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RESEARCH ARTICLE

Methyl-CpG-binding protein 2 gene mutations and its association with epilepsy: a single centre study from the Indian subcontinent

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Abstract. Rett syndrome (RTT) is an X-linked disorder caused by mutations in *MECP2* in majority of cases. It is characterized by arrested development between 6 and 18 months of age, regression of acquired hand skills and speech, stereotypic hand movements, gait abnormalities and seizures. There are a very few studies in India which illustrates mutation spectrum in RTT. None of the studies have correlated seizures with the genotype. This study describes the phenotype and genotype spectrum in children with RTT syndrome and analyses the association of epilepsy with various clinical features and molecular findings. All children with RTT in our cohort had global developmental delay. Genetic diagnosis identified mutations of the *MECP2* in all 25 children where RTT was suspected. We have identified point mutations in 20 patients, one insertion and four deletions by Sanger sequencing, namely c.1164_1207 (44 bp), c.1165_1207 (43 bp), c.1157_1197 (41 bp) del and c.1157_1188 (32 bp). Clinically, none of the patients with deletion had seizures. We identified one novel insertion variant c.337_338 (p.S113Ffs*9). All the deletions were located in the C-terminal region. Majority of the mutations (22/25) were identified in exon 4 which comprised of nonsense and missense types. Screening of hotspot mutations in exon 4 should be the first line evaluation in diagnosis of RTT. Molecular testing could help in specific management of seizures in RTT.

Keywords. *MECP2* gene; seizure; C-terminal deletion; point mutation; exon 4.

Introduction

Rett syndrome (RTT, OMIM: 312750) was first described by Andreas Rett (Amir *et al*. 1999) as an X-linked neurodevelopmental disorder caused by mutations in methyl-CpG-binding protein 2 gene (*MECP2*) in 80% of typical RTT cases. The estimated prevalence is one in 10,000 live female births. Majority of the cases (>99%) are sporadic (Amir and Zoghbi 2000) and characterized by arrest in the development between 6 and 18 months of age, regression of acquired hand skills, loss of acquired speech, stereotypic hand movements, gait abnormalities, microcephaly and seizures (Hagberg *et al*. 1983; Amir *et al*. 1999; Amir and Zoghbi 2000). Bruxism, breathing abnormalities and cold hands and feet are the other key features suggestive of diagnosis (Amir *et al*. 1999; Amir and Zoghbi 2000; Kyle *et al*. 2018). Diagnostic criteria were established for RTT in 2002 (OMIM: 312750) which was revised in 2010 (Neul *et al*. 2010). Initial criteria included four main and seven supportive features. The revised criteria need only four main criteria for the diagnosis of typical RTT, which includes partial or complete loss of purposeful hand skills and language, dyspraxic gait and stereotypic hand wringing movements (Neul *et al*. 2010).

The *MECP2* gene was first described in 1992 (Amir *et al*. 1999). The gene is located on chromosome Xq28 and comprises of four exons and three functionally important domains, namely a methylated-DNA binding domain (MBD; residues 78–163), a transcription repression domain (TRD; residues 206–310) and a C-terminal domain (CTD-alpha; residues 311–355 and CTD-beta; residues 356–486) (Mushtaq *et al*. 2018). *MECP2* encodes 486 amino acids and highly expressed in the neuronal cells of the brain. It binds to methylated CpG as well as nonCG methylated DNA and interacts with Sin3A-histone corepressor complex and histone deacetylases and

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hence removes acetyl groups from histones. These result in the inactivation of chromatin and transcription repression (Nan *et al*. 1998; Laccone *et al*. 2001). *MECP2* dysfunction causes impairment in neurogenesis, neuronal differentiation and maturation. It also affects the connectivity of circuit in the brain, which causes imbalance between excitation and inhibition. This contributes to the neuro developmental defect in RTT (Ip *et al.* 2018).

There are some case reports from India and a few collaborative larger case series from the Indian subcontinent which illustrates the spectrum of *MECP2* mutations in RTT (Das *et al*. 2013; Lallar *et al*. 2018). We retrospectively analysed the data of 25 patients with RTT from a tertiary care centre in south India, who were confirmed by molecular testing. In this study, we aim to describe the phenotypic and genotypic spectrum in children with RTT and determine the association of epilepsy with various clinical features and *MECP2* mutations.

