

Phenotype and Genotype in Nicolaides–Baraitser Syndrome

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Nicolaides–Baraitser syndrome (NCBRS) is an intellectual disability (ID)/multiple congenital anomalies syndrome caused by non-truncating mutations in the ATPase region of SMARCA2, which codes for one of the two alternative catalytic subunits of the BAF chromatin remodeling complex. We analyzed 61 molecularly confirmed cases, including all previously reported patients ($n = 47$) and 14 additional unpublished individuals. NCBRS is clinically and genetically homogeneous. The cardinal features (ID, short stature, microcephaly, typical face, sparse hair, brachydactyly, prominent interphalangeal joints, behavioral problems and seizures), are almost universally present. There is variability however, as ID can range from severe to mild, and sparse hair may be present only in certain age groups. There may be a correlation between the severity of the ID and presence of seizures, absent speech, short stature and microcephaly. SMARCA2 mutations causing NCBRS are likely to act through a dominant-negative effect. There may be some genotype–phenotype correlations (mutations at domain VI with severe ID and seizures; mutations affecting residues Pro883, Leu946, and Ala1201 with mild phenotypes) but numbers are still too small to draw definitive conclusions. © 2014 Wiley Periodicals, Inc.

KEY WORDS: Nicolaides–Baraitser syndrome; natural history; intellectual disability; phenotype; genotype; SMARCA2; BAF (SWI/SNF) complex

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INTRODUCTION

Nicolaides–Baraitser syndrome (NCBRS, OMIM #601358) is characterized by severe intellectual disability (ID), seizures, short stature, sparse hair, typical face, brachydactyly, and prominent interphalangeal joints. It was first described in 1993 by pediatric neurologist Paola Nicolaides and clinical geneticist Michael Baraitser, when they reported a 16-year-old British girl with this unusual combination of features

[Nicolaides and Baraitser, 1993]. In the first 15 years thereafter five further cases were reported [Krajewska–Walasek et al., 1996; Morin et al., 2003; Witters and Fryns, 2003; Castori et al., 2008]. In 2009, the follow-up of these patients and description of 18 additional patients allowed the establishment of NCBRS as a discrete syndrome [Sousa et al., 2009]. Follow-up of the original patient at 32 years of age showed her to have had seizures refractory to treatment with multiple drugs, a gradual decline of her

motor and mental abilities, and more marked physical features: of note her hair

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became more sparse, her facial features coarsened and the joint anomalies became prominent. She died at 33 years of age as a consequence of *status epilepticus* and subsequent respiratory complications. It became clear by the evolution of this and several other patients, that certain features of NCBRS can be progressive. The patients and families that participated in that publication [Sousa et al., 2009] started an international NCBRS parents support group, which has met yearly since 2010.

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All the known NCBRS affected individuals were sporadic and unrelated cases, from various ancestry groups, with no difference in severity or prevalence

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between the sexes. Neither familial recurrence (except for one pair of monozygotic twins) nor consanguinity has been reported. These characteristics, added to the specificity of the phenotype, led to the hypothesis that NCBRS could be caused by a small microdeletion and/or by *de novo* heterozygous dominant mutations in a single gene. We performed classical cytogenetic studies and array-based comparative genomic hybridization in 12 NCBRS patients and were unable to detect pathogenic chromosomal abnormality. The phenotypic overlap with Coffin–Siris syndrome (CSS, OMIM #135900) was recognized [Coffin and Siris, 1970; Schrier Vergano et al., 2013] and it was suggested that either the entities were allelic or caused by genes with related functions [Sousa et al., 2009].

Following our above-mentioned work, we set out to identify the genetic cause of NCBRS using whole-exome sequencing and filtering variants according with the predicted mode of inheritance. In a joint project between Amsterdam and Leuven, *SMARCA2*, coding for one of the components of the “BRG1/*SMARCA4* and hBRM/*SMARCA2*-associated factor” (BAF) complex, was identified in 2012 as the

causative gene of NCBRS [Van Houdt et al., 2012]. At the time, 44 patients were studied, 22 of the 26 previously reported patients [Nicolaidis and Baraitser, 1993; Krajewska-Walasek et al., 1996; Morin et al., 2003; Witters and Fryns, 2003; Castori et al., 2008; Sousa et al., 2009; Gana et al., 2011], and 22 additional cases. We classified these patients in two categories of 37 and seven patients, respectively, with high (group 1) and low (group 2) certainty by which the diagnosis could be retained. Heterozygous mutations in the ATPase region of *SMARCA2* were identified in 34 out of 37 patients of group 1 and on two of the seven individuals of group 2.

In the present review we focused on mutation positive patients only and collected information on a total of 61 cases (Table I), aiming to better characterize the NCBRS clinical spectrum and evaluate possible genotype–phenotype correlations.

METHODS

We contacted the physicians of all patients known to us with NCBRS in whom a *SMARCA2* mutation had been found, and invited them to collaborate by filling out a detailed questionnaire. The format of this questionnaire was developed to retrieve data, not only on NCBRS, but also on the related entities CSS, DOOR(S) syndrome and Van der Aa–Kooy syndrome (caused by *ADNP* mutations). We supplemented the data with information previously gathered by us, and available clinical pictures for all affected individuals. The genotype of the study participants was obtained either through publications [Van Houdt et al., 2012; Wolff et al., 2012; Kosho et al., 2013; Santen et al., 2013; Wiczorek et al., 2013] or collected from the molecular genetics Diagnostic Laboratories from the Academic Medical Center in Amsterdam, the Centre for Human Genetics in Leuven and the Great Ormond Street Hospital in London, using standard Sanger sequencing.

PATIENTS

In total, 61 cases have been analyzed, including all previously reported

SMARCA2 mutation-positive patients ($n = 47$) [Van Houdt et al., 2012; Wolff et al., 2012; Kosho et al., 2013; Santen et al., 2013; Wiczorek et al., 2013] and 14 additional unpublished individuals. Patients originate from the five continents and several ethnic backgrounds, although the majority (2/3) are from European countries. The male to female ratio is 35:26. The median age at the last clinical evaluation is 10 years (range 2–33 years). Mean paternal age at birth is 32.9 years and mean maternal age is 30.1 years. Due to the mixed origin of families we are unsure whether this falls within normal limits or not.

One hitherto unreported patient is discussed separately as the *SMARCA2* mutation was identified by whole-exome sequencing in a patient not suspected to have NCBRS, and is located outside the ATPase domain (see below).

PHENOTYPE

Growth

One-third of NCBRS patients (19/57) are small for gestational age. Length at birth is < -2 SD in 21% (8/38) but $> +2$ SD in 13% (5/38) of the newborns. Postnatally, short stature is common, present in over half (30/56) of the patients. Almost all have a height below the 50th centile. Adult height in adult males varies between 1.44 and 1.80 m ($n = 5$, mean 1.58 m), and in adult females between 1.30 and 1.69 m ($n = 6$, mean 1.57 m). Body proportions are normal. Microcephaly at birth is present in 23% (7/30), and later on in 65% (34/52). Mean adult head size is 52.8 cm in males ($n = 4$, range 50–56 cm) and 53.2 cm ($n = 6$, range 50.5–54 cm) in females. Body weight is < 50 th centile in three-quarter (36/46) of patients and < -2 SD in half of them (24/46). No patient is overweight.

