

Genetic Testing for Rett Syndrome

Policy Number: AHS – M2088 – Genetic Testing for Rett Syndrome	Prior Policy Name and Number, as applicable:
Initial Policy Effective Date: 12/01/2024	

[POLICY DESCRIPTION](#) | [RELATED POLICIES](#) | [INDICATIONS AND/OR LIMITATIONS OF COVERAGE](#) | [TABLE OF TERMINOLOGY](#) | [SCIENTIFIC BACKGROUND](#) | [GUIDELINES AND RECOMMENDATIONS](#) | [APPLICABLE STATE AND FEDERAL REGULATIONS](#) | [APPLICABLE CPT/HCPCS PROCEDURE CODES](#) | [EVIDENCE-BASED SCIENTIFIC REFERENCES](#) | [REVISION HISTORY](#)

I. Policy Description

Rett syndrome (RTT) is a rare X-linked neurodevelopmental disorder that occurs almost exclusively in females and is usually caused by mutations in the methyl CpG binding protein 2 (*MECP2*) gene (Amir et al., 1999). It is characterized by normal early growth and development, followed by regressions in development, walking, language, and purposeful use of the hands, along with slowed brain and head growth, distinctive hand movements, seizures, and intellectual disability (Colvin et al., 2004; Hagberg et al., 1983; Leonard et al., 2017; Naidu et al., 1986; Neul et al., 2010; Rett, 1966).

Terms such as male and female are used when necessary to refer to sex assigned at birth.

II. Related Policies

Policy Number	Policy Title
AHS – M2176	Testing for Autism Spectrum Disorder and Developmental Delay

III. Indications and/or Limitations of Coverage

Application of coverage criteria is dependent upon an individual’s benefit coverage at the time of the request. Specifications pertaining to Medicare and Medicaid can be found in the “Applicable State and Federal Regulations” section of this policy document.

- 1) For a child with developmental delay/intellectual disability **and** signs/symptoms of Rett syndrome (RTT), but for whom there is uncertainty in the clinical diagnosis, genetic testing of *MECP2*, *CDKL5*, and/or *FOXG1* to confirm a diagnosis of RTT **MEETS COVERAGE CRITERIA**.

The following does not meet coverage criteria due to a lack of available published scientific literature confirming that the test(s) is/are required and beneficial for the diagnosis and treatment of an individual’s illness.

- 2) For all other situations not described above (e.g., prenatal screening, testing of family members), genetic testing for RTT **DOES NOT MEET COVERAGE CRITERIA**.

IV. Table of Terminology

Term	Definition
AAN	American Academy of Neurology
AAP	American Academy of Pediatrics
ACMG	American College of Medical Genetics
ASD	Autism spectrum disorder
BDNF	Brain-derived neurotrophic factor
CDKL5	Cyclin-dependent kinase-like 5
CMS	Centers for Medicare and Medicaid
CNS	Child Neurology Society
CPS	Canadian Pediatric Society
<i>CTNNB1</i>	<i>Catenin beta 1</i>
DNA	Deoxyribonucleic acid
FDA	Food and Drug Administration
<i>FOXP1</i>	<i>Forkhead box g1</i>
GABA	Gamma-aminobutyric acid
GDD	Global developmental delay
LDT	Laboratory developed test
MDS	<i>Methyl CPG binding protein duplication syndrome</i>
<i>MECP</i>	<i>Methyl CPG binding protein</i>
miRNA	Micro ribonucleic acid
MLPA	Multiplex ligation-dependent probe amplification
NGS	Next generation sequencing
qPCR	Quantitative polymerase chain reaction
REST	Rett evaluation of symptoms and treatments
RNA	Ribonucleic acid
RTT	Rett Syndrome
<i>SCN1A</i>	<i>Sodium voltage-gated channel alpha subunit 1</i>
<i>WDR45</i>	<i>WD repeat domain 45</i>
XLID	X-linked intellectual disability

V. Scientific Background

Rett syndrome (RTT) is a severe neurodevelopmental disorder which affects approximately 1:10,000 live female births in the United States annually (Hagberg, 1985; NORD, 2019). It is a prominent cause of severe intellectual disability in women, accounting for up to 10% of cases inherited genetically (Armstrong, 1997). Originally thought to be lethal in males (Amir et al., 1999; Chahil et al., 2018; Franco & Ballabio, 2006), RTT has been identified in up to 1.3% of male patients with mental retardation (Villard, 2007) and can be associated with a more severe phenotype (Zhang et al., 2017). These males have either an extra X-chromosome (Klinefelter

syndrome) or somatic mosaicism of the *MECP2* variant. Reichow et al. (2015) claim to have published the first review of male RTT data in 2015, and they only identified a total of 57 published cases.

Rett syndrome can be inherited as an X-linked dominant disorder; however, more than 99% of cases result from a de novo pathogenic mutation in the *methyl CpG binding protein 2 (MECP2)* gene (Amir et al., 1999; Christodoulou & Ho, 1993), a transcriptional regulator located on the X chromosome. More than 200 mutations in *MECP2* have been associated with RTT (Suter et al., 2014). Analysis of parental origin of the mutated *MECP2* gene in sporadic cases of RTT showed that 94.4% of mutations were from paternal origin, 90.6% of which were point mutations; further, 5.6% of mutations were from maternal origin (Zhang et al., 2012). This may explain the high occurrence of RTT in females. *MECP2* is a multifunctional protein which interprets DNA methylation and regulates chromatin architecture, gene transcription, and RNA splicing (Sun et al., 2018). The complex upstream and downstream pathways of *MECP2* involve microRNAs and neurotrophic factors, such as GABA and BDNF (Kang et al., 2014). Transcriptome level analysis in tissues derived from RTT patients report dysregulations in dendritic connectivity and synapse maturation, mitochondrial dysfunction, and glial cell activity (Shovlin & Tropea, 2018). Researchers have identified two individuals with an RTT diagnosis who lacked a mutation in the *MECP2* gene but had a mutation in other genes previously unassociated with RTT: *CTNNB1* and *WDR45* (Percy et al., 2018).