Materials and methods

This retrospective study was approved by the Institutional Review Board of our institution. All children with a clinical suspicion of RTT from the departments of Medical Genetics, Neurological Sciences and Developmental Paediatrics, considered as outpatients during the period from January 2013 to December 2018, were included in the study.

All of them had a mutation screening of *MECP2* gene, as a part of the routine diagnostic evaluation. The procedure is as follows: 2 mL of blood was collected in an EDTA vacutainer tube. Genomic DNA was extracted by Qiagen Mini kit method (QIAamp DNA minikit; Qiagen, Hilden, Germany) and then quantified using Nanodrop (Nanodrop 2000c Spectrophotometer, Thermo Fischer Scientific, Asheville). All the four exons of *MECP2* gene were amplified by uniplex PCR (GoTaq green mastermix, Promega) followed by Sanger sequencing in ABI 3500 sequencer (ABI 3500 Genetic Analyser, Applied Biosystems Thermo Fischer Scientific). All the identified mutations were confirmed with the Human Genome Mutation Database (GRCh37 assembly in Ensembl)/Clinvar to confirm the novelty of variants. Mutations were described according to the HGVS guidelines. All the 25 children were confirmed to have RTT.

The clinical and genetic data of these children were retrieved from the hospital's electronic database. Statistical analysis was performed based on 2000 bootstrap replicates, as it is a very rare genetic disease with a small sample size (Lin *et al*. 2015).

Results

Clinical phenotype

Among the 25 children diagnosed with RTT, 56% had symptoms from as early as 1–2 years of age. All the children with RTT had global developmental delay. The features of neuroregression of either motor skills or language or both (22/25), loss of hand grip (22/25), stereotypic hand wringing movements (24/25), gait ataxia (23/25) and bruxism (13/24) were frequently recorded. Epilepsy was documented in 14/25 patients. Generalized tonic-clonic seizures being the most common semiology and two patients were reported with absence of seizures. Key features such as sleep disturbances and laughter spells were noticed in 9/19 and 7/19 patients, respectively. Features of hyperventilation were observed in 6/21 patients.

Genotype findings

Genetic testing identified mutations in all 25 children with a clinical diagnosis of RTT. Point mutations were reported in 20/25 patients. The remaining four patients had deletions in the C-terminal region and only one patient had an insertion. Maximum number of mutations were detected in exon 4. The spectrum of mutations in our cohort is shown in figure 1. In four patients, four different deletions were identified, namely 44-bp deletion (c.1164_1207del, p.Pro389*), 43-bp deletion (c.1165_1207del, p.P389Ifs*6), 41-bp deletion (c.1157_1197del, p.L386Hfs*5) and 32-bp deletion (c.1157_1188del, p.L386Rfs*8). Nonsense and missense mutations were identified in 11 and 9 cases, respectively, and were observed to be the most common type of mutations. One novel mutation, c.337_338insT (p.S113Ffs*9) was identified in exon 3 which resulted in a frame shift and premature truncation of protein 9 amino acids downstream to codon 113 (figures 1–2).

Genotype–phenotype correlation

Of the three patients with R133C mutation, two had bruxism and behavioural abnormalities which manifested in the form of compulsive hand biting. There was no breathing abnormality in any of these patients. One of the patients did not have repetitive hand wringing movements, and was initially suspected to be a case of Angelman syndrome but subsequent DNA methylation test disproved the diagnosis. Computerized tomography (CT) brain of this child showed mild cerebral activity with the prominence of cerebrospinal fluid (CSF) spaces.

Of the three patients with R306C mutation, one had delay in developmental milestones since the onset of disease. All of them had bruxism and two of them had behavioural abnormalities such as aggressiveness apart from self-biting. Two of these patients had features of hyperventilation.