Face

The facial characteristics are typically not easily recognized in younger patients (Fig. 1a,b). Several patients have initially been suspected to have Williams

TABLE I. Major Signs and Symptoms of 61 Individuals with Nicolaides–Baraitser Syndrome

	Age groups (yrs)				All patients (n = 61)	
	0–6.9 (n = 14)	7–11.9 (n = 23)	12–16.9 (n = 14)	≥17 (n = 10)		%
Growth at birth						
Length (cm): mean vs. median (range) ^a					48.7 vs. 49 (42–54)	
Length at birth <–2 SD					8/38	21.1
Length at birth >+2 SD					5/38	13.2
Weight (kg): mean vs median (range) ^a					2732 vs. 2700 (1774–3850)	
Weight <–2 SD					19/57	33.3
Head circumference (cm): mean vs. median (range) ^a					33.1 vs. 33.0 (30–34.5)	
Head circumference <–2 SD					7/30	23.3
Growth post-natal						
Weight <–2 SD	7/12	5/16	9/11	3/7	24/46	52.2
Stature <–2 SD	9/14	9/19	9/14	3/9	30/56	53.6
Head circumference <–2 SD	8/12	12/19	9/13	5/8	34/52	65.4
Neurodevelopment						
ID – Mild vs. Moderate vs. Severe ^b	2 vs. 6 vs. 6/14	5 vs. 11 vs. 7/23	1 vs. 4 vs. 9/14	3 vs. 1 vs. 6/10	11 vs. 22 vs. 28/61	18.0 vs. 36.1 vs. 45.9
Sitting age (months) — mean vs. median (range)					9 vs. 8 (6–20)	
Walking independently age (months) mean vs. median (range)					21 vs. 18 (10–60)	
Hypotonia					19/51	37.3
Absent speech					19/60	31.7
First words age (months) mean vs. median (range)					29.5 vs. 24 (10–96)	
Speech decline at a later age					9/42	21.4
Seizures						
Age of first seizure (months) — mean vs. median (range)	8/14	16/23	10/14	5/10	39/61	63.9
Craniofacial features						
Coarse face	9/11	14/22	13/13	7/10	43/56	76.6
Progressive coarse features	6/7	4/10	4/8	4/6	18/31	58.0
Low anterior hairline	7/11	15/21	10/14	7/10	39/56	69.6
Sparse hair	14/14	22/23	13/14	10/10	59/61	96.7
Narrow forehead	1/11	7/21	8/11	2/10	18/53	40.0
Prominent eyelashes	12/14	19/23	8/14	5/10	44/61	86.2
Ptosis	1/13	5/23	4/12	2/10	12/58	20.7
Synophrys	2/11	4/22	4/12	2/10	12/55	21.8
Thick eyebrows	10/14	14/23	10/12	6/10	40/59	67.8
Increased skin wrinkling	7/14	14/23	8/14	4/10	33/61	54.1
Sagging periorbital skin	9/11	16/22	8/14	5/10	38/57	66.7
Narrow palpebral fissures	0/11	5/22	2/14	2/10	9/57	15.8
Upward vs. Downward slant palpebral fissures	1 vs. 4/11	0 vs. 15/22	1 vs. 10/13	0 vs. 2/10	2 vs. 31/56	3.5 vs. 55.4
Wide vs. Narrow nasal bridge	7 vs. 1/11	6 vs. 9/22	1 vs. 10/12	4 vs. 4/10	18 vs. 24/55	32.7 vs. 43.6
Upturned nasal tip	11/13	15/23	9/14	5/10	40/60	66.7
Short nose	7/11	4/22	4/11	5/8	20/52	38.5
Broad nasal tip	9/11	13/22	9/13	5/10	36/56	64.3
Thick alae nasi	11/13	17/23	11/13	8/10	47/59	79.7
Broad nasal base	8/11	14/22	11/14	8/10	41/57	71.9
Choanal stenosis	0/13	0/22	0/14	1/10	1/59	1.7
Broad philtrum	11/14	12/22	12/14	9/10	44/60	73.3
Long vs. Short philtrum	7 vs. 3/13	17 vs. 0/23	8 vs. 3/13	4 vs. 2/10	36 vs. 8/59	61.0 vs. 13.6
Large mouth	11/13	18/23	10/14	8/10	47/60	78.3
Thin upper vermillion	11/14	20/22	10/14	6/10	47/60	78.3
Thick lower vermillion	12/13	19/23	11/14	8/10	50/60	83.3
Drooping lower lip	5/11	11/21	6/13	8/10	30/55	54.5
Cleft palate	0/14	0/23	1/14	0/10	1/61	1.6
Widely spaced teeth	8/10	11/22	8/14	5/9	32/53	58.2
Abnormal enamel	0/8	1/16	1/10	1/8	3/42	7.1
Hypo/oligodontia	2/8	2/16	3/11	1/8	8/43	18.6
Gum hypertrophy	2/8	0/21	0/10	9/10	2/48	4.0
Malformed ears	4/14	9/22	3/13	1/10	17/59	28.8
Ear tags	1/14	1/22	0/13	0/9	2/58	3.4
Trunk and limbs						
Broad neck	3/11	1/21	1/12	5/10	10/54	18.5
Scoliosis	0/14	5/22	8/14	4/10	17/60	28.3
Widely spaced nipples	1/8	0/18	2/11	1/9	4/45	8.8
Pectus excavatum	0/11	0/19	1/8	2/9	3/47	6.4
Cryptorchidism	3/8	9/13	6/9	2/4	20/34	58.8
Umbilical/inguinal hernia	6/11	11/22	7/14	2/10	26/57	45.6
Small 5th finger	3/14	4/21	0/13	0/9	7/57	12.3
Prominent interphalangeal joints	10/13	18/22	14/14	8/10	50/59	84.7
Prominent distal phalanges	9/14	17/21	9/14	5/10	40/59	67.8
Fetal finger pads	2/11	11/20	6/13	3/9	22/53	41.5
Dislocated hips	0/10	2/16	1/12	1/7	4/45	8.9
Small patellae	0/11	0/15	0/12	1/6	1/44	2.3
Joint laxity	5/14	6/12	1/18	4/8	16/52	30.8
Sandal gap	5/10	10/19	7/14	4/8	26/51	50.1

TABLE I. (Continued)

	Age groups (yrs)				All patients (n = 61)	
	0–6.9 (n = 14)	7–11.9 (n = 23)	12–16.9 (n = 14)	≥17 (n = 10)		%
Nail anomalies						
Small nails 5th finger/toe only					0/34	0
Small nails thumbs/halluces					2/33	6.1
Small nails all fingers/toes					6/34	17.6
Radiology						
Bone age (Delayed vs. Advanced)					16 vs. 2/39	41.0 vs. 5.1
Short Metacarpals vs. Metatarsals					16 vs. 4/41	39.0 vs. 9.8
Short phalanges					9/43	20.9
Cone shaped epiphyses					9/34	26.5
Other features						
Behavioral problems					19/?	
Hypertrichosis (not scalp)					22/50	44.0
Eczema					22/58	37.9
Feeding problems					23/49	46.9
Frequent infections					13/48	27.1
Cardiac defect (see text)					6/61	9.8
Hearing loss					4/59	6.8
Myopia					10/?	
Astigmatism					4/?	
Hypospadias					1/36	2.8
Malignancy					0/61	0

^aFor these calculations, only babies born between 38–42 weeks were considered: n = 47 for weight; n = 30 for length; and n = 28 for head circumference.

^b+ mild (IQ50–69); ++ moderate (IQ35–49); +++ severe (IQ < 35); IQ levels are estimates as formal testing has often not been performed.

syndrome. The face in NCBRS is characterized by a triangular shape, dense and prominent eyelashes, broad nasal base, thick nares, upturned nasal tip, rounded premaxilla, broad philtrum, thin upper vermilion, thick and everted lower vermilion, and wide mouth. The palpebral fissures are sometimes narrow and/or downslanting. The broad philtrum is often associated with a protrusion of the central region of the upper lip, evoking a cupid bow–shape of the upper vermilion, especially in younger children. With increasing age the amount of subcutaneous fat tissue tends to decrease, making the skin below the orbits sagging and wrinkled, especially at the cheeks when smiling. However, some individuals retain full cheeks. Facial characteristics are typically more pronounced with age (Fig. 1b). The facial features are coarse in three-quarter (43/56) and progressive coarsening is noted in 58% (18/31) of cases. In some adults the lower third of the face becomes markedly broad, especially at the angle of the mandible, and may involve the neck. A single patient had congenital choanal atresia, and one other had a cleft palate.

