = 5), symptomatology of peripheral neuropathy (n = 3), tremor (n = 3),
13 pyramidal syndrome (n = 2) and right upper limb monoparesis (n = 1).

14
15 Brain magnetic resonance imaging (MRI) was performed for 75% (18/24) of the
16 individuals, and 72% (13/18) of these patients had abnormalities (Figure 2E). The
17 most frequent abnormality was a thin or dysplastic corpus callosum in 44% of the
18 individuals (n = 8). Other abnormalities were nonspecific: thick cranial vault bones (n
19 = 3), brain atrophy (n = 2), white matter hyperintensities (n = 2), pineal cysts (n = 2),
20 arachnoid cysts (n = 1), choroidal fissure cysts (n = 1), ventriculomegaly (n = 1), a
21 narrow posterior fossa (n = 1) and olfactory bulb hypoplasia (n = 1). None of the
22 individuals had Chiari malformations, as previously described for TBRS individuals;
23 however, protrusion of the cerebellar amygdala was observed in one individual, and
24 basilar impressions were observed in another individual.

25

1 **Additional symptoms**

2 Twenty-eight percent (9/24) of the individuals had sleep disorders, including poor
3 sleep and nocturnal awakening (n = 6), mild obstructive sleep apnea (OSA) or
4 snoring (n = 4), night sweats (n = 1) and nocturnal enuresis (n = 1). Thirteen percent
5 (3/24) of patients had recurrent infections. Forty-two percent (10/24) had refractive
6 disorders. Thirteen percent (3/24) had other ophthalmic abnormalities, such as
7 bilateral cataracts with surgical treatment (n = 1), poor central vision (n = 1), or
8 papillary excavation (n = 1). Twenty-five percent (6/24) had a history of
9 cardiovascular disease, including nonobstructive ventricular septal defect (n = 1),
10 atrial septal defect requiring surgery (n = 1), malformation of the aortic arch with the
11 retroesophageal subclavian artery (n = 1), recurrent episodes of pericarditis (n = 1),
12 familial Wolff–Parkinson–White syndrome (n = 1), and ventricular extrasystoles (n =
13 1). A total of 33% (8/24) had a respiratory or pulmonary history of mild obstructive
14 sleep apnea (OSA) or snoring (n = 4), history of repeated bronchitis and/or
15 pneumonia (n = 2), or asthma (n = 2) with recurrent pneumothorax leading to surgical
16 treatment in one individual. Twenty-one percent (5/24) had an umbilical hernia.

17

18 Seventy-five percent (18/24) of patients presented with abnormalities of the
19 extremities (hands and/or feet), including long and thin fingers (n = 9), short and/or
20 widely spaced toes (n = 10), flat feet (n = 10) and, most often, a large shoe size for
21 their age (Figure 2B). Forty-six percent (11/24) had joint hypermobility. Forty-two
22 percent (10/24) had scoliosis. Twenty-one percent (5/24) had other skeletal
23 abnormalities, including major pectus excavatum (n = 1), patellar dislocation (n = 1),
24 Scheuermann's disease (n = 1), plagiocephaly (n = 1) and dorsal gibbosity (n = 1)
25 (Figure 2D).

1

2 Patient 5, who was previously reported [14], presented at the age of 17, with
3 unsteady gait, headaches, rotatory vertigo, stage 1 papilledema on funduscopy
4 examination, ataxic gait, nystagmus, and dysdiadochokinesia. MRI revealed a fourth
5 ventricular mass causing obstructive hydrocephalus. She underwent a craniotomy,
6 and uncomplicated gross total resection. Further investigations revealed the lesion to
7 be a localized medulloblastoma in the standard risk category. She received adjuvant
8 craniospinal radiotherapy under general anesthesia. Her clinical follow-up revealed
9 no clinical or radiological evidence of disease recurrence. She continued to have a
10 cerebellar syndrome on follow-up [14].

11

12 **Genotype–phenotype association**

13 We compared the ID severity between individuals with truncating variants and
14 individuals with missense variants located in one of the three known functional
15 domains of the DNMT3A protein in this series and in the literature. The chi-square
16 independence test did not reveal a correlation between genotype and severity of
17 intellectual disability, suggesting that the mechanism of pathogenicity of missense
18 variants is a loss of function of the protein.

19

20 **DISCUSSION**

21

22 Katrina Tatton-Brown and Nazneen Rahman clinically evaluated a total of 55 TBRS
23 individuals [2]. Tlemsani et al. [18] and Tenorio et al. [8] reported 7 individuals, Balci
24 et al. [5] reported 6 individuals, and Xin et al. [6] reported 6 individuals from two
25 families. To our knowledge, the present series of 24 French TBRS individuals is the

1 second largest series described to date. Our study confirmed the main features of the
2 syndrome and provided further molecular and clinical data on this rare overgrowth
3 condition. We confirmed that the phenotypic spectrum is very broad, ranging from
4 nonsyndromic ID for one of our individuals (Patient 19) to very severe and syndromic
5 ID for two individuals (Patients 5 and 15).

