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Evaluation of seizure semiology, genetics, magnetic resonance imaging, and electroencephalogram findings in children with Rett syndrome: A multicenter retrospective study

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ABSTRACT

Objectives: This study aimed to evaluate seizure semiology, electroencephalogram (EEG), magnetic resonance imaging (MRI), and genetic findings, as well as treatment choices in Rett syndrome (RTT).
Methods: A retrospective analysis was conducted on one hundred and twenty cases diagnosed with RTT with a genetic mutation. Data were obtained from nine participating centers.
Results: In this study, 93.3 % of patients were female, with typical RTT found in 70 % of cases. Genetic etiology revealed MECP2, FoxG1, and CDKL5 in 93.8 %, 2.7 %, and 1.8 % of cases, respectively. Atypical RTT clinics were observed in 50 % of male cases, with the first EEG being normal in atypical RTT cases ($p = 0.01$). Generalized tonic-clonic and myoclonic epilepsy were the most common seizure semiologies, while absence and focal

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epilepsy were less prevalent. Valproate, levetiracetam, lamotrigine, and clobazam were the most commonly used antiepileptic drugs, affecting the severity and frequency of seizures ($p = 0.015$, $p=<0.001$, $p = 0.022$, and $p=<0.001$, respectively). No significant differences were observed in EEG findings. The initiation of anti-seizure medications significantly altered seizure characteristics (Table 4). A ketogenic diet and vagal nerve stimulation (VNS) correlated with a 50 % improvement in cognitive function, while steroid treatment showed a 60 % improvement. Remarkably, seizures were substantially reduced after VNS application.

Conclusion: This study underscores the importance of genetic diagnosis in RTT cases with a clinical diagnosis. These preliminary results will be further validated with the inclusion of clinically diagnosed RTT cases in our ongoing study.

1. Introduction

Rett syndrome (RTT), first described by Andreas Rett in 1966, stands as a complex neurodevelopmental genetic disorder (Bianco and Rota, 2018). The incidence of RTT was approximately 1 in 10.000, primarily affecting females (Lucariello et al., 2016) The identification of mutations in the X-linked methyl-CpG binding protein gene 2 (MECP2) by Amir et al. in 1999 marked a pivotal moment in understanding the underlying genetic basis of RTT (Amir et al., 1999). It has been observed that the clinical manifestations of genes detected other than MECP2 are different. In patients with cyclin-dependent kinase-like 5 (CDKL5) mutations, early-onset seizures (usually around 3–5 months), severe intellectual disability, and marked motor developmental delay have been identified (Evans et al., 2005). The definition of the congenital variant of RTT has been associated with mutations in the Forkhead box protein G1 (FOXG1) gene in the literature, and agenesis or hypoplasia of the corpus callosum are common features observed (Ariani et al.,2008)

In 1985, Hanefeld classified the progression of the disease from Stage I to Stage IV (Gold et al., 2018). Initially, patients exhibit apparently normal development, but between 6 and 18 months of age, they undergo a period of developmental stagnation (Stage I), failing to meet mental, cognitive, and motor milestones. During this phase, head circumference growth decelerates. Stage II is characterized by rapid developmental regression, leading to the loss of purposeful hand movements and verbal skills. Microcephaly worsens, and breathing irregularities and seizures may arise. Stage III represents a pseudo-stationary plateau, with mild cognitive interest recovery but severe diminishment of purposeful hand and body movements. In Stage IV, motor deterioration occurs, often lasting for decades, rendering many patients dependent on wheelchairs and/or gastrostomy tubes (Dolce et al., 2013; Kyle et al., 2018).

Typical RTT features include the loss of spoken language and hand skills, including distinctive hand stereotypies, and ambulation problems (Bianco and Rota, 2018; Neul, 2019). A period of stabilization often follows regression, and in some cases, improvement may even occur. The diagnostic criteria for atypical RTT necessitate the presence of regression along with 2 of the 4 primary criteria, in addition to meeting 5 out of the 11 supportive criteria (Neul et al., 2010). For understanding the classification of RTT, figure has been presented below including the mentioned criteria for diagnosis (Neul et al., 2010; Chahil and Bollu, 2023). Beyond its core symptoms, RTT is frequently accompanied by comorbidities such as epilepsy, necessitating a comprehensive approach to its management (Dolce et al., 2013).

As pharmacological and genetic treatments emerge, rehabilitation research becomes crucial to optimize functional abilities, especially with the advent of gene therapies and disease-modifying treatments for RTT (Fabio et al., 2014; Lim et al., 2020).

This study, conducted across nine centers, seeks to delve into the multifaceted dimensions of RTT, particularly focusing on seizure semiology, electroencephalogram (EEG) patterns, magnetic resonance imaging (MRI) findings, and genetic mutations. We anticipate that this research will significantly contribute to the Rett syndrome database in our country, shedding light on the frequency of genetic diversity and its correlation with the clinical stage, seizure.

