Newborn Screening Advisory Committee Meeting
Tuesday, July 29, 2025, from 8:30 a.m. – 12:00 p.m.
Nebraska Children's Home Society – Omaha

Committee Members	Specialty	Present	Absent
Peter Abasolo, MD	Pathologist	X	
Jill Allen, RN	Nebraska Hospital Association Rep.		X
Khalid Awad, MD	Chair, Neonatologist	X	
Jill Beck, MD, MS	UNMC Pediatric Hematology/Oncology		X
Angela Brennan, MD	Family Physician	X	
Zoe González-García, MD	Pediatric Endocrinologist		X
Jessica Hansen, MSN, FNP-C	APRN	X	
James Harper, MD	Pediatric Hematologist	X	
Kathryn Heldt, RD	Dietitian/Metabolic Specialty	X	
Isabella Herman, MD	Pediatric Neurogenetics		X
Claire Jackson, MD	Hematology	X	
Alyssa Keller, MS, CGC	Certified Genetic Counselor	X	
Tiffany Moore, PhD, RN	Parent Advocate	X	
Hana Niebur, MD	Vice-Chair, Pediatric Immunology		X
Elizabeth Null, MD, FAAP, FACMG	Department of Pediatrics, Division of Inherited Metabolic Diseases, UNMC		X
Erin Pierce, MD	General Pediatrics	X	
Robert Rauner	X-ALD Advocate	X	
William Rizzo, MD	Pediatric Genetic/Metabolic	X	
Jill Skrabal, PhD, RD	Dietitian/Metabolic	X	
Heather Thomas, MD	CF Clinic Director	X	
Jennifer Wallin	Parent Advocate	X	
Emeritus Members			
Richard Lutz, MD	Pediatric Genetic/Endo/Metabolic	X	
Deborah Perry, MD	Pathologist	X	
Laboratory			
PJ Borandi	Revvity NBS Lab	X	
Meredith Patik	Revvity NBS Lab	X	
Staff			
Erin Bryant, RN, BSN	Nurse Navigator, Hematology Sickle Cell Disease & Infant Hemoglobinopathies	X	
Jillian Chance, DNP, RN, APRN	NBS Program Manager	X	
Michaela Howard, RD	NBS Inherited Diseases Clinical Specialist	X	
Yousif Ibrahim	NBS Inherited Diseases Clinical Specialist	X	
Tyra Mills, MPH	Bioinformatics Fellow	X	
Derek Ross	Birth Defects Registry Health Data Coordinator	X	
Krizia Sanoy	Newborn Screen Systems Coordinator	X	
Tim Tesmer, MD	Nebraska DHHS CMO	X	
Sarah Ward, MPH	NBS Inherited Diseases Clinical Specialist	X	
Guests & Speakers			

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Amy Brower, PhD	American College of Medical Genetics and Genomics (ACMG)	**X**	
Catherine Jirikowic	Genetic Counselor	**X**	
Craig Baker	UNMC Biochemical Geneticist	**X**	
Chandler Morton	Pediatric Resident UNMC and Children's	**X**	

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Open Meeting Act Announcement

Dr. Khalid Awad, the chair, called the July 29, 2025, quarterly meeting for the Nebraska Newborn Screening Advisory Committee to order at 8:30 a.m. He announced that, under the Nebraska Statute (Opening Meeting Act), a notice of this public meeting is posted on the State of Nebraska Public Meeting Calendar, and a copy of the Open Meeting Act and the agenda for the meeting are posted on the wall by the entrance of the meeting space.

Introduction

Dr. Khalid Awad asked the guests, personnel, advisory committee members, and new advisory committee members to introduce themselves. Amy Brower, PhD (American College of Medical Genetics and Genomics (ACMG)), Catherine Jirikowic (Genetic Counselor), Craig Baker, MD (UNMC Biochemical Geneticist), and Chandler Morton (Pediatric Resident, UNMC and Children's) all attended as guests.

Review/Approval of Minutes from the April 2025 Meeting

The approval for April 29, 2025, was brought up by Dr. Khalid Awad, who presented the minutes. The motion to approve was made by Dr. William Rizzo, seconded by Dr. Angela Brennan, and passed unanimously by all committee members.

