European Journal of Medical Genetics 63 (2020) 104042

Contents lists available at ScienceDirect

European Journal of Medical Genetics

journal homepage: www.elsevier.com/locate/ejmg

Neurofibromatosis type 2 in Phelan-McDermid syndrome: Institutional experience and review of the literature

Catherine A. Ziats [a,*], Lavanya Jain [a,b], Brittany McLarney [c], Emily Vandenboom [c], Barbara R. DuPont [a], Curtis Rogers [a], Sara Sarasua [b], Julian Nevado [d], Emanuela Lucci Cordisco [e,f], Katy Phelan [g], Luigi Boccuto [a,h]

[a] J.C. Self Research Institute of Human Genetics, Greenwood Genetic Center, Greenwood, SC, USA
[b] School of Nursing, College of Behavioral, Social and Health Sciences, Clemson University, Clemson, SC, USA
[c] Phelan-McDermid Syndrome Foundation, Osprey, FL, USA
[d] INGEMM -Instituto de Genética Médica y Molecular/Hospital Universitario La Paz, IdiPAZ- Instituto de Investigación Sanitaria del Hospital Universitario La Paz, Universidad Autónoma de Madrid, Madrid, Spain, and CIBERER (Centro de Investigación Básica en RED de Enfermedades Raras), Madrid, Spain
[e] Fondazione Policlinico Universitario A. Gemelli IRCCS, UOC Genetica Medica, Rome, Italy
[f] Istituto di Medicina Genomica, Università Cattolica del Sacro Cuore, Rome, Italy
[g] Cytogenetics Laboratory, Florida Cancer Specialists and Research Institute, Fort Myers, FL, USA
[h] Clemson University School of Health Research, Clemson, SC, USA

ARTICLE INFO

Keywords:
Phelan-McDermid syndrome
Neurofibromatosis type 2
NF2
Ring 22
r(22)

ABSTRACT

Phelan-McDermid syndrome (PMS) is a rare neurodevelopmental disorder caused by rearrangements on chromosome 22q13.3 or sequence variants in *SHANK3*. Individuals with PMS caused by a 22*q* terminal deletion and a ring chromosome are at increased risk for Neurofibromatosis type 2 (NF2). However, the prevalence of NF2 in individuals with PMS and a r (22) is unknown.

Individuals with PMS and a r (22) chromosome evaluated at the Greenwood Genetic Center (GGC) or by international collaborators, or identified through the PMS International Registry (PMSIR) were contacted and participated in a clinical questionnaire. Forty-four families completed the questionnaire and consented for the study. Of the individuals with a r (22), 7 (16%) carried a diagnosis of NF2. The average age of diagnosis of r (22) was 18 years old in individuals with NF2 and three years old in individuals without NF2 (p-value <0.001). Clinical findings were similar among all individuals in our sample with the exception of hearing loss, present in 57% of individuals with NF2 and 8% of individuals without NF2 (p-value <0.01).

This is the largest clinical report of individuals with PMS and a r (22) chromosome. We show a diagnosis of NF2 in individuals with r (22) is not uncommon and may be under ascertained. Moreover, the presentation of NF2 in this cohort is variable and lifelong routine screening for features of NF2 in this population should be considered.

1. Introduction

Phelan-McDermid Syndrome (PMS) is a rare neurodevelopmental condition caused by rearrangements on chromosome 22q13.3 or sequence variants in the *SHANK3* gene, and characterized clinically by neonatal hypotonia, delayed to absent speech development, seizures, autistic traits, minor dysmorphic facial features, and variable intellectual disability (ID) (Phelan and McDermid, 2012; De Rubeis et al., 2018; Droogmans et al., 2020; Ponson et al., 2018). The diagnosis is usually made in a proband with typical clinical features and detection of a chromosome 22q13.3 variant that encompasses *SHANK3* (Phelan and McDermid, 2012). Alterations in chromosome 22q13.3 in PMS can be caused by terminal/interstitial deletions, balanced or unbalanced translocations, mosaicism of the region, and ring 22 (r (22)) chromosomes (Bonaglia et al., 2011). Ring chromosomes are rare cytogenetic aberrations that are caused by loss of the distal regions of the p and q arms of the chromosome followed by fusion at the points of fracture or breakage (Kistenmacher and Punnett, 1970). It is estimated that up to 14% of individuals with PMS have a r (22) chromosome, however this is likely an underestimation of the true prevalence as many individuals

* Corresponding author. Greenwood Genetic Center, 106 Gregor Mendel Drive, Greenwood, SC, 29646. USA.
E-mail address: cziats@ggc.org (C.A. Ziats).

https://doi.org/10.1016/j.ejmg.2020.104042
Received 13 April 2020; Received in revised form 14 August 2020; Accepted 16 August 2020
Available online 19 August 2020
1769-7212/

with PMS are now diagnosed using chromosomal microarray alone (CMA) (Bonaglia et al., 2011).

Ring 22 chromosomes are important to identify in individuals with PMS as this diagnosis carries the added risk of developing Neurofibromatosis type 2 (NF2; OMIM 607379) (Tommerup et al., 1992; Tsilchorozidou et al., 2004; Zirn et al., 2012). NF2 is a noncancerous tumor syndrome characterized by several benign tumor types such as schwannomas, meningiomas, and ependymomas; affected individuals often develop bilateral vestibular schwannomas and multiple meningiomas, and diagnosis is confirmed with molecular analysis demonstration biallelic variants in the *NF2* gene (Evans et al., 1992). The *NF2* gene is a tumor suppressor gene, and in accordance with the two-hit model, tumorigenesis is thought to occur when both alleles are lost (Knudson, 1971). Although the terminal 22*q* deletion in individuals with a ring chromosome 22 usually does not include the more proximally located *NF2* gene, ring chromosomes are mitotically unstable and so are frequently lost with replication leading to complete monosomy 22 within the cell (Kistenmacher and Punnett, 1970; Tsilchorozidou et al., 2004). Tumorigenesis is thought to be triggered by spontaneous loss of the second normal *NF2* allele in somatic cells with monosomy 22, which is confirmed via molecular analysis of the tumor tissue showing biallelic loss of *NF2* (Zirn et al., 2012).