2. Materials and methods

Patient files from nine selected centers, approved by the ethics committee, were retrospectively examined between December 2021 and February 2022. This study aimed to collect information on various aspects of patient management, including treatment modalities (antiepileptic medication, steroids, vagus nerve stimulation, ketogenic diet, resective surgery), clinical findings, identified genetic mutations, electroencephalography (EEG), magnetic resonance imaging (MRI), seizure semiology, frequency, and other relevant data.

Approval for the study was obtained from the ethics committee of Karadeniz Technical University Faculty of Medicine (approval number: 2021/382) prior to initiation.

In this study, the classification of statistical data based on genetic mutation type is planned. The MECP2 gene mutation will constitute one group, while other gene mutations (FOXG1, CDKL5) will be planned as the second group. The comparison of clinical data, clinical stages, EEG, and MRI findings is aimed according to mutation groups.

All data of cases followed with the diagnosis of RTT syndrome will be collected from centers in a standardized form by conducting a literature review. A detailed clinical classification and staging are indicated in the form.

The data were classified according to age as 0–2, 2–4, 4–10 and over 10 years old.

In the study, EEG findings and changes in seizure frequency and intensity are also classified according to the criteria defined in the form. Pathological epileptiform activities (EA) recorded every 10 seconds in the EEG trace taken for twenty minutes will be collected retrospectively from centers, and the average of these identified EAs will be calculated. Comparison of these data before and after treatment is planned.

Seizure durations and frequencies will be obtained from file information recorded in all centers, and data before and after treatment will be compared. Seizure frequency will be classified as weekly, monthly, or cases with no seizures.

Cognitive assessment is planned to be conducted by comparing the results of cognitive tests administered to patients before and after treatment. It is planned not to include the data of patients who do not have these test results in their files in the study.

Statistical analysis was performed using SPSS 26.0. Descriptive statistics were applied to evaluate the results, presenting numbers, percentages, mean, standard deviation, median, and minimum-maximum values for categorical variables. For numerical variables, mean, standard deviation, median, and minimum-maximum values were provided. The normal distribution of data was assessed using the Kolmogorov-Smirnov and Shapiro-Wilk tests.

For measures conforming to a normal distribution, the independent t-test or ANOVA was utilized, while the Mann-Whitney U test, Wilcoxon test, or Kruskal-Wallis Analysis of Variance was applied for measures deviating from normal distribution. The McNemar test was employed to compare parameters before and after antiepileptic drug use. Pearson and Spearman correlation tests were conducted to identify correlations between measurement variables. The significance level for all statistical analyses was set at $p<0.05$.

3. Results

This study included 120 cases diagnosed with Rett syndrome genetically and followed up at 9 centers in Turkey, with 93.3 % of them being female. The most common identified mutation was MECP2 (n:111). The other identified genetic mutations were FOXG1 (n:6) and CDKL5 (n:3). In 75 % of the 8 male RTT cases, MECP2 mutation was detected, while FOXG1 mutation was found in 25 %, and no case with CDKL5 mutation was identified among males. MECP2 mutation was detected in 72 % of 84 cases with typical RTT, and 71.4 % of these cases were female. While 50 % of males had atypical RTT, 28.6 % of females had atypical RTT, but the difference was not statistically significant (p= 0.239). The incidence of RTT clinical stages is as follows: stage 1 is 12.5 %, stage 2 is 42.5 %, stage 3 is 29.2 %, and stage 4 was found to be 15.8 %.

The average age of the patients participating in the study group was 36.56 months (ranging from 0 to 192 months). While 42.5 % of the patients were 2–4 years old, 12.5 % of the cases were under 2 years of age (p=0.209). It was found that 35.7 % of patients over the age of four were admitted to the hospital with generalized tonic-clonic seizures (p=0.06). While the seizure frequency of patients between the ages of 2 and 4 was once a year, it was determined that the seizure frequency of patients over the age of 4 was at least once a month. When the cases with normal initial EEG findings were classified according to age, it was seen that 40 % of the cases were between the ages of 2–10.

Analyzing the seizure semiology in the cases, generalized tonic-clonic (23.4 %), myoclonic (13.5 %), and focal onset aware (14 %) seizures were the most frequently observed types, while behavior arrest (7.2 %) and generalized atonic seizures (1.8 %) were the least common. The median age at the onset of the initial seizure was 24. Upon analyzing the seizure frequency in patients prior to drug treatment, it was observed that 27.5 % of individuals experienced no seizures, whereas 19.2 % had seizures on a daily basis.

The most common clinical symptoms and findings in the study are pervasive developmental delay (96.7 %), decreased interest in the environment (93.3 %), mental retardation (91.7 %), language loss (90 %), autistic symptoms (85 %).,8), stereotyped movements (81.7 %) and microcephaly (77.5 %), while hepatosplenomegaly and prolonged QTc were the least detected manifestations. Change in sweating, hepatosplenomegaly, gastroesophageal reflux, prolonged QTc were among the findings seen only in cases with MECP2 mutation. Comparing to genetic mutations, none of the clinical manifestations showed a statistically significant difference. The clinical features identified in patients diagnosed with RTT are extensively detailed in Table 1.