6

7 We analyzed the mutational landscape of *DNMT3A*. Seventeen pathogenic or likely
8 pathogenic variants observed in our cohort have never been described before,
9 including an in-frame duplication affecting six highly conserved amino acids in
10 proximity to the PWWP domain for which the DNA methylation profile showed a
11 concordant epismutation [35]. Furthermore, these positions have been shown to be
12 crucial in the DNMT1-DNMT3A interaction [36]. The majority of the variants are
13 missense variants localized in one of the three described functional domains of the
14 DNMT3A protein. Importantly, *DNMT3A* is one of the most common genes
15 associated with clonal hematopoiesis of indeterminate potential (CHIP) [37]. Indeed,
16 some somatic mutations in *DNMT3A* provide a preneoplastic growth advantage that
17 can result in the accumulation, over a period of years, of lymphocytes bearing the
18 somatic variant. Therefore, some pathogenic germline variants are common in the
19 general population due to age-related clonal hematopoiesis, thus limiting the utility of
20 databases such as gnomAD in the pathogenicity stratification of *DNMT3A* germline
21 variants [2] since the gnomAD-derived variants were largely derived from lymphocyte
22 DNA and many gnomAD participants were middle-aged or older. GnomAD missense
23 variants occurring in the three main DNMT3A functional domains are represented by
24 histograms in figure 1.

25

1 As previously shown [19], we found no significant correlation between the type of
2 variant and ID severity.

3

4 There is relatively little pregnancy and neonatal data in the literature. We collected
5 only 10 TBRS individuals with abnormalities during the fetal period, including 4 with
6 macrosomia (one individual in our cohort) and 4 with preeclampsia (absent in our
7 cohort). In comparison, preeclampsia was described in 17% of individuals with Sotos
8 syndrome, another genetic overgrowth syndrome [18]. In conclusion, pregnancy and
9 neonatal data in patients with TBRS vary and are nonspecific.

10

11 The analysis of antenatal ultrasound data, birth parameters and their evolution favors
12 overgrowth with constitutional macrocephaly present from birth or early childhood,
13 with an evolution in the range of +2 SDs consistent with the literature [2, 38].

14 Physical examination revealed facial features already described in the literature,
15 including a round face with coarse features, thick horizontal eyebrows, narrow
16 palpebral fissures and dental abnormalities. As previously described, facial features
17 are more apparent in early adolescence and less specific in early childhood and
18 adulthood [2, 38]. Among dental abnormalities, malocclusion, caries and double teeth
19 have been reported [38], and a recent work highlighted the importance of appropriate
20 dental care for individuals with TBRS [12].

21 We observed hair abnormalities in 11 individuals with hypertrichosis in our cohort.
22 This manifestation has not been previously noted and could aid in the clinical
23 diagnosis of TBRS. The early onset of puberty observed in 4 female individuals in our
24 cohort could also be a new feature of TBRS syndrome.

25

1 In our cohort, psychomotor development was delayed, with regression in 4
2 individuals. One hundred percent of individuals had mild to severe ID, with moderate
3 ID being most common, and 29% of individuals had ASD. These data are consistent
4 with the literature [38, 39]. We noted compulsive eating and/or hyperphagia in 6 of
5 our individuals, as previously reported [2, 5]. This could contribute to the obesity
6 observed in this syndrome.

7 At least one epileptic event occurred in 42% of our individuals; most were isolated
8 febrile seizures, with afebrile events occurring in only 4 individuals. EEG profiles have
9 rarely been described in previous studies. We collected EEG data for 12 TBRS
10 individuals, with focal abnormalities in 3 symptomatic individuals, generalized
11 abnormalities in one symptomatic individual, and focal abnormalities in 1 individual
12 without clinical seizures.

13 Fourteen individuals (58%) had neurological features, with hypotonia, cerebellar
14 syndrome, a decrease in fine motor coordination, tremor and symptomatology of
15 peripheral neuropathy being the most frequent findings. Neurological findings are
16 poorly described in the literature, highlighting the need for systematic examination of
17 TBRS individuals by an experienced neurologist to confirm and validate these
18 findings in further studies. Recently, the description of an individual with congenital
19 myopathy and rhabdomyolysis and another individual with axonal polyneuropathy led
20 to the extension of the TBRS phenotype to include neuromuscular features and
21 polyneuropathy [24, 26].

22 On brain imaging, we found the same various nonspecific abnormalities as previously
23 described in other overgrowth syndromes. However, corpus callosum abnormalities
24 were present in 8 individuals (44%). This finding is consistent with a recent study
25 suggesting that structural neuroimaging abnormalities have been previously

1 underestimated in TBRS patients and revealing the presence of corpus callosum and
2 brain tract alterations, likely caused by microstructural cortical defects and/or primary
3 anomalies in connection development [23]. In contrast to the findings of previous
4 studies, ventriculomegaly or Chiari malformation were rarely observed in our cohort.

5

6 We observed a cardiovascular phenotype in 25% of our individuals. Cardiovascular
7 phenotypes include congenital malformations diagnosed early in life and aortic root
8 dilatation, dilated cardiomyopathy or arrhythmia diagnosed in late childhood or
9 adulthood; periodic surveillance with echocardiography is needed to assess aortic
10 root size [2, 4, 6, 11, 15, 17, 18, 40]. Among respiratory abnormalities, sleep apnea is
11 the most common manifestation, as previously described [2, 5]. Musculoskeletal
12 pathologies are common and include joint hypermobility and scoliosis.