Of the three patients with R294* mutation, one patient did not have neuroregression but there was developmental delay since infancy and also mild hearing loss. One of these patients had breath holding spells and another had kyphosis, and a short neck. Three patients were identified to have R255* mutation. Two of them had bruxism and one had

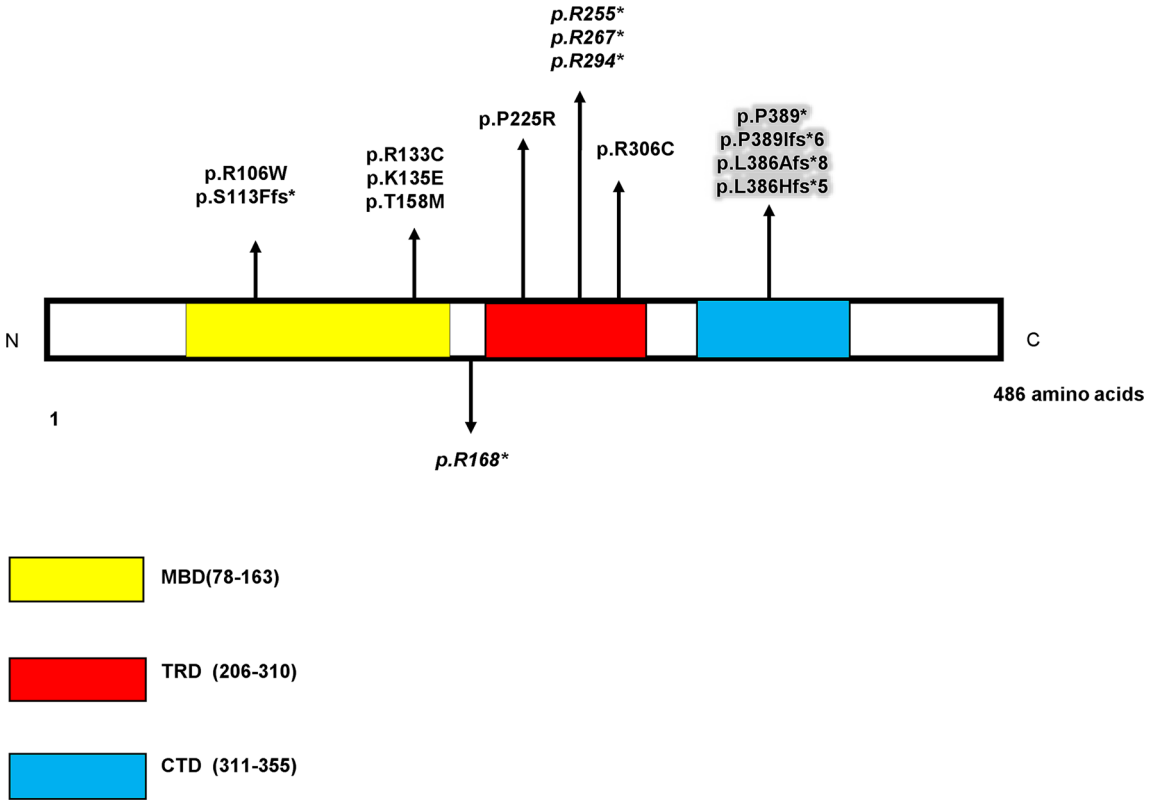

Figure 1. Schematic representation of mutations identified in MECP2 protein product. Domains are shown in different colours based on the legend. Types of mutations are represented as follows: missense, bold; nonsense, italic; deletion, shaded; insertion, normal text. Mutations which are part of domain are mentioned above and the one which is not part of any domain is labelled below the diagram.