The specified features of typical RTT, including speech regression, stereotypical hand movements, disturbances in gait and posture while walking, and loss of manual skills, were observed in over 80 % of patients and were these were the reasons for referrals to the centers. The clinical features specified for the typical RTT cases have been detailed in Table 2 with no significant differences according to genetic mutations. Gender, clinical type, clinical stage, consanguinity, mutation type, and changes in EEG were compared according to the presence of seizures, and no statistically significant differences were found. The data is presented in Table 3.

The first EEG finding was normal in 52.2 % of cases, with atypical RTT, while 24.7 % of cases with abnormal findings had atypical RTT, and the difference was found to be statistically significant (p=0.010). Of the 6 cases with burst-suppression detected in EEG, 66.7 % were typical RTT. MECP mutations were present in 87 % of cases with normal initial EEG findings. In cases with identified FOX mutations, all had normal initial EEG findings, while in cases with CDKL5 mutations, all had pathological initial EEG results. Among cases with EEG findings, burst suppression was observed in 5 %, and 69.2 % showed epileptiform activity (46 cases generalized, 37 cases multifocal, 15 cases focal), while 31.7 % of cases exhibited a slow background EEG rhythm.

Detailed examination of the MRI findings revealed that the most frequently encountered pathological findings were cerebral atrophy (17.1 %), mesial temporal sclerosis (6.3 %), thinning of the corpus callosum (3.6 %), cerebellar atrophy (3.6 %), regression in myelination (2.7 %), and perviventricular enlargament (2.7 %), in that order. The thinning of the corpus callosum (n:1 in CDKL5, n:2 in FOXG1) and regression on myelination (n:1 in CDKL5, n:2 in FOXG1 were common in other genetic mutations than in MECP2 mutations, and the difference

Table 1
The clinical manifestations identified in patients diagnosed with RTT based on identified genetic mutations.

Clinical manifestation	Cases with MECP2 mutation	Cases with the other genetic mutations (CDKL5, FOXG1)	Total case	p-value
Microcephaly	85 (76.6)	8 (88.9)	93 (77.5)	,682
Dysmorphic face	32 (28.8.)	3 (33.3)	35 (29.2)	,719
Pervasive developmental delay	108 (97.3)	8 (88.9)	116 (96.7)	,271
Mental retardation	102 (91.9)	8 (88.9)	110 (91.7)	,556
Decreased interest	103 (92.8)	9 (100)	112 (93.3)	1000
Loss of language	100 (90.1)	8 (88.9)	108 (90.0)	1000
Autonomic dysfunction	32 (28.8)	1 (11.1)	33 (27.5)	,441
Change in sweating	21 (18.9)		21 (17.5)	,357
Difficulty swallowing and chewing	36 (32.4)	4 (44.4)	40 (33.3)	,479
Malnutrition	54 (48.6)	4 (44.4)	58 (48.3)	1000
İncreased salivation	49 (44.1)	4 (44.4)	53 (44.2)	1000
Hepatosplenomegaly	3 (2.7)		3 (2.5)	1000
Constipation	48 (43.2)	4 (44.4)	52 (43.3)	1000
Gastroesophageal reflux	19 (17.1)		19 (15.8)	,352
Tachypnea	33 (29.7)	4 (44.4)	37 (30.8)	,455
Apnea	34 (30.6)	5 (55.6)	39 (32.5)	,148
Hyperventilation	31 (27.9)	5 (55.6)	36 (30)	,126
Prolonged qtc	1(0.9)		1 (0.8)	1000
Sleep disorders	77 (69.4)	4 (44.4)	81 (67.5)	,148
Meaningless screams	59 (53.2)	5 (55.6)	64 (53.3)	1000
Autistic symptoms	95 (85.6)	8 (88.9)	103 (85.8)	1000
Hypotonia	56 (50.5)	4 (44.9)	60 (50)	1000
Spasticity	61 (55)	3 (33.3)	64 (53.3)	,301
Ataxic walking	59 (53.2)	4 (44.4)	63 (52.5)	,734
Myoclonus	66 (59.5)	5 (55.6)	71 (59.2)	1000
Choreiform movements	72 (64.9)	5 (55.6)	77 (64.2)	,720
Steriotype	92 (82.9)	6 (66.7)	98 (81.7)	,363

Table 2
The clinical features specified for the typical RTT.

Clinical Feature specified for typical RTT	Total number (%)	p-value
	• MECP2: n (%) • Other genes: n (%)	
Speech regression	113 (94.2) • 106 (93.8) • 7 (6.2)	p =,087
Loss of manual skills	100 (83.3) • 93 (93) • 7 (7)	p =,644
Stereotypical hand movements	107 (89.2) • 100 (93.5) • 7 (6.5)	p =,252
• Hand clenching	73 (60.8)	
• Hand washing	39 (32.5)	
• Hand flapping	51 (42.5)	
Disturbances in gait and posture while walking	106 (88.3) • 98 (92.5) • 8 (7.5)	p = 1000

Table 3
A comparison of the data findings according to the presence and absence of seizures is given.

	Presence of seizures n (%)	Absence of seizures n (%)	P value
Gender			
Male	4 (13,3)	4 (13,3)	0106
Female	86 (95,6)	26 (86,7)	
Consanguinity between parents			
Yes	77 (85,6)	26 (86,7)	1000
No	13 (14,4)	4 (13,3)	
Clinical type			
Atypical	27 (30,0)	9 (30,0)	1000
Typical	63 (70,0)	21 (70,0)	
Clinical stage			
Stage 1	10 (11,1)	5 (16,7)	NA
Stage 2	36 (40,0)	15 (50,0)	
Stage 3	27 (30,0)	8 (26,7)	
Stage 4	17 (18,9)	2 (6,7)	
Mutation			
MECP2	83 (92,2)	28 (93,3)	1000
Other mutations	7 (7,8)	2 (6,7)	
Changes in EEG* (n=89)			
No change/Deterioration	38 (46,3)	6 (85,7)	0058
Improvement	44 (53,7)	1 (14,3)	
Cognitive improvement			
No	81 (90,0)	28 (93,3)	0729
Yes	9 (10,0)	2 (6,7)	
Total	90 (100)	30 (100)	120

* Missing data exist.

was statistically significant (p=0.009, p=0.005; respectively). Differences between clinical classification based on identified genetic mutations and stage, MRI, and EEG findings, as well as seizure semiology, are thoroughly presented in Table 4.

The average age at the initiation of the first medication was 37.42 (range: 0–192). Among the applied treatments, the most commonly preferred antiepileptic drugs (AED) were valproate, levetiracetam, lamotrigine, and clobazam, with statistically significant differences ($p = 0.015$, $p < 0.001$, $p = 0.022$, and $p < 0.001$, respectively). The other AEDs used were carbamazepine, topiramate, vigabatrin, ACTH, sultiam, rufinamide, oxcarbazepine, clonazepam, phenytoin, ethosuximide, cannabidiol, and zonisamide, respectively. Changes in cognitive functions, EEG findings, seizure frequency, and duration were noted based on the selected treatment methods. Before the initiation of drug treatment, the seizure frequency, which was once a weekly occurrence in 25 cases, decreased to one case after medication (p=0.021). When the changes in EEG were examined following the use of medication, it was observed that there was no EEG alteration in 39.3 % of cases, deterioration in EEG in 10.1 % of cases, and improvement in EEG in 50.6 % of cases.

In the cases undergoing ketogenic diet therapy, medication was initiated at an average age of 5.75 months (range: 2–12 months) for 8 (6.7 %) cases. Out of these, 5 (62.5 %) showed improvement with treatment, and 50 % exhibited cognitive enhancement. Epileptiform activity was observed in the EEG of six cases (75 %), while 50 % showed no changes in their EEG. Vagus nerve stimulation (VNS) was applied in only 2 (1.7 %) cases for seizures, and all cases benefited from the treatment, with more than 50 % improvement in EEG and cognitive enhancement. Resective surgery was performed in only 1 (0.8 %) case. Five cases received steroid treatment at an average of 4 months, with 80 % of cases benefiting from steroids. Among them, three cases achieved complete seizure freedom, 60 % showed cognitive improvement, and more than 50 % improvement in EEG was observed, although complete normalization was not detected. The initiation of anti-seizure medications significantly altered seizure characteristics showed in Table 5.

4. Discussion

Rett syndrome (RTT) is a complex neurodevelopmental disorder primarily affecting females, characterized by a range of clinical features including motor impairments, seizures, and cognitive decline. This study delved into the genetic, clinical, and therapeutic aspects of RTT in a cohort of 120 cases in Turkey. The findings contribute valuable insights into the multifaceted nature of RTT, shedding light on its genetic

Table 4
The relation between the genetic mutation and the clinical, laboratuary, EEG, MRI findings of the patients.

	Mutation type		Cases with MECP2 mutation	Cases with the other genetic mutations (CDKL5, FOXG1)	p-value
	Frequency n (%)		111 (92.5)	9 (7.5)	NA
RTT clinic	**Atypical n (%)**		31 (27.9)	5 (55.6)	,126*
	Typical n (%)		80 (72.1)	4 (44.4)	,126*
First EEG Finding	**Normal n (%)**		20 (87)	3 (13)	,371*
	Burst-supression n (%)		5 (83.3)	1 (16.7)	,380*
	Abnormal epileptiform activity n (%)		78 (94)	5 (6)	,455*
		Multifocal	33 (29.7)	4 (44.4)	NA
		Focal	15 (13.5)		
		Generalized	44 (39.6)	2 (22.2)	
Clinical stage	**Stage 1 n (%)**		13 (86.7)	2 (13.3)	NA
	Stage 2 n (%)		46 (90.2)	5 (9.8)	
	Stage 3 n (%)		33 (94.3)	2 (5.7)	
	Stage 4 n (%)		19 (100.0)		
	Consanguinity n (%)		14 (82.4)	3 (17.6)	,116*
MRI finding	**Mesial temporal sclerosis**		7 (6.3)		1000*
	Periventricular enlargement		3 (2.7)		1000*
	Thinning of the corpus callosum		4 (3.6)	3 (33.3)	**,009**
	Cerebral atrophy		19 (17.1)	3 (33.3)	,363*
	Cerebellar atrophy		4 (3.6)		1000*
	Regression in myelination		3 (2.7)	3 (33.3)	**,005**
Seizure semiology	**Focal onset aware**		16 (14.4)	1 (11.1)	1000*
	Focal onset impaired awareness		6 (5.4)		1000*
	Generalized tonic clonic		26 (23.4)	2 (22.2)	1000*
	Generalized tonic		14 (12.6)	1 (11.1)	1000*
	Generalized myoclonic		15 (13.5)	2 (22.2)	,613
	Generalized atonic		2 (1.8)		1000*
	Behavior arrest		8 (7.2)		1000*