The *MECP2* gene is critical for neuronal maturation (Fukuda et al., 2005; Smrt et al., 2007), and its deficiency results in impaired dendritic morphogenesis and reduced dendritic spine numbers (Chapleau et al., 2009; Kishi & Macklis, 2010). This results in dysfunctional synaptic transmission and neural network activity (Sun et al., 2018), affecting successive stages of brain development, including prenatal neurogenesis, postnatal development of synaptic connections and function, experience-dependent synaptic plasticity, and maintenance of adult neural function, including sensory integration (Feldman et al., 2016).

The clinical picture of RTT is characterized by a broad clinical spectrum of signs and symptoms (Pini et al., 2016) and a distinctive course of apparent normal development for the first six to 18 months of life, followed by characteristic developmental stagnation and loss of acquired skills, including loss of intellectual functioning, loss of acquired fine and gross motor skills and communication (Colvin et al., 2004; Dolce et al., 2013; Hagberg et al., 1983; Leonard et al., 2017; Naidu et al., 1986; Neul et al., 2010; Rett, 1966). Purposeful use of the hands is often replaced by repetitive stereotypical hand movements (Dy et al., 2017; Elian & de, 1996; Goldman & Temudo, 2012). Other clinical observations include deceleration of head growth, seizures, disturbed breathing patterns, scoliosis, growth retardation, and gait apraxia (Cianfaglione et al., 2015).

Despite a period of apparently normal early development, the profound neurological regressions characteristic of RTT have been found to result from *MECP2*-related defects in the establishment and refinement of early neural circuits and, later, cortical plasticity (Feldman et al., 2016). Subtle signs, such as hypotonia, jerkiness in limb movement, and limited social interaction can be present during early infancy (Ip et al., 2018).

The severity and rate of progression of this disease can vary with several recognized atypical variants. The milder forms (Zappella) present with less severe regression and milder expression of the clinical characteristics of RTT. In the most severe forms, there is no normal development period (Neul et al., 2010). Both genetic and clinical variants of RTT are associated with distinct electrophysiological profiles reflecting how genetic dysregulation of synapse formation results in differences in neuronal network architecture (Sun et al., 2018) and varying clinical phenotypes (Keogh et al., 2018). The pattern of X-chromosome inactivation can also influence the severity of the clinical disease (Archer et al., 2007; Weaving et al., 2003).

Mutations in the upstream cyclin-dependent kinase-like five (*CDKL5*) gene cause an early seizure (Hanefield) variant of the RTT phenotype (Bahi-Buisson et al., 2008), and mutations in the forkhead box G1 (*FOXG1*) gene have been found in the congenital variant (Rolando) (Ariani et al., 2008). Two cases of females with pathogenic de novo mutations in *SCN1A*, which usually leads to Dravet syndrome, but fulfill the diagnostic criteria for classic RTT have also been reported (Henriksen et al., 2018). In males, *MECP2* duplication phenotypically presents with infantile hypotonia, recurrent respiratory infections, and severe mental retardation (Villard, 2007).

Fu et al. (2020) published a set of “consensus guidelines” with input from several clinical sites, Rett Syndrome-focused centers, two patient advocacy groups, and Rett Syndrome clinical specialists. Although this guideline focuses on “management” of Rett Syndrome, the guideline does comment on the genetics of Rett Syndrome. The guideline remarks that “nearly” all individuals with Rett Syndrome (RTT) have a loss-of-function mutation on the *MECP2* and that these mutations are “almost always” *de novo* (and thereby not expected to recur in families). Two other genes (*CDKL5* and *FOXG1*) are named as possible causes of RTT. The guideline does not note any specific treatments based on type of mutation, though two other genes (*CDKL5* and *FOXG1*) are named as possible causes of RTT. However, the guideline states that “Alterations in *MECP2*, *CDKL5* and *FOXG1* should be considered in all individuals, male and female, with developmental delays and intellectual disability” (Fu et al., 2020). This consensus guideline also notes there is hope for disease-modifying therapy as reversing symptoms in mice has occurred in a clinical research context (Fu et al., 2020).

Banerjee et al. (2019) published a paper titled “Towards a Better Diagnosis and Treatment of Rett Syndrome: A Model” summarizing the developments in the diagnosis and treatment of Rett syndrome over the past 50 years. They note that the first gene therapy trial for *MECP2* “was modelled after the successful (i.e., improved survival and motor functions) single dose intravenous adeno-associated virus serotype 9 delivery of complementary DNA.” Even with promising gene therapy techniques, the authors note that the field has challenges. “The Rett syndrome field is experiencing the same challenges as other neurodevelopmental disorders pursuing neurobiologically based treatments: inadequate outcomes and measures of response.” The authors also comment on the complexity of the *MECP2* mutation’s role in the syndrome, “*MECP2* mutations is supportive, but not confirmatory because of the limited genotype-phenotype correlations in Rett syndrome” (Banerjee et al., 2019).

Confirmation of the genetic diagnosis can improve the medical management of the patient. It can also end the diagnostic odyssey, provide a general idea of prognosis for the patient, and/or provide closure to the family (Mroch et al., 2012). Complex neurodevelopmental disorders need

multi-disciplinary treatment approaches for optimal care. The clinical effectiveness of treatments is limited in patients with rare genetic syndromes and multisystem morbidity such as RTT; single drug strategies may not be sufficient, due to the multiple overlapping physiological systems affected (Singh & Santosh, 2018).

Functional performance for self-care, upper extremity function, and mobility in RTT patients may relate to the type of mutation. Knowledge of these relationships is useful for developing appropriate rehabilitation strategies and prognosis (Pidcock et al., 2016).

Of the clinical criteria for RTT, loss of hand skills was the most significant clinical predictor of a positive genetic test for mutations of a *MECP* gene in females. Gait abnormalities and stereotypic hand movements were also strong predictors of a positive genetic test for mutations of *MECP*. Language delay is the least specific of the major criteria (Knight et al., 2016). A reliable and single multidimensional questionnaire, the Rett Evaluation of Symptoms and Treatments (REST) Questionnaire, is being developed to combine physiological aspects of the disease obtained using wearable sensor technology, along with genetic and psychosocial data to stratify patients and streamline the care pathway (Santosh et al., 2017).

Clinical Utility and Validity

Lallar et al. (2018) used Sanger sequencing to diagnose suspected RTT cases. Participants were divided into two groups: Group 1 was comprised of females with symptoms of classical and atypical RTT, and Group two was comprised of females with other “Rett like features” that did not fit into the first category. *MECP2* mutations were identified in 74% of females in Group one and in zero percent of females in Group two; females in Group one with classical RTT had a mutation detection rate of 93% (Lallar et al., 2018). This shows that Sanger sequencing is efficient in detecting RTT in patients with the classical form of the disease.

Sheinerman et al. (2019) used brain-enriched microRNAs (miRNAs) to identify miRNA biomarkers of RTT; for this study, 30 patients with RTT were matched with 30 healthy controls of similar age (Sheinerman et al., 2019). Results showed that miRNAs identified RTT patients with 85-100% sensitivity when compared to controls; further, the researchers determined that “the dynamics in levels of miRNAs appear to be associated with disease development (involvement of liver, muscle and lipid metabolism in the pathology)” (Sheinerman et al., 2019). These results suggest that circulating miRNAs could be used to measure RTT disease progression or individual response to treatment.

In at least 95% of Rett syndrome cases, the cause is a de novo mutation in the child; *MECP2* variants are rarely inherited from a carrier mother with a germline mutation in *MECP2*, in whom favorable skewing of X-chromosome inactivation results in minimal to no clinical findings. When the mother is a known carrier, inheritance follows an X-linked dominant pattern with a 50% risk to her offspring of inheriting the *MECP2* variant (Christodoulou & Ho, 1993).

A mutation in *MECP2* does not necessarily equate to a clinical diagnosis of RTT. *MECP2* mutations have also been reported in other clinical phenotypes, including individuals with an Angelman-like picture, nonsyndromic X-linked intellectual disability, autism, in males as PPM-X syndrome (an X-linked genetic disorder characterized by psychotic disorders,

parkinsonism, and intellectual disability), and most commonly as neonatal encephalopathy (Liyanage & Rastegar, 2014; Suter et al., 2014; Williamson & Christodoulou, 2006).

Recent expert opinion in the United Kingdom concluded that genetic testing for all children with unexplained global developmental delay (GDD) should be first-line if an exogenous cause is not already established. All patients, irrespective of severity of GDD, should have investigations for treatable conditions. The yield for treatable conditions is higher than previously thought and that investigations for these conditions should be considered as first-line. Additional second-line investigations can be led by history, examination, and developmental trajectories (Mithyantha et al., 2017).

Vidal et al. (2017) have utilized next generation sequencing (NGS) in a total of 1577 patients with RTT-like clinical diagnoses or patients with potential RTT genetic mutations as determined previously by Sanger Sequencing. Of the 1577 patients with RTT-like clinical diagnoses, the NGS method was able to confirm the RTT diagnosis in 477 patients (about 30%). Further, “Positive results were found in 30% by Sanger sequencing, 23% with a custom panel, 24% with a commercial panel and 32% with whole exome sequencing,” suggesting that NGS is a competitive diagnostic RTT tool compared to the aforementioned methods (Vidal et al., 2017).

Vidal et al. (2019) used multiplex ligation-dependent probe amplification (MLPA) in the *MECP2* gene of 21 RTT patients to identify deletions of varying sizes; these researchers identified both total and partial deletions of the *MECP2* gene in each patient, with identified partial deletions ranging from 1,235 bp to 85 kb. Breakpoints were delineated by DNA-qPCR; the results have allowed the researchers to “propose a genotype–phenotype correlation” which will assist in appropriate genetic counseling (Vidal et al., 2019).

Seventy-two classical Rett syndrome (RTT) female patients were included in a cohort study by Khajuria et al. (2020) to analyze exons two-four of *MECP2* gene by Sanger sequencing for sequence variations followed by deletion/duplication analysis using Multiplex Ligation-dependent Probe Amplification (MLPA). Patients were defined as classical when they showed signs of partial or complete loss of acquired purposeful hand skills, partial or complete loss of acquired spoken language, gait abnormalities, impaired or absence of ability to walk, and stereotypic hand movements. Through Sanger Sequencing, *MECP2* sequence variations were identified in 90.3% of patients. With further evaluation using MLPA, large deletions of *MECP2* were identified in 9.7% of the patients, which were negative on DNA sequencing. MLPA analysis increased the detection rate of *MECP2* sequence variants identified in patients from 90.3% to 98.6%. The authors emphasize that “MLPA analysis of *MECP2* is crucial and needs to be performed in classical RTT patients. Large deletions can be missed using DNA sequencing and reaffirms the view that large *MECP2* deletions are an important cause of classical RTT (Khajuria et al., 2020).” Xioli et al. (2021) performed a clinical review of technological advances in RTT genetics. The authors review summarizes that our understanding of Rett syndrome has evolved “towards a spectrum of overlapping phenotypes with great genetic heterogeneity.” The authors note that advances in genetic diagnosis have been impacted by the rise in next generation sequencing (NGS) and whole genome sequencing. Of note are the “90 causative genes” and “significantly overlapping phenotypes” involved in RTT spectrum disorders. To achieve an accurate and quick diagnosis of Rett syndrome, the authors strongly recommend simultaneous multiple gene testing and thorough phenotypic characterization.

Bassuk (2021) published a paper concerning methyl-CpG-binding protein 2 (*MECP2*) and its encoding of an epigenetic reader, MeCP2. The author notes that loss of function of the epigenetic reader may be a factor in RTT, but also that “locus duplications also cause a severe neurodevelopmental disorder, *MECP2* duplication syndrome (MDS).” This suggests that MeCP2 (the protein) could be what is called a “Goldilocks protein,” that is, one that requires an activity level that is precise. Using the re-expression of the MeCP2 protein in mouse models, the author presents a case for the development of therapeutic interventions in people and the restoration of the desirable phenotypes. However, gene therapy must be approached with caution, as restoring function to the protein still carries the risk of “MDS overexpression phenotypes” (Bassuk, 2021).

VI. Guidelines and Recommendations

American Academy of Pediatrics (AAP)

A 2014 policy statement from the AAP recommends *MECP2* mutation analysis for females with microcephaly or deceleration of head growth and other features of Rett syndrome, or who present with stereotypical hand-wringing movements and developmental regression. *MECP2* gene mutations are extremely rare in males but may be considered in boys who present with clinical features of Rett syndrome or severe developmental regression (Moeschler & Shevell, 2014).

Complete *MECP2* deletion, duplication, and sequencing study is also recommended for females with intellectual disability or global developmental delay for whom the chromosomal microarray, specific metabolic testing, and fragile X genetic testing did not produce a diagnosis (Moeschler & Shevell, 2014).

The above guideline was reaffirmed in 2019 (AAP, 2019).

The AAP also published a guideline focusing on children with autism spectrum disorder (ASD). In it, they note that other disorders may meet certain criteria for ASD. However, the AAP notes that these disorders should prompt the “appropriate targeted testing” (or referral to a specialist). The AAP lists an example of Rett Syndrome, stating that “for example, a girl with significant developmental delays, deceleration in head growth velocity, and characteristic midline hand movements should prompt genetic testing for a mutation or deletion or duplication of *MECP2*, the gene implicated in Rett syndrome.” In Supplemental Table 13, they list the following findings as representative of Rett Syndrome: “Deceleration of head growth velocity, acquired microcephaly, loss of purposeful hand use, prominent hand stereotypies (especially hand wringing or clasping), apraxia, hyperventilation or breath-holding, seizures” (Hyman et al., 2020).

Canadian Pediatric Society (CPS)

The CPS supports the guidelines mentioned above by the AAP. The CPS stated that “According to the AAP. . . *MECP2* molecular analysis should be ordered when characteristic symptomatology is present (i.e., initially normal development followed by loss of speech and

purposeful hand use, stereotypical hand movement, gait abnormalities) or for moderately-to-severely affected girls” (Belanger & Caron, 2018).

RettSearch

The AAP has not provided recommendations on when to use *CDKL5* or *FOXP1* testing. RettSearch members, representing the majority of the international clinical RTT specialists, “participated in an iterative process to come to a consensus on a revised and simplified clinical diagnostic criteria for [RTT]” (Neul et al., 2010). This group provided clarifications for diagnosis of classic or typical RTT and atypical RTT and provided guidelines for molecular evaluation of specific variant forms of RTT. The authors define RTT as a clinical diagnosis based on distinct clinical criteria, independent of molecular findings. Presence of a *MECP2* mutation is not sufficient for the diagnosis of RTT. Neul et al. (2010) proposed three distinct criteria for diagnosis of variant forms of RTT: preserved speech variant (Zapella variant), early seizure variant (Hanefeld variant) and congenital variant (Rolando variant); identifying the molecular genetics of each variant was also recommended. In the Zapella variant, the molecular analysis for *MECP2* was recommended. In Hanefeld and Rolando variants, recommended mutations for analysis were in the *CDKL5* and *FOXP1* genes, respectively. Further, it was stated that patients found negative for *MECP2* mutations and who have a strong clinical diagnosis of RTT should be considered for further screening for the *CDKL5* gene if early onset seizures or *FOXP1* gene congenital features (e.g., severe postnatal microcephaly) are present (Neul et al., 2010).

American College of Medical Genetics (ACMG)

In 2013, the ACMG revised its evidence-based guidelines for clinical genetics evaluation of autism spectrum disorders. Testing for *MECP2* mutations is recommended as part of the diagnostic workup of females who present with an autistic phenotype. Routine *MECP2* testing in males with autistic spectrum disorders is not recommended. However, when features of *MECP2* duplications (e.g., drooling, recurrent respiratory infections, hypotonic facies) are present, *MECP2* duplication testing in boys with autism and such features may be considered (Schaefer & Mendelsohn, 2013).

VII. Applicable State and Federal Regulations

DISCLAIMER: If there is a conflict between this Policy and any relevant, applicable government policy for a particular member [e.g., Local Coverage Determinations (LCDs) or National Coverage Determinations (NCDs) for Medicare and/or state coverage for Medicaid], then the government policy will be used to make the determination. For the most up-to-date Medicare policies and coverage, please visit the Medicare search website: <https://www.cms.gov/medicare-coverage-database/search.aspx>. For the most up-to-date Medicaid policies and coverage, visit the applicable state Medicaid website.

Food and Drug Administration (FDA)

Many labs have developed specific tests that they must validate and perform in house. These laboratory-developed tests (LDTs) are regulated by the Centers for Medicare and Medicaid (CMS) as high-complexity tests under the Clinical Laboratory Improvement Amendments of 1988 (CLIA '88). LDTs are not approved or cleared by the U. S. Food and Drug Administration; however, FDA clearance or approval is not currently required for clinical use.

VIII. Applicable CPT/HCPCS Procedure Codes

CPT	Code Description
81302	MECP2 (methyl CpG binding protein 2) (eg, Rett syndrome) gene analysis; full sequence analysis
81303	MECP2 (methyl CpG binding protein 2) (eg, Rett syndrome) gene analysis; known familial variant
81304	MECP2 (methyl CpG binding protein 2) (eg, Rett syndrome) gene analysis; duplication/deletion variants
81404	Molecular pathology procedure, Level 5 (eg, analysis of 2-5 exons by DNA sequence analysis, mutation scanning or duplication/deletion variants of 6-10 exons, or characterization of a dynamic mutation disorder/triplet repeat by Southern blot analysis)
81405	Molecular pathology procedure, Level 6 (eg, analysis of 6-10 exons by DNA sequence analysis, mutation scanning or duplication/deletion variants of 11-25 exons, regionally targeted cytogenomic array analysis)
81406	Molecular pathology procedure, Level 7 (eg, analysis of 11-25 exons by DNA sequence analysis, mutation scanning or duplication/deletion variants of 26-50 exons, cytogenomic array analysis for neoplasia)
0234U	MECP2 (methyl CpG binding protein 2) (eg, Rett syndrome), full gene analysis, including small sequence changes in exonic and intronic regions, deletions, duplications, mobile element insertions, and variants in non-uniquely mappable regions Proprietary test: Genomic Unity® MECP2 Analysis Lab/Manufacturer: Variantyx Inc

Current Procedural Terminology© American Medical Association. All Rights reserved.

Procedure codes appearing in Medical Policy documents are included only as a general reference tool for each policy. They may not be all-inclusive.

IX. Evidence-based Scientific References

AAP. (2019). AAP Publications Reaffirmed or Retired. *Pediatrics*, 145(3), e20193991. <https://doi.org/10.1542/peds.2019-3991>

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(2), 185-188. <https://doi.org/10.1038/13810>

Archer, H., Evans, J., Leonard, H., Colvin, L., Ravine, D., Christodoulou, J., Williamson, S., Charman, T., Bailey, M. E., Sampson, J., de Klerk, N., & Clarke, A. (2007). Correlation between clinical severity in patients with Rett syndrome with a p.R168X or p.T158M

- MECP2 mutation, and the direction and degree of skewing of X-chromosome inactivation. *J Med Genet*, 44(2), 148-152. <https://doi.org/10.1136/jmg.2006.045260>
- 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). FOXP1 is responsible for the congenital variant of Rett syndrome. *Am J Hum Genet*, 83(1), 89-93. <https://doi.org/10.1016/j.ajhg.2008.05.015>
- Armstrong, D. D. (1997). Review of Rett syndrome. *J Neuropathol Exp Neurol*, 56(8), 843-849. <https://www.ncbi.nlm.nih.gov/pubmed/9258253>
- Bahi-Buisson, N., Nectoux, J., Rosas-Vargas, H., Milh, M., Boddaert, N., Girard, B., Cances, C., Ville, D., Afenjar, A., Rio, M., Heron, D., N'Guyen Morel, M. A., Arzimanoglou, A., Philippe, C., Jonveaux, P., Chelly, J., & Bienvenu, T. (2008). Key clinical features to identify girls with CDKL5 mutations. *Brain*, 131(Pt 10), 2647-2661. <https://doi.org/10.1093/brain/awn197>
- Banerjee, A., Miller, M. T., Li, K., Sur, M., & Kaufmann, W. E. (2019). Towards a better diagnosis and treatment of Rett syndrome: a model synaptic disorder. *Brain*, 142(2), 239-248.
- Bassuk, A. G. (2021). Gene therapy for Rett syndrome. *Genes, Brain and Behavior, Early View*.
- Belanger, S. A., & Caron, J. (2018). Evaluation of the child with global developmental delay and intellectual disability. *Paediatr Child Health*, 23(6), 403-419. <https://doi.org/10.1093/pch/pxy093>
- Chahil, G., Yelam, A., & Bollu, P. C. (2018). Rett Syndrome in Males: A Case Report and Review of Literature. *Cureus*, 10(10), e3414. <https://doi.org/10.7759/cureus.3414>
- Chapleau, C. A., Calfa, G. D., Lane, M. C., Albertson, A. J., Larimore, J. L., Kudo, S., Armstrong, D. L., Percy, A. K., & Pozzo-Miller, L. (2009). Dendritic spine pathologies in hippocampal pyramidal neurons from Rett syndrome brain and after expression of Rett-associated MECP2 mutations. *Neurobiol Dis*, 35(2), 219-233. <https://doi.org/10.1016/j.nbd.2009.05.001>
- Christodoulou, J., & Ho, G. (1993). MECP2-Related Disorders. In M. P. Adam, H. H. Ardinger, R. A. Pagon, S. E. Wallace, L. J. H. Bean, K. Stephens, & A. Amemiya (Eds.), *GeneReviews(R)*. <https://www.ncbi.nlm.nih.gov/pubmed/20301670>
- Cianfaglione, R., Clarke, A., Kerr, M., Hastings, R. P., Oliver, C., & Felce, D. (2015). A national survey of Rett syndrome: age, clinical characteristics, current abilities, and health. *Am J Med Genet A*, 167(7), 1493-1500. <https://doi.org/10.1002/ajmg.a.37027>
- Colvin, L., Leonard, H., de Klerk, N., Davis, M., Weaving, L., Williamson, S., & Christodoulou, J. (2004). Refining the phenotype of common mutations in Rett syndrome. *J Med Genet*, 41(1), 25-30. <https://www.ncbi.nlm.nih.gov/pubmed/14729826>
- 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. <https://doi.org/10.1016/j.pediatrneurol.2012.11.001>
- Dy, M. E., Waugh, J. L., Sharma, N., O'Leary, H., Kapur, K., D'Gama, A. M., Sahin, M., Urion, D. K., & Kaufmann, W. E. (2017). Defining Hand Stereotypies in Rett Syndrome: A Movement Disorders Perspective. *Pediatr Neurol*, 75, 91-95. <https://doi.org/10.1016/j.pediatrneurol.2017.05.025>
- Elian, M., & de, M. R. N. (1996). Observations on hand movements in Rett syndrome: a pilot study. *Acta Neurol Scand*, 94(3), 212-214. <https://www.ncbi.nlm.nih.gov/pubmed/8899055>

- Feldman, D., Banerjee, A., & Sur, M. (2016). Developmental Dynamics of Rett Syndrome. *Neural Plast*, 2016, 6154080. <https://doi.org/10.1155/2016/6154080>
- Franco, B., & Ballabio, A. (2006). X-inactivation and human disease: X-linked dominant male-lethal disorders. *Curr Opin Genet Dev*, 16(3), 254-259. <https://doi.org/10.1016/j.gde.2006.04.012>
- Fu, C., Armstrong, D., Marsh, E., Lieberman, D., Motil, K., Witt, R., Standridge, S., Nues, P., Lane, J., Dinkel, T., Coenraads, M., von Hehn, J., Jones, M., Hale, K., Suter, B., Glaze, D., Neul, J., Percy, A., & Benke, T. (2020). Consensus guidelines on managing Rett syndrome across the lifespan. *BMJ Paediatr Open*, 4(1), e000717. <https://doi.org/10.1136/bmjpo-2020-000717>
- Fukuda, T., Itoh, M., Ichikawa, T., Washiyama, K., & Goto, Y. (2005). Delayed maturation of neuronal architecture and synaptogenesis in cerebral cortex of Mecp2-deficient mice. *J Neuropathol Exp Neurol*, 64(6), 537-544. <https://www.ncbi.nlm.nih.gov/pubmed/15977646>
- Goldman, S., & Temudo, T. (2012). Hand stereotypies distinguish Rett syndrome from autism disorder. *Mov Disord*, 27(8), 1060-1062. <https://doi.org/10.1002/mds.25057>
- Hagberg, B. (1985). Rett's syndrome: prevalence and impact on progressive severe mental retardation in girls. *Acta Paediatr Scand*, 74(3), 405-408. <https://www.ncbi.nlm.nih.gov/pubmed/4003065>
- Hagberg, B., Aicardi, J., Dias, K., & 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(4), 471-479. <https://doi.org/10.1002/ana.410140412>
- Henriksen, M. W., Ravn, K., Paus, B., von Tetzchner, S., & Skjeldal, O. H. (2018). De novo mutations in SCN1A are associated with classic Rett syndrome: a case report. *BMC Med Genet*, 19(1), 184. <https://doi.org/10.1186/s12881-018-0700-z>
- Hyman, S. L., Levy, S. E., & Myers, S. M. (2020). Identification, Evaluation, and Management of Children With Autism Spectrum Disorder. *Pediatrics*, 145(1), e20193447. <https://doi.org/10.1542/peds.2019-3447>
- Ip, J. P. K., Mellios, N., & Sur, M. (2018). Rett syndrome: insights into genetic, molecular and circuit mechanisms. *Nat Rev Neurosci*, 19(6), 368-382. <https://doi.org/10.1038/s41583-018-0006-3>
- Kang, S. K., Kim, S. T., Johnston, M. V., & Kadam, S. D. (2014). Temporal- and Location-Specific Alterations of the GABA Recycling System in Mecp2 KO Mouse Brains. *J Cent Nerv Syst Dis*, 6, 21-28. <https://doi.org/10.4137/JCNSD.S14012>
- Keogh, C., Pini, G., Dyer, A. H., Bigoni, S., DiMarco, P., Gemo, I., Reilly, R., & Tropea, D. (2018). Clinical and genetic Rett syndrome variants are defined by stable electrophysiological profiles. *BMC Pediatr*, 18(1), 333. <https://doi.org/10.1186/s12887-018-1304-7>
- Khajuria, R., Gupta, N., Roozendaal, K. V., Sapra, S., Ghosh, M., Gulati, S., Smeets, E., Curfs, L., & Kabra, M. (2020). Spectrum of MECP2 mutations in Indian females with Rett Syndrome - a large cohort study.
- Kishi, N., & Macklis, J. D. (2010). MeCP2 functions largely cell-autonomously, but also non-cell-autonomously, in neuronal maturation and dendritic arborization of cortical pyramidal neurons. *Exp Neurol*, 222(1), 51-58. <https://doi.org/10.1016/j.expneurol.2009.12.007>
- Knight, V. M., Horn, P. S., Gilbert, D. L., & Standridge, S. M. (2016). The Clinical Predictors That Facilitate a Clinician's Decision to Order Genetic Testing for Rett Syndrome. *Pediatr Neurol*, 63, 66-70. <https://doi.org/10.1016/j.pediatrneurol.2016.06.016>

- Lallar, M., Rai, A., Srivastava, P., Mandal, K., Gupta, N., Kabra, M., & Phadke, S. R. (2018). Molecular Testing of MECP2 Gene in Rett Syndrome Phenotypes in Indian Girls. *Indian Pediatr*, 55(6), 474-477. <https://www.ncbi.nlm.nih.gov/pubmed/29428920>
- Leonard, H., Cobb, S., & Downs, J. (2017). Clinical and biological progress over 50 years in Rett syndrome. *Nat Rev Neurol*, 13(1), 37-51. <https://doi.org/10.1038/nrneurol.2016.186>
- Liyanage, V. R., & Rastegar, M. (2014). Rett syndrome and MeCP2. *Neuromolecular Med*, 16(2), 231-264. <https://doi.org/10.1007/s12017-014-8295-9>
- Mithyantha, R., Kneen, R., McCann, E., & Gladstone, M. (2017). Current evidence-based recommendations on investigating children with global developmental delay. *Arch Dis Child*, 102(11), 1071-1076. <https://doi.org/10.1136/archdischild-2016-311271>
- Moeschler, J. B., & Shevell, M. (2014). Comprehensive evaluation of the child with intellectual disability or global developmental delays. *Pediatrics*, 134(3), e903-918. <https://doi.org/10.1542/peds.2014-1839>
- Mroch, A. R., Flanagan, J. D., & Stein, Q. P. (2012). Solving the puzzle: case examples of array comparative genomic hybridization as a tool to end the diagnostic odyssey. *Curr Probl Pediatr Adolesc Health Care*, 42(3), 74-78. <https://doi.org/10.1016/j.cppeds.2011.10.003>
- Naidu, S., Murphy, M., Moser, H. W., & Rett, A. (1986). Rett syndrome--natural history in 70 cases. *Am J Med Genet Suppl*, 1, 61-72. <https://www.ncbi.nlm.nih.gov/pubmed/3087204>
- 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, C. (2010). Rett syndrome: revised diagnostic criteria and nomenclature. *Ann Neurol*, 68(6), 944-950. <https://doi.org/10.1002/ana.22124>
- NORD. (2019). *Rett Syndrome*. <https://rarediseases.org/rare-diseases/rett-syndrome/>
- Percy, A. K., Lane, J., Annese, F., Warren, H., Skinner, S. A., & Neul, J. L. (2018). When Rett syndrome is due to genes other than MECP2. *Transl Sci Rare Dis*, 3(1), 49-53. <https://doi.org/10.3233/trd-180021>
- Pidcock, F. S., Salorio, C., Bibat, G., Swain, J., Scheller, J., Shore, W., & Naidu, S. (2016). Functional outcomes in Rett syndrome. *Brain Dev*, 38(1), 76-81. <https://doi.org/10.1016/j.braindev.2015.06.005>
- Pini, G., Bigoni, S., Congiu, L., Romanelli, A. M., Scusa, M. F., Di Marco, P., Benincasa, A., Morescalchi, P., Ferlini, A., Bianchi, F., Tropea, D., & Zappella, M. (2016). Rett syndrome: a wide clinical and autonomic picture. *Orphanet J Rare Dis*, 11(1), 132. <https://doi.org/10.1186/s13023-016-0499-7>
- Reichow, B., George-Puskar, A., Lutz, T., Smith, I. C., & Volkmar, F. R. (2015). Brief report: systematic review of Rett syndrome in males. *J Autism Dev Disord*, 45(10), 3377-3383. <https://doi.org/10.1007/s10803-015-2519-1>
- Rett, A. (1966). [On a unusual brain atrophy syndrome in hyperammonemia in childhood]. *Wien Med Wochenschr*, 116(37), 723-726. <https://www.ncbi.nlm.nih.gov/pubmed/5300597> (Über ein eigenartiges hirnatrophisches Syndrom bei Hyperammonämie im Kindersalter.)
- Santosh, P., Lievesley, K., Fiori, F., & Singh, J. (2017). Development of the Tailored Rett Intervention and Assessment Longitudinal (TRIAL) database and the Rett Evaluation of Symptoms and Treatments (REST) Questionnaire. *BMJ Open*, 7(6), e015342. <https://doi.org/10.1136/bmjopen-2016-015342>
- Schaefer, G. B., & Mendelsohn, N. J. (2013). Clinical genetics evaluation in identifying the etiology of autism spectrum disorders: 2013 guideline revisions. *Genet Med*, 15(5), 399-407. <https://doi.org/10.1038/gim.2013.32>

- Sheinerman, K., Djukic, A., Tsivinsky, V. G., & Umansky, S. R. (2019). Brain-enriched microRNAs circulating in plasma as novel biomarkers for Rett syndrome. *PLoS One*, *14*(7), e0218623. <https://doi.org/10.1371/journal.pone.0218623>
- Shovlin, S., & Tropea, D. (2018). Transcriptome level analysis in Rett syndrome using human samples from different tissues. *Orphanet J Rare Dis*, *13*(1), 113. <https://doi.org/10.1186/s13023-018-0857-8>
- Singh, J., & Santosh, P. (2018). Key issues in Rett syndrome: emotional, behavioural and autonomic dysregulation (EBAD) - a target for clinical trials. *Orphanet J Rare Dis*, *13*(1), 128. <https://doi.org/10.1186/s13023-018-0873-8>
- Smrt, R. D., Eaves-Egenes, J., Barkho, B. Z., Santistevan, N. J., Zhao, C., Aimone, J. B., Gage, F. H., & Zhao, X. (2007). Mecp2 deficiency leads to delayed maturation and altered gene expression in hippocampal neurons. *Neurobiol Dis*, *27*(1), 77-89. <https://doi.org/10.1016/j.nbd.2007.04.005>
- Sun, Y., Gao, Y., Tidei, J. J., Shen, M., Hoang, J. T., Wagner, D., & Zhao, X. (2018). Loss of MeCP2 in immature neurons leads to impaired network integration. *Hum Mol Genet*. <https://doi.org/10.1093/hmg/ddy338>
- Suter, B., Treadwell-Deering, D., Zoghbi, H. Y., Glaze, D. G., & Neul, J. L. (2014). Brief report: MECP2 mutations in people without Rett syndrome. *J Autism Dev Disord*, *44*(3), 703-711. <https://doi.org/10.1007/s10803-013-1902-z>
- Vidal, S., Brandi, N., Pacheco, P., Gerotina, E., Blasco, L., Trotta, J. R., Derdak, S., Del Mar O'Callaghan, M., Garcia-Cazorla, A., Pineda, M., & Armstrong, J. (2017). The utility of Next Generation Sequencing for molecular diagnostics in Rett syndrome. *Sci Rep*, *7*(1), 12288. <https://doi.org/10.1038/s41598-017-11620-3>
- Vidal, S., Pascual-Alonso, A., Rabaza-Gairi, M., Gerotina, E., Brandi, N., Pacheco, P., Xiol, C., Pineda, M., & Armstrong, J. (2019). Characterization of large deletions of the MECP2 gene in Rett syndrome patients by gene dosage analysis. *Mol Genet Genomic Med*, *7*(8), e793. <https://doi.org/10.1002/mgg3.793>
- Villard, L. (2007). MECP2 mutations in males. *J Med Genet*, *44*(7), 417-423. <https://doi.org/10.1136/jmg.2007.049452>
- Weaving, L. S., Williamson, S. L., Bennetts, B., Davis, M., Ellaway, C. J., Leonard, H., Thong, M. K., Delatycki, M., Thompson, E. M., Laing, N., & Christodoulou, J. (2003). Effects of MECP2 mutation type, location and X-inactivation in modulating Rett syndrome phenotype. *Am J Med Genet A*, *118A*(2), 103-114. <https://doi.org/10.1002/ajmg.a.10053>
- Williamson, S. L., & Christodoulou, J. (2006). Rett syndrome: new clinical and molecular insights. *Eur J Hum Genet*, *14*(8), 896-903. <https://doi.org/10.1038/sj.ejhg.5201580>
- Xiol, C., Heredia, M., Pascual-Alonso, A., Oyarzabal, A., & Armstrong, J. (2021). Technological Improvements in the Genetic Diagnosis of Rett Syndrome Spectrum Disorders. *Int J Mol Sci*, *22*(19). <https://doi.org/10.3390/ijms221910375>
- Zhang, Q., Zhao, Y., Bao, X., Luo, J., Zhang, X., Li, J., Wei, L., & Wu, X. (2017). Familial cases and male cases with MECP2 mutations. *Am J Med Genet B Neuropsychiatr Genet*, *174*(4), 451-457. <https://doi.org/10.1002/ajmg.b.32534>
- Zhang, X., Bao, X., Zhang, J., Zhao, Y., Cao, G., Pan, H., Zhang, J., Wei, L., & Wu, X. (2012). Molecular characteristics of Chinese patients with Rett syndrome. *Eur J Med Genet*, *55*(12), 677-681. <https://doi.org/10.1016/j.ejmg.2012.08.009>

X. Review/Revision History

Effective Date	Summary
12/01/2024	Initial Policy Implementation