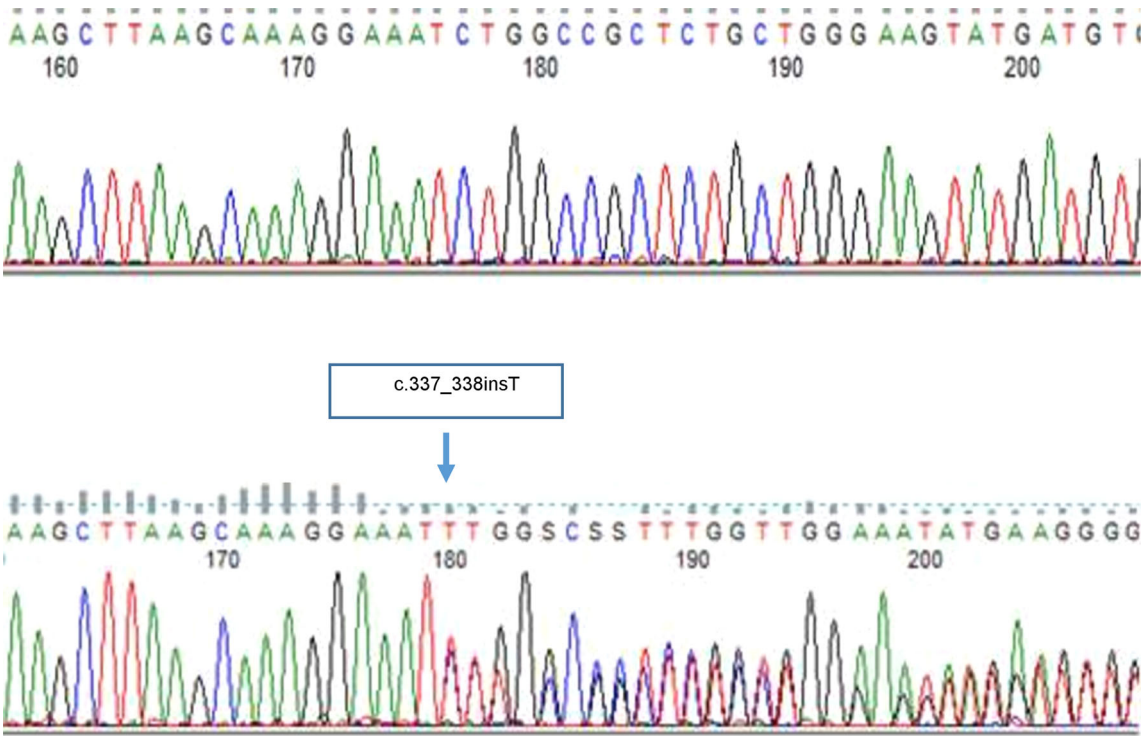

Figure 2. Upper panel showing normal sequence and lower panel showing the novel mutation detected in one patient.

kyphosis. Two patients had R106W mutation, of which one had kyphosis and another one had bruxism. One patient with T158M mutation had bruxism and small hands and feet. Another patient with P225L had cold hands and feet and mild scoliosis.

There were four patients identified with deletion mutations in the C-terminal region of *MECP2*. These patients presented with behavioural abnormality in the form of hyperactivity, aggressiveness or self-injury. None of them had clinical seizures irrespective of EEG findings. A novel insertion mutation c.337_338insT (p.S113Ffs*9) was identified in a patient who had a classic phenotype.

Association of seizures with molecular findings

We have described the semiology of seizures and EEG findings with identified *MECP2* mutation (table 1). In the bivariate analysis, those children who had initial normal development prior to neuro-regression had a seven times odds (OR: 7.43; 95% CI = 1.28 – 43.11) of having epileptic activity as compared to those who did not have an initial normal development. Also, genotypically, those who were found to have point mutation had a seven times odds (OR: 7.43; 95% CI = 1.28 – 43.11) of having epileptic activity as compared to those who had insertions or deletions at the C-terminal region. These have been elaborated in table 2.

Discussion

This study highlights the common clinical presentations and mutation spectrum in a series of children with RTT from a single centre of south India. Based on the revised Rett criteria by Hadgeberg, clinical features such as absent or delayed speech, neuroregression, gait abnormality and repetitive stereotypic behaviour were identified to be the hallmark features of the disease (Neul *et al*. 2010; Pintaudi *et al*. 2010).

We have observed that seizures are one of the core symptoms of RTT and the type of mutations in *MECP2* gene has a distinct impact on epilepsy. With regards to seizures, we observed two groups of patients, one with clinical seizure with associated EEG abnormalities (9/10) and another group of patients without clinical seizures but with abnormal EEG. It is important to differentiate true epilepsy from nonepilepsy as reported by Tarquinio *et al.* (2016) who stated that EEG is usually abnormal in RTT after three years of age. Hence, antiepileptic medicines should be started only after confirmation of clinical seizures or correlation with video EEG monitoring (d'Orsi *et al*. 2012; Tarquinio *et al*. 2016).