The association between r (22) chromosomes and NF2 is well recognized, however over the last 30 years there have only been a small number of reported cases of NF2 in r (22) with the majority of the individuals included in the original reports evaluated before PMS was a defined clinical disorder, and so the prevalence of NF2 in the PMS population specifically is unknown (Arinami et al., 1986; Denayer et al., 2009; Kehrer-Sawatzki et al., 1997; Lyons-Warren et al., 2017; Petrella et al., 1993; Tommerup et al., 1992; Tsilchorozidou et al., 2004; Zirn et al., 2012). Moreover, as management of medical comorbidities associated with PMS improve and the lifespan of these individuals approach those of the unaffected population, we expect the number of individuals with and without a r (22) will increase, emphasizing the importance of understanding the co-incidence of NF2 in this population. In an attempt to better understand the clinical characteristics associated with NF2 in the PMS population, we reviewed all individuals with PMS and a r (22) evaluated at the Greenwood Genetic Center (GGC) and by international collaborators, and additionally all individuals with a r (22) reported through the PMS International Registry (PMSIR). This observational study describes the association of NF2 in individuals with PMS and a r (22) highlighting the importance of karyotype testing in individuals with PMS with a terminal deletion on chromosome 22. Associated clinical and genetic findings as well as recommendations for diagnosis, screening and management in the r (22) PMS population are discussed. Additionally, we review prior reports describing the r (22) phenotype and the association with NF2 showing how our understanding of this phenotype and of the mechanisms of tumorigenesis have evolved.

2. Methods

Forty-four families of individuals with PMS and a r (22) chromosome were contacted and agreed to participate in a questionnaire developed to identify the presence of risk factors associated with development of NF2 (questionnaire included in the supplementary materials). Seven individuals had previously been evaluated at the GGC in Greenwood, SC and the remaining were recruited either through national meetings, international collaborators from Canada, Italy, Spain, Australia, and New Zealand, or through the PMSIR email server. Presence of clinical features of NF2, imaging results, family history of NF2, and genetic testing results were recorded via phone interview conducted by a physician or by a staff scientist trained in clinical genetics (Supplemental Table 1, Fig. 1). It was also noted if an individual had received a previous diagnosis of NF2 by their treating physician or if they had an NF2-associated tumor (meningioma, ependymoma, or schwannoma). All families of individuals with PMS provided consent to participate in the questionnaire. This study was approved by the Self Regional Healthcare IRB committee (project number Pro00081462). All statistical analyses were completed with Microsoft Excel software; two tailed student's t-test was used to analyze differences in age and in deletion size between NF2 and non-NF2 patients, and Chi-squared analysis was used to compare clinical phenotypes between the groups (Supplemental Table 1). All new and exceptional variants identified in individuals with PMS and included in this article were uploaded to the ClinVar database (http://www.ncbi.nlm.nih.gov/clinvar) under submission ID SUB7924559 (Supplemental Table 1).

Additionally, in an effort to determine the overall prevalence of r (22) in the PMS population in a larger cohort, the PMSIR was independently reviewed and a total of 442 genetic reports from individuals with PMS were analyzed. Individuals with sequence variants, an unclear diagnosis of PMS, a result that would not require a karyotype (such as an interstitial deletion), or with unknown and/or unclear karyotype results were removed to leave a sample of 340 individuals with a known terminal 22q13.3 deletion. Notably, several of the individuals and families that participated in the questionnaire reported uploading their genetic results into the PMSIR.

3. Results

Forty-four individuals with a diagnosis of PMS and r (22) and their families completed the clinical questionnaire and were included in the analysis (Supplemental Table 1). Seven individuals of 44 total (16%) had received a diagnosis of NF2 or had a NF2-associated tumor at the time of the interview. One individual with a diagnosis of NF2 had two deletions on chromosome 22 one of which encompassed the *NF2* gene and therefore was considered to have an inborn form of NF2, while the remaining individuals in the cohort had terminal deletions that did not include the *NF2* gene. The average age of diagnosis of r (22) in the entire cohort was six years old. However, the average age of diagnosis of r (22) differed significantly when differentiated by NF2 status; 18 years old in individuals with NF2 versus three years old in individuals without an NF2 diagnosis (p-value <0.0001). The average age of all individuals in the cohort at the time of data collection, including those individuals with and without NF2, was 15 years old; the average age of the individuals without NF2 was 12 years old and the average age of individuals with a diagnosis of NF2 was 30 years old (p-value <0.0001). The average deletion size carried by individuals in the cohort was 4.07 megabases (Mb) and was not significantly different between individuals with NF2 and individuals without NF2, 4.88 versus 3.79 Mb, respectively. Six individuals (four without NF2 and two with NF2) were mosaic for the r (22) chromosome on karyotype testing. The unique variants identified in this cohort are available on the ClinVar database (SUB7924559).