13

14 Although neoplasia was rarely observed in our cohort, several cases of cancer of
15 various types have been described in the literature, including acute myeloid leukemia
16 and other hematologic or solid tumors [10, 14, 41]. As there are no consensus
17 guidelines regarding screening for hematologic malignancy in individuals with TBRS
18 [33, 38, 42], we recommend that hematological malignancies such as easy bruising
19 and petechiae, recurrent bleeding, pallor, persistent fatigue, fever and recurrent
20 infections be assessed at each follow-up visit [2]. Furthermore, TBRS individuals and
21 their families should be educated about the signs and symptoms that would warrant
22 medical evaluation. Complete blood count and further investigations should be
23 performed in the event of signs of hematologic malignancy. In addition, any other
24 change in signs and symptoms in these patients should be investigated further to
25 exclude the development of tumors. Natural history studies of a larger number of

1 TBRS individuals would be needed to enable surveillance guidelines for
2 malignancies particularly hematological malignancies to be established.

3

4 In conclusion, this national collaborative study included the second largest cohort of
5 individuals with TBRS. Our findings are consistent with known clinical features
6 described in the literature, including ID of varying severity, overgrowth and
7 recognizable facial features. It is essential to keep in mind the limits induced by
8 reporting bias, which can lead to probable overestimation of the frequency and
9 severity of neurodevelopmental disorders. However, we have highlighted the lack of
10 specific antenatal signs of Tatton-Brown-Rahman syndrome. Constitutional
11 overgrowth, although not typically detected in the antenatal period, seems to be
12 present at birth or in early childhood. We broadened the phenotype by reporting
13 localized congenital hypertrichosis and early onset of puberty in females. We
14 highlight the neurological phenotype and specific EEG abnormalities. Although
15 malignancies seem to be rare, monitoring blood parameters is essential. We did not
16 identify any phenotype–genotype correlations.

17 This study may contribute to improving the care of TBRS individuals. Furthermore,
18 given the involvement of the *DNMT3A* gene in methylation, current therapeutic
19 advances targeting chromatin marks may benefit TBRS patients in the future.

20

21 **Data availability**

22 All the data underlying the results are available as part of the article.

23

24 **Funding** No financial assistance was received in support of this study

25

1 **Acknowledgments** The authors gratefully thank the patients and their families for
2 their participation in this study and the French National AnDDI-Rares (*Developmental*
3 *Abnormalities with or without Intellectual Deficiency of Rare Causes*) health network.
4 The sponsor was CHRU de Nancy (Direction de la Recherche et de l'Innovation).

5

6 **Competing interests** None declared

7

8 **Ethics approval**

9 This study was approved by the ethics committee of Nancy University Hospital
10 (referral n°429) and was conducted in accordance with the Declaration of Helsinki. All
11 individuals or legal guardians provided written informed consent for genetic analyses.
12 As required by French law, we informed all participants of their right to oppose the
13 use of their data. Individuals or legal guardians have given their consent for the use
14 of their photographs in the publication.

15

16 **Contributorship**

17 CB is the guarantor.

18 LL, MR and CB conceptualized and designed the study.

19 CB collected the samples.

20 LL, MR, ER, JW, SZ, BL, DG, MW, CC, ES, SB, MV, MN, LR, EP, PE, LF, JLA, ML,
21 CQ, SO, CM, AA, SJ, AG, ASM, SN, MD, CM and VCD clinically characterized the
22 individuals.

23 TA, CP, ND, JMR, MW, MB, AB, VR, MM, MD, GV, AP, BG, BC, GL, NC, MT, FT,
24 VD, MF, NC, BK, SR, EB, GB, TH, AO and CB performed the genetic studies and
25 data analysis and interpretation.

- 1 TH, DC, FL, GN and CC generated DNA methylation data from patient 13 and
- 2 interpreted the DNMT3A epesignature.
- 3 HT collected the clinical data.
- 4 TA and CB collected the molecular data.
- 5 HT wrote the first draft of the manuscript.
- 6 TA, ER, JMR, LL and CB revised the manuscript.
- 7 CB supervised the project and edited the manuscript.
- 8 All the authors critically revised and approved the final manuscript.
- 9