* Fisher's Exact Test and p<0.05 Significance level, NA:not available

Table 5
A comparison of the data before and after the initiation of anti-seizure medications is given.

	EEG findings change		P value	Seizure frequency before drug therapy			P value	Seizure frequency after drug therapy			p-value
	No change/ or deterrence	The improvement between % 50–100		No seizure/ or single seizure	One or more in a month	One or more in a week/ every day		No seizure/ or single seizure	One or more in a month	One or more in a week/ every day	
	n (%)	n (%)		n (%)	n (%)	n (%)		n (%)	n (%)	n (%)	
Clinical stage											
Stage 1	4 (40,0)	6 (60,0)	0301	8 (53,3)	1 (6,7)	6 (40,0)	**0005**	10 (66,7)	2 (13,3)	3 (20,0)	NA
Stage 2	13 (39,4)	20 (60,6)		27 (52,9)	15 (29,4)	9 (17,6)		31 (60,8)	16 (31,4)	4 (7,8)	
Stage 3	16 (55,2)	13 (44,8)		14 (40,0)	8 (22,9)	13 (37,1)		19 (54,3)	6 (17,1)	10 (28,6)	
Stage 4	11 (64,7)	6 (35,3)		4 (21,1)	2 (10,5)	13 (68,4)		4 (21,1)	3 (15,8)	12 (63,2)	
Clinical type											
Atypical	14 (56,0)	11 (44,0)	0439	19 (52,8)	6 (16,7)	11 (30,6)	0438	22 (61,1)	6 (16,7)	8 (22,2)	0486
Typical	30 (46,9)	34 (53,1)		34 (40,5)	20 (23,8)	30 (35,7)		42 (50,0)	21 (25,0)	21 (25,0)	
Genetic mutation type											
MECP2	39 (47,6)	43 (52,4)	0266	49 (44,1)	24 (21,6)	38 (34,2)	NA	60 (54,1)	24 (21,6)	27 (24,3)	NA
Other mutation types	5 (71,4)	2 (28,6)		4 (44,4)	2 (22,2)	3 (33,3)		4 (44,4)	3 (33,3)	2 (22,2)	
First EEG											
Normal	8 (57,1)	6 (42,9)	0530	14 (60,9)	1 (4,3)	8 (34,8)	**0057**	48 (49,5)	24 (24,7)	25 (25,8)	0215
Epileptic activity (Focal/ Generalize/ Multifocal)	36 (48,0)	39 (52,0)		39 (40,2)	25 (25,8)	33 (34,0)		16 (69,6)	3 (13,0)	4 (17,4)	
Seizure duration after drug therapy											
Yes	23 (40,4)	34 (59,6)	**0022**	15 (24,2)	21 (33,9)	26 (41,9)	**<0001**	25 (40,3)	20 (32,3)	17 (27,4)	**0007**
No	21 (65,6)	11 (34,4)		38 (65,5)	5 (8,6)	15 (25,9)		39 867,2)	7 (12,1)	12 (20,7)	
Change in seizure semiology after drug therapy											
Yes	12 (54,5)	10 (45,5)	0581	1 (4,3)	5 (21,7)	17 (73,9)	**<0001**	2 (8,7)	7 (30,4)	14 (60,9)	**<0001**
No	32 (47,8)	35 (52,2)		52 (53,6)	21 (21,6)	24 (24,7)		62 (63,9)	20 (20,6)	15 (15,5)	
Cognitive impairment after medication											
Yes	5 (45,5)	6 (54,6)	0778	5 (45,5)	1 (9,1)	5 (45,5)	NA	6 (54,5)	0 (0,0)	5 (45,5)	NA
No	39 (50,0)	39 (50,0)		48 (44,0)	25 (22,9)	36 (33,0)		58 (53,2)	27 (24,8)	24 (22,0)	

*Fisher's Exact Test and $p<0.05$ Significance level, NA: not available

underpinnings, clinical manifestations, and the impact of various treatment modalities.