Among those patients who did not have a clinical seizure, four of them had a C-terminal deletion. Our study showed a seven-fold increase in the occurrence of epilepsy if there was a point mutation, as compared to a C-terminal deletion or insertion. The literature shows a lower prevalence of epilepsy (Hoffbuhr *et al.* 2001; Jian *et al*. 2006; Pintaudi *et al*. 2010; Cardoza *et al.* 2011; Bao *et al*. 2013; Nissenkorn *et al.* 2015; Operto *et al*. 2019), as well as less severe phenotype in the C-terminal deletion (Hoffbuhr *et al.* 2001; Bao *et al*. 2013; Nissenkorn *et al.* 2015; Operto *et al*. 2019), as compared to point mutations found in our cohort. Probably, C-terminal deletions alter stability and not the function of the MECP2 protein (Guy *et al*. 2018).

One patient with the C-terminal insertion mutation had generalized tonic seizures from the age of 1 year 6 months. Seizures were controlled and EEG became normal after one year of valproate therapy. Patients with R294X mutation had no seizures till 5 years of age concordant with a previous

Table 1. Classification of mutations based on seizure semiology.

	Epileptic spasms (n = 1)	GTCS (n = 3)	GTS (n = 3)	CPS (n = 1)	GTCS and absence seizures (n = 1)	Absence seizures (n = 1)	No seizures (n = 15)
Age of onset in years	3	0.9–5	1.6–5	1.9	3	1.8	NA
EEG findings	Abnormal	Abnormal	Normal EEG-1 Abnormal-2	Abnormal	Abnormal	Abnormal	Normal EEG-3 Abnormal-10 Not done-2
Variants	p.R106W	p.R225X p.R294* p.R133C	p.P225R p.S113Ffs* p.R306C	p.R133C	p.T158M	p.R168*	p.R168*-3 p.R294*-2 p.R255*-2 p.R306C-2 p.R133C p.R106W p.P389* p.L386Hfs*5 p.P389Ifs*6s p.L386Afs*8

GTCS, generalized tonic-clonic seizures; GTS, generalized tonic seizures; CPS, complex partial seizures; EEG, electroencephalography.

Table 2. Statistical analysis for association of seizures with various clinical features and identified mutations.

Variables	Groups	Presence of epilepsy (n = 14)	Absence of epilepsy (n = 11)	P value	Odds ratio	95% CI
Age of onset (years)	Toddlers (n = 15)	60% (9)	40% (6)	1.00	1.12	0.18–6.93
	Infants (n = 7)	57.1% (4)	42.9% (3)			
Initial normal development	Yes (n = 20)	65% (13)	35% (7)	0.025*	7.43	1.28–43.11
	No (n = 5)	20% (1)	80% (4)			
Neuro regression	Yes (n = 22)	59.1% (13)	40.9% (9)	0.55	3.25	0.25–41.91
	No (n = 3)	33.3% (1)	66.7% (2)			
Sleep disturbances	Yes (n = 8)	75% (6)	25% (2)	1.00	1.71	0.23–12.89
	No (n = 11)	63.6% (7)	36.4% (4)			
Laughter spells	Yes (n = 8)	62.5% (5)	37.5% (3)	0.65	2.00	0.31–12.8
	No (n = 11)	45.5% (5)	54.5% (6)			
Breath holding spells	Yes (n = 6)	66.7% (4)	33.3% (2)	0.36	3.00	0.41–21.88
	No (n = 15)	40% (6)	60% (9)			
Bruxism	Yes (n = 13)	61.5% (8)	38.5% (5)	1.00	1.33	0.26–6.80
	No (n = 11)	54.5% (6)	45.5% (5)			
Type of mutation	Point mutation (n = 20)	65% (13)	35% (7)	0.025*	7.43	1.28–43.11
	Insertion/deletion (n = 5)	20% (1)	80% (4)			

*Based on the bootstrap resampling at 2000 times, the P value is significant (0.025).

study (Jian *et al*. 2006). Besides, only one of the four patients with R168X mutation had seizures since 4 years of age similar to the previous reports (Jian *et al.* 2006). These mutations are not part of the MBD and TBD. More functional studies are required to confirm whether these domains play a role in epileptogenesis. The role of CTD region mutation in RTT also warrants functional evidence.

