Appendix 4 Characteristics of included studies

Author	Boucher
Year	2015
Country	USA
Ref #	[1]
Study design	Retrospective review
Recruitment	All patients undergoing HSCT for the diagnosis of MLD were identified from the prospectively maintained University of Minnesota Blood and Marrow Transplant Database
Population	Not applicable
Inclusion criteria	MLD diagnosis could be retrospectively confirmed were included
Follow up	Twenty-one patients (53%) are alive at a median posttransplant follow-up of 10 years.
Intervention	Hematopoietic stem cell transplantation (HSCT)
Participants (n)	3: 1 late infantile, 2 early juvenile
Drop-outs (n)	
Comparison	Natural history of disease
Participants (n)	3: 1 late infantile 2 early juvenile
Drop-outs (n)	1 late infantile, 2 early juvenile
Outcomes	GMFC-MLD, ELFC-MLD, ADL, VABS, mortality
Comments	
Risk of bias	High

ADL = Age at loss of independent performance of common activities of daily livning; **ELFC** = Expressive Language Function Classification; **GMFC** = Gross Motor Function Classification; **HSCT** = Hematopoietic stem-cell transplantation; **MLD** = metachromatic leukodystrophy**; VABS** = Vineland Adaptive Behavior Scales

Author	Chen
Year	2016
Country	China
Ref #	[2]
Study design	Case-control
Recruitment	Pre symptomatic siblings of affected children
Population	Not applicable
Inclusion criteria	Diagnosed based on the family history. Two patients underwent targeted gene tests, which confirmed the diagnosis.
Follow up	6 years for one sibling group and 14 years for other sibling pair
Intervention	Allogenic HSCT
Participants (n)	Early juvenile: 3 (2 pre-symptomatic, 1 possible early symptomatic)
Drop-outs (n)	
Comparison	Natural history of disease
Participants (n)	Early juvenile: 2
Drop-outs (n)	
Outcomes	WPPSI-III, WISC-IV, clinical assessment
Comments	
Risk of bias	High

WISC-IV = Wechsler Intelligence Scale for Children, Fourth Edition; **WPPSI-III** = Wechsler Preschool and Primary Scale of Intelligence, Third Edition

Author	Fumagalli
Year	2022
Country	Italy
Ref #	[3]
Study design	Prospective, non-randomised
Recruitment	No information
Population	Patients having presymptomatic early-juvenile-onset metachromatic leukodystrophy who underwent treatment with umbilical cord blood transplantation
Inclusion criteria	Eligible patients had a molecular and biochemical diagnosis of MLD of either pre-symptomatic late infantile or pre-symptomatic or early-symptomatic early-juvenile variants
Follow up	Median 3,2 years for intervention group (patients totally followed between 0,64–7,51 years)
Intervention	Autologous haematopoietic stem and progenitor cell (HSPC) population transduced ex vivo gene therapy (Arsa-cel) (HSCT-GT)
Participants (n)	16 late infantile, 13 Early juvenile
Drop-outs (n)	3
Comparison	Non-interventional natural history
Participants (n)	Late infantile: 19, Early juvenile: 12
Drop-outs (n)	
Outcomes	Total GMFM score, sMFS, Mortality
Comments	
Risk of bias	High

GMFM score = Gross Motor Function Measure Score; **SMFS** = severe motor impairment-free survival

Author	Hong
Year	2021
Country	USA
Ref #	[4]
Study design	Screening study
Recruitment	Existing DBS in ongoing NBS program
Population	Newborns
Inclusion criteria	Not specified
Follow up	
Indextest	Dried blood-spot test, two-tier screening (Sulfatide screening, ARSA enzyme testing), C16:0. Genetic sequencing
Participants (n)	Tier 1: 27 335, Tier 2: 122
Drop-outs (n)	Drop out tier 1 to 2: 73 of 195 viable
stepwise	
Reference test	Not specified how reference group was diagnosed
Participants (n)	15
Drop-outs (n)	
Outcomes	
Comments	
Risk of bias	Not applicable