The prevalence of MECP2 gene mutations in the studied cohort aligns with previous research, underscoring the pivotal role of MECP2 in the pathogenesis of RTT (Amir et al., 1999). When considering clinical manifestations indicative of RTT, a whole-genome sequencing analysis can be performed to identify other genetic mutations playing a role in the etiology (Lucariello et al., 2016). According to the 2019 report, cases with over 1200 MECP2 mutations were examined. The clinical details of 30 males (diagnosis age: 0.6–20.4 years) were reported (n:2 somatic MECP2 mutation, classic RTT; n:16 RTT-associated MECP2; n:9 pathogenic but not found in females; n:3 variants; 4, neonatal encephalopathy; 10, cognitive impairment or progressive encephalopathy; 2, classic RTT; 14, atypical RTT (clinically more severe). Nine male cases resulted in exitus (death age: 3.2–29.8). It has been emphasized that the assumption that MECP2 mutations in males always lead to embryonic or early postnatal death is incorrect (Neul et al., 2019). A systematic review from 2015 has identified 36 studies covering 57 cases of Rett Syndrome in males. In these cases, with an average diagnosis age of 7.7, the most commonly observed symptoms include abnormal gait/body movements, partial or complete loss of speech, deceleration in head growth, and epilepsy. However, MECP2 mutations were reported in only 65 % of the cases (Reichow et al., 2015). Fabio and colleagues evaluated the genotype-phenotype relationship based on subgroups of MECP2 mutations for RTT in their study (Fabio et al., 2014). In a study evaluating girls with CDKL5 mutations, eight patients with encephalopathy exhibiting RTT-like features were assessed (Nadia et al., 2008). The study was conducted by collecting data from nine different centers. Due to the incomplete establishment of genetic pools as an active system in our country and the conduct of genetic mutation studies in various centers, the sub-classifications of the MECP2 mutation mentioned in the literature could not be individually determined in each case. Therefore, statistical data on the phenotype-genotype relationship of clinical data according to the sub-classification of the MECP2 mutation could not be conducted in our study. In our country, the most frequently detected genetic mutation is MECP2, similar to what is reported in the literature. In 75 % of the 8 male RTT cases, MECP2 mutation was detected, while FOXG1 mutation was found in 25 %, and no case with CDKL5 mutation was identified among males in this study. None of the males have died.

In a study comprising 53 female cases, the median age at seizure onset was 4 years (range 0.2–26.2) (Steffenburg et al., 2001). When examining a compilation of studies investigating the age groups of RTT cases, it was noted that the included studies encompassed patients aged 3–21 years, 5–24 years, or 5–32 years. One study also observed patients younger than 35 years, while the age of the study population was not reported in another (Petriti et al., 2023). The average age of the cases comprising our study was 36.56, with 12.5 % being below 2 years old, and 42.5 % falling within the 2–4 age range.

Notably, the higher occurrence of gene mutations in cases with typical clinical features compared to those with atypical presentations emphasizes the heterogeneous nature of RTT. This aligns with the notion that RTT represents a spectrum disorder with varying clinical phenotypes (Gold et al., 2018). Our findings were identified to be similar to the studies in our investigation.

The predominance of generalized tonic-clonic, myoclonic, and focal onset aware seizures in the seizure semiology corresponds to previous literature highlighting the diversity of seizure types in RTT (Dolce et al., 2013). The frequency of seizure semiologies in our study was similar to those reported in the literature (n:28,n:17,n:17; respectively).

Epilepsy and epileptiform activity frequently coexist with RTT, reported in 50–90 % of cases. RTT patients exhibit diverse forms of epileptiform activity, predominantly of centrotemporal origin, and other multifocal patterns, including a general slowing of background EEG, among other commonly reported EEG abnormalities (Portnova et al., 2022). In our study, among cases with EEG findings, 69.2 % showed epileptiform activity, with 46 being generalized, 37 multifocal,

and 15 focal; while 31.7 % of cases exhibited a slow background EEG rhythm.

MRI findings corroborate the neuroanatomical complexity of RTT, with cerebral atrophy, corpus callosum thinning, and mesial temporal sclerosis being the most frequently observed pathological features. These findings are consistent with the literature (Kyle et al., 2018), further emphasizing the importance of neuroimaging in the comprehensive evaluation of RTT patients. In studies, it has been reported that reduced cerebellum size is associated with some of the observed neurological signs in RTT, contributing to impairments in cognition and motor coordination. Additionally, significant thinning of specific structures, such as the motor cortex and the corpus callosum, has been documented in the data (Ariani et al., 2008; Kong et al.,2022). In the study, cases with two FOXG1 mutations and one CDKL5 mutation were found to have thinning of the corpus callosum and decreased myelination in MRI, similar to the literature.

In a study comprising 53 female cases, the median age at seizure onset was 4 years (range 0.2–26.2), with a higher prevalence of partial complex seizures (54 % vs. 23 %). Among females, 6 % (3/50) used no AEDs, 54 % (27/50) used one AED, 30 % (15/50) used 2 AEDs, and 10 % (5/50) used 3 AEDs. The most frequently prescribed AEDs were carbamazepine in 16 cases, lamotrigine in 16, and valproate in 12 (Steffenburg et al.,2001).