Laboratory Update

Revvity provided an update on a new three-tiered approach for Metachromatic Leukodystrophy (MLD) screening, designed to reduce the need for sequencing. The tiers include sulfatides, ARSA, and sequencing. The ARSA tier aims to minimize cases needing sequencing by resolving most abnormalities early. The goal is to begin implementation on January 1, 2026, with data collection throughout the year. Revvity is following standard assay validation for the approach before moving into clinical testing, without a separate pilot phase.

Questions were raised about the sufficiency of dried blood spots. It was clarified that no extra spot is needed if samples are collected correctly, although limits are being tested through multi-tier retesting. Multiplexing is being explored to increase efficiency.

A comparison was made to tandem mass spectrometry, highlighting potential efficiency gains if assays are compatible. Nebraska was commended for having one of the lowest unsatisfactory specimen rates, which reduces the risk of insufficient samples.

It was noted that MLD was scheduled for a federal vote, but the committee was dissolved before the meeting, leaving future disorder nominations uncertain. Duchenne Muscular Dystrophy (DMD) was also on the agenda.

Concerns were raised about an increase in flagged variants, such as Barts, in recent months. It was confirmed that a standard six-mutation panel is used across all individual screens.

Recurring shipping delays were discussed. Nearly all disorders were reported just within or beyond time-critical limits. Delays were partly due to the inability to print Saturday delivery labels unless they were created on Fridays, and Next Day Air shipping arrangements were not being fulfilled. Efforts are underway to improve coordination with the carrier, including reinstating weekly meetings with the dispatch team and working toward allowing Saturday delivery codes on any day's label. UPS established meetings about a year ago, and follow-up on label coding is scheduled soon. Committee members raised the possibility of alternative courier services, but a definitive answer wasn't available. Revvity staff clarified that courier services are bundled in the existing contract the program has with Revvity, so issues are addressed directly with the contracted partner.

Legislation, Regulation, and Policy

There have been ongoing efforts to reform or reestablish a national committee structure for newborn screening. One major initiative is being led by ACMG, which has expressed interest in forming a body similar to the former Secretary's Advisory Committee. ACMG has begun reaching out to stakeholders and former committee members as part of this initiative.

The organization also supports revisiting the Newborn Screening Saves Lives Act, which initially established the advisory committee in statute. Although the act was scheduled for reauthorization in 2019, it has since stalled in Congress. While the House of Representatives passed it at one point, it failed in the Senate despite ongoing advocacy and movement through various committees. The absence of the national advisory committee has renewed calls to close this legislative gap.

Further updates confirmed the existence of a grassroots movement in support of restoring the committee. This includes a significant advocacy event held in Washington, D.C., where numerous disease-focused organizations met with legislators to emphasize the importance of the committee's role in improving patient outcomes. There is widespread recognition among advocates that the committee plays a critical role in the newborn screening community. There was also discussion of emerging priorities, such as advocating for the inclusion of Metachromatic Leukodystrophy (MLD) in the Recommended Uniform Screening Panel (RUSP), with several states, including Nebraska, leading the charge. The National Organization for Rare Disorders (NORD) continues to play an active role in supporting national newborn screening efforts.

Additional advocacy efforts included participation in Rare Disease Week, where advocates were trained and mobilized across Capitol Hill. Discussions with congressional representatives focused on the importance of the advisory committee in guiding federal research investments, particularly for conditions such as SCID, SMA, and Duchenne Muscular Dystrophy. These engagements aimed to underscore the committee's influence in expanding and improving screening and treatment programs. In May, advocates participated in the Rare Disease Caucus to

address the consequences of the advisory committee's dissolution and the risks posed to the infrastructure that the ACHDNC developed over decades.

Looking ahead, a virtual workshop hosted by Every Life Foundation is scheduled for August 21. The event will feature former advisory committee members and will focus on MLD, Duchenne muscular dystrophy, and ongoing efforts to maintain momentum in newborn screening advancements. Although not a replacement for the former committee, the workshop is intended to carry forward its essential work. It was also noted that the National Academies recently completed a comprehensive review of newborn screening, highlighting areas for improvement and calling for the broader inclusion of diverse voices in shaping the program's future.