Seizures are responsive to antiepileptic medicines in most of the patients. However, our patient having T158M mutation showed drug resistance correlating with previous studies (Bao *et al*. 2013; Operto *et al*. 2019). Nissenkorn *et al*. (2015) describes T158M as having statistically significant risk of severe epilepsy compared to the C-terminal deletion and requires aggressive management.

We noticed that 44% of mutations were missense and 33% were nonsense, as reported previously in the literature (Laccone *et al*. 2001; Lee *et al*. 2001). In our study, R168X, R306C, R294X, R133C, R255X were the most frequent point mutations and proposed to be hotspots as missense and nonsense mutations as described (De Bona *et al*. 2000; Dragich *et al*. 2000; Xiang 2000; Laccone *et al*. 2001; Lee *et al*. 2001; Monrós *et al*. 2001; Neul *et al*. 2008; Bao *et al*. 2013; Lallar *et al*. 2018). The recurrent mutations identified in the Indian subcontinent were similar to those in other populations. R168X was the most common mutation among them which is shown to be more prevalent in America, United Kingdom, Germany, Italy, Sweden and Spain (Cheadle 2000; De Bona *et al*. 2000; Xiang 2000; Laccone *et al*. 2001; Monrós *et al*. 2001; Neul *et al*. 2008). Even though this mutation is not part of any functional domain, it is reported to lead to decreased synthesis of functional protein.

There are 16% deletion mutations in the C-terminal region, and among them, 7% were frame-shift mutations correlating with previous studies (Amir and Zoghbi 2000; Dragich *et al*. 2000; Laccone *et al*. 2001; Lee *et al*. 2001; Neul *et al*. 2008; Lyst and Bird 2015; Lallar *et al*. 2018). Deletion mutations are clustered between nucleotide positions 1140 and 1197 with nucleotide position 1154 being the proximal breakpoint of the deletion prone region. It has been previously described that this region contains multiple palindromic repeats, which may lead to deletion mutations due to slippage of DNA polymerase (Cheadle 2000; De Bona *et al*. 2000; Dragich *et al*. 2000; Laccone *et al*. 2001; Lee *et al*. 2001; Miltenberger-Miltenyi and Laccone 2003).

Most of the patients had mutations (22/25) in exon 4 and a few of them had mutations in exon 3 (3/25). This is similar to the previously reported mutation in India, as well as other populations (Laccone *et al*. 2001; Das *et al*. 2013; Lallar *et al*. 2018). Exon 4 is likely to be more susceptible to mutation in RTT patients (Laccone *et al*. 2001). This shows exon 4 as a mutation hotspot which might facilitate a cost-effective approach for the diagnosis of *MECP2* gene in RTT patients (Das *et al*. 2013).

We have found that nonsense mutations were prevalent within TRD or in the region between MBD and TRD. Missense mutations were located more commonly in MBD than TRD, similar to previously described literature (Cheadle 2000; Xiang 2000; Laccone *et al*. 2001; Miltenberger-Miltenyi and Laccone 2003; Das *et al*. 2013). We observed 70% of mutation in one of the CpG hotspots, particularly C>T transitions. This is correlating with the literature described (Cheadle 2000; De Bona *et al*. 2000; Xiang 2000; Laccone *et al*. 2001; Lee *et al*. 2001; Miltenberger-Miltenyi and Laccone 2003; Das *et al*. 2013).