Clinical characteristics of individuals with r (22) included in our analysis are shown in Supplemental Table 1. Briefly, 11 (25%) patients had a history of seizures, 16 (33%) had a history of hypotonia, and 13 (30%) had a history of motor delay. Fourteen (32%) individuals had visual symptoms, with the most common being strabismus, present in ten individuals in the sample. One individual had a history of cataracts. Skin findings were uncommon in the cohort with only ten (23%) individuals reporting presence of at least one *café-au-lait* macule. Hearing loss and/or hearing problems were present in seven (16%) individuals in the cohort. Thirty (81%) individuals without a known diagnosis of NF2 reported undergoing cranial CT or MRI imaging as part of their clinical management of PMS. Thirteen of the 30 (43%) individuals without NF2 and with cranial imaging reported presence of an abnormality on cranial imaging results. The most common abnormality reported was an arachnoid or other benign intracranial cyst, which was reported by nine individuals. Overall, neurologic and NF2-related clinical findings tended to be more common in individuals with NF2 as compared to individuals without NF2, only presence of *café-au-lait* macules more common in individuals without NF2 (Fig. 1). Additionally, only percentage of

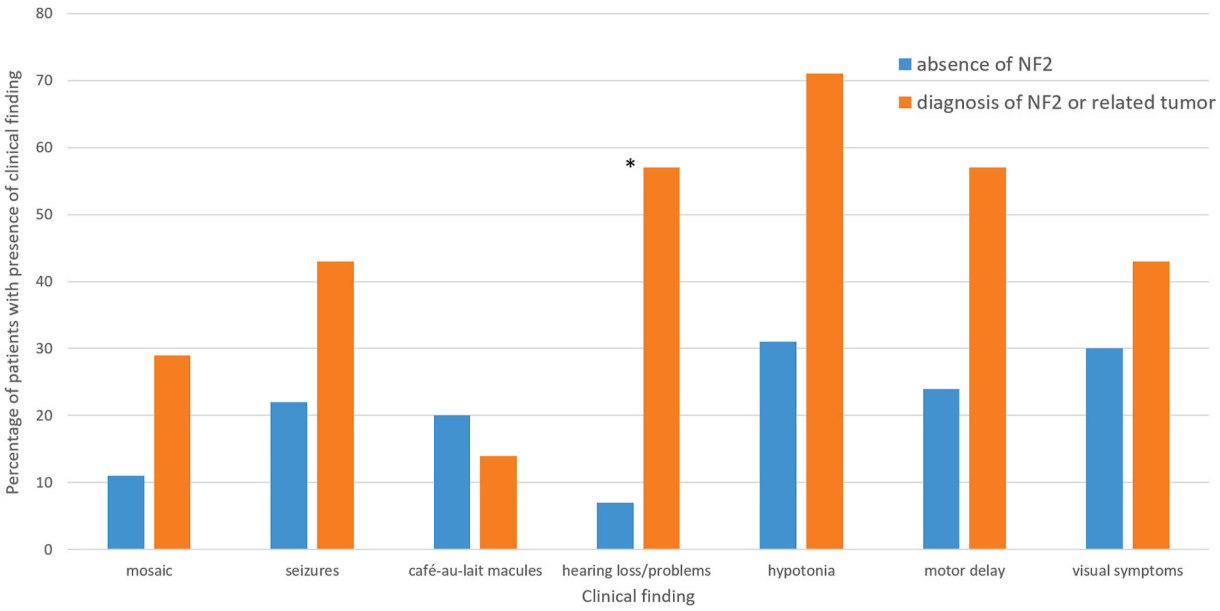

Fig. 1. Clinical characteristics of PMS patients with a ring 22 chromosome. The asterisk indicates a *p*-value of <0.01.

patients with presence of hearing problems, reported in four (57%) NF2 patients and three (8%) patients without NF2 (p-value < 0.01) reached statistical significance (Fig. 1, Supplemental Table 1).

Table 1 details clinical, genetic and imaging findings of the seven r (22) individuals identified in this cohort with NF2 or an NF2-associated tumor. The average age of NF2-associated tumor presentation in this sample was 25 years old, however the range was variable with the earliest tumor diagnosis given at three years old and the latest at 39 years old. Notably, the individual presenting with an NF2-associated tumor at age three had an interstitial deletion on chromosome 22q12.1q12.2 incorporating the *NF2* gene, in addition to a distal terminal chromosome deletion incorporating the PMS region and thus was considered to have inborn NF2. The most common tumor reported in the NF2 cohort was a vestibular schwannoma, which was present in six individuals. Three individuals reported surgical removal of an NF2-associated tumor.

A summary of genetic information reported in the PMSIR by individuals with PMS is shown in Supplemental Table 2. Sixty-two percent (211/340) of individuals with PMS and a terminal deletion on 22*q* who provided genetic reports for PMSIR review did not report karyotype testing as part of the genetic work-up for PMS. Of the 129 (41%) individuals that provided documentation of karyotype testing for PMSIR review, 42 (33%) had a r (22) chromosome.

4. Discussion

In this manuscript we describe the largest sample of individuals with PMS and a r (22) chromosome. Interestingly, before PMS was a clinically recognized syndrome, ID in addition to motor delay, and several minor craniofacial and skeletal anomalies were appreciated as common features of the r (22) phenotype (Hunter et al., 1977). However, the association of r (22) with PMS specifically, was not made until 2005, when it was noted that phenotypes in individuals with r (22) overlapped with those described in individuals with 22q13 deletions and PMS (Jeffries et al., 2005).

Previous reports have estimated the presence of r (22) in up to 14% of individuals with PMS, however based on our analysis this may be an underestimation of the true prevalence of this chromosome anomaly in this sample of individuals with PMS (Bonaglia et al., 2011). Our review of the PMSIR cohort showed that 33% of individuals with PMS and a terminal deletion confirmed on microarray report also had a r (22) based on karyotype testing. This number does not account for the prevalence of r (22) among all individuals with PMS as individuals not requiring karyotype testing, such as individuals with missense changes or interstitial deletions, were not included in this analysis. Additionally, the prevalence of r (22) among the population sampled does not account for the expected ascertainment bias associated with self-reporting of genetic information. Specifically, over half (62%) of individuals with PMS in the PMSIR cohort with a terminal deletion on 22q13.3 did not report having karyotype testing, and although it was assumed this was because karyotype testing was not performed, we cannot exclude the possibility that karyotype testing was simply not reported, especially in cases of a normal result. Despite this, our analysis draws attention to the under reporting of karyotype results overall, and highlights the importance of this testing in the work-up for PMS as karyotype testing is required for accurate identification of large structural abnormalities such as translocation and ring chromosomes, which would change recommendations regarding surveillance and management in this population.

Ring 22 chromosomes are important to recognize because of their association with central nervous system tumors and the benign tumor syndrome NF2 (Evans et al., 1992). This association was first reported in 1986 in a 27 year old male with a r (22), multiple meningiomas, severe ID, dysmorphic facies, hearing loss, and microcephaly (Arinami et al., 1986). Since this original description several other reported cases of r (22) in association with other benign tumorous lesions and the tumor syndrome NF2 have emerged (Table 2).

Neurofibromatosis type 2 is caused by pathogenic variants in the *NF2* tumor suppressor gene located at 22q12.2 and is autosomal dominant (AD) in the inherited form with complete penetrance usually by the 6th decade of life (Ardern-Holmes et al., 2017). In individuals with r (22) patients specifically, it was initially thought that mosaic loss of the r (22) and resulting monosomy 22 were the inciting events triggering tumorigenesis as several genes on chromosome 22, in addition to *NF2*, are known tumor-suppressor genes (Kehrer-Sawatzki et al., 1997). Sequencing of tumor tissue from individuals with a r (22) questioned this theory when several groups showed that similar to inherited cases of NF2, it was actually bi-allelic loss of *NF2* that likely triggered tumorigenesis, in accordance with Knudson's two-hit model of tumorigenesis, with mosaic loss of the r (22) representing the first hit and the second hit a spontaneous structural or sequence variant on the second *NF2* allele resulting in a non-functional *NF2* gene product (Denayer et al., 2009; Knudson, 1971; Tsilchorozidou et al., 2004; Zirn et al., 2012). However,

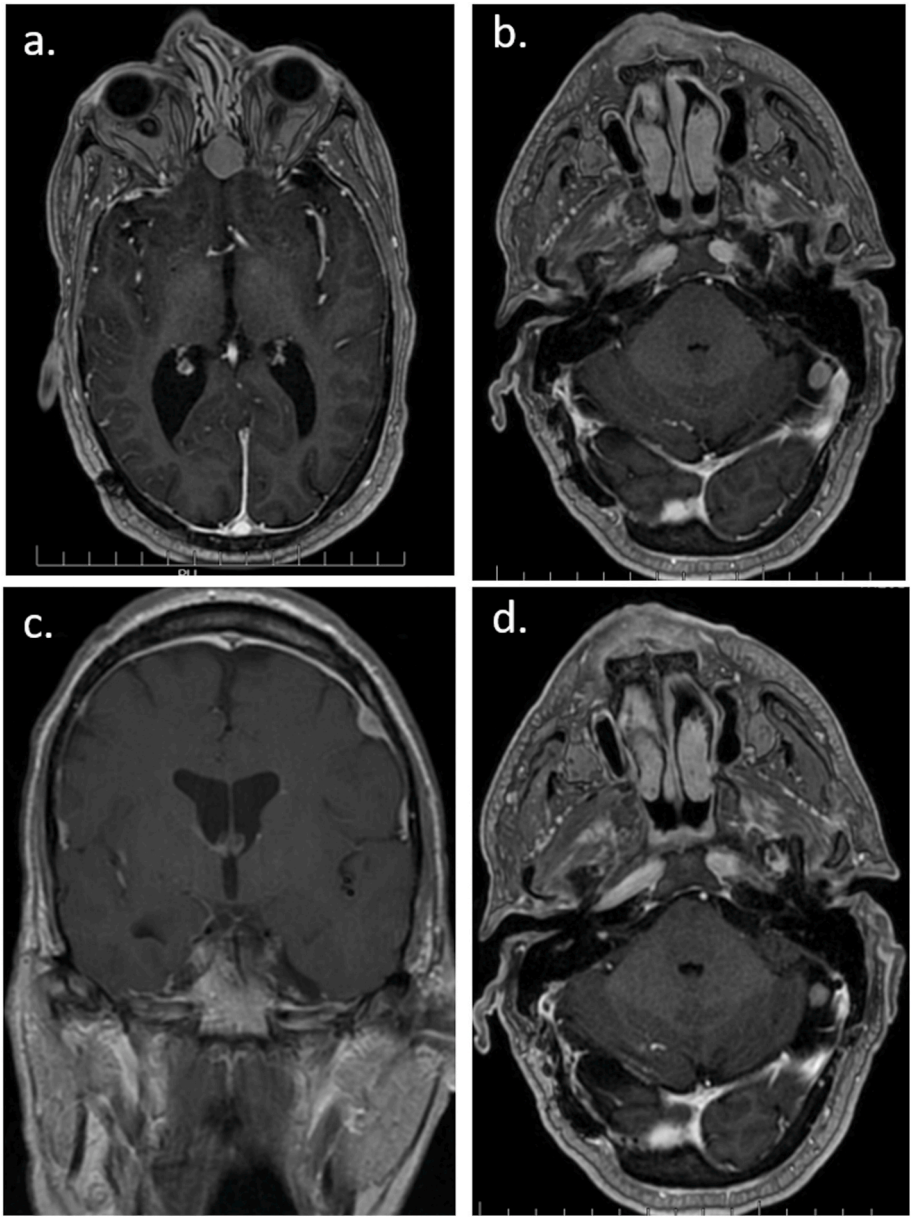

Fig. 2. T1 weighted post-gadolinium contrast MRI of patient # 4 in our cohort showing several small enhancing lesions favored to represent small meningiomas on the cribriform plate (a), left tentorium (b), left convexity (c), and a schwannoma along the right internal auditory canal (d).

despite these reports documenting increased risk for tumors in individuals with r (22), the prevalence, and clinical and radiologic findings associated with development of NF2 in this population has not been previously well-defined. Moreover, despite PMS being a recognized clinical syndrome for several decades, the specific risks associated with NF2 related tumors in PMS have not been well documented.

This report adds to the growing body of literature describing the association between r (22) chromosomes and risk for NF2. In our cohort, seven (16%) of 44 total individuals interviewed with PMS and r (22) either carried a diagnosis of NF2 or had tumors characteristic of NF2. However, the prevalence of NF2 in this sample may still be an underestimation of the true prevalence of NF2 in the PMS population as NF2 is typically diagnosed during the third decade of life and the average age of individuals without NF2 in our cohort was 12 years old (Ardern-Holmes et al., 2017). Notably, one of the individuals with NF2 in our cohort had a proximal deletion that incorporated the *NF2* gene and so was considered to have an inborn form of NF2 and thus potentially a different risk profile as compared to individuals with r (22) and a functional copy of *NF2* present on the ring chromosome.

The average age of diagnosis of a NF2-associated tumor in our cohort was 25 years old however the range was wide with the youngest reported age, not including the individual with a germline *NF2* deletion, being 17 years old. Additionally, while there were several individuals who were diagnosed with NF2 in their fourth decade of life, it is likely these individuals developed NF2-associated tumors several years earlier based on the severity of clinical exam findings and the presence of multiple intracranial tumors at the time of diagnosis. Further work is needed to better assess if monosomy 22 with loss of additional tumor suppressor genes on chromosome 22 contributes to tumorigenesis in individuals with r (22) and NF2 and how the natural history of disease differs as compared to individuals with NF2 due to bi-allelic hits involving only the *NF2* gene (Kehrer-Sawatzki et al., 1997).

Attempts at identifying early clinical screening markers indicative of tumor development in individuals with a family history of NF2 have been disappointing and therefore screening is dependent on MRI imaging primarily. Development of cataracts has been implicated by some

Table 1
Clinical characteristics of Phelan-McDermid syndrome patients with a ring 22 chromosome and NF2 or an NF2-associated tumor.

Current age (years)	Age at tumor presentation (years)	Neurologic symptoms	Clinical features of NF2	Deletion size	Breakpoints	NF2- associated tumor	Clinical course
45	39	epilepsy, hypotonia, motor delay	bilateral sensorineural hearing loss	4.4 Mb	p11.2q13.31*	bilateral vestibular schwannomas, multiple intracranial meningiomas **	has undergone surgery for removal of multiple meningiomas and currently undergoing radiosurgery for skull base meningioma
9	3 ***	epilepsy, hypotonia, motor delay	bilateral sensorineural hearing loss	3.3 Mb interstitial deletion incorporating *NF2*, 7.8 Mb distal terminal deletion in PMS region	unknown	multiple subcutaneous nodules of upper extremities, unilateral vestibular schwannoma	has undergone surgery for removal of several subcutaneous skin nodules
45	32	hypotonia, motor delay	3 *café-au-lait* macules	2.12 Mb	p13q11.2	bilateral vestibular schwannomas, 2 intracranial meningiomas	followed clinically, no history of surgery or radiosurgery for lesions
20	18	motor delay	bilateral sensorineural hearing loss	6.7 Mb	pterq13.31	bilateral vestibular schwannomas, spinal meningioma	recently developed hydrocephalus and plan for surgical removal of intracranial tumors pending
20	15	hypotonia	hearing problems without complete loss of hearing	2.83 Mb	p11q13	unilateral vestibular schwannoma, multiple intracranial meningiomas	has undergone radiosurgery for vestibular schwannoma, meningiomas followed clinically
29	*29*	*hypotonia, epilepsy, motor delay*	*none*	*9.5 Mb*	*p11.2q13*	*unilateral vestibular schwannoma*	*plan for surgical removal in near future*
39	*39*	*none*	*none*	*0.8 Mb*	*p11q11.2*	*posterior fossa meningioma*	*developed hydrocephalus and underwent surgical removal of tumor*

* mosaic for ring 22 chromosome; ** MRI of schwannoma shown in Fig. 2, *** inherited NF2 diagnosis.
Abbreviations: Mb = megabases Italics: Have not been formally diagnosed with NF2.

as an early clinical marker for disease development and moreover is thought to be the most promising clinical screening tool to date (Ardern-Holmes et al., 2017). While only one individual in our cohort reported presence of cataracts, given the high prevalence of cataracts and other ophthalmologic issues in individuals with inherited NF2, yearly eye examinations should be considered in all individuals with r (22) (Evans, 1993). Notably, several individuals (10/44, 23%) in our cohort reported presence of *café-au-lait* macules, however it is unclear how useful this clinical finding is for predicting risk for NF2 in individuals with r (22) as several groups have found that *café-au-lait* macules in particular, when present in individuals with inborn NF2, usually present later in the disease course (Evans, 1993). Additionally, *café-au-lait* macules have been implicated by many groups in patients with other ring chromosomes, and further evidence that this feature is less likely to be a specific clinical sign for increased risk of NF2- associated tumor development in our population of r (22) patients (Fagan et al., 1988; Morava et al., 2003; Zen et al., 2005). Clinician re-assessment with deep phenotyping is needed to confirm the above trends as our analysis relies on parental recall alone, and many of the families interviewed either could not recall presence or absence of certain clinical features, or provided medical records that did not note presence of these clinical findings specifically. Additional analysis will be especially important to further evaluate for differences in neurological symptoms between individuals with PMS and a r (22) with NF2 and those without NF2. Moreover, several families interviewed did not provide information about PMS-associated neurological symptoms such as hypotonia, seizures, or other motor delays, and we suspect this inconsistent reporting contributed to the lower than expected prevalence of these symptoms in this cohort. Additionally, the clinical trends observed may have been confounded by ascertainment bias and possible overrepresentation of NF2 and NF2 features in a cohort identified through a self-reported and voluntary survey, where in most cases a diagnosis of NF2 was made clinically. Similarly, ascertainment bias may have also contributed, at least in part, to higher rates of PMS-associated symptoms, especially neurological symptoms such as hypotonia and motor delay in individuals with NF2, who in general tended to be older in age and therefore perhaps be expected to have a more complex phenotype as compared to younger children with a recent diagnosis of PMS.

Interestingly while few (16%) of the individuals in our cohort reported hearing loss and or hearing problems, a significantly higher proportion of individuals with NF2 as compared to individuals without an NF2-associated tumor reported hearing loss (57% compared to 7% respectively, $p = 0.005$). As hearing loss is an important feature of NF2, it is critical to assess for in our sample of individuals without prior diagnosis of NF2 (Evans, 1993). However, hearing loss in the PMS population is particularly difficult to evaluate without formal audiology testing as the majority of these individuals have severely delayed or absent speech in addition to varying degrees of ID. Therefore, in the PMS population specifically, yearly audiology evaluation including formal tympanometry (or otoacoustic emissions) testing could be considered.

Previous reports have encouraged conservative screening for NF2 in individuals with PMS patients and a r (22) chromosome, and guidelines for monitoring for development of related signs and symptoms of disease are similar to those recommended for children with a parent with a germline variant in *NF2* or inborn NF2 (Lyons-Warren et al., 2017; Phelan et al., 1993). This report supports those prior recommendations however, in the PMS population with a r (22) we would encourage cranial MRI imaging at least every two years starting at 10 years old, and would caution decreasing the frequency of cranial MRI imaging in individuals over the age of 30, which is the current recommendation in individuals at risk for NF2 based on family history alone (Ardern-Holmes et al., 2017). This is because in individuals over the age of 30 at risk for inborn NF2, the risk of NF2 reduces with each normal scan, increasing the likelihood that the individual has not inherited NF2. In contrast, in PMS the risk is inherent in the condition and so imaging should continue throughout life. Moreover, baseline spinal MRI imaging at time of initial cranial imaging should be also considered as the limitations in

Table 2
Published cases of patients with a ring 22 and a diagnosis of a NF2-associated tumor.

Case	Inheritance	ID/DD	Other features	NF2- associated tumor	Age at tumor presentation (years)	Other clinical features of NF2	Clinical course
1: (Tommerup et al., 1992)	*de-novo*	severe ID and DD	unsteady gait, hypotonia, absent speech, hydronephrosis, cardiac defect, dysmorphic facies, testicular seminoma with cryptorchidism	unilateral vestibular schwannoma at CP angle	22	cutaneous schwannomas	unilateral deafness, optic atrophy, progressive hydrocephalus
2: (Arinami et al., 1986)*	unknown	severe ID and DD	microcephaly, dysmorphic facies, increased tone, hearing loss	multiple meningiomas near falx, large cystic meningioma at CP angle	27 (diagnosis on autopsy, however symptoms present at 25 yo)	none	-progressive insomnia, apathy, tremor, gait instability, seizures -death at 27 yo caused by obstructive hydrocephalus
3: (Petrella et al., 1993)	unknown	severe ID and DD	hypotonia, cryptorchism, coloboma, cataract, seizures, short stature, multiple skeletal anomalies	multiple meningiomas of tentorium cerebelli, and thoracic spine with compression of spinal cord	16 (at autopsy)	none	death at 16 yo from unknown allergic reaction
4: (Kehrer-Sawatzki et al., 1997)	unknown	severe ID and DD, regression starting at 4 yo	delayed speech, dysarthria, parkinsonian symptoms (akinetic episodes), seizures, paranoid/psychotic episodes	bilateral vestibular schwannomas, multiple intracranial meningiomas, thoracic tumor	17	multiple small peripheral subcutaneous nodules on extremities	- surgical removal of infratenorial fibroblastic meningioma and fibrillary neurinoma progression of tumors - post-surgery: worsening of seizures and ultimate tetraparesis
5: (Tsilchorozidou et al., 2004)	unknown	moderate DD and severe ID	large goiter, bilateral hearing loss, delayed speech, dysmorphic facies, pes cavus	bilateral vestibular schwannomas, 4+ extradural extramedullary thoracic spine tumors	20	none	unknown
6: (Tsilchorozidou et al., 2004)	unknown	severe ID and DD	seizures, absent speech, lower extremity lymphedema, pleomorphic leiomyoma	bilateral vestibular schwannomas, multiple meningiomas of falx and middle cranial fossa	52	none	- no surgical management recommended for tumors - progressive catatonic episodes, headaches, status epilepticus leading to death at 52 yo
7: (Tsilchorozidou et al., 2004)	unknown	severe ID and DD	motor and speech delay, dysmorphic facies	bilateral vestibular schwannomas, multiple meningiomas (unknown location)	39	several subcutaneous nodules	unknown
8: (Denayer et al., 2009)	unknown	severe ID and DD	delayed psychomotor development, hypotonia, autistic features, microcephaly, aggressive behavior, dysmorphic facial features, several skeletal anomalies	unilateral vestibular schwannoma (First to demonstrate loss of r (22) and second hit in NF2 in tumor cells)	20	multiple *café-au-lait* macules, several subcutaneous nodules	progressive gait instability, abducens nerve palsy and hydrocephalus leading to surgical removal of schwannoma
9: (Zirn et al., 2012)	*de novo*	severe ID and DD	speech and motor delay, behavior problems, some mild dysmorphic facial features, microcephaly	extramedullary spinal thoracic tumor with compression of cord, intramedullary cervical spinal tumor (tumor with loss of *NF2* and sequence variant on second *NF2* allele)	15	2 *café-au-lait* macules	progressive spastic paresis and gait abnormality leading to surgical removal of thoracic meningioma
10: (Lyons-Warren et al., 2017)**	unknown	mild ID and DD	developmental regression, mild dysmorphic features, delayed speech, loss of fine motor skills, repetitive movement and obsessive behaviors	bilateral vestibular schwannomas, multiple spinal extramedullary tumors	16	several painless growths on arms (likely schwannomas)	progressive seizures, staring spells and change in personality. Surgery not recommended. Started on bevacizumab with improvement in mood

*first description; **diagnosed with PMS.
Abbreviations: ID = intellectual disability, DD = developmental delay, yo = years old.

communication in the PMS population make it difficult to evaluate symptoms such as a back pain and decreased sensation elicited on history and clinical exam, which may suggest presence of a spinal or paraspinal lesion and prompt imaging. Finally, as clinical ocular examinations are also historically difficult in the PMS population, formal ophthalmological assessment with slit-lamp examination should be strongly considered, especially as changes in vision and early cataract development may represent early stigmata of NF2.

5. Conclusion

In this short report we present the largest cohort of individuals with PMS patients and with documented NF2 and add to the growing body of literature describing the association of r (22) and NF2. However, despite the 16% prevalence of NF2 in our sample of individuals with PMS and r (22), this may still be an underestimation of the true prevalence of NF2 given incomplete karyotyping in the larger PMS population as shown in our review of the PMSIR database. Moreover, the average age of individuals in our cohort with a documented r (22) on karyotype was below the average age of tumor presentation and diagnosis of NF2 in individuals with inherited NF2, which may also be influencing the prevalence of this association. Therefore, continued follow-up and appropriate screening of individuals with r (22) over time is essential to determine the true prevalence of NF2 in the PMS population. Importantly, our report supports prior recommendations for conservative tumor screening in individuals with PMS and r (22) including cranial MRI imaging starting at 10 years old and formal clinical examinations with particular attention to skin, eye, hearing, and neurologic systems beginning at two years old. However, further characterization of tumorigenesis and natural history of disease in individuals with NF2 and a r (22) is still needed.

Funding

This study was supported in part by the Greenwood Genetic Center Foundation and the Hope for 22q13 Gala.

CRediT authorship contribution statement

Catherine A. Ziats: Investigation, Data curation, contributed to investigation and data curation, Writing - original draft, contributed to writing- original draft preparation. **Lavanya Jain:** Investigation, Data curation, contributed to investigation and data curation. **Brittany McLarney:** Investigation, Data curation, contributed to investigation and data curation. **Emily Vandenboom:** Investigation, Data curation, contributed to investigation and data curation. **Barbara R. DuPont:** Writing - review & editing, assisted with writing-reviewing and editing. **Curtis Rogers:** Writing - review & editing, assisted with writing-reviewing and editing. **Sara Sarasua:** Writing - review & editing, assisted with writing-reviewing and editing. **Julian Nevado:** Investigation, Data curation, contributed to investigation and data curation. **Emanuela Lucci Cordisco:** Investigation, Data curation, contributed to investigation and data curation. **Katy Phelan:** Writing - review & editing, assisted with writing-reviewing and editing. **Luigi Boccuto:** Investigation, Data curation, contributed to investigation and data curation, Writing - review & editing, assisted with writing-reviewing and editing, Conceptualization, Methodology, contributed to conceptualization and methodology.

Declaration of competing interest

The authors declare no conflict of interest.

Acknowledgements

The authors express their deep gratitude to the families who participated in this study for their kind availability and support and acknowledge the precious help of the family foundations, specifically the Phelan-McDermid syndrome Foundation (PMSF), the Canadian PMS Foundation, the Italian Association for Phelan-McDermid Syndrome (AISPHEM ONLUS), Spanish Association for Phelan-McDermid syndrome, and the Australian PMS Foundation.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ejmg.2020.104042.

References

Ardern-Holmes, S., Fisher, G., North, K., 2017. Neurofibromatosis type 2. J. Child Neurol. 32 (1), 9–22.

Arinami, T., Kondo, I., Hamaguchi, H., Nakajima, S., 1986. Multifocal meningiomas in a patient with a constitutional ring chromosome 22. J. Med. Genet. 23 (2), 178–180.

Bonaglia, M.C., Giorda, R., Beri, S., De Agostini, C., Novara, F., Fichera, M., Grillo, L., Galesi, O., Vetro, A., Ciccone, R., Bonati, M.T., Giglio, S., Guerrini, R., Osimani, S., Marelli, S., Zucca, C., Grasso, R., Borgatti, R., Mani, E., Motta, C., Molteni, M., Romano, C., Greco, D., Reitano, S., Baroncini, A., Lapi, E., Cecconi, A., Arrigo, G., Patricelli, M.G., Pantaleoni, C., D'Arrigo, S., Riva, D., Sciacca, F., Dalla Bernardina, B., Zoccante, L., Darra, F., Termine, C., Maserati, E., Bigoni, S., Priolo, E., Bottani, A., Gimelli, S., Bena, F., Brusco, A., di Gregorio, E., Bagnasco, I., Giussani, U., Nitsch, L., Politi, P., Martinez-Frias, M.L., Martinez-Fernandez, M.L., Martinez Guardia, N., Bremer, A., Anderlid, B.M., Zuffardi, O., 2011. Molecular mechanisms generating and stabilizing terminal 22q13 deletions in 44 subjects with Phelan/McDermid syndrome. PLoS Genet. 7 (7), e1002173.

Denayer, E., Brems, H., de Cock, P., Evans, G.D., Van Calenbergh, F., Bowers, N., Sciot, R., Debiec-Rychter, M., Vermeesch, J.V., Fryns, J.P., Legius, E., 2009. Pathogenesis of vestibular schwannoma in ring chromosome 22. BMC Med. Genet. 10, 97.

De Rubeis, S., Siper, P.M., Durkin, A., Weissman, J., Muratet, F., Halpern, D., Del Pilar Trelles, M., Frank, Y., Lozano, R., Wang, A.T., Holder Jr., J.L., Betancur, C., Buxaum, J.D., Kolevzon, A., 2018. Delineation of the genetic and clinical spectrum of Phelan-McDermid syndrome caused by *SHANK3* point mutations. Mol. Autism. 9, 31.

Droogmans, G., Swillen, A., Van Buggenhout, G., 2020. Deep phenotyping of development, communication and behaviour in phelan-McDermid syndrome. Molecular Syndromology 10 (6), 294–305.

Evans, D.G., 1993. Neurofibromatosis 2. In: Adam, M.P., Ardinger, H.H., Pagon, R.A., Wallace, S.E., Bean, L.J.H., Stephens, K., Amemiya, A. (Eds.), GeneReviews((R)). University of Washington, Seattle University of Washington, Seattle. GeneReviews is a registered trademark of the University of Washington, Seattle. All rights reserved., Seattle (WA).

Evans, D.G., Huson, S.M., Donnai, D., Neary, W., Blair, V., Newton, V., Harris, R., 1992. A clinical study of type 2 neurofibromatosis. Q. J. Med. 84 (304), 603–618.

Fagan, K., Suthers, G.K., Hardacre, G., 1988. Ring chromosome 11 and cafe-au-lait spots. Am. J. Med. Genet. 30 (4), 911–916.

Hunter, A.G., Ray, M., Wang, H.S., Thompson, D.R., 1977. Phenotypic correlations in patients with ring chromosome 22. Clin. Genet. 12 (4), 239–249.

Jeffries, A.R., Curran, S., Elmslie, F., Sharma, A., Wenger, S., Hummel, M., Powell, J., 2005. Molecular and phenotypic characterization of ring chromosome 22. Am. J. Med. Genet. 137 (2), 139–147.

Kehrer-Sawatzki, H., Udart, M., Krone, W., Baden, R., Fahsold, R., Thomas, G., Schmucker, B., Assum, G., 1997. Mutational analysis and expression studies of the neurofibromatosis type 2 (NF2) gene in a patient with a ring chromosome 22 and NF2. Hum. Genet. 100 (1), 67–74.

Kistenmacher, M.L., Punnett, H.H., 1970. Comparative behavior of ring chromosomes. Am. J. Hum. Genet. 22 (3), 304–318.

Knudson Jr., A.G., 1971. Mutation and cancer: statistical study of retinoblastoma. Proc. Natl. Acad. Sci. U.S.A. 68 (4), 820–823.

Lyons-Warren, A.M., Cheung, S.W., Holder Jr., J.L., 2017. Clinical Reasoning: a common cause for Phelan-McDermid syndrome and neurofibromatosis type 2: one ring to bind them. Neurology 89 (17), e205–e209.

Morava, E., Bartsch, O., Czako, M., Frensel, A., Karteszi, J., Kosztolanyi, G.Y., 2003. A girl with cutaneous hyperpigmentation, cafe au lait spots and ring chromosome 15 without significant deletion. Genet. Counsel. 14 (3), 337–342.

Petrella, R., Levine, S., Wilmot, P.L., Ashar, K.D., Casamassima, A.C., Shapiro, L.R., 1993. Multiple meningiomas in a patient with constitutional ring chromosome 22. Am. J. Med. Genet. 47 (2), 184–186.

Phelan, K., McDermid, H.E., 2012. The 22q13.3 deletion syndrome (phelan-McDermid syndrome). Molecular syndromology 2 (3–5), 186–201.

Phelan, K., Rogers, R.C., Boccuto, L., 1993. Phelan-McDermid syndrome. In: Adam, M.P., Ardinger, H.H., Pagon, R.A., Wallace, S.E., Bean, L.J.H., Stephens, K., Amemiya, A. (Eds.), GeneReviews((R)). University of Washington, Seattle University of Washington, Seattle. GeneReviews is a registered trademark of the University of Washington, Seattle. All rights reserved., Seattle (WA.

Ponson, L., Gomot, M., Blanc, R., Barthelemy, C., Roux, S., Munnich, A., Romana, S., Aguillon-Hernandez, N., Malan, V., Bonnet-Brilhault, F., 2018. 22q13 deletion

syndrome: communication disorder or autism? Evidence from a specific clinical and neurophysiological phenotype. Transl. Psychiatry 8 (1), 146.
Tommerup, N., Warburg, M., Gieselmann, V., Hansen, B.R., Koch, J., Petersen, G.B., 1992. Ring chromosome 22 and neurofibromatosis. Clin. Genet. 42 (4), 171–177.
Tsilchorozidou, T., Menko, F.H., Lalloo, F., Kidd, A., De Silva, R., Thomas, H., Smith, P., Malcolmson, A., Dore, J., Madan, K., Brown, A., Yovos, J.G., Tsaligopoulos, M., Vogiatzis, N., Baser, M.E., Wallace, A.J., Evans, D.G., 2004. Constitutional rearrangements of chromosome 22 as a cause of neurofibromatosis 2. J. Med. Genet. 41 (7), 529–534.
Zen, P.R., Pinto, L.L., Graziadio, C., Pereira, V.B., Paskulin, G.A., 2005. Association of microcephaly and cafe-au-lait spots in a patient with ring chromosome 12 syndrome. Clin. Dysmorphol. 14 (3), 141–143.
Zirn, B., Arning, L., Bartels, I., Shoukier, M., Hoffjan, S., Neubauer, B., Hahn, A., 2012. Ring chromosome 22 and neurofibromatosis type II: proof of two-hit model for the loss of the NF2 gene in the development of meningioma. Clin. Genet. 81 (1), 82–87.