1 References

2

- 3 1 Tatton-Brown K, Seal S, Ruark E, Harmer J, Ramsay E, del Vecchio Duarte S,
4 Zachariou A, Hanks S, O'Brien E, Aksglaede L, Baralle D, Dabir T, Gener B,
5 Goudie D, Homfray T, Kumar A, Pilz DT, Selicorni A, Temple IK, Van Maldergem
6 L, Yachelevich N, van Montfort R, Rahman N. Mutations in the DNA
7 methyltransferase gene DNMT3A cause an overgrowth syndrome with intellectual
8 disability. *Nat Genet* 2014;**46**:385–8.
- 9 2 Tatton-Brown K, Zachariou A, Loveday C, Renwick A, Mahamdallie S, Aksglaede
10 L, Baralle D, Barge-Schaapveld D, Blyth M, Bouma M, Breckpot J, Crabb B, Dabir
11 T, Cormier-Daire V, Fauth C, Fisher R, Gener B, Goudie D, Homfray T, Hunter M,
12 Jorgensen A, Kant SG, Kirally-Borri C, Koolen D, Kumar A, Labilloy A, Lees M,
13 Marcelis C, Mercer C, Mignot C, Miller K, Neas K, Newbury-Ecob R, Pilz DT,
14 Posmyk R, Prada C, Ramsey K, Randolph LM, Selicorni A, Shears D, Suri M,
15 Temple IK, Turnpenny P, Van Maldergem L, Varghese V, Veenstra-Knol HE,
16 Yachelevich N, Yates L, Rahman N. The Tatton-Brown-Rahman Syndrome: A
17 clinical study of 55 individuals with de novo constitutive DNMT3A variants.
18 *Wellcome Open Res* 2018;**3**:46.
- 19 3 Okamoto N, Toribe Y, Shimojima K, Yamamoto T. Tatton-Brown-Rahman
20 syndrome due to 2p23 microdeletion. *Am J Med Genet A* 2016;**170**:1339–42.
- 21 4 Hage C, Sabini E, Alsharhan H, Fahrner JA, Beckers A, Daly A, Salvatori R.
22 Acromegaly in the setting of Tatton-Brown-Rahman Syndrome. *Pituitary*
23 2020;**23**:167–70.
- 24 5 Balci TB, Strong A, Kalish JM, Zackai E, Maris JM, Reilly A, Surrey LF, Wertheim
25 GB, Marcadier JL, Graham GE, Carter MT. Tatton-Brown-Rahman syndrome: Six
26 individuals with novel features. *Am J Med Genet A* 2020;**182**:673–80.
- 27 6 Xin B, Cruz Marino T, Szekely J, Leblanc J, Cechner K, Sency V, Wensel C,
28 Barabas M, Therriault V, Wang H. Novel DNMT3A germline mutations are
29 associated with inherited Tatton-Brown-Rahman syndrome. *Clin Genet*
30 2017;**91**:623–8.
- 31 7 Lee CG, Jang J-H, Seo J-Y. First identified Korean family with Tatton-Brown-
32 Rahman Syndrome caused by the novel DNMT3A variant c.118G>C
33 p.(Glu40Gln). *Ann Pediatr Endocrinol Metab* 2019;**24**:253–6.
- 34 8 Tenorio J, Alarcón P, Arias P, Dapía I, García-Miñaur S, Palomares Bralo M,
35 Campistol J, Climent S, Valenzuela I, Ramos S, Monseny AM, Grondona FL, Botet
36 J, Serrano M, Solís M, Santos-Simarro F, Álvarez S, Teixidó-Tura G, Fernández
37 Jaén A, Gordo G, Bardón Rivera MB, Nevado J, Hernández A, Cigudosa JC, Ruiz-
38 Pérez VL, Tizzano EF, Rosa AL, Blanquer A, Alix AG, Santana A, Delicado A,
39 Alonso A, Rodríguez A, Sanchis A, Moreno A, García AP, Vega A, Bredani A, Solari
40 AP, Villavicencio A, Acosta A, Nieto A, González AMC, Baldellon A, Meneses AG,
41 Carrascal AM, de Bustamante AD, Ocasar A, Gener B, González B, Groisman B,
42 Coffee B, Martín CA, Fernández CA, Benito C, Armengod CG, Seisedos CM,
43 Roche C, Arberas C, Perandones C, Pacheco CT, Contessotto C, Olivás C,
44 Armenta D, Cavalcanti D, Elorza D, Castilla E, Zamora E, Zambrano E, Steichen
45 E, Navarro EG, Cruz EC, Gómez EG, Román E, Goldschmidt E, Marfil E, Gean E,
46 Antolín E, Jiménez FJG, Sarries FJM, Ramos F, Grondona FL, Córdoba F, Santos