The novel mutation detected in this study is a null variant (frame shift) affecting the *MECP2* gene, which is a known

mechanism of disease. In addition, insertion of missense residues following a frame shift might cause improper folding or disrupt the stability of *MECP2* (Lyst and Bird 2015). This position is conserved which is indicated by genomic evolutionary rate profiling (GERP) score >4. This finding directs that the novel variant is pathogenic as per ACMG guidelines. This study shows that there are a number of patients with similar mutations but variation in phenotype indirectly indicates no genotype–phenotype correlation in our study. We had only girl children with RTT syndrome, however recently, male patients with *MECP2* mutations had also been reported (Soffer and Sidlow 2016).

In conclusion, in our series of children identified with RTT, majority of the mutations were seen in exon 4 of the *MECP2* gene. Clinical seizures were absent in patients with the C-terminal deletions. However, large cohorts of patients are required to conclude this finding derived from the present study. This study reaffirms that prioritizing targeted sequencing of exon 4 of *MECP2* gene can help in the effective identification of mutation in majority of the cases. RTT syndrome has a characteristic phenotype; however, molecular analysis would help the clinicians for prognostication of the outcome and plan specific management.

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References

Amir R. E. and Zoghbi H. Y. 2000 Rett syndrome: Methyl-CpG-binding protein 2 mutations and phenotype-genotype correlations. *Am. J. Med. Genet*. **97**, 147–152.

Amir R. E., Van den Veyver I. B., Wan M., Tran C. Q., Francke U. and Zoghbi H. Y. 1999 Rett syndrome is caused by mutations in X-linked MECP2, encoding methyl-CpG-binding protein 2. *Nat. Genet.* **23,** 185–188.

Bao X., Downs J., Wong K., Williams S. and Leonard H. 2013 Using a large international sample to investigate epilepsy in Rett syndrome. *Dev. Med. Child Neurol*. **55**, 553-558.

Cardoza B., Clarke A., Wilcox J., Gibbon F., Smith P., Archer H. *et al.* 2011 Epilepsy in Rett syndrome: Association between phenotype and genotype, and implications for practice. Seizure: *J. Br. Epilepsy Assoc.* **20**, 646–649.

Cheadle J. P. 2000 Long-read sequence analysis of the MECP2 gene in Rett syndrome patients: correlation of disease severity with mutation type and location. *Hum. Mol. Genet*. **9**, 1119–1129.

d'Orsi G., Trivisano M., Luisi C., Demaio V., Claudio M., Pascarella M *et al*. 2012 Epileptic seizures, movement disorders, and breathing disturbances in Rett syndrome: diagnostic relevance of video-polygraphy. *Epilepsy Behav.* **25**, 401–407.

Das D. K., Raha S., Sanghavi D., Maitra A. and Udani V. 2013 Spectrum of MECP2 gene mutations in a cohort of Indian patients with Rett syndrome: report of two novel mutations. *Gene* **515**, 78–83.

De Bona C., Zappella M., Hayek G., Meloni I., Vitelli F., Bruttini M *et al*. 2000 Preserved speech variant is allelic of classic Rett syndrome. *Eur. J. Hum. Genet*. **8**, 325–330.

Dragich J., Houwink-Manville I. and Schanen C. 2000 Rett syndrome: a surprising result of mutation in MECP2. *Hum. Mol. Genet*. **9**, 2365–2375.

Guy J., Alexander-Howden B., FitzPatrick L., DeSousa D., Koerner M. V., Selfridge J. *et al*. 2018 A mutation-led search for novel functional domains in MeCP2. *Hum. Mol. Genet.* **27**, 2531–2545.

Hagberg B., Aicardi J., Dias K. and Ramos O. 1983 A progressive syndrome of autism, dementia, ataxia, and loss of purposeful hand use in girls: Rett's syndrome: report of 35 cases. *Ann. Neurol*. **14**, 471–479.

Hoffbuhr K., Devaney J. M., LaFleur B., Sirianni N., Scacheri C., Giron J. *et al*. 2001 MeCP2 mutations in children with and without the phenotype of Rett syndrome. *Neurology* **56**, 1486–1495.

Ip J. P. K., Mellios N. and Sur M. 2018 Rett syndrome: Insights into genetic, molecular and circuit mechanisms. *Nat. Rev. Neurosci*. **19**, 368–382.

Jian L., Nagarajan L., Klerk N., Ravine D., Bower C., Anderson A. *et al*. 2006 Predictors of seizure onset in Rett syndrome. *J. Pediatr.* **149**, 542–547.

Kyle S. M., Vashi N. and Justice M. J. 2018 Rett syndrome: A neurological disorder with metabolic components. *Open Biol*. **8**, 170216.

Laccone F., Huppke P., Hanefeld F. and Meins M. 2001 Mutation spectrum in patients with Rett syndrome in the German population: Evidence of hot spot regions. *Hum. Mutat*. **17**, 183–190.

Lallar M., Rai A., Srivastava P., Mandal K., Gupta N., Kabra M *et al*. 2018 Molecular testing of MECP2 gene in Rett syndrome phenotypes in Indian girls. *Indian J. Pediatr.* **55**, 474–477.

Lee S. S. J., Wan M. and Francke U. 2001 Spectrum of MECP2 mutations in Rett syndrome. *Brain Dev.* **23** suppl. **1**, S138–S143.

Lin J.-J., Chang C.-H. and Pal N. 2015 A revisit to contingency table and tests of independence: Bootstrap is preferred to Chi-square approximations as well as Fisher's exact test. *J. Biopharm. Stat.* **25**, 438–458.

Lyst M. J. and Bird A. 2015 Rett syndrome: a complex disorder with simple roots. *Nat. Rev. Genet.* **16**, 261–275.

Miltenberger-Miltenyi G. and Laccone F. 2003 Mutations and polymorphisms in the human methyl CpG-binding protein MECP2. *Hum. Mutat*. **22**, 107–115.

Monrós E., Armstrong J., Aibar E., Poo P., Canós I. and Pineda M. 2001 Rett syndrome in Spain: Mutation analysis and clinical correlations. *Brain Dev.* **23**, S251–S253.

Mushtaq A. U., Lee Y., Hwang E., Bang J. K., Hong E., Byun Y *et al*. 2018 Biophysical characterization of the basic cluster in the transcription repression domain of human MeCP2 with AT-rich DNA. *Biochem. Biophys. Res. Commun*. **495**, 145–150.

Nan X., Ng H. H., Johnson C. A., Laherty C. D., Turner B. M., Eisenman R. N. *et al*. 1998 Transcriptional repression by the methyl-CpG-binding protein MeCP2 involves a histone deacetylase complex. *Nature* **393**, 386–389.

Neul J. L., Fang P., Barrish J., Lane J., Caeg E. B., Smith E. O. *et al.* 2008 Specific mutations in methyl-CpG-binding protein 2 confer different severity in Rett syndrome. *Neurology* **70**, 1313–1321.

Neul J. L., Kaufmann W. E., Glaze D. G., Christodoulou J., Clarke A. J., Bahi-Buisson N. *et al*. 2010 Rett syndrome: revised diagnostic criteria and nomenclature. *Ann. Neurol*. **68**, 944–950.

Nissenkorn A., Levy-Drummer R. S., Bondi O., Renieri A., Villard L., Mari F. *et al.* 2015 Epilepsy in Rett syndrome-Lessons from the Rett networked database. *Epilepsia* **56**, 569–576.

Operto F. F., Mazza R., Pastorino G. M. G., Verrotti A. and Coppola G. 2019 Epilepsy and genetic in Rett syndrome: a review. *Brain Behav.* **9**, e01250.

Pintaudi M., Calevo M., Vignoli A., Parodi E., Aiello F., Baglietto M. *et al*. 2010 Epilepsy in Rett syndrome: Clinical and genetic features. *Epilepsy Behav.* **19**, 296–300.

Soffer O. D. and Sidlow R. 2016 A rare MeCP2_e1 mutation first described in a male patient with severe neonatal encephalopathy. *Am. J. Med. Genet. Part A.* **170**, 1881–1883.

Tarquinio D. C., Hou W., Berg A., Kaufmann W. E., Lane J. B., Skinner S. A. *et al*. 2016 Longitudinal course of epilepsy in Rett syndrome and related disorders. *Brain* **140**, 306–318.

Xiang F. 2000 Mutation screening in Rett syndrome patients. *J. Med. Genet*. **37**, 250–255.

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