BRIEF REPORT

Neurofibromatosis type 1: A comparison of the 1997 NIH and the 2021 revised diagnostic criteria in 75 children and adolescents

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Introduction

Neurofibromatosis type 1 (NF1; OMIM 613113) is a complex, genetic multisystem condition manifesting in childhood and adolescence, affecting approximately 1 in 3000 live births worldwide.[1,2] Penetrance is virtually complete after adolescence.[1] NF1 is associated with a wide range of clinical signs and a significantly increased risk of

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Table 1 Overview of diagnostic criteria for neurofibromatosis type 1 according to NIH consensus statement 1987 and revised diagnostic criteria, diagnostic criteria for Legius syndrome and criteria for CMMRD counseling and testing

Diagnostic Criteria for Neurofibromatosis Type 1—NIH Consensus Statement 1987	Revised Diagnostic Criteria for Neurofibromatosis Type 1 (Legius et al)[9]
The diagnostic criteria for NF-1 are met in an individual if 2 or more of the following are found.	1. The diagnostic criteria for NF1 are met in an individual, who does not have a parent diagnosed with NF1, if 2 or more of the following are present
• ≥6 café-au-lait macules >5 mm in greatest diameter in prepubertal individuals and >15 mm in greatest diameter in postpubertal individuals	
• Freckling in the axillary or inguinal region	• Freckling in the axillary or inguinal region At least 1 of the 2 pigmentary findings (café-au-lait macules or freckling) should be bilateral
• ≥2 neurofibromas of any type or 1 plexiform neurofibroma	
• Optic pathway glioma	
• ≥2 Lisch nodules (iris hamartomas)	• ≥2 iris Lisch nodules identified using slitlamp examination or 2 or more choroidal abnormalities—defined as bright, patchy nodules imaged using optical coherence tomography/near-infrared reflectance imaging
• A distinctive osseous lesion such as sphenoid dysplasia or thinning of long bone cortex with or without pseudarthrosis	• A distinctive osseous lesion such as sphenoid dysplasia, anterolateral bowing of the tibia, or pseudarthrosis of a long bone; sphenoid wing dysplasia is not a separate criterion in case of an ipsilateral orbital plexiform neurofibroma
	• A heterozygous pathogenic *NF1* variant with a variant allele fraction of 50% in apparently normal tissue such as white blood cells
• A first-degree relative (parent, sibling, or offspring) with NF-1 by the earlier mentioned criteria	2. A child of a parent, who meets the diagnostic criteria specified in 1, merits a diagnosis of NF1 if 1 or more of the criteria in 1 are present

CMMRD, constitutional mismatch repair deficiency; *NF1*, Neurofibromatosis type 1; *NIH*, National Institutes of Health.

malignancy. Tumor surveillance is recommended in affected children and adolescents starting at birth and/or NF1 diagnosis.

The National Institutes of Health (NIH) Consensus Conference on Neurofibromatosis established diagnostic criteria in 1987,[3] which were reviewed and confirmed in 1997[4] (Table 1 and Box 1). The NIH criteria include the most frequent disease manifestations (café-au-lait macules [CALMs], freckling, neurofibromas, and Lisch nodules), specific disease complications (optic pathway glioma [OPG], sphenoid dysplasia, cortical thinning of long bones with/without pseudarthrosis), and a first-degree relative with

Box 1. Criteria for CMMRD counseling and testing in a child suspected to have NF1/Legius syndrome without malignancy (Suerink et al)[10]

Patient fulfills criteria for CMMRD testing when all 3 prerequisites are fulfilled and at least 1 additional feature (either in the family or in the patient) is present

Prerequisites fulfilled
- Suspicion of NF1 due to the presence of at least 1 diagnostic NF1 feature (according to the NIH consensus statement 1987), including ≥ 2 hyperpigmented skin patches reminiscent of café-au-lait macules
- No *NF1* and *SPRED1* germline mutations detected using comprehensive and highly sensitive mutation analysis protocols
- Absence of diagnostic NF1 sign(s) in both parents

Additional features in the family
- Consanguineous parents
- Sibling with diagnostic NF1 sign(s)
- A (deceased) sibling with any type of childhood malignancy
- Genetic diagnosis of Lynch syndrome in 1 or both of the parental families
- Carcinoma(s) from the Lynch syndrome spectrum (colorectal, endometrial, ovarian, gastric, small bowel, bile duct or gallbladder, pancreatic, urothelial cancer) before the age of 60 years in first-degree or second-degree relative

Additional features in the patient
- Atypical café-au-lait-macules (irregular borders and/or pigmentation)
- Hypopigmented skin patches
- One or more pilomatricoma(s) in the patient
- Brain MRI in the patient: multiple developmental vascular abnormalities in separate regions of the brain
- Agenesis of the corpus callosum
- Non–therapy-induced cavernoma

NF1. NF1 is generally accepted as a cancer predisposition syndrome. A number of conditions phenotypically overlapping NF1 have been recognized, among them are various RASopathies such as Legius syndrome and constitutional mismatch repair deficiency (CMMRD). These conditions differ in their natural history and their risk for tumor development. Thus, the precise distinction between these conditions is important for predicting the individual clinical course and determining the need and extent of tumor surveillance. For patients with Legius syndrome, no surveillance is recommended owing to the low likelihood of childhood cancer.[5] In contrast, patients with CMMRD may develop a large variety of neoplasms in childhood or adolescence and require a comprehensive surveillance regimen.[6] However, both conditions may be clinically diagnosed as NF1 using the NIH criteria.[7,8]

An international panel of neurofibromatosis experts published revised diagnostic criteria for NF1 and Legius syndrome in 2021 incorporating recent advances in clinical phenotyping and genetic testing.[9] (Table 1 and Box 1) We evaluated these proposed revised diagnostic criteria in children and adolescents who were referred to undergo evaluation regarding NF1 in a tertiary referral hospital (referred to as pediatric cohort). We asked whether the use of the proposed revised diagnostic criteria improved the distinction between NF1, Legius syndrome, and CMMRD.

Materials and Methods

We performed a database search in the hospital information system (ORBIS v. 08.043.302.11210 DACHL, Agfa Healthcare) of the University Children's Hospital Augsburg with International Classification of Diseases-10 code Q85.0 (phakomatoses, not elsewhere classified). Results were checked for plausibility reviewing text entries (NF1, Legius syndrome, CALM, OPG, Neurofibroma, Lisch nodule). Patients aged <18 years at first contact and in inpatient and/or outpatient care between January 1, 2017 and December 31, 2020 were included. The study was approved by the responsible ethics committee of the Ludwig Maximilian University of Munich (approval number, 21-1103), Germany.

We evaluated the clinical phenotypes through retrospective chart review. Clinical signs were classified according to the NIH diagnostic criteria for NF1[3] and the revised diagnostic criteria for NF1.[9] If applicable, alternative diagnostic criteria for Legius syndrome[9] and CMMRD testing (https://www.i-med.ac.at/tumorgenetik-erbliche-tumoren/cmmrd.html) were reviewed (Table 1 and Box 1). We analyzed the diagnostic criteria for NF1 according to those clinical signs documented at the first visit or reported age (referred to as first suspicion of NF1) and at last follow up. An item was considered not present for an individual patient if no information was provided in the patient's chart and/or if magnetic resonance imaging (MRI) and/or ophthalmologic evaluation was not performed. Follow up ended on June 30, 2021.

Results

A total of 75 children and adolescents with suspected or clinically diagnosed NF1 were identified at the University Children's Hospital Augsburg with a median age of 11.0 years at last follow up (range 1.1-22.6 years) (Supplemental Table 1). Sex ratio showed a small male predominance (40 [53.3%] males; 35 [46.7%] females).

At first suspicion (Table 2 and Supplemental Figure 1), 44 of 75 (58.7%) patients met the NIH criteria at a median age of 4.1 years (range 0.0-13.6 years). In contrast, 56 of 75 (74.7%) patients would have been diagnosed with NF1 on the basis of the revised diagnostic criteria at a median age of 3.5 years (range 0.0-13.6 years) (Fisher exact test: $P = .0562$). At last follow up, 53 (70.7%) patients met the NIH criteria and 57 (76.0%) patients met the revised diagnostic criteria ($P = .58$). In total, 9 patients who had only pigmentary changes developed additional nonpigmentary NF1 signs (neurofibromas and/or OPG). Of these patients, 3 fulfilled the revised criteria before the development of nonpigmentary signs owing to the identification of *NF1* pathogenic variants. In total, 4 patients who had only CALMs at first suspicion developed skinfold freckling at last follow up; all of these patients fulfilled the revised criteria owing to the presence of *NF1* pathogenic variants.

Next, we took a closer look at the distinguishing factors between the 12 patients who fulfilled the revised criteria but not the NIH criteria at their first visit. We found that a pathogenic *NF1* variant was identified in 11 of the 12 patients with CALMs and in 1 clinically unsuspicious patient with a parent diagnosed with NF1. The median age of those patients was 1.9 years (range 0.0-7.2 years). Subsequently, 8 of those 12 patients met the NIH criteria. However, the remaining 4 patients (presenting with a variable number and size of CALMs only) still did not meet the NIH criteria at last follow up (median age 4.9 years; range 1.6-7.9 years).

At last follow up (Supplemental Table 1), 6 or more CALMs were detected in 69 (92.0%) patients and freckling in the axillary or inguinal region in 28 (37.3%) and 21 (28.0%) patients, respectively. Two or more neurofibromas of any type were diagnosed in 27 (36.0%) patients, 22 had at least 1 plexiform neurofibroma. In 43 patients, brain MRI with or without contrast was performed for various reasons (eg, visual disorder, screening), 18 of them were diagnosed with OPG. A total of 5 (6.7%) patients were diagnosed with Lisch nodules and 2 (2.7%) patients with sphenoid dysplasia. In 20 of 65 (30.8%) patients with documented family history, a first-degree relative was diagnosed with NF1 (the father in 9 patients, the mother in 11 patients). Of these 20 patients, 6 had affected siblings: 1 sibling was diagnosed with NF1, another with Lisch nodules, and 4 siblings presented with CALMs only. Genetic testing was performed in 31 of 75 (41.3%) patients; in 26 of 31 patients (83.9%), a pathogenic *NF1* variant was identified.

Further analysis of the data collected during the last follow up revealed that 48 of 75 (64.0%) patients met both

Table 2 Revised and NIH diagnostic criteria in 75 children and adolescents suspicious of neurofibromatosis type 1 at first suspicion of NF1

Pat Nr	Age at First Suspicion	Sex	CALM ≥6	Freckling	OPG	Neurofibromas ≥2 of Any Type	Neurofibromas ≥1 Plexiform	Parent Fulfilling NF1 Criteria	Lisch Nodules ≥2	Choroidal Abnormalities ≥2	Sphenoid Dysplasia	Pseudarthrosis	Anterolateral Bowing of the Tibia	Heterozygous Pathogenic *NF1* Variant	Revised Criteria Fulfilled	NIH Criteria Fulfilled
2021 revised and 1997 NIH diagnostic criteria fulfilled (patients with pigmentary findings only excluded)																
16	3.5	F	+	+	+	–	–	+	–	Not done	–	–	Not done	+	+	+
3	5.3	F	+	+	–	–	–	+	–	Not done	–	–	Not done	+	+	+
50	5.0	M	+	–	–	–	–	+	+	Not done	–	–	Not done	+	+	+
27	0.4	F	+	–	–	–	–	+	–	Not done	–	–	Not done	+	+	+
31	0.2	F	+	–	–	–	–	+	–	Not done	–	–	Not done	+	+	+
12	0.0	F	+	–	–	–	–	+	–	Not done	–	–	Not done	+	+	+
68	Unknown	M	+	–	–	–	–	+	–	Not done	–	–	Not done	+	+	+
71	Unknown	M	+	–	–	–	–	+	–	Not done	–	–	Not done	+	+	+
73	Unknown	M	+	–	–	–	–	+	–	Not done	–	–	Not done	+	+	+
66	Unknown	M	+	–	–	–	–	+	–	Not done	–	–	Not done	+	+	+
22	1.1	F	+	+	–	+	–	–	–	Not done	–	–	Not done	+	+	+
11	13.1	M	+	–	–	+	–	–	–	Not done	–	–	Not done	+	+	+
60	5.1	F	+	+	–	–	–	+	–	Not done	–	–	Not done	Not done	+	+
41	Unknown	F	+	–	–	–	+	+	–	Not done	–	–	Not done	Not done	+	+
1	0,4	M	+	–	–	–	–	+	–	Not done	–	–	Not done	Not done	+	+
15	Unknown	M	+	–	–	–	–	+	–	Not done	–	–	Not done	Not done	+	+
18	0.3	M	+	–	–	–	–	+	–	Not done	–	–	Not done	Not done	+	+
20	Unknown	F	+	–	–	–	–	+	–	Not done	–	–	Not done	Not done	+	+
36	0.4	F	+	–	–	–	–	+	–	Not done	–	–	Not done	Not done	+	+
40	Unknown	F	+	–	–	–	–	+	–	Not done	–	–	Not done	Not done	+	+
74	Unknown	F	+	–	–	–	–	+	–	Not done	–	–	–	Not done	+	+
8	1.7	F	+	–	+	–	–	Unknown	–	Not done	–	–	Not done	Not done	+	+
5	2.0	M	+	–	+	–	+	–	–	Not done	+	–	Not done	Not done	+	+
9	4.0	M	+	–	–	–	+	Unknown	–	Not done	–	–	Not done	Not done	+	+
45	0.2	F	+	–	–	–	+	–	–	Not done	–	–	Not done	Not done	+	+
57	0.6	M	+	–	–	–	+	–	–	Not done	–	–	Not done	Not done	+	+
25	5.6	F	+	–	–	+	+	–	–	Not done	–	–	Not done	Not done	+	+
32	Unknown	F	+	–	–	+	+	–	–	Not done	–	–	Not done	Not done	+	+
54	Unknown	M	+	–	–	+	+	Unknown	–	Not done	–	–	Not done	Not done	+	+
55	Unknown	F	+	–	–	+	+	Unknown	–	Not done	+	–	Not done	Not done	+	+
4	1.8	F	+	+	–	+	–	–	–	Not done	–	–	Not done	Not done	+	+
30	3.9	M	+	–	–	+	–	–	–	Not done	–	–	Not done	Not done	+	+
33	8.4	M	+	–	–	–	–	–	+	Not done	–	–	Not done	Not done	+	+
44	4.4	M	+	+	+	–	–	–	–	Not done	–	–	Not done	Not done	+	+
52	Unknown	F	+	–	+	–	–	–	–	Not done	–	–	Not done	Not done	+	+
53	7.0	F	+	–	+	–	–	–	–	Not done	–	–	Not done	Not done	+	+
61	13.6	F	–	+	+	–	–	Unknown	–	Not done	–	–	Not done	–	+	+
28	1.7	F	+	–	+	–	–	–	–	Not done	–	–	Not done	–	+	+

(continued)

Table 2 Continued

Pat Nr	Age at First Suspicion	Sex	CALM ≥6	Freckling	OPG	Neurofibromas ≥2 of Any Type	Neurofibromas ≥1 Plexiform	Parent Fulfilling NF1 Criteria	Lisch Nodules ≥2	Choroidal Abnormalities ≥2	Sphenoid Dysplasia	Pseudarthrosis	Anterolateral Bowing of the Tibia	Heterozygous Pathogenic *NF1* Variant	Revised Criteria Fulfilled	NIH Criteria Fulfilled
Pigmentary findings only																
13	7.4	F	+	+	–	–	–	–	–	Not done	–	–	Not done	+	+	+
14	4.4	M	+	+	–	–	–	–	–	Not done	–	–	Not done	+	+	+
26	5.2	F	+	+	–	–	–	–	–	Not done	–	–	Not done	Not done	+	+
59	7.7	F	+	+	–	–	–	–	–	Not done	–	–	Not done	Not done	+	+
70	3.0	M	+	+	–	–	–	Unknown	–	Not done	–	–	Not done	Not done	+	+
63	10.4	M	+	+	–	–	–	–	–	Not done	–	–	Not done	–	+	+
19	0.5	F	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
38	7.2	F	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
39	0.7	M	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
21	2.5	M	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
34	0.6	M	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
42	1.2	F	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
46	4.5	M	+	–	–	–	–	Unknown	–	Not done	–	–	Not done	+	+	–
48	0.3	F	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
56	0.3	M	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
69	3.0	M	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
72	1.4	F	+	–	–	–	–	–	–	Not done	–	–	Not done	+	+	–
2	2.4	M	+	–	–	–	–	Unknown	–	Not done	–	–	Not done	Not done	–	–
6	1.1	F	+	–	–	–	–	Unknown	–	Not done	–	–	Not done	Not done	–	–
10	Unknown	M	+	–	–	–	–	–	–	Not done	–	–	Not done	Not done	–	–
17	3.4	M	+	–	–	–	–	–	–	Not done	–	–	Not done	Not done	–	–
24	8.0	M	+	–	–	–	–	–	–	Not done	–	–	–	Not done	–	–
29	0.6	M	+	–	–	–	–	–	–	Not done	–	–	Not done	Not done	–	–
49	5.6	M	+	–	–	–	–	–	–	Not done	–	–	Not done	Not done	–	–
58	2.9	M	+	–	–	–	–	–	–	Not done	–	–	Not done	Not done	–	–
62	6.9	M	+	–	–	–	–	Unknown	–	Not done	–	–	Not done	Not done	–	–
64	4.3	F	+	–	–	–	–	–	–	–	–	–	Not done	Not done	–	–
65	7.6	F	+	–	–	–	–	–	–	–	–	–	–	Not done	–	–
75	2.2	M	+	–	–	–	–	–	–	Not done	–	–	–	Not done	–	–
7	5.4	M	+	–	–	–	–	–	–	Not done	–	–	Not done	–	–	–
47	1.3	M	+	–	–	–	–	–	–	Not done	–	–	Not done	–	–	–
Segmental clinical findings																
23	8.5	M	–	–	–	–	–	–	–	Not done	–	–	–	Not done	–	–
37	13.0	F	–	–	–	–	+	–	–	Not done	–	–	–	Not done	–	–
67	11.2	M	–	–	–	–	+	–	–	–	–	–	Not done	Not done	–	–
2021 revised and/or 1997 NIH diagnostic criteria not fulfilled																
43	12.2	M	–	–	–	–	–	–	–	Not done	–	–	Not done	Not done	–	–
35	1.1	F	–	–	–	–	–	–	–	Not done	–	–	Not done	Not done	–	–
51	0.0	M	–	–	–	–	–	+	–	Not done	–	–	Not done	+	+	–

+, present; –, not present; *CALM*, café-au-lait macules; *F*, female; *M*, male; *NF1*, neurofibromatosis type 1; *NIH*, National Institutes of Health; *Nr*, number; *OPG*, optic pathway glioma; *Pat*, patient.

diagnostic criteria, the NIH and the revised, including at least 1 criterion other than pigmentary findings. In 21 of those (21/48; 43.8%), genetic testing was performed and identified pathogenic *NF1* variants in 19 (19/21; 90.5%) individuals.

Within our pediatric cohort, 41.3% (31/75) of patients presented with only pigmentary findings (CALMs and/or skinfold freckling) at first suspicion of NF1. Genetic testing was performed in 16 of those 31 patients. Pathogenic *NF1* variants were identified in 13 patients (13/16; 81.3%). No pathogenic *NF1* variant was identified in the remaining 3 individuals (3/16; 18.8%). One of those 3 patients fulfilled requirements for CMMRD testing (CALMs, *NF1*/*SPRED1* germline variant negative, absence of NF1 signs in both parents) but no additional feature of CMMRD was present in the patient or family. In another one of those 3 patients, a heterozygous variant of uncertain significance was identified in *SPRED1*. *SPRED1* analysis was not performed in the third patient.

Three (3/75; 4.0%) other patients presented with segmental clinical findings (solitary [$n = 2$] and multiple neurofibroma [$n = 1$] with or without CALMs with localized distribution) and clinically unaffected parents and siblings. Thus, we suspected segmental mosaic NF1.

Three (3/75; 4.0%) additional patients did not meet the NIH and/or the revised diagnostic criteria at first suspicion of NF1. So far, 1 of these 3 patients underwent genetic testing confirming NF1. In a second patient, 5 CALMs were documented and NF1 was suspected but the NIH and revised criteria were not fulfilled. Yet, a sibling was diagnosed with Hodgkin lymphoma and presented with more than 6 CALMs. Although this patient met 1 additional feature (ie, sibling with a diagnostic NF1 sign and any childhood malignancy), requirements for CMMRD testing (ie, no *SPRED1* germline variant detected) were not fulfilled. Both individuals did not undergo genetic testing.

Focal areas of signal intensity (FASI), nevus anemicus, and juvenile xanthogranuloma have been proposed as diagnostic criteria but were not incorporated in the revised criteria. Brain MRI were performed in 43 patients and FASI were detected in 67.4% (29/43). In total, 26 (26/29; 89.7%) of those patients met the revised diagnostic criteria at first suspicion of NF1, whereas 2 (2/29; 6.9%) did not meet the criteria until the last follow up. A total of 11 (11/29; 37.9%) patients with FASI underwent genetic testing. Heterozygous pathogenic *NF1* variants were identified in all of them. Juvenile xanthogranuloma was documented in 1 of 75 patients. Nevus anemicus was not documented in any of the patients.

Discussion

Clinical diagnosis of NF1 in children and adolescents holds numerous challenges, eg, the low sensitivity of NIH NF1 clinical diagnostic criteria in young patients. In addition, there are limitations to differentiate between phenotypically overlapping conditions, particularly in children presenting with pigmentary findings only. On these grounds, international experts proposed revised diagnostic criteria in 2021.

Our data mirror these challenges. We identified 75 children and adolescents with suspected NF1 in a pediatric cohort at the University Children's Hospital Augsburg. At first suspicion of NF1, 58.7% of patients met the NIH criteria, whereas 74.7% would have been diagnosed on the basis of the revised criteria. This difference was due to the molecular genetic testing of 11 of the 75 (14.7%) patients. The revised criteria incorporated genetic testing, which is useful especially in young children who would have not fulfilled the NIH criteria. In line with this observation, the difference decreased (70.7% NIH, 76.0% revised) at last follow up upon increasing penetrance of clinical NF1-signs (freckling, neurofibromas, OPG) and increasing age.

Importantly, the revised criteria merely include an affected parent and not an affected sibling. The expert panel for CMMRD diagnostic genetic testing decided that a diagnosis of CMMRD should be considered, if only siblings were affected.[10] In our patient cohort, all affected first-degree relatives were affected parents.

CALMs were documented in 4 siblings from unaffected parents. It is noteworthy that 1 sibling with CALMs and Hodgkin lymphoma was reported, warranting further evaluation for CMMRD in 1 of those patients.

We identified 3 patients with a segmental distribution of neurofibroma and/or CALMs. According to the expert panel, genetic testing for mosaic NF1 is recommended.

The consensus recommendations advocate genetic testing for children who meet the NF1 diagnosis on the basis of only pigmentary findings and consideration of alternatives such as Legius syndrome and CMMRD.[9] In our cohort, 31 patients presented with isolated pigmentary findings, of which only 51.6% underwent genetic testing. So far, genetic testing confirmed NF1 in most of these patients. In 1 patient, a variant of uncertain significance in *SPRED1* was identified and in other 2 patients, genetic testing needs to be complemented by *SPRED1*. One of those patients, however, presented with additional features suspicious of CMMRD. Based on the documented clinical items/phenotype without supplemental genetic analyses, we were not able to consider alternative diagnoses such as other RASopathies and McCune-Albright syndrome in those children.

FASI, nevus anemicus, and juvenile xanthogranuloma were proposed diagnostic items but were not included in the revised criteria because of insufficient data on specificity and/or sensitivity. FASI were detected in 69.0% of our patients in the cranial MRI. Genetic testing was performed and heterozygous pathogenic *NF1* variants were identified in 37.9% of those patients. If FASI would have been a diagnostic criterion, an additional 6.9% of patients would have been diagnosed with NF1.

Our study has several limitations.

1. Our approach was based on a retrospective chart review. Important information may not have been fully documented.
2. The cohort has a high prevalence of OPG (18/75 and 18/43 with MRI data) and probably reflects the pediatric oncology bias of the University Children's Hospital Augsburg.
3. Choroidal abnormalities have a high specificity and sensitivity for NF1[11] and allow for the differentiation between NF1 and Legius syndrome.[11,12] However, we were not able to evaluate this new ophthalmologic criterion because choroidal abnormalities were not routinely documented and/or assessed.
4. We may not have been aware of anterolateral bowing of the lower limbs.
5. Finally, the detection rate and specificity of *NF1* genetic testing depend on the approach including dosage analysis to detect copy-number variants and DNA- and RNA-based sequencing approaches.

In our study, genetic testing was performed in various laboratories with different and possibly not all technical approaches. Thus, we may have missed pathogenic *NF1* variants in some patients. Two patients met the NIH and the revised diagnostic criteria (including bilateral pigmentary findings) but pathogenic *NF1* variants were not identified in their blood samples. This raises suspicion of generalized (postzygotic) mosaic NF1 and the analysis of affected tissue may be necessary. In addition, sensitivity of the technology for detecting mosaicism needs to be considered.

In our pediatric cohort, the 2021 revised diagnostic criteria established an NF1 diagnosis in more patients than the 1997 NIH diagnostic criteria. Criteria were fulfilled using genetic testing in all 12 patients diagnosed by revised criteria before NIH criteria. Particularly young children with pigmentary findings only were detected by the new diagnostic criterion "genetic diagnosis". To identify patients with segmental mosaic NF1, patients with pigmentary findings only, patients with additional features suggestive of CMMRD, and/or patients with alternative diagnosis, thorough clinical evaluations are necessary and may be complemented by genetic testing.

Data Availability

Data are available individually upon request.

Acknowledgments

This work is supported by the PLGA Fund at the Pediatric Brain Tumor Foundation of the United States, Inc. The research of M.K. on hereditary cancer predisposition is supported by research funding of the Medical Faculty, University of Augsburg, Germany.

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Ethics Declaration

The study was approved by the ethics committee of Ludwig Maximilian University Munich, Germany (approval number, 21-1103). Owing to the retrospective manner of the study, written informed consent was not obtained.

Conflict of Interest

The authors declare no conflicts of interest.

Additional Information

The online version of this article (https://doi.org/10.1016/j.gim.2022.05.013) contains supplementary material, which is available to authorized users.

References

1. Lammert M, Friedman JM, Kluwe L, Mautner VF. Prevalence of neurofibromatosis 1 in German children at elementary school enrollment. *Arch Dermatol*. 2005;141(1):71–74. http://doi.org/10.1001/archderm.141.1.71.
2. Evans DG, Howard E, Giblin C, et al. Birth incidence and prevalence of tumor-prone syndromes: estimates from a UK family genetic register service. *Am J Med Genet A*. 2010;152A(2):327–332. http://doi.org/10.1002/ajmg.a.33139.
3. Neurofibromatosis. Conference statement. National Institutes of Health Consensus Development Conference. *Arch Neurol*. 1988;45(5):575–578.
4. Gutmann DH, Aylsworth A, Carey JC, et al. The diagnostic evaluation and multidisciplinary management of neurofibromatosis 1 and neurofibromatosis 2. *JAMA*. 1997;278(1):51–57.
5. Villani A, Greer MC, Kalish JM, et al. Recommendations for cancer surveillance in individuals with RASopathies and other rare genetic conditions with increased cancer risk. *Clin Cancer Res*. 2017;23(12):e83–e90. http://doi.org/10.1158/1078-0432.CCR-17-0631.
6. Tabori U, Hansford JR, Achatz MI, et al. Clinical management and tumor surveillance recommendations of inherited mismatch repair deficiency in childhood. *Clin Cancer Res*. 2017;23(11):e32–e37. http://doi.org/10.1158/1078-0432.CCR-17-0574.
7. Messiaen L, Yao S, Brems H, et al. Clinical and mutational spectrum of neurofibromatosis type 1-like syndrome. *JAMA*. 2009;302(19):2111–2118. Published correction appears in *JAMA*. 2010;303(24):2477. https://doi.org/10.1001/jama.2009.1663.
8. Wimmer K, Rosenbaum T, Messiaen L. Connections between constitutional mismatch repair deficiency syndrome and neurofibromatosis type 1. *Clin Genet*. 2017;91(4):507–519. http://doi.org/10.1111/cge.12904.
9. Legius E, Messiaen L, Wolkenstein P, et al. Revised diagnostic criteria for neurofibromatosis type 1 and Legius syndrome: an international

consensus recommendation. *Genet Med*. 2021;23(8):1506–1513. http://doi.org/10.1038/s41436-021-01170-5.

10. Suerink M, Ripperger T, Messiaen L, et al. Constitutional mismatch repair deficiency as a differential diagnosis of neurofibromatosis type 1: consensus guidelines for testing a child without malignancy. *J Med Genet*. 2019;56(2):53–62. http://doi.org/10.1136/jmedgenet-2018-105664.
11. Cassiman C, Casteels I, Jacob J, et al. Choroidal abnormalities in cafe-au-lait syndromes: a new differential diagnostic tool? *Clin Genet*. 2017;91(4):529–535. http://doi.org/10.1111/cge.12873.
12. Vagge A, Camicione P, Capris C, et al. Choroidal abnormalities in neurofibromatosis type 1 detected by near-infrared reflectance imaging in paediatric population. *Acta Ophthalmol*. 2015;93(8):e667–e671. http://doi.org/10.1111/aos.12750.