- 1 F, Vargas F, Martínez F, Uzielli G, Gacio G, Mercado G, Cassinelli H, Orioli I,
2 Arroyo I, López ID, Ramírez IO, Castroviejo IP, Pascual IP, Rio IV, Bueno I,
3 Portero IE, Sánchez IL, del Pozo JS, Campistol J, Arcas J, Fernández J, Planells
4 JG, Pisón JL, Barreiro J, Núñez J del V, Toral JF, Ramírez J, Rosell J, Vilaplana J,
5 de Almeida JCC, Labarta JI, Herranz JL, Luna JLF, Fuster JL, Díaz JM, Gairi JM,
6 Sagredo JMG, Piñero JA, Gutiérrez JCL, Fernández JM, Siguero JPL, Tovar J,
7 Armstrong J, Lara J, Soriano LRL, Cardoso L, Arranz L, De Alba L, Cimbaliene
8 L, Martorell L, Solana LGG, Jurado LP, Ariztegui MAL, Molina MA, García MC,
9 Lozano MF, Merillas MJA, Martínez-Frías ML, Jadraque MR, Pérez MAG, Pedreira
10 MMR, Guardia MNM, Ribate M^aP, López MTG, Pumarega MTM, Segovia M,
11 Lizama M, Lorente MJ, Pombo M, Martínez M, Tabernero M, Ramos MA, Ballesta
12 M, Belar M, Lautre MJ, Cruz M, Artigas M, Villanueva M, Torrabías M, del Campo
13 M, Vila MT, Urioste M, Rosello M, Rahman N, Kantaputra N, Matos PP, Dorao P,
14 Casano P, Mallada PL, Olivares P, Tirado P, Bernardi P, León RC, Cañete R,
15 Gaztañaga R, Velazquez R, Samahuja RV, Delgado RP, Villaverde RS, Gracia R,
16 Scott R, Valdez R, Arteaga R, Cedeño R, Cazorla R, Iglesias RM, Bronberg R,
17 Climent S, Barreiro SC, Kapoor S, Martín SV, Miñaur SG, Kleppe S, Santillán S,
18 Calvo T, Vendrell T, González VL, López V, Lotersztejn V, Méndez V, Albiach V,
19 López VMN, Soler V, Cosentino V, Aoki Y, Lapunzina P. Further delineation of
20 neuropsychiatric findings in Tatton-Brown-Rahman syndrome due to disease-
21 causing variants in DNMT3A: seven new patients. *European Journal of Human*
22 *Genetics* 2020;**28**:469–79.
- 23 9 Lennartsson O, Lodefalk M, Wehtje H, Stattin E-LL, Sävendahl L, Nilsson O. Case
24 Report: Bilateral Epiphysiodesis Due to Extreme Tall Stature in a Girl With a De
25 Novo DNMT3A Variant Associated With Tatton-Brown-Rahman Syndrome. *Front*
26 *Endocrinol (Lausanne)* 2021;**12**:3–8.
- 27 10 Hollink IHIM, van den Ouweland AMW, Beverloo HB, Arentsen-Peters STCJM,
28 Zwaan CM, Wagner A. Acute myeloid leukaemia in a case with Tatton-Brown-
29 Rahman syndrome: the peculiar DNMT3A R882 mutation. *J Med Genet*
30 2017;**54**:805–8.
- 31 11 Kosaki R, Terashima H, Kubota M, Kosaki K. Acute myeloid leukemia-associated
32 DNMT3A p.Arg882His mutation in a patient with Tatton-Brown-Rahman
33 overgrowth syndrome as a constitutional mutation. *Am J Med Genet A*
34 2017;**173**:250–3.
- 35 12 Paz-Alegría M, Gómez-Forero D, Osorio-Patiño J, Jaramillo-Echeverry A.
36 Behavioral and dental management of a patient with Tatton-Brown-Rahman
37 syndrome: Case report. *Special Care in Dentistry* 2020;**40**:597–604.
- 38 13 Tovy A, Rosas C, Gaikwad AS, Medrano G, Zhang L, Reyes JM, Huang Y-H,
39 Arakawa T, Kurtz K, Conneely SE, Guzman AG, Aguilar R, Gao A, Chen C-W, Kim
40 JJ, Carter MT, Lasa-Aranzasti A, Valenzuela I, Van Maldergem L, Brunetti L, Hicks
41 MJ, Marcogliese AN, Goodell MA, Rau RE. Perturbed hematopoiesis in individuals
42 with germline DNMT3A overgrowth Tatton-Brown-Rahman syndrome.
43 *Haematologica* 2021;**107**:0–0.
- 44 14 Sweeney KJ, Mottolise C, Belot A, Szathmari A, Frappaz D, Lesca G, Putoux A, Di
45 Rocco F. The first case report of medulloblastoma associated with Tatton-Brown-
46 Rahman syndrome. *Am J Med Genet A* 2019;**179**:1357–61.
- 47 15 Shen W, Heeley JM, Carlston CM, Acuna-Hidalgo R, Nillesen WM, Dent KM,
48 Douglas G V., Levine KL, Bayrak-Toydemir P, Marcelis CL, Shinawi M, Carey JC.