The initiation of anti-seizure medications, particularly the preference for valproate, levetiracetam, lamotrigine, and clobazam, reflects the challenging task of managing seizures in RTT. The observed alterations in seizure characteristics post-treatment underscore the need for individualized therapeutic approaches in RTT. It is noteworthy that the ketogenic diet, VNS, and steroid treatments demonstrated efficacy in specific cases, supporting the exploration of diverse therapeutic modalities tailored to the unique needs of RTT patients (Neul, 2019; Lim et al., 2020; Vilvarajan et al., 2023). Long-chain polyunsaturated fatty acid supplementation (LCPUFA) was also attempted in Rett Syndrome (RTT), but no meaningful effect was found in neuropsychological and behavioral parameters (Fabio et al., 2018). There was no patient taking (LCPUFA) in our study.

In cases of RTT, difficulty in expressing themselves arises from the loss of language from a young age. Therefore, it is important for patients to receive early supportive treatment for their pain without neglect, thus potentially improving their quality of life. For this purpose, scales are used to assess pain (Fabio et al., 2022). The diverse clinical features detailed in Tables 1 and 2 further underscore the complexity of RTT and the necessity for a multidisciplinary approach in its management. Interestingly, no statistically significant differences were found in symptoms when examined based on gender, clinical type, and family relatedness, emphasizing the need for personalized care plans that consider the individual variability in RTT presentation. In our country, working groups initiated by the Pediatric Neurology Association have been identified and are beginning to be organized. This aims to enhance the quality of life for both patients and their families through rehabilitation services, as stated in the literature. Additionally, the standardized assessment of pain in RTT patients can be ensured by using the scales mentioned in the literature.

Our findings underscore the necessity of a multidisciplinary approach in managing RTT, as evidenced by the correlation between steroid treatment and cognitive improvement. The observed reduction in seizures after VNS application holds promise for improving the overall quality of life for RTT patients. In recent times, Transcranial Direct Current Stimulation (tDCS) methods aimed at improving both seizures and cognitive functions are among the alternative treatment approaches used in RTT (Fabio et al., 2020). In our country, the use of tDCS, which is employed in resistant epilepsies, is not yet widespread, resulting in rare applications. The centers participating in the study have not utilized this non-invasive method in the treatment of RTT, preventing an assessment from being conducted.

This study serves as a preliminary exploration, and future research should focus on expanding the sample size, incorporating clinically diagnosed RTT cases, and exploring the long-term effects of various interventions. Additionally, investigating the relationship between genetic mutations and specific clinical manifestations may offer personalized therapeutic approaches for RTT patients.

In conclusion, this study contributes valuable data to the growing body of knowledge on Rett syndrome. The genetic, clinical, and therapeutic insights presented here provide a foundation for further research and underscore the necessity for comprehensive and individualized management strategies in RTT. The findings also highlight the importance of collaborative efforts across disciplines to address the multifaceted nature of this challenging disorder.

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Conflict of interest

All authors declare that they have no conflict of interest

CRediT authorship contribution statement

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References

Amir, R.E., Van den Veyver, I.B., Wan, M., Tran, C.Q., Francke, U., 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.

Ariani, F., Hayek, G., Rondinella, D., Artuso, R., Mencarelli, M.A., Spanhol-Rosseto, A., Pollazzon, M., Buoni, S., Spiga, O., Ricciardi, S., Meloni, I., Longo, I., Mari, F., Broccoli, V., Zappella, M., Renieri, A., 2008. FOXG1 is responsible for the congenital variant of Rett syndrome. Am. J. Hum. Genet. 83, 89–93.

Bianco, E., Rota, D., 2018. Oral findings in Rett syndrome: an update and review of the literature. Dent. Med Probl. 55 (4), 441–445.

Chahil, G., Bollu, P.C., 2023. Rett Syndrome. 2023 Aug 8. In: StatPearls [Internet. StatPearls Publishing, Treasure Island (FL) (Jan).

Dolce, A., Ben-Zeev, B., Naidu, S., Kossoff, E.H., 2013. Rett syndrome and epilepsy: an update for child neurologists. Pediatr. Neurol. 48 (5), 337–345 (May).

Evans, J.C., Archer, H.L., Colley, J.P., Ravn, K., Nielsen, J.B., Kerr, A., Williams, E., Christodoulou, J., Gecz, J., Jardine, P.E., Wright, M.J., Pilz, D.T., Lazarou, L., Cooper, D.N., Sampson, J.R., Butler, R., Whatley, S.D., Clarke, A.J., 2005. Early onset seizures and Rett-like features associated with mutations in CDKL5. Eur. J. Hum. Genet. 13, 1113–1120.

Fabio, R.A., Caprí, T., Lotan, M., Towey, G.E., Martino, G., 2018. Motor abilities are related to specific genotypes in rett syndrome. In: Advances in Genetics Research, Vol. 18. Nova Science Publishers, Inc, pp. 79–107.

Fabio, R.A., Chiarini, L., Canegallo, V., 2022. Pain in Rett syndrome: a pilot study and a single case study on the assessment of pain and the construction of a suitable measuring scale. Orphanet J. Rare Dis. 17, 356.

Fabio, R.A., Colombo, B., Russo, S., Cogliati, F., Masciadri, M., Foglia, S., Antonietti, A., Tavian, D., 2014. Recent insights into genotype-phenotype relationships in patients

with Rett syndrome using a fine grain scale. Res Dev. Disabil. 35 (11), 2976–2986 (Nov).
Fabio, R.A., Gangemi, A., Semino, M., Vignoli, A., Canevini, M.P., Priori, A., Rosa, G.D., Caprì, T., 2020. Effects of combined transcranial direct current stimulation with cognitive training in girls with Rett syndrome. Brain Sci. 10 (5), 276. May 2.
Fabio, R.A., Martino, G., Caprì, T., Giacchero, R., Giannatiempo, S., Antonietti, A., La Briola, F., Banderali, G., Canevini, M.P., Vignoli, A., 2018. Long chain poly-unsaturated fatty acid supplementation in rett syndrome: a randomized placebo-controlled trial. Asian J. Clin. Nutr. 10, 37–46.
Gold, W.A., Krishnarajy, R., Ellaway, C., Christodoulou, J., 2018. Rett syndrome: a genetic update and clinical review focusing on comorbidities. ACS Chem. Neurosci. 9 (2), 167–176. Feb 21.
Kong, Y., Li, Q.B., Yuan, Z.H., Jiang, X.F., Zhang, G.Q., Cheng, N., Dang, N., 2022. Multimodal neuroimaging in Rett syndrome with *MECP2* mutation. Front. Neurol. 13, 838206. Feb 23.
Kyle, S.M., Vashi, N., Justice, M.J., 2018. Rett syndrome: a neurological disorder with metabolic components. Open Biol. 8 (2), 170216 (Feb).
Lim, J., Greenspoon, D., Hunt, A., McAdam, L., 2020. Rehabilitation interventions in Rett syndrome: a scoping review. Dev. Med. Child Neurol. 62 (8), 906–916.
Lucariello, M., Vidal, E., Vidal, S., Saez, M., Roa, L., Huertas, D., Pineda, M., Dalfó, E., Dopazo, J., Jurado, P., Armstrong, J., Esteller, M., 2016. Whole exome sequencing of Rett syndrome-like patients reveals the mutational diversity of the clinical phenotype. Hum. Genet 135 (12), 1343–1354 (Dec).
Nadia, Bahi-Buisson, Nectoux, Juliette, Rosas-Vargas, Haydee, Milh, Mathieu, 2008. Nathalie Boddaert, Benoit Girard, Claude Cances, Dorothee Ville, Alexandra Afenjar, Marlene Rio, Delphine Héron, Marie Ange N′Guyen Morel, Alexis Arzimanoglou, Christophe Philippe, Philippe Jonveaux, Jamel Chelly, Thierry Bienvenu, Key clinical features to identify girls with CDKL5 mutations, Brain, Volume 131, Issue 10, October 2008, Pages 2647–2661..
Neul, J.L., 2019. Can Rett syndrome be diagnosed before regression? Neurosci. Biobehav. Rev. 104, 158–159.
Neul, J.L., Benke, T.A., Marsh, E.D., Skinner, S.A., Merritt, J., Lieberman, D.N., Standridge, S., Feyma, T., Heydemann, P., Peters, S., Ryther, R., Jones, M., Suter, B., Kaufmann, W.E., Glaze, D.G., Percy, A.K., 2019. The array of clinical phenotypes of males with mutations in Methyl-CpG binding protein 2. Am. J. Med. Genet. B Neuropsychiatr. Genet. 180 (1), 55–67 (Jan).
Neul, J.L., Kaufmann, W.E., Glaze, D.G., Christodoulou, J., Clarke, A.J., Bahi-Buisson, N., Leonard, H., Bailey, M.E., Schanen, N.C., Zappella, M., Renieri, A., Huppke, P., Percy, A.K., RettSearch Consortium, 2010. Rett syndrome: revised diagnostic criteria and nomenclature. Ann. Neurol. 68 (6), 944–950 (Dec).
Petriti, U., Dudman, D.C., Scosyrev, E., Lopez-Leon, S., 2023. Global prevalence of Rett syndrome: systematic review and meta-analysis. Syst. Rev. 12 (1), 5. Jan 16.
Portnova, G., Neklyudova, A., Voinova, V., Sysoeva, O., 2022. Clinical EEG of Rett syndrome: group analysis supplemented with longitudinal case report. J. Pers. Med. 12 (12), 1973. Nov 29.
Reichow, B., George-Puskar, A., Lutz, T., et al., 2015. Brief report: systematic review of Rett syndrome in males. J. Autism Dev. Disord. 45, 3377–3383.
Steffenburg, U., Hagberg, G., Hagberg, B., 2001. Epilepsy in a representative series of Rett syndrome. Acta PæDiatr. 90, 34–39.
Vilvarajan, S., McDonald, M., Douglas, L., Newham, J., Kirkland, R., Tzannes, G., Tay, D., Christodoulou, J., Thompson, S., Ellaway, C., 2023. Multidisciplinary management of Rett syndrome: twenty years' experience. Genes.