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Hair

Sparse scalp hair is a major sign of NCBRS that often gradually becomes more pronounced with age and is present in almost all (59/61) individuals (Fig. 1b). At birth, there may be facial hypertrichosis and a low anterior hairline. The sparseness of scalp hair can be present in the first months of life, but can also start to become evident in the second half of the first decade and in the

second decade. The growth and texture of hair is usually normal, some patients being noted to have increased thickness. Microscopic evaluation does not show significant abnormalities. In some NCBRS individuals the sparseness of the scalp hair improves with time. The eyebrows are normal or even dense at first but usually follow the same reduction in density over time. Eyelashes remain prominent. Pubic hair develops normally but facial hair is very limited in adult males.

Skin and Teeth

The skin is wrinkled in half of the cases (33/61), being more noticeable at the distal limbs. Subcutaneous veins are very visible, likely also due to poor development of subcutaneous fat tissue. A total of 44% of the patients have hypertrichosis mainly on their neck and back. Eczema is present in one third of the patients, involves mainly the distal limbs and face, is severe in some patients, and almost invariably decreases in the second half of the first decade. The skin is often sensitive and somewhat pale. Six patients have generally small nails. No individual

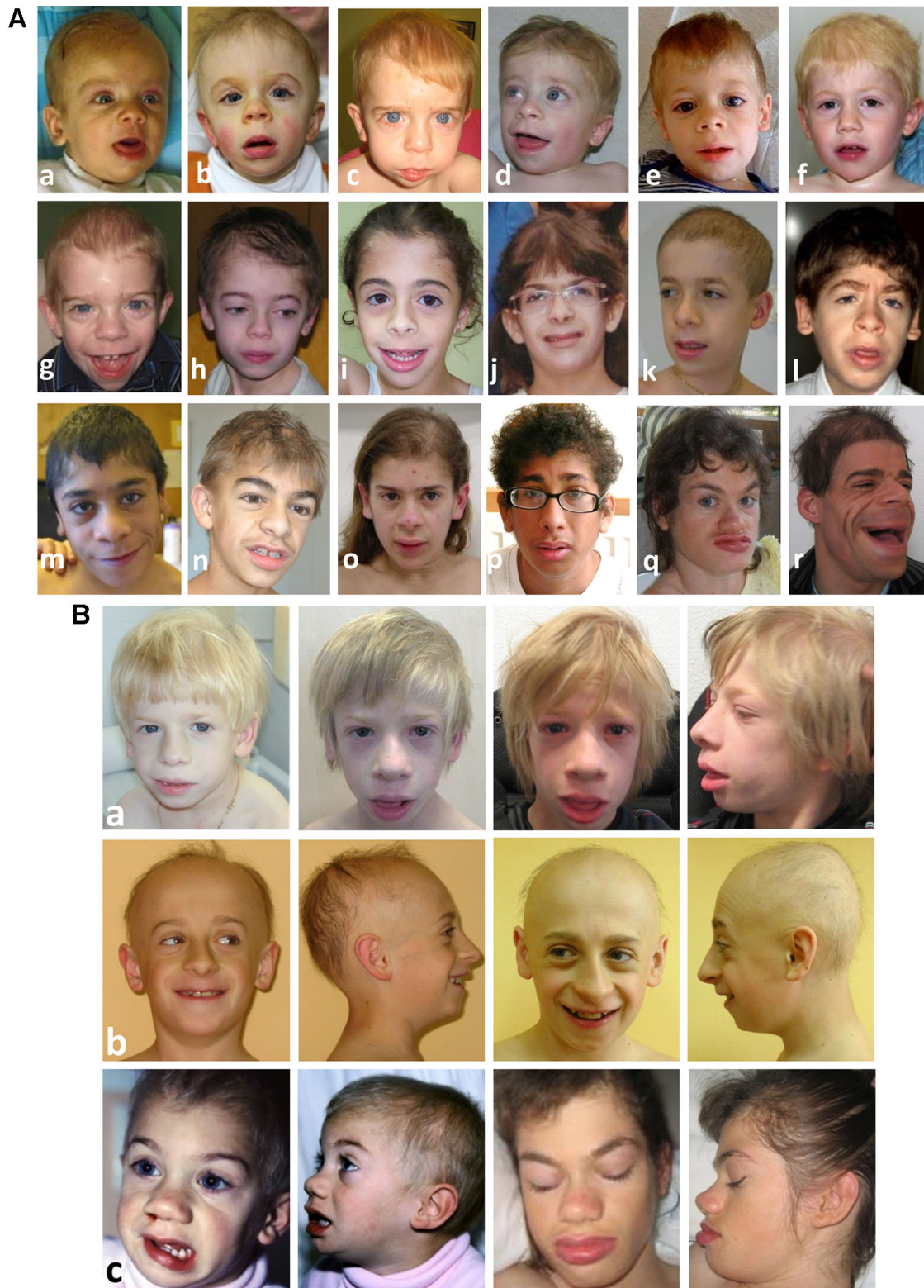


Figure 1. Clinical pictures of some of the present individuals with Nicolaides–Baraitser syndrome. (A) Faces at various ages: a. male, 8 months; b. male, 12 months; c. male, 14 months; d. male, 21 months; e. male, 2 years; f. male, 3 years; g. male, 3 years (same person as in c); h. female, 6 years; i. female, 9 years; j. female, 10 years; k. male, 10 years; l. male, 11 years; m. male, 12 years; n. male, 13 years; o. female, 13 years; p. female, 16 years; q. female, 19 years (same person also depicted in B row c); r. male, 26 years. (B) Changes in facial characteristics in three individuals with Nicolaides–Baraitser syndrome. a: male, left 5 years and 9 years, right 11 years. b: male, left 9 years, right 15 years. c: female, left 2 years, right 14 years (same person also depicted in A, patient q).

has small nails of only the fifth fingers. Unusual skin manifestations are hypohidrosis, spots of hypopigmentation, a single café-au-lait spot, and patchy skin hyperpigmentation, each present in a single individual. One patient has an extra nipple.

Teeth are widely spaced in 58.2% of cases, and hypodontia is reported in 18.6%. Dental eruption is delayed in one-third of cases, which may involve primary dentition, but more frequently involved secondary dentition. Surgical interventions to allow eruption have not been uncommon.

Limbs

At birth, hands and feet are usually unremarkable. With time, distal phalan-

ges broaden in all directions in three-quarters of the cases. Almost half of the patients have increased fetal pads. Prominent inter-phalangeal joints are the most characteristic sign, present in 84.7% (Fig. 2). At first, mobility is normal and in several NCBRS children fingers are described as hypermobile. But later on mobility often decreases, and some older individuals dislike passive movements of their fingers. Prominence of the other large joints has been reported in a few cases. True arthritis has not been found in any patient. Sandal gaps are present in 50% of the patients, and although thickening of the distal toes occurs, it is less pronounced compared to the fingers (Fig. 3). Cone-shaped epiphyses, at first thought to be a major characteristic

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[Nicolaidis and Baraitser, 1993] is found in nine individuals. A few patients have ivory epiphyses. Other radiological findings of the hands include short metacarpals and/or short phalanges, which



Figure 2. Variability of some of the hands of the present series of Nicolaidis–Baraitser syndrome. Note the thickening of the distal phalanges in all directions, prominence of interphalangeal joints, and short metacarpals.



Figure 3. Variability of some of the feet of the present series of Nicolaidis–Baraitser syndrome. Note the variable but usually mild shortening of the toes, sandal gaps, and some thickening of the distal phalanx of mainly the hallux.

may be present in all fingers but especially in the 4th and 5th rays. Only a few patients have small 5th fingers or toes but no individual has small or absent distal phalanges and none had absent nails of the fifth fingers or toes. Bone age can vary remarkably, from normal (53.9%), delayed (41%) or advanced (5.1%). Osteoporosis is not uncommon and indeed several patients develop fractures in puberty or thereafter. Congenital hip dislocation is present occasionally, a single patient has patella dislocations, and a few patients have generalized joint laxity. Scoliosis is present in 28.3%.

Other Physical Features

Hearing loss is found in four patients, three with conductive and one with congenital sensorineural deafness. Myopia was diagnosed in 10 patients and astigmatism in four. Easy choking occurs very frequently. Indeed, feeding problems are common (46.9%) but usually mild and not requiring nasogastric tube. Many like their food to be completely blended, a preference which continues into adulthood. Cryptorchidism is present in 58.8% of the males. One boy has hypospadias. Inguinal and/or umbilical hernias were present in 45.6% of patients, and one patient

was born with an omphalocele. Three patients have a pectus excavatum. Six patients had cardiac anomalies (double aortic arch; small ostium secundum atrial septal defect and mild stenosis of the left pulmonary artery [spontaneous resolution]; mild pulmonary stenosis and mild left ventricular hypertrophy [spontaneous resolution]; small ostium secundum atrial septal defect and persistent ductus arteriosus; mild aortic coarctation and persistent ductus arteriosus; truncus brachiocephalica crossing the midline and compressing the trachea). Menarche occurred at a median age of 14.8 years (range: 11–19 years) and menses were normal except for a single female who had oligomenorrhea. At least half of the adult females have poor mammary development. Recurrent infections do occur, mainly urinary tract infections, but are not frequent. Vesico-ureteral reflux, IgA nephritis, marked constipation, villous atrophy, mild dyslipidemia, abnormal carnitine profile, spastic paraplegia, are each present in a single individual. No patient is known to have developed a malignancy.

Neurodevelopment and Behavior

Impaired cognition is ubiquitous in NCBRS. For the majority of patients,

no formal assessment was performed but in most the delay was considered to be severe (45.9%). The delay can also be moderate (36.1%) or mild (18%). Language is particularly limited, and indeed at least 30% of patients never develop speech. In nine patients, their initial words were lost or significantly reduced later in life. In some patients the loss of speech co-occurred with their first seizure. Hypotonia is reported in one third of patients, but major motor milestones such as sitting (mean 9 months) and walking independently (mean 21 months) are usually not very delayed. Brain imaging was performed in at least 42 patients and usually yielded normal results. Three patients had large ventricles and two had a small corpus callosum. Typically patients are happy and very friendly, but may have temper tantrums and periods of aggression as well. Often their behavior also shows symptoms that can be found in autism, although in none of the patients described was a formal diagnosis of autism made. Several patients have a short attention span and high threshold for pain. Many show a remarkable sensitivity for loud noises, which tends to decrease with age. Several parents have noticed that their children have oral sensitivity. Still they do like salty and spicy food. In older individuals, slowing

down of movements has been reported by parents.

Epilepsy

Epilepsy occurs in two-third of patients. The type of seizure is variable, even within the same individual. Mean and median ages at the first seizure are 23.9 and 18 months, respectively. Seizures can show increased frequency in some patients despite multiple anti-epileptic drugs, and indeed treatment can be very difficult. Sodium valproate is the medication of first choice for many. Some parents have noticed a co-occurrence of decreasing mental abilities with the onset of seizures. It seems unlikely that this is the case: in affected identical twins a cognitive decline co-occurred with the start of seizures in one, while in the other the same decline occurred but seizures only started two years later [Sousa et al., 2009]. We have suggested that the same process that causes the decrease in

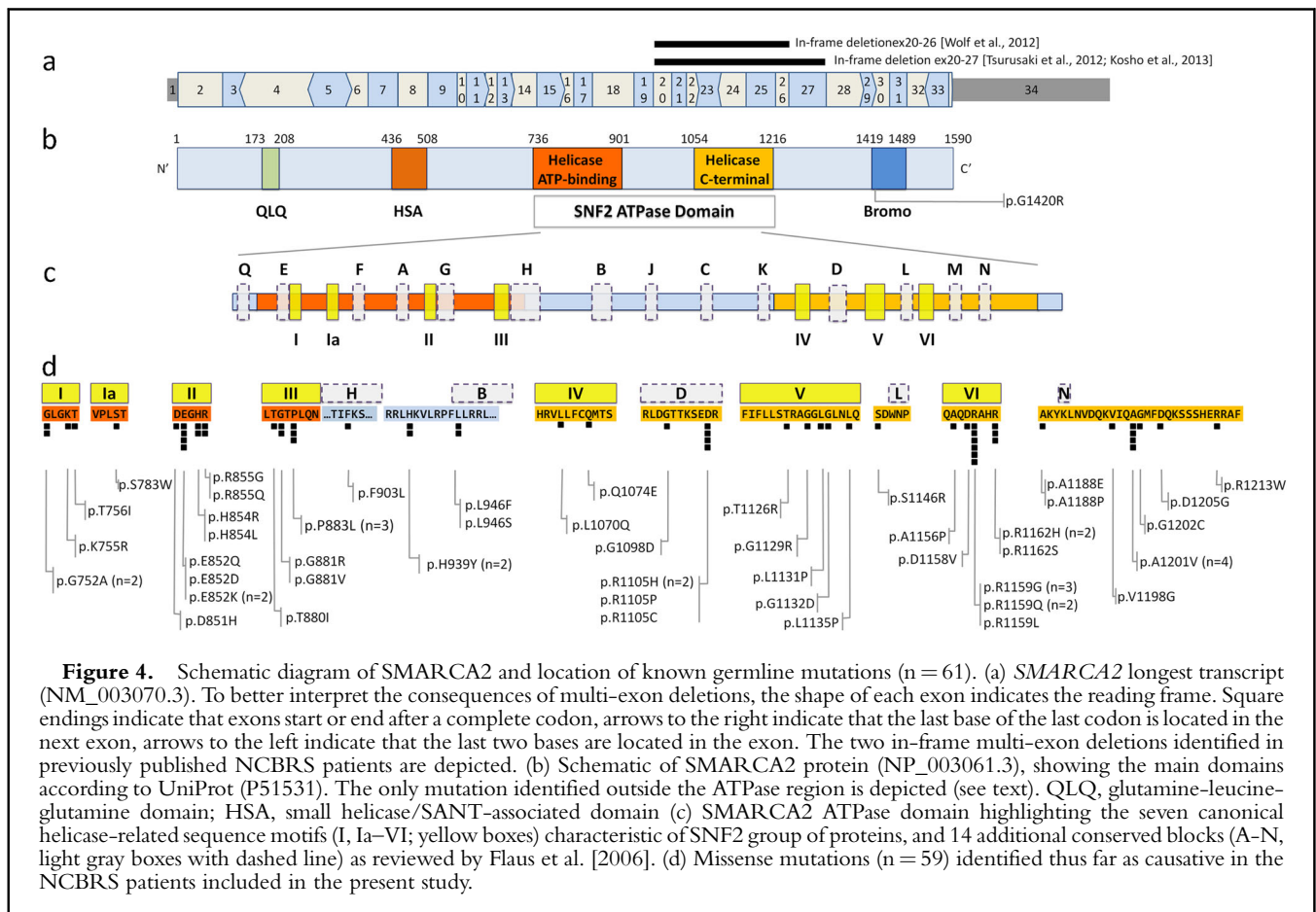
cognitive abilities is also responsible for the seizures [Sousa et al., 2009]. The electroencephalograms do not show specific abnormalities.

GENOTYPE

SMARCA2

SMARCA2, also known as *hBRM*, is located on chromosome 9p24.3. The longest transcript (NM_003070) contains 34 exons corresponding to an 1,590 amino acids protein (NP_003061), which is one of the two alternative homologues ATPase subunits (the other being *SMARCA4*) that constitute the catalytic core of the BAF complexes [Khavari et al., 1993; Muchardt and Yaniv, 1993; Wang et al., 1996] (Fig. 4). Both proteins are members of the Swi2/Snf2 family, which share a helicase-like ATPase domain [Gorbalenya et al., 1988; Flaus et al., 2006], and which has a translocation module consisting of

two similar protein domains that resemble the fold of the recombination protein RecA. The common feature of the Snf2 family proteins is a region of sequence similarity that includes seven canonical helicase-related sequence motifs (labeled sequentially I, Ia - VI), also found in DExx box helicases, which line at the inter-domain cleft separating the two RecA-like domains and are involved in ATP-binding/hydrolysis as well as DNA-binding [Dürr et al., 2006; Fairman-Williams et al., 2010]. In this region, 14 additional conserved blocks (A-N) were also described [Flaus et al., 2006] (Fig. 4). *SMARCA2* and *SMARCA4* proteins are highly similar with a sequence identity of 74% in humans and they display similar enzymatic properties [Khavari et al., 1993; Muchardt and Yaniv, 1993; Chiba et al., 1994]. Despite the similarities between these two proteins and that in certain circumstances they can potentially compensate for each other



[Strobeck et al., 2000], a growing number of studies have shown that these two alternative ATPase subunits have different and even antagonist roles in the regulation of differentiation, transcriptional control, and other important cell processes [Reyes et al., 1998; Bultman et al., 2000; Kadam and Emerson, 2003; Flowers et al., 2009].

Mutations in NCBRS

In the present review, we have only incorporated patients in whom a mutation was found. Indeed, in the last two years in all patients in whom the present authors (SBS, RCH) were convinced the diagnosis NCBRS was correct, a mutation was present, so we have no knowledge of NCBRS patients without *SMARCA2* mutation. Still, no *SMARCA2* mutation was found in 3/37 cases that we had classified as convincing NCBRS phenotype [Van Houdt et al., 2012], and in two of these (the patient described by Krajewska-Walasek et al. [1996] and patient 18 from Sousa et al. [2009]) a pathogenic *ARID1B* mutation was identified. Clinically the patients were reclassified as having Coffin–Siris syndrome [Santen et al., 2013]. There is also a patient described by Wiczorek et al. [2013], classified as NCBRS without mutation identified in any of the genes from BAF complex. The clinical manifestations in this patient are not completely convincing for NCBRS however.

All presently known germline *SMARCA2* mutations are depicted in Figure 4. All mutations identified in NCBRS are missense ($n = 59$) or in-frame deletions ($n = 2$) affecting the ATPase *SMARCA2* domain. All parents who were available for molecular analysis did not have the same mutation as their child, which is in accordance with the predicted genetic etiology. The exonic mutations were clustered in exons 15 ($n = 5$), 18 ($n = 16$), 19 ($n = 4$), 23 ($n = 2$), 24 ($n = 11$), and 25 ($n = 21$), so in the region encoding the ATPase domain (exon 15–25; 490 amino acids). Several mutations were recurrent; we identified in total 48 different missense mutations in 59

patients, affecting 34 different ultra-conserved amino acids. All mutations were predicted to be damaging by *in silico* analysis. The ATPase domain is 100% conserved in chimpanzee and mouse and 94.7% conserved in zebra and fish compared to the human protein and was shown to be functionally conserved [Khavari et al., 1993; Elfring et al., 1994]. Considering that deletions encompassing the whole human *SMARCA2* gene do not cause NCBRS [Christ et al., 1999], that mice lacking functional m*SMARCA2* do not present with major developmental abnormalities [Reyes et al., 1998; Bultman et al., 2000; Koga et al., 2009], that NCBRS mutations cluster at the *SMARCA2* ATPase region and that none of these variants are truncating, we have predicted that the mutations identified in NCBRS do not lead to haploinsufficiency, but rather have a dominant-negative or gain-of-function effect [Van Houdt et al., 2012].

All mutations identified in NCBRS are missense ($n = 59$) or in-frame deletions ($n = 2$) affecting the ATPase *SMARCA2* domain.

Mutations of yeast *SNF2* affecting the ATPase domain conserved motifs resulted in dominant-negative activity in a functional assay [Richmond and Peterson, 1996], and two of these mutations are identical to *SMARCA2* mutations in individuals with NCBRS, while at least an additional four map to the same motifs [Van Houdt et al., 2012]. This clustering of mutations provides genetic evidence that abolishing the ATP hydrolyzing engine, which provides energy directed toward the repositioning of histones on DNA, causes functional inactivation [Hall and Matson, 1999; De La Serna et al., 2000]. Several human *SMARCA2* (and *SMARCA4*) mutants involving the helicase domains were produced and

studied [Khavari et al., 1993; Muchardt and Yanivl, 1993; De La Serna et al., 2000; Magnani and Cabot, 2009]. In particular, a missense mutation affecting the highly conserved lysine residue on the ATP-binding site GKT (motif I), also studied in yeast [Laurent et al., 1993; Richmond and Peterson, 1996] and affecting the same residue as p.Lys755-Arg identified in one NCBRS patient, was transfected in C33 [Muchardt and Yanivl, 1993] and in NIH 3T3 cells [De La Serna et al., 2000]. These studies showed that these mutant proteins had normal nuclear localization, co-immunoprecipitated with other units of the BAF complex, and impaired the complex function.

These data support a model for NCBRS cellular/nuclear pathogenesis, in which non functional but structurally undamaged *SMARCA2* generates BAF complexes that may be intact with respect to their composition and proper positions in the chromatin; but, nonetheless, are functionally inactive resulting in a dominant-negative effect. As the mutant *SMARCA2* are able to recruit the other “targeting” subunits of the complex in the mentioned studies, these complexes are likely to be targeted to appropriate places in the genome, where they would fail to function, presumably in some event involving the remodeling of nucleosome structure [De La Serna et al., 2000]. As a consequence and through competitive inhibition, at each moment, wild-type endogenous BAF complexes would not have access to the genomic positions already taken by the mutant complexes. As previously suggested [De La Serna et al., 2000], other mechanisms may contribute to this effect, mainly the formation of incomplete BAF complexes around the mutant ATPase subunits depleting the wild-type endogenous *SMARCA2* and *SMARCA4* proteins of one or more of their associated subunits. Another question that arises from this proposed mechanism is which type of BAF complexes are affected by these mutant proteins, whether it is only the ones that use *SMARCA2* or also the ones that use *SMARCA4*. A combination of these effects is likely to occur.

As for CSS [Santen et al., 2012], there is no evidence of increased risk of malignancy in NCBRS patients: This might suggest that the effect of BAF complex subunits germline mutations identified in neurodevelopmental disorders, such as NCBRS and CSS, is different from the effect of somatic mutations involving the same subunits in cancer (and germline mutations associated with tumor predisposition syndromes). Nevertheless, the present cohort is in general still young, and occurrence of tumors at older ages cannot yet be excluded.

GENOTYPE-PHENOTYPE CORRELATIONS

As we did not include mutation-negative patients in our study, we cannot compare the phenotype of mutation-positive *versus* mutation-negative NCBRS patients. We have evaluated potential correlations between the various parts of the phenotype. There is no correlation between the facial characteristics and the neurodevelopmental level. There is a correlation between the severity of ID and epilepsy, speech impairment, short stature and microcephaly; individuals with severe ID are more likely to have postnatal short stature (17/25 vs. 12/22 vs. 1/9 patients with severe, moderate and mild ID, respectively), microcephaly (20/23, vs. 11/20 vs. 3/9 patients with severe, moderate and mild ID, respectively), absent speech (15/27 vs. 4/22 vs. 0/11 patients with severe, moderate and mild ID, respectively) and seizures (23/28, vs. 13/22 vs. 3/11 patients with severe, moderate and mild ID, respectively).

Comparing the groups with (39/59) and without (20/59) seizures, there is a correlation with ID severity (23/39 patients with seizures have severe ID vs. 13/39 having moderate ID and 3/39 having mild ID). Severe ID and seizures are especially prevalent in mutations located in motif VI (and II), and less in mutations affecting alanine 1201, proline 883 and leucine 946. Patients with seizures also have higher prevalence of microcephaly (25/35, vs. 10/35 in patients without seizures).

As illustrated in Figure 4, the missense mutations identified in the present NCBRS cohort locate in helicase motifs I, Ia, II, III, IV, V, and VI and in the SNF2 conserved blocks B, D, and H. In addition, eight highly conserved amino acid residues affected by mutations in 13 NCBRS patients are located in areas not previously identified as especially conserved or associated with a particular role, suggesting that these may also constitute functionally important motifs/residues of the ATPase domain. This seems particularly interesting in the area distal to motif VI and blocks M/N, where mutations are found in seven patients (especially residues 1201–1205).

More than half of NCBRS patients (36/61) have mutations affecting the C-terminal helicase region (Helicase_C) from the SMARCA2 ATPase domain (Fig. 4). This group tends to have severe ID and higher chance of developing seizures. Conserved motif VI (coded by exon 25) constitutes a hotspot with 11 patients having mutations located in it, six of which affect the arginine in position 1,159. This highly conserved residue interacts with the γ -phosphate of the bound ATP, stabilizing the domain's close arrangement, and is also necessary for the ATP hydrolysis [Dürr et al., 2006]. A mutation affecting this residue in the yeast was shown to have a more effective dominant-negative effect than mutations affecting the ATP-binding site [Richmond and Peterson, 1996]. These studies corroborate the phenotype severity of the six identified patients with mutations affecting this residue: all have severe ID, all have epilepsy (first episode at 1–2.5 years of life), three patients never developed speech and further two lost speech when seizures started, three have short stature and four have microcephaly. Four patients with moderate-severe phenotypes have mutations affecting the highly conserved arginine in position 1,105 located at conserved block D.

Two patients are described with multi-exon deletions affecting the C-terminal helicase domain (Fig. 4a) and which are also reviewed in the present

study. Patient 1 from Wolff et al. [2012] (Fig. 1Ab) has a 32 kb *de novo* in-frame deletion of exons 20–26 and subject 19 from Tsurusaki et al. [2012] (later re-described as SMARCA2-1 by Kosho et al. [2013]) has a 55 kb in-frame deletion of exons 20–27. Both patients have typical physical features of NCBRS associated with severe ID, absent speech, and seizures, but the Japanese patient has additional unusual features such as hypospadias and an episode of gross haematuria caused by immunoglobulin A nephropathy.

Twenty patients have mutations at the N-terminal/ATP-binding helicase region, nine of which located at the motif II (Fig. 4), which is also involved in ATP hydrolysis. However, in here we were not able to identify any consistent correlation.

Mutations p.Ala1201Val (n = 4), p.Pro883Leu (n = 3), and p.Leu946Ser/p.Leu946Phe (n = 2) are associated in the present study with a mild-moderate degree of intellectual disability, less speech impairment and a reduced chance of developing epilepsy. However, given the small number of patients and the fact that severely affected patients harbor mutations located in adjacent residues, one must be cautious in interpreting these observations.

All aforementioned patients with SMARCA2 mutations have been identified by targeted screening motivated by a NCBRS phenotype. We must assume that the phenotypic spectrum of SMARCA2 mutations can be wider. We reviewed a single patient (Fig. 5) in whom there was no suspicion for NCBRS and in whom exome sequencing identified a *de novo* SMARCA2 heterozygous variant (c.4258G > A, p.Gly1420Arg). This variant is not located at the ATPase helicase-like domain but at the evolutionarily conserved acetyllysine binding bromodomain (Fig. 4). This patient has phenotype distinct from NCBRS (no sparse hair or typical face, no independent walking and severe feeding problems) but still demonstrates overlap with NCBRS (severe ID, seizures, absent speech, rounded premaxilla, decreased subcutaneous fat, and slight prominence of interphalangeal



Figure 5. Clinical photographs of the patient with mutation p.Gly1420Arg located at the SMARCA2 bromodomain and identified by whole exome sequencing (male: left six years, middle seven years and right facial and hand photograph at 17 years). This patient was not included in the analyzed cohort (see text). Note the distinct but overlapping phenotype with NCBRS.

joints in the second decade of life) (Fig. 5). Although this variant is most likely causative for the phenotype in this patient, we did not include this patient in the NCBRS group analysis due to the differences in phenotype and mutation location. In our opinion, the diagnosis of NCBRS should be kept for patients with sufficient clinical resemblance to the typical NCBRS phenotype, which almost invariably can be caused by a *de novo* mutation affecting the SMARCA2 ATPase domain. Only by checking large groups of unselected individuals with ID will we learn the phenotype(s) that can be caused by mutations in SMARCA2. In comparison, mutations in SMARCA4 in individuals with Coffin–Siris syndrome are typically missense or in-frame deletions, and most are also located at the ATPase domain. A few CSS patients have been described with similar mutations outside this domain [Kosho et al., 2013; Santen et al., 2013] (but not at the bromodomain) while inactivating germline SMARCA4 mutations have been identified as causing specific tumor predisposition syndromes (not associated with ID) [Schneppenheim et al., 2010; Jelinic et al., 2014; Ramos et al., 2014; Witkowski et al., 2014].

Proposed Diagnostic Approach

In countries where whole exome sequencing is not available on a diagnostic basis, the molecular approach will be through Sanger sequencing. If a patient presents with a phenotype suggestive of

NCBRS, a careful clinical evaluation is needed, especially of the limbs and including radiographs of the hands. Small distal phalanges or small nails of the 5th finger has never been reported in NCBRS, and such a finding points to the presence of Coffin–Siris syndrome in the patient. If the diagnosis NCBRS remains likely, sequencing of the exons 15–25 of SMARCA2 should be the first diagnostic test (proposed sequence: 18 + 24 + 25 > 15 + 19 + 23 > 16 + 17 + 20 + 21 + 22). If this is negative, heterozygous in-frame deletions of this region of SMARCA2 should be investigated by MLPA or by detailed array CGH. If no SMARCA2 variants are identified, screening of the whole coding region of ARID1B gene and, if negative, exclusion of heterozygous whole gene or partial deletions should be the following step. If no mutation is found, re-evaluation of the clinical manifestations of the patient is suggested. If still compatible with NCBRS, untargeted whole exome sequencing can be considered on a research basis. Alternatively, if locally available, one can perform whole exome sequencing on a diagnostic basis *ab initio*, either targeted for all genes coding for proteins in the BAF complexes subunits [Wieczorek et al., 2013], or targeted for all genes known to cause intellectual disability. If no mutations are identified in these genes, untargeted whole exome sequencing on a research basis in the affected patient and both parents searching for *de novo* mutations may be indicated.

In the near future, it is predictable that whole exome sequencing will become part of routine diagnostics in many centers and the above stepwise Sanger approach will probably become less utilized. Only then we will be able to assess the true phenotypic variability of this group of conditions and evaluate how to improve the clinical classification discussed here. A molecularly based classification of patients allows a more detailed understanding of the disease, its phenotyping (“next-generation phenotyping” [Hennekam and Biesecker, 2012]) and its natural history. The publication of a significant number of additional variants involving the BAF complex genes and the phenotype present in the corresponding individuals will bring insight into the mechanisms underlying the pathophysiological process and facilitate the discrimination between non-pathogenic and pathogenic variants. Santen et al. [2013] extracted and analyzed all documented variants in the BAF subunits from the Exome Variant Server to verify if these putatively non-pathogenic variants could be classified rightfully as such or might have been classified as pathogenic variants if they would have been found in patients with ID. They concluded that variants can be reclassified reliably for most genes. However, for SMARCA2 (as for SMARCA4 and SMARCE1), it is difficult to discriminate between non-pathogenic and pathogenic variants without access to parental DNA [Santen et al., 2013]. Exome data on larger cohorts of phenotyped individuals,

confirmatory studies in variants present in databases to exclude false positives and functional studies will be needed to improve our ability to accurately classify variants in these genes [MacArthur et al., 2014]. It will also allow us to assess the phenotypic effect of second or further variants in other BAF complex subunits or elsewhere in the genome.

CONCLUSION

In conclusion, NCBRS has a specific phenotype that has a much more narrow spectrum than the phenotype in Coffin–Siris syndrome. There is a minority of patients in whom differentiation between NCBRS and Coffin–Siris syndrome is difficult, but with increasing experience of clinicians this will become easier. NCBRS is not only distinct but also genetically homogeneous as all genuine patients have de novo mutations in a single gene, *SMARCA2*. Indeed, in reverse, *SMARCA2* mutations have not been detected in individuals with other ID syndromes (except a single case detected by whole exome sequencing, as described above). Our study on the consequences of having a *SMARCA2* mutation is biased, as only patients with syndromic forms of intellectual disability in which NCBRS is considered are screened for mutations using classical Sanger sequencing approaches. With the increasing use of whole exome sequencing targeted for genes that cause intellectual disability, we will recognize *SMARCA2* mutations in individuals with phenotypes that are less clearly diagnostic of NCBRS. The consequences of such mutations for patients may, temporarily, remain uncertain, so detailed phenotyping will be mandatory to elucidate this. These studies, together with long-term follow-up data of cohorts of NCBRS patients should provide additional information that can be used to optimize care to patients and support to their families.

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REFERENCES

Bultman S, Gebuhr T, Yee D, La Mantia C, Nicholson J, Gilliam A, Randazzo F, Metzger D, Chambon P, Crabtree G, Magnuson T. 2000. A Brg1 null mutation in the mouse reveals functional differences among mammalian SWI/SNF complexes. *Mol Cell* 6:1287–1295.

Castori M, Covaciu C, Rinaldi R, Grammatico P, Paradisi M. 2008. A rare cause of syndromic hypotrichosis: Nicolaidis–Baraitser syndrome. *J Am Acad Dermatol* 59:S92–98.

Chiba H, Muramatsu M, Nomoto A, Kato H. 1994. Two human homologues of *Saccharomyces cerevisiae* SWI2/SNF2 and *Drosophila* brahma are transcriptional coactivators cooperating with the estrogen receptor and the retinoic acid receptor. *Nucleic Acids Res* 22:1815–1820.

Christ LA, Crowe CA, Micale MA, Conroy JM, Schwartz S. 1999. Chromosome breakage hotspots and delineation of the critical region for the 9p–deletion syndrome. *Am J Hum Genet* 65:1387–1395.

Coffin GS, Siris E. 1970. Mental retardation with absent fifth fingernail and terminal phalanx. *Am J Dis Child* 119:433–439.

De La Serna IL, Carlson Ka, Hill Da, Guidi CJ, Stephenson RO, Sif S, Kingston RE, Imbalzano aN. 2000. Mammalian SWI–SNF complexes contribute to activation of the *hsp70* gene. *Mol Cell Biol* 20:2839–2851.

Dürr H, Flaus A, Owen–Hughes T, Hopfner KP. 2006. Snf2 family ATPases and DExx box helicases: Differences and unifying concepts from high–resolution crystal structures. *Nucleic Acids Res* 34:4160–4167.

Elfring LK, Deuring R, McCallum CM, Peterson CL, Tamkun JW. 1994. Identification and characterization of *Drosophila* relatives of the yeast transcriptional activator SNF2/SWI2. *Mol Cell Biol* 14:2225–2234.

Fairman–Williams ME, Guenther UP, Jankowsky E. 2010. SF1 and SF2 helicases: Family matters. *Curr Opin Struct Biol* 20:313–324.

Flaus A, Martin DM, Barton GJ, Owen–Hughes T. 2006. Identification of multiple distinct Snf2 subfamilies with conserved structural motifs. *Nucleic Acids Res* 34:2887–2905.

Flowers S, Nagl NG, Beck GR, Moran E. 2009. Antagonistic roles for BRM and BRG1 SWI/SNF complexes in differentiation. *J Biol Chem* 284:10067–10075.

Gana S, Panizzon M, Fongaro D, Selicorni A, Memo L, Scandurra V, Vannucci C, Bigozzi M, Scordo MR. 2011. Nicolaidis–Baraitser syndrome: Two new cases with autism spectrum disorder. *Clin Dysmorphol* 20:38–41.

Gorbalenya AE, Koonin EV, Donchenko AP, Blinov VM. 1988. A novel superfamily of nucleoside triphosphate–binding motif containing proteins which are probably involved in duplex unwinding in DNA and RNA replication and recombination. *FEBS Lett* 235:16–24.

Hall MC, Matson SW. 1999. Helicase motifs: the engine that powers DNA unwinding. *Mol Microbiology* 34: 867–877.

Hennekam RC, Biesecker LG. 2012. Next–generation sequencing demands next–generation phenotyping. *Hum Mutat* 33: 884–886.

Van Houdt JKJ, Nowakowska BA, Sousa SB, van Schaik BDC, Seuntjens E, Avonce N, Sifrim A, Abdul–Rahman OA, van den Boogaard MJH, Bottani A, Castori M, Cormier–Daire V, Deardorff MA, Filges I, Fryer A, Fryns JP, Gana S, Garavelli L, Gillessen–Kaesbach G, Hall BD, Horn D, Huylebroeck D, Klapęcki J, Krajewska–Walasek M, Kuechler A, Lines