- 1 The spectrum of DNMT3A variants in Tatton-Brown-Rahman syndrome overlaps
2 with that in hematologic malignancies. *Am J Med Genet A* 2017;**173**:3022–8.
- 3 16 Lemire G, Gauthier J, Soucy JF, Delrue MA. A case of familial transmission of the
4 newly described DNMT3A-Overgrowth Syndrome. *Am J Med Genet A*
5 2017;**173**:1887–90.
- 6 17 Cecchi AC, Haidar A, Marin I, Kwartler CS, Prakash SK, Milewicz DM. Aortic root
7 dilatation and dilated cardiomyopathy in an adult with
8 <scp>Tatton-Brown-Rahman</scp> syndrome. *Am J Med Genet A*
9 2022;**188**:628–34.
- 10 18 Tlemsani C, Luscan A, Leulliot N, Bieth E, Afenjar A, Baujat G, Doco-Fenzy M,
11 Goldenberg A, Lacombe D, Lambert L, Odent S, Pasche J, Sigaudy S, Buffet A,
12 Violle-Poirsier C, Briand-Suleau A, Laurendeau I, Chin M, Saugier-Veber P, Vidaud
13 D, Cormier-Daire V, Vidaud M, Pasmant E, Burglen L. SETD2 and DNMT3A screen
14 in the Sotos-like syndrome French cohort. *J Med Genet* 2016;**53**:743–51.
- 15 19 Yokoi T, Enomoto Y, Naruto T, Kurosawa K, Higurashi N. Tatton-Brown-Rahman
16 syndrome with a novel DNMT3A mutation presented severe intellectual disability
17 and autism spectrum disorder. *Hum Genome Var* 2020;**7**:6–8.
- 18 20 Jiang YH, Yuen RKC, Jin X, Wang M, Chen N, Wu X, Ju J, Mei J, Shi Y, He M,
19 Wang G, Liang J, Wang Z, Cao D, Carter MT, Chrysler C, Drmic IE, Howe JL, Lau
20 L, Marshall CR, Merico D, Nalpathamkalam T, Thiruvahindrapuram B, Thompson
21 A, Uddin M, Walker S, Luo J, Anagnostou E, Zwaigenbaum L, Ring RH, Wang J,
22 Lajonchere C, Wang J, Shih A, Szatmari P, Yang H, Dawson G, Li Y, Scherer SW.
23 Detection of clinically relevant genetic variants in autism spectrum disorder by
24 whole-genome sequencing. *Am J Hum Genet* 2013;**93**:249–63.
- 25 21 Yuen RKC, Merico D, Bookman M, Howe JL, Thiruvahindrapuram B, Patel R V.,
26 Whitney J, Deflaux N, Bingham J, Wang Z, Pellecchia G, Buchanan JA, Walker S,
27 Marshall CR, Uddin M, Zarrei M, Deneault E, D'Abate L, Chan AJS, Koyanagi S,
28 Paton T, Pereira SL, Hoang N, Engchuan W, Higginbotham EJ, Ho K, Lamoureux
29 S, Li W, MacDonald JR, Nalpathamkalam T, Sung WWL, Tsoi FJ, Wei J, Xu L,
30 Tasse AM, Kirby E, Van Etten W, Twigger S, Roberts W, Drmic I, Jilderda S, Modi
31 BM, Kellam B, Szego M, Cytrynbaum C, Weksberg R, Zwaigenbaum L, Woodbury-
32 Smith M, Brian J, Senman L, Iaboni A, Doyle-Thomas K, Thompson A, Chrysler C,
33 Leef J, Savion-Lemieux T, Smith IM, Liu X, Nicolson R, Seifer V, Fedele A, Cook
34 EH, Dager S, Estes A, Gallagher L, Malow BA, Parr JR, Spence SJ, Vorstman J,
35 Frey BJ, Robinson JT, Strug LJ, Fernandez BA, Elsabbagh M, Carter MT,
36 Hallmayer J, Knoppers BM, Anagnostou E, Szatmari P, Ring RH, Glazer D,
37 Pletcher MT, Scherer SW. Whole genome sequencing resource identifies 18 new
38 candidate genes for autism spectrum disorder. *Nat Neurosci* 2017;**20**:602–11.
- 39 22 Kumps C, D'haenens E, Kerkhof J, McConkey H, Alders M, Sadikovic B, Vanakker
40 OM. Methylation signatures in clinically variable syndromic disorders: a familial
41 DNMT3A variant in two adults with Tatton-Brown–Rahman syndrome. *European*
42 *Journal of Human Genetics* Published Online First: 2023. doi:10.1038/s41431-
43 023-01459-w
- 44 23 Jiménez de la Peña M, Rincón-Pérez I, López-Martín S, Albert J, Martín
45 Fernández-Mayoralas D, Fernández-Perrone AL, Jiménez de Domingo A, Tirado P,
46 Calleja-Pérez B, Porta J, Álvarez S, Fernández-Jaén A. Tatton-Brown–Rahman
47 syndrome: Novel pathogenic variants and new neuroimaging findings. *Am J Med*
48 *Genet A* 2023;:1–7.

- 1 24 Ghaoui R, Ha TT, Kerkhof J, McConkey H, Gao S, Babic M, King R, Ravenscroft G,
2 Koszyca B, Otto S, Laing NG, Scott H, Sadikovic B, Kassahn KS. Expanding the
3 phenotype of DNMT3A as a cause a congenital myopathy with rhabdomyolysis.
4 *Neuromuscular Disorders* 2023;**33**:484–9.
- 5 25 Kalil GH, Christopher WO, Brister KA, Orr WS. Treatment of Primary
6 Hyperparathyroidism in the Setting of Tatton-Brown-Rahman Syndrome. *Am*
7 *Surg* 2023;**89**:3203–4.
- 8 26 AlSabah A-A, Alsalmi M, Massie R, Bilodeau M-C, Campeau PM, McGraw S,
9 D’Agostino MD. An adult patient with Tatton-Brown-Rahman syndrome caused by
10 a novel DNMT3A variant and axonal polyneuropathy. *Am J Med Genet A*
11 Published Online First: 1 December 2023. doi:10.1002/ajmg.a.63484
- 12 27 Lyko F. The DNA methyltransferase family: a versatile toolkit for epigenetic
13 regulation. *Nat Rev Genet* 2018;**19**:81–92.
- 14 28 Rinaldi L, Datta D, Serrat J, Morey L, Solanas G, Avgustinova A, Blanco E, Pons
15 JI, Matallanas D, Von Kriegsheim A, Di Croce L, Benitah SA. Dnmt3a and Dnmt3b
16 Associate with Enhancers to Regulate Human Epidermal Stem Cell Homeostasis.
17 *Cell Stem Cell* 2016;**19**:491–501.
- 18 29 Kaneda M, Okano M, Hata K, Sado T, Tsujimoto N, Li E, Sasaki H. Essential role
19 for de novo DNA methyltransferase Dnmt3a in paternal and maternal imprinting.
20 *Nature* 2004;**429**:900–3.
- 21 30 Fukuda A, Hazelbaker DZ, Motosugi N, Hao J, Limone F, Beccard A, Mazzucato P,
22 Messana A, Okada C, San Juan IG, Qian M, Umezawa A, Akutsu H, Barrett LE,
23 Eggan K. De novo DNA methyltransferases DNMT3A and DNMT3B are essential
24 for XIST silencing for erosion of dosage compensation in pluripotent stem cells.
25 *Stem Cell Reports* 2021;**16**:2138–48.
- 26 31 Russler-Germain DA, Spencer DH, Young MA, Lamprecht TL, Miller CA, Fulton R,
27 Meyer MR, Erdmann-Gilmore P, Townsend RR, Wilson RK, Ley TJ. The R882H
28 DNMT3A Mutation Associated with AML Dominantly Inhibits Wild-Type DNMT3A
29 by Blocking Its Ability to Form Active Tetramers. *Cancer Cell* 2014;**25**:442–54.
- 30 32 Kim SJ, Zhao H, Hardikar S, Singh AK, Goodell MA, Chen T. A DNMT3A mutation
31 common in AML exhibits dominant-negative effects in murine ES cells. *Blood*
32 2013;**122**:4086–9.
- 33 33 Ferris MA, Smith AM, Heath SE, Duncavage EJ, Oberley M, Freyer D, Wynn R,
34 Douzgou S, Maris JM, Reilly AF, Wu MD, Choo F, Fiets RB, Koene S, Spencer DH,
35 Miller CA, Shinawi M, Ley TJ. DNMT3A overgrowth syndrome is associated with
36 the development of hematopoietic malignancies in children and young adults.
37 *Blood* 2022;**139**:461–4.
- 38 34 Richards S, Aziz N, Bale S, Bick D, Das S. ACMG Standards and Guidelines
39 Standards and guidelines for the interpretation of sequence variants : a joint
40 consensus recommendation of the American College of Medical Genetics and
41 Genomics and the Association for Molecular Pathology. 2015;**17**:405–24.
- 42 35 Husson T, Lecoquierre F, Nicolas G, Richard AC, Afenjar A, Audebert-Bellanger S,
43 Badens C, Bilan F, Bizaoui V, Boland A, Bonnet-Dupeyron MN, Brischoux-Boucher
44 E, Bonnet C, Bournez M, Boute O, Brunelle P, Caumes R, Charles P, Chassaing N,
45 Chatron N, Cogné B, Colin E, Cormier-Daire V, Dard R, Dauriat B, Delanne J,
46 Deleuze JF, Demurger F, Denommé-Pichon AS, Depienne C, Dieux A, Dubourg C,
47 Edery P, El Chehadeh S, Faivre L, Fergelot P, Fradin M, Garde A, Geneviève D,
48 Gilbert-Dussardier B, Goizet C, Goldenberg A, Gouy E, Guerrot AM, Guimier A,

- 1 Harzalla I, Héron D, Isidor B, Lacombe D, Le Guillou Horn X, Keren B, Kuechler
2 A, Lacaze E, Lavillaureix A, Lehalle D, Lesca G, Lespinasse J, Levy J, Lyonnet S,
3 Morel G, Jean-Marçais N, Marlin S, Marsili L, Mignot C, Nambot S, Nizon M, Olaso
4 R, Pasquier L, Perrin L, Petit F, Pingault V, Piton A, Prieur F, Putoux A, Planes M,
5 Odent S, Quélin C, Quemener-Redon S, Rama M, Rio M, Rossi M, Schaefer E,
6 Rondeau S, Saugier-Weber P, Smol T, Sigaudy S, Touraine R, Mau-Them FT,
7 Trimouille A, Van Gils J, Vanlerberghe C, Vantalón V, Vera G, Vincent M, Ziegler
8 A, Guillin O, Champion D, Charbonnier C. Episignatures in practice: independent
9 evaluation of published episignatures for the molecular diagnostics of ten
10 neurodevelopmental disorders. *European Journal of Human Genetics* Published
11 Online First: 2023. doi:10.1038/s41431-023-01474-x
- 12 36 Kim G-D, Ni J, Kelesoglu N, Roberts RJ, Pradhan S. Co-operation and
13 communication between the human maintenance and de novo DNA (cytosine-5)
14 methyltransferases. *EMBO J* 2002;**21**:4183–95.
- 15 37 Kessler MD, Damask A, O’Keeffe S, Banerjee N, Li D, Watanabe K, Marketta A,
16 Van Meter M, Semrau S, Horowitz J, Tang J, Kosmicki JA, Rajagopal VM, Zou Y,
17 Houvras Y, Ghosh A, Gillies C, Mbatchou J, White RR, Verweij N, Bovijn J,
18 Parikshak NN, LeBlanc MG, Jones M, Regeneron Genetics Center, GHS-RGC
19 DiscovEHR Collaboration, Glass DJ, Lotta LA, Cantor MN, Atwal GS, Locke AE,
20 Ferreira MAR, Deering R, Paulding C, Shuldiner AR, Thurston G, Ferrando AA,
21 Salerno W, Reid JG, Overton JD, Marchini J, Kang HM, Baras A, Abecasis GR,
22 Jorgenson E. Common and rare variant associations with clonal haematopoiesis
23 phenotypes. *Nature* 2022;**612**:301–9.
- 24 38 Ostrowski PJ, Tatton-Brown K. *Tatton-Brown-Rahman Syndrome*. 1993.
25 <http://www.ncbi.nlm.nih.gov/pubmed/35771960>
- 26 39 Lane C, Tatton-Brown K, Freeth M. Tatton-Brown-Rahman syndrome: cognitive
27 and behavioural phenotypes. *Dev Med Child Neurol* 2020;**62**:993–8.
- 28 40 Tenorio J, Alarcón P, Arias P, Dapía I, García-Miñaur S, Palomares Bralo M,
29 Campistol J, Climent S, Valenzuela I, Ramos S, Monseny AM, Grondona FL, Botet
30 J, Serrano M, Solís M, Santos-Simarro F, Álvarez S, Teixidó-Tura G, Fernández
31 Jaén A, Gordo G, Bardón Rivera MB, Nevado J, Hernández A, Cigudosa JC, Ruiz-
32 Pérez VL, Tizzano EF, Lapunzina P. Further delineation of neuropsychiatric
33 findings in Tatton-Brown-Rahman syndrome due to disease-causing variants in
34 DNMT3A: seven new patients. *European Journal of Human Genetics*
35 2020;**28**:469–79.
- 36 41 Mellid S, Coloma J, Calsina B, Monteagudo M, Roldán-Romero JM, Santos M,
37 Leandro-García LJ, Lanillos J, Martínez-Montes AM, Rodríguez-Antona C,
38 Montero-Conde C, Martínez-López J, Ayala R, Matias-Guiu X, Robledo M, Cascón
39 A. Novel DNMT3A Germline Variant in a Patient with Multiple Paragangliomas and
40 Papillary Thyroid Carcinoma. *Cancers (Basel)* 2020;**12**.
41 doi:10.3390/cancers12113304
- 42 42 Tovy A, Rosas C, Gaikwad AS, Medrano G, Zhang L, Reyes JM, Huang Y-H,
43 Arakawa T, Kurtz K, Conneely SE, Guzman AG, Aguilar R, Gao A, Chen C-W, Kim
44 JJ, Carter MT, Lasa-Aranzasti A, Valenzuela I, Van Maldergem L, Brunetti L, Hicks
45 MJ, Marcogliese AN, Goodell MA, Rau RE. Perturbed hematopoiesis in individuals
46 with germline DNMT3A overgrowth Tatton-Brown-Rahman syndrome.
47 *Haematologica* 2022;**107**:887–98.
48

1 **Figure Legends**

2

3 **Figure 1. Molecular data and genomic context** (A) The DNMT3A protein structure
4 and details of the 24 disease-causing variants described in this work. The three
5 DNMT3A domains are shaded in blue: the proline-tryptophan-tryptophan-proline
6 (PWWP) domain, the ATRX-DNMT3-DNMT3L (ADD) domain and the
7 methyltransferase (MTase) domain. The missense variants and the *in-frame*
8 duplications are represented in the upper panel. Truncating and splice variants are
9 represented in the lower panel. Orange line: deletion of exons 7-23. GnomAD
10 missense variants (except variants annotated in ClinVar as pathogenic, likely
11 pathogenic or of uncertain significance; n=309) are displayed in blue as controls from
12 the general population. (B) The blue histogram indicates GnomAD missense variants
13 from the general population occurring in the three main DNMT3A functional domains
14 (except for variants annotated in ClinVar as pathogenic, likely pathogenic or of
15 uncertain significance), and the red stars indicate the positions of the disease-
16 causing missense variants described in this work. None of the variants reported in
17 this work are present without annotation in GnomAD. Missense variants affecting the
18 same protein position result in a change in different amino acids.

19

20 **Figure 2. Morphological features of Tatton-Brown-Rahman syndrome.** (A) Teeth
21 abnormalities. (B) Hands and feet. (C) Hypertrichosis in individuals 1 and 3. (D)
22 Pectus excavatum in individual 2. Hyperlordosis and elbow hyperextension in
23 individual 3. (E) Brain MR image of individual 11: sagittal T1-weighted image:
24 complete but thin corpus callosum. Normal posterior fossa. There was no cerebellar
25 tonsillar herniation.

1 **Table 1.** Clinical features of the 24 patients in the French cohort by frequency compared to those of previously published patients
 2 [38]
 3

Features	French cohort (n = 24) (%)	Ostrowski, P. J., & Tatton-Brown, K. in GeneReviews® [38] (n = 90) (%)
Intellectual disability (ID)	100%	100%
Mild ID	29%	28%
Moderate ID	57%	58%
Severe ID	14%	14%
Distinctive facial features	96%	ND
Overgrowth*	87%	>80%
Tall stature (height ≥ 2 SD above mean)	65%	$\sim 70\%$
Macrocephaly (head circumference ≥ 2 SD above mean)	68%	$\sim 50\%$
Overweight (BMI)	46%	ND
Weight ≥ 2 SD	76%	$\sim 65\%$
Joint hypermobility	46%	$\sim 75\%$
Hypotonia	25%	$\sim 55\%$
Behavioral/psychiatric issues	63%	$\sim 50\%$
Autistic traits	29%	44%
Kyphoscoliosis	42%	$\sim 30\%$
Seizures	42%	$\sim 20\%$
Cryptorchidism	30% of males	$\sim 20\%$ of males
Cardiovascular disease	25%	$\sim 10\%$
Corpus callosum abnormalities	44%	ND
Ventriculomegaly	4%	<10%
Chiari malformation	0%	<10%
Malignant tumors	4%	$\sim 5\%$

1
2 * defined as height and/or head circumference at least 2 standard deviations above the population mean
3
4