It was reported that a larger publication from the National Academies will be distributed, following an earlier release of the executive summary a few months ago. The Nebraska Newborn Screening Program is currently dependent on the Secretary's Advisory Committee for oversight of conditions grandfathered into the Affordable Care Act. These conditions fall under preventive care, meaning insured individuals should be provided these services at no cost, an essential factor in keeping newborn screening affordable. At present, this arrangement remains in place; however, there is uncertainty about its long-term sustainability, as the Secretary's Advisory Committee, which previously updated condition lists and guided insurance and CMS-related decisions, is no longer active. There is hope for a replacement or revitalization of the committee, given its critical role.

A committee member noted that the Nebraska Advisory Committee has the authority, within the state, to recommend additions to the newborn screening panel. At the national level, ACMG is leveraging its expertise in methodology to publish clinical guidelines and identify candidate conditions for review. One challenge with the former committee's process was that conditions were evaluated individually, often resulting in long delays. With advancements in genomics, there may now be dozens or even hundreds of potential conditions for review. ACMG is working to contribute its expertise to streamline evaluations and provide recommendations.

During state updates, staff members stated that it is likely another bill will be introduced in the next legislative session, again seeking to remove the mandate for newborn screening. The legislator, who previously introduced the bill twice in the past, has one remaining term. Additionally, the Early Hearing Detection and Intervention (EHDI) program successfully secured CDC funding for another year. However, the program's long-term budget outlook is uncertain. The presidential budget proposal for 2026 includes cuts that would eliminate HRSA EHDI funding, which represents the most significant portion of the program's grant support in Nebraska. If this occurs, the Department of Health and Human Services may need to request an increase in the newborn screening fee from the legislature. EHDI currently operates without a fee or state funding; therefore, the loss of federal support would necessitate the use of state dollars to continue operations. A fee increase could also help prepare for the anticipated addition of more screening conditions, which would require additional resources.

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Review of Completed Evidence Review of GAMT and Krabbe

Dr. Brower provided a comprehensive overview of Nebraska's evidence review concerning two conditions recently added to the federal Recommended Uniform Screening Panel (RUSP). She reflected on her earlier involvement with the committee in 2005, noting that both conditions had been first discussed nearly two decades ago, and remarked on the significant progress made since then. Advances in technology and treatment options have reignited interest in incorporating these conditions into newborn screening panels.

Regarding Krabbe disease, key developments have included enhancements in diagnostic technology, advancements in hematopoietic stem cell transplantation techniques, and the emergence of novel cell and gene therapies. In the case of GAMT deficiency, the committee's primary challenge was the rarity of the disorder. Although limited case reports and complicated assessments, as well as family studies, have demonstrated the substantial benefits of early detection, often identifying affected younger siblings before symptoms develop, thereby enabling pre-symptomatic treatment. While supplementation through medical foods poses particular challenges for families, it remains a relatively straightforward and highly effective intervention for GAMT deficiency.

Dr. Brower reviewed the Secretary's advisory committee's decisions to add Krabbe disease in 2024 and GAMT deficiency in 2023. She conducted an extensive literature review and consulted national experts to evaluate developments since the federal evidence reviews were completed. She also assessed the potential impact of including these conditions on Nebraska's newborn screening panel.

In discussing Krabbe disease, Dr. Brower explained that it is a rare, often fatal disorder most effectively treated within the first 30 days of life through hematopoietic stem cell transplantation. Newborn screening has improved the understanding of its incidence. Earlier estimates suggested a rate of approximately 1 in 100,000 live births; however, population-based screening in states such as New York now indicates a U.S. incidence closer to 1 in 250,000 to 310,000. For Nebraska, this translates to roughly one case every 13 to 14 years, though variation is possible.

Krabbe disease manifests in two subtypes: a severe infantile form with onset before 12 months of age, and a later-onset form characterized by slower progression. Timely diagnosis is critical; transplantation within the first 30 to 45 days of life can slow but not cure the infantile form. A significant factor contributing to Krabbe's recent inclusion on the RUSP was the development of a reliable two-tier screening approach incorporating a second-tier psychosine measurement, which improved test specificity and reduced false positives.

Nebraska currently utilizes the Mayo Clinic's laboratory, which offers confirmatory Krabbe disease screening. However, the state lacks an in-state pediatric stem cell transplant center with sufficient experience, making referrals to out-of-state centers essential. Established centers of excellence, including the University of Minnesota and facilities in Pennsylvania, already collaborate with Nebraska on other transplant-requiring conditions. Implementation would

necessitate a rapid referral process, 24/7 notification protocols, insurance coordination, and strong partnerships with transplant centers.