- MA, Maas S, Macdermot KD, McKee S, Magee A, de Man SA, Moreau Y, Morice-Picard F, Obersztyn E, Pilch J, Rosser E, Shannon N, Stolte-Dijkstra I, Van Dijck P, Vilain C, Vogels A, Wakeling E, Wieczorek D, Wilson L, Zuffardi O, van Kampen AHC, Devriendt K, Hennekam R, Vermeesch JR. 2012. Heterozygous missense mutations in SMARCA2 cause Nicolaides-Baraitser syndrome. *Nat Genet* 44:445–449S1.
- Jelinic P, Mueller JJ, Olvera N, Dao F, Scott SN, Shah R, Gao J, Schultz N, Gonen M, Soslow RA, Berger MF, Levine DA. 2014. Recurrent SMARCA4 mutations in small cell carcinoma of the ovary. *Nat Genet* 46:424–426.
- Kadam S, Emerson BM. 2003. Transcriptional specificity of human SWI/SNF BRG1 and BRM chromatin remodeling complexes. *Mol Cell* 11:377–389.
- Khavari PA, Peterson CL, Tamkun JW, Mendel DB, Crabtree GR. 1993. BRG1 contains a conserved domain of the SY12/SNF2 family necessary for normal mitotic growth and transcription. *Nature* 366:170–174.
- Koga M, Ishiguro H, Yazaki S, Horiuchi Y, Arai M, Niizato K, Iritani S, Itokawa M, Inada T, Iwata N, Ozaki N, Ujike H, Kunugi H, Sasaki T, Takahashi M, Watanabe Y, Someya T, Kakita A, Takahashi H, Nawa H, Muchardt C, Yaniv M, Arinami T. 2009. Involvement of SMARCA2/BRM in the SWI/SNF chromatin-remodeling complex in schizophrenia. *Hum Mol Genet* 18:2483–2494.
- Kosho T, Okamoto N, Ohashi H, Tsurusaki Y, Imai Y, Hibi-Ko Y, Kawame H, Homma T, Tanabe S, Kato M, Hiraki Y, Yamagata T, Yano S, Sakazume S, Ishii T, Nagai T, Ohta T, Niikawa N, Mizuno S, Kaname T, Naritomi K, Narumi Y, Wakui K, Fukushima Y, Miyatake S, Mizuguchi T, Saitu H, Miyake N, Matsumoto N. 2013. Clinical correlations of mutations affecting six components of the SWI/SNF complex: Detailed description of 21 patients and a review of the literature. *Am J Med Genet A* 161:1221–1237.
- Krajewska-Walasek M, Chrzanoska K, Czermska-Kowalska A. 1996. Another patient with an unusual syndrome of mental retardation and sparse hair? *Clin Dysmorphol* 5:183–186.
- Laurent BC, Treich I, Carlson M. 1993. The yeast SNF2/SWI2 protein has DNA-stimulated ATPase activity required for transcriptional activation. *Genes Dev* 7:583–591.
- MacArthur DG, Manolio TA, Dimmock DP, Rehm HL, Shendure J, Abecasis GR, Adams DR, Altman RB, Antonarakis SE, Ashley EA, Barrett JC, Biesecker LG, Conrad DF, Cooper GM, Cox NJ, Daly MJ, Gerstein MB, Goldstein DB, Hirschhorn JN, Leal SM, Pennacchio LA, Stamatoyanopoulos JA, Sunyaev SR, Valle D, Voight BF, Winckler W, Gunter C. 2014. Guidelines for investigating causality of sequence variants in human disease. *Nature* 508:469–476.
- Magnani L, Cabot RA. 2009. Manipulation of SMARCA2 and SMARCA4 transcript levels in porcine embryos differentially alters development and expression of SMARCA1, SOX2, NANOG, and EIF1. *Reproduction* 137:23–33.
- Morin G, Villemain L, Baumann C, Mathieu M, Blanc N, Verloes A. 2003. Nicolaides-Baraitser syndrome: Confirmatory report of a syndrome with sparse hair, mental retardation, and short stature and metacarpals. *Clin Dysmorphol* 12:237–240.
- Muchardt C, Yaniv M. 1993. A human homologue of *Saccharomyces cerevisiae* SNF2/SWI2 and *Drosophila* brm genes potentiates transcriptional activation by the glucocorticoid receptor. *EMBO J* 12:4279–4290.
- Nicolaides P, Baraitser M. 1993. An unusual syndrome with mental retardation and sparse hair. *Clin Dysmorphol* 2:232–236.
- Ramos P, Karnezis AN, Craig DW, Sekulic A, Russell ML, Hendricks WPD, Corneveaux JJ, Barrett MT, Shumansky K, Yang Y, Shah SP, Prentice LM, Marra MA, Kiefer J, Zismann VL, McEachron TA, Kalia B, Prat J, D'Angelo E, Clarke BA, Pressey JG, Farley JH, Anthony SP, Roden RBS, Cunliffe HE, Huntsman DG, Trent JM. 2014. Small cell carcinoma of the ovary, hypercalcemic type, displays frequent inactivating germline and somatic mutations in SMARCA4. *Nat Genet* 46:427–429.
- Reyes JC, Barra J, Muchardt C, Camus A, Babinet C, Yaniv M. 1998. Altered control of cellular proliferation in the absence of mammalian brahma (SNF2alpha). *EMBO J* 17:6979–6991.
- Richmond E, Peterson CL. 1996. Functional analysis of the DNA-stimulated ATPase domain of yeast SWI2/SNF2. *Nucleic Acids Res* 24:3685–3692.
- Santen G, Kriek M, Van Attikum H. 2012. SWI/SNF complex in disorder, SWItching from malignancies to intellectual disability. *Epigenetics* 1219–1224.
- Santen GWE, Aten E, Vulto-van Silfhout AT, Pottinger C, van Bon BWM, van Minderhout IJHM, Snowdowne R, van der Lans CaC, Boogaard M, Linsen MML, Vijfhuisen L, van der Wielen MJR, Vollebregt MJE, the Coffin-Siris consortium, Breuning MH, Kriek M, van Haeringen A, den Dunnen JT, Hoischen A, Clayton-Smith J, de Vries BBA, Hennekam RCM, van Belzen MJ. 2013. Coffin-Siris Syndrome and the BAF Complex: Genotype-Phenotype Study in 63 Patients. *Hum Mutat* 34:1519–1528.
- Schneppenheim R, Frühwald MC, Gesk S, Hasselblatt M, Jeibmann A, Kordes U, Kreuz M, Leuschner I, Martin Subero JI, Obser T, Oyen F, Vater I, Siebert R. 2010. Germline nonsense mutation and somatic inactivation of SMARCA4/BRG1 in a family with rhabdoid tumor predisposition syndrome. *Am J Hum Genet* 86:279–284.
- Schrier Vergano S, Santen G, Wieczorek D, Wollnik B, Matsumoto N, Deardorff M. 2013. Coffin-Siris Syndrome. RA, Pagon MP, Adam TD, Bird, al, Ed GeneReviews™ [Internet] Seattle. Univ Washington. Seattle: 1993–2013 Available from <http://www.ncbi.nlm.nih.gov/books/NBK131811/>
- Sousa SB, Abdul-Rahman OA, Bottani A, Cormier-Daire V, Fryer A, Gillissen-Kaesbach G, Horn D, Josifova D, Kuechler A, Lees M, MacDermot K, Magee A, Morice-Picard F, Rosser E, Sarkar A, Shannon N, Stolte-Dijkstra I, Verloes A, Wakeling E, Wilson L, Hennekam RC. 2009. Nicolaides-Baraitser syndrome: Delineation of the phenotype. *Am J Med Genet A* 149A:1628–1640.
- Strobeck MW, Fribourg AF, Puga A, Knudsen ES. 2000. Restoration of retinoblastoma mediated signaling to Cdk2 results in cell cycle arrest. *Oncogene* 19:1857–1867.
- Tsurusaki Y, Okamoto N, Ohashi H, Kosho T, Imai Y, Hibi-Ko Y, Kaname T, Naritomi K, Kawame H, Wakui K, Fukushima Y, Homma T, Kato M, Hiraki Y, Yamagata T, Yano S, Mizuno S, Sakazume S, Ishii T, Nagai T, Shiina M, Ogata K, Ohta T, Niikawa N, Miyatake S, Okada I, Mizuguchi T, Doi H, Saitu H, Miyake N, Matsumoto N. 2012. Mutations affecting component of the SWI/SNF complex cause Coffin-Siris syndrome. *Nat Genet* 44:376–8.
- Wang W, Côté J, Xue Y, Zhou S, Khavari PA, Biggar SR, Muchardt C, Kalpana GV, Goff SP, Yaniv M, Workman JL, Crabtree GR. 1996. Purification and biochemical heterogeneity of the mammalian SWI-SNF complex. *EMBO J* 15:5370–5382.
- Wieczorek D, Bögershausen N, Beleggia F, Steiner-Haldenstatt S, Pohl E, Li Y, Milz E, Martin M, Thiele H, Altmüller J, Alanay Y, Kayserili H, Klein-Hitpass L, Böhringer S, Wollstein A, Albrecht B, Buduroglu K, Caliebe A, Chrzanoska K, Cogulu O, Cristofoli F, Czeschik JC, Devriendt K, Dotti MT, Elcioglu N, Gener B, Goecke TO, Krajewska-Walasek M, Guillén-Navarro E, Hayek J, Houge G, Kilic E, Simsek-Kiper PÖ, López-González V, Kuechler A, Lyonnet S, Mari F, Marozza A, Mathieu Dramard M, Mikat B, Morin G, Morice-Picard F, Ozkynay F, Rauch A, Renieri A, Tinschert S, Utine GE, Vilain C, Vivarelli R, Zweier C, Nürnberg P, Rahmann S, Vermeesch J, Lüdecke HJ, Zeschmgk M, Wollnik B. 2013. A comprehensive molecular study on Coffin-Siris and Nicolaides-Baraitser syndromes identifies a broad molecular and clinical spectrum converging on altered chromatin remodeling. *Hum Mol Genet* 22(25):5121–35.22: 5121–5135.
- Witkowski L, Carrot-Zhang J, Albrecht S, Fahiminiya S, Hamel N, Tomiak E, Grynspan D, Saloustros E, Nadaf J, Rivera B, Gilpin C, Castellsagué E, Silva-Smith R, Plourde F, Wu M, Saskin A, Arseneault M, Karabakhtsian RG, Reilly EA, Ueland FR, Margiolaki A, Pavlakis K, Castellino SM, Lamovec J, Mackay HJ, Roth LM, Ulbright TM, Bender TA, Georgoulas V, Longy M, Berchuck A, Tischkowitz M, Nagel I, Siebert R, Stewart CJ, Arseneau J, McCluggage WG, Clarke BA, Riazalhosseini Y, Hasselblatt M, Majewski J, Foulkes WD. 2014. Germline and somatic SMARCA4 mutations characterize small cell carcinoma of the ovary, hypercalcemic type. *Nat Genet* 46:438–443.
- Witters I, Frysns JP. 2003. Mental retardation, sparse hair, facial dysmorphism with a prominent lower lip, and lipodystrophy. A variant example of Nicolaides-Baraitser syndrome? *Genet Couns* 14:245–247.
- Wolff D, Ende S, Azzarello-Burri S, Hoyer J, Zweier M, Schanze I, Schmitt B, Rauch A, Reis A, Zweier C. 2012. In-frame deletion and missense mutations of the C-Terminal helicase domain of SMARCA2 in three patients with Nicolaides-Baraitser syndrome. *Mol Syndromol* 237–244.