DBS = Dried Blood Spot; **NBS** = New Born Screening

Author	Kasapkara
Year	2024
Country	Turkey
Ref #	[5]
Study design	Retrospective cohort study
Recruitment	MLD who were diagnosed in the centre
Population	Not applicable
Inclusion criteria	MLD diagnosis through proven biochemical and genetic defects, ARSA gene sequencing and ASA activity
Follow up	Intervention: 6 months for early juvenile and 30 months for late infantile Comparison: 12-44 months
Intervention	Allogenic HSCT
Participants (n)	Late infantile: 1; Early juvenile: 1
Drop-outs (n)	
Comparison	Natural history of disease, Symptomatic treatment
Participants (n)	Late infantile: 5, Early juvenile: 1
Drop-outs (n)	1 death
Outcomes	Mortality, GMFC-MLD
Comments	
Risk of bias	High

GMFC = Gross Motor Function Classification

Author	Laugwitz
Year	2024
Country	Germany
Ref #	[6]
Study design	Cohort screening study
Setting	Hospitals
Recruitment	
Population	Neonates
Inclusion criteria	(1) the legal guardian´s consent, (2) the collection of dried blood spot (DBS) samples within the first 36–72 hours of life following the national guidelines for NBS in Germany, and (3) the availability of residual DBS specimen after completing the regular national NBS program.
Follow up	
Index test	Three-tiered screening (Sulfatide screening, ARSA enzyme testing, Genetic testing)
Participants (n)	Tier 1: 109 259; Tier 2: 230; Tier 3: 381
Drop-outs (n)	Drop out tier 1 to 2: 151 of 381 viable. All viable from tier 1 was used in tier 3.
Reference test	Confirmatory diagnostics, clinical assessment and subtype prediction by a qualified treatment centre + positive controls from other concurrent pilot studies
Participants (n)	3 (screen-positive in third-tier test)
Drop-outs (n)	
Outcomes	Confirmed MLD diagnosis
Comments	
Risk of bias	Not applicable

DBS = Dried Blood Spot**; NBS** = New Born Screening

Author	Pridjian
Year	1994
Country	USA
Ref #	[7]
Study design	Case-control
Recruitment	No information for comparison, intervention participant recruited through diagnosed sibling
Population	Not applicable
Inclusion criteria	MLD diagnosis through leukocyte ASA, fibroblast ASA, un-metabolized sulfatide findings
Follow up	42 months
Intervention	Allogenic HSCT
Participants (n)	Late infantile: 1 sibling pre-symptomatic
Drop-outs (n)	
Comparison	Natural history of disease
Participants (n)	Late infantile: 1 sibling
Drop-outs (n)	1 death
Outcomes	Mortality and clinical assessment of psychometric development
Comments	
Risk of bias	High

ASA = Acetylsalicylic Acid

Author	Van Rappard
Year	2016
Country	The Netherlands
Ref #	[8]
Study design	Retrospective cohort
Recruitment	In order to assess HCT efficacy, we evaluated all 35 consecutive MLD patients presenting between 2004 and 2015 in our department, the Dutch Leukodystrophy Referral Center.
Population	Not applicable
Inclusion criteria	Patients with a total intelligence quotient (IQ) above 70 and without gross neurological signs (ie, ambulation without support, no dysphagia) were considered HCT candidates
Follow up	Transplanted patients were followed for a mean duration of 4.7 years
Intervention	Allogenic HSCT
Participants (n)	5 total, 2 late infantile and 3 early juvenile
Drop-outs (n)	1 death late infantile
Comparison	Natural history of disease
Participants (n)	9 total, 6 late infantile and 3 early juvenile
Drop-outs (n)	4 deaths late infantile, 2 death early juvenile
Outcomes	Mortality, GMFC-MLD, cognition (BSID-II-NL)
Comments	Early juvenile patients MLD-12, MLD-17, MLD-18; controls MLD-4, MLD-16, MLD-37.
Risk of bias	High

BSID-11-NL = Bayley Scales of Infant Development, Second Edition, Dutch Version

Author	Wu
Year	2024
Country	United Kingdom
Ref #	[9]
Study design	Pre-Pilot Newborn Screening Study
Recruitment	From newborn screening laboratory
Population	Neonates
Inclusion criteria	Not specified but exclusion criteria: Parental decline of any research be performed on baby's residual NBS bloodspot. Bloodspots collected from babies ≤4 days or > 12 months of age, rejected due to blood transfusion, or of poor quality
Follow up	
Index test	Two-tiered screening (Sulfatide screening, ARSA enzyme testing)
Participants (n)	Tier 1: 3697; Tier 2: 11; Tier 3: 0
Drop-outs (n)	
Reference test	Not specified
Participants (n)	
Drop-outs (n)	
Outcomes	
Comments	
Risk of bias	Not applicable

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