Dr. Brower emphasized that some families may decline transplantation, underscoring the importance of preparing for ongoing support regardless of treatment decisions. She recommended Nebraska:

- Adopt the two-tier infantile Krabbe disease screening method, including psychosine measurement.
- Establish a rapid, coordinated referral system for out-of-state treatment.
- Develop partnerships with transplant centers to ensure long-term follow-up.
- Provide training for staff and clinicians on Krabbe disease management and psychosine level interpretation.
- Track long-term outcomes for all identified cases, including those that do not undergo transplantation.
- Ensure equitable access to information and care, addressing social determinants of health.

In conclusion, although the expected incidence of infantile Krabbe disease in Nebraska is low, the benefits of early detection and intervention are substantial. Effective implementation will require robust infrastructure, staffing, and interstate collaboration. By incorporating federal recommendations and adapting them to Nebraska's context, the state can maintain its leadership in newborn screening and optimize outcomes for affected families.

In response, Dr. Chance outlined the considerable challenges families face when a child requires an out-of-state transplant, including prolonged separation from home, loss of income, and the cost of temporary housing. She noted that these burdens can lead some families to decline treatment. Dr. Chance reported that discussions with Judy Thomas, service line and nursing leader, confirmed active efforts to reestablish stem cell transplantation services in Nebraska, with implementation targeted for 2026 or early 2027 at the Children's Hospital. She emphasized both the current hardship resulting from the absence of in-state services and the prospect that this issue may be resolved before the panel's next review.

Dr. Brower then presented on the second condition, GAMT deficiency. This rare disorder affects creatine biosynthesis and, if untreated, results in intellectual disability, epilepsy, and behavioral challenges. After an extensive review, GAMT deficiency was recommended for inclusion in the RUSP in 2022. The recommendation process involves an evidence review, committee vote, and submission to the Secretary of Health and Human Services (HHS), who then endorses or rejects the addition. In January 2023, the Secretary endorsed the inclusion of GAMT deficiency. Dr. Brower's detailed review reflects this endorsement.

Similar to the process for Krabbe disease, several key factors support the inclusion of GAMT deficiency in Nebraska's newborn screening panel. Early detection enables prompt treatment with creatine supplementation, medical foods, and dietary modifications, preventing neurological symptoms when initiated pre-symptomatically. The first-tier mass spectrometry test is low-cost and can be multiplexed with screening for other disorders. Diagnosis is often delayed for years

and can be mistaken for autism spectrum disorder, intellectual disability, or epilepsy, resulting in nonspecific clinical labels. While treatments can address neurological symptoms, the absence of supplementation leads to inevitable neurological decline.

Laboratory methods include a second-tier sequencing approach supported by Revvity, which may also enable expanded first-tier biochemical testing in the future.

Several considerations for implementation were discussed. The incidence of GAMT deficiency is extremely low; published data and the RUSP nomination suggest Nebraska might expect only one case every 20 years. This rarity presents challenges related to long-term follow-up, cost-effectiveness, and maximizing the benefits of newborn screening. Discussions with Mayo Clinic colleagues confirmed that, although clinical cases are rare, substantial follow-up is conducted to interpret screening results and assess their clinical significance. Family studies highlight the benefits of early diagnosis and treatment in younger siblings, who demonstrate markedly improved neurological outcomes compared to older siblings diagnosed later.

Confirmatory testing requires access to specialized metabolic providers and follow-up care, which Nebraska currently possesses. However, psychosocial, logistical, and insurance barriers exist that may delay timely treatment. Medical foods and supplements are often not covered by insurance, and families bear most responsibility for preparation and administration.

Laboratory validation and personnel training would be necessary if the state laboratory does not currently have a validated first-tier test. Short-term follow-up is critical to ensure parents are prepared to initiate treatment within the pre-symptomatic window. Without early supplementation of creatine and ornithine, neurological decline will persist.

Educational materials, including GAMT deficiency fact sheets, algorithms, and videos produced by ACMG, are available and have been utilized in Nebraska for other conditions. Statewide education for primary care providers will be essential, especially given the condition's rarity and the likelihood that many pediatricians may never encounter a case, yet must be prepared to manage it.

Challenges include the very low incidence and the impact of prematurity and total parenteral nutrition (TPN) on mass spectrometry screening results. Additionally, arginase deficiency can produce similar biochemical markers, which can complicate interpretation. Mayo Clinic confirmed the ultra-rare status of GAMT deficiency but emphasized the importance of thorough follow-up for suspected cases.

Actionable recommendations for Nebraska include considering the addition of GAMT deficiency to the newborn screening panel, validating laboratory protocols and cutoff values using existing dried blood spot quality assurance materials from the CDC, and establishing confirmatory testing protocols with in-state metabolic genetics and biochemical experts. Coordination with Revvity Omics and partner laboratories to offer second-tier GAMT gene sequencing is advised. Given the autosomal recessive inheritance pattern, cascade testing within families should also be considered. Educational materials can be tailored for use by clinical and newborn screening staff.

As Nebraska continues to develop its long-term follow-up system, consensus on minimal data elements for monitoring children with GAMT deficiency can be incorporated.

A significant challenge will be preparing for the rare positive screens and ensuring equitable access to care and treatment throughout the state of Nebraska.

In conclusion, although GAMT deficiency is ultra-rare, federal endorsement through the RUSP supports its inclusion in newborn screening programs. A recent Canadian study, published in May 2023, examined four sibling pairs affected by GAMT deficiency alongside eight age-matched healthy siblings. Utilizing mixed methods, including parental reports on behavior and development, the study compared outcomes between siblings diagnosed via newborn screening and those diagnosed clinically at a later stage. Findings demonstrated that early supplementation leads to neurotypical development, highlighting the profound impact of timely intervention.

This evolving clinical evidence underscores the importance of ongoing review and adaptation of newborn screening recommendations to maximize benefits for affected families.

Dr. Brower invited questions and highlighted the availability of valuable resources and videos illustrating the positive impact of early treatment on children's outcomes, despite challenges such as supplement taste and administration.

An update on laboratory screening was provided by PJ, who confirmed that a two-tiered approach has been in place since January 2024. The first-tier screens for key metabolites (guanidinoacetate and creatine) by mass spectrometry, while the second tier involves single-gene sequencing of the GAMT gene. Approximately 250,000 first-tier screens have been conducted to date, indicating a low incidence thus far. The increasing diagnoses of autism and epilepsy make this screening valuable in clarifying underlying causes of neurodevelopmental delays and reducing the diagnostic odyssey experienced by families.

Although the condition remains rare, the straightforward intervention involving medical foods aligns well with the benefits of newborn screening. However, some states report that despite active screening, cases are infrequently detected in clinical practice.

Finally, a proposal to add GAMT deficiency to the Nebraska Newborn Screening Panel was motioned by Dr. William Rizzo, seconded by Alyssa Keller, and passed unanimously with 12 votes in favor and a quorum present.

Please see attached Krabbe and GAMT executive summaries and evidence reviews.

Bioinformatics Fellow Update

Tyra Mills, the bioinformatics fellow, presented data comparing cystic fibrosis (CF) cases in 2023 and 2024, noting a shift from 10 carriers and 9 disease cases in 2023 to 14 carriers and 1 disease case in 2024. Preterm births decreased from 19 in 2023 to 8 in 2024. Tyra analyzed sweat test values and birth weights, finding a significant change in CF indicators. Tyra then

discussed a hematology research project on referral rates, provider specialties, and the impact of letter changes on the referral process. The meeting concluded with updates on birth defects data dashboards and long-term follow-up projects.

Case Management System Update

Dr. Chance provided an update regarding the Natus system, acknowledging that the implementation has encountered delays. As this is her first experience collaborating with a software vendor, she noted that the IT team has assured her that such delays are common in projects of this nature. The current target for the system's go-live date is June 2026.

The new database will serve as the case management system for the Hearing Program, encompassing Critical Congenital Heart Disease (CCHD), dried blood spot screening, and the congenital disabilities registry. A notable advancement was mentioned, including Nebraska Medicine's agreement to begin supplying CCHD case data, potentially including raw data, to facilitate the application of the new algorithm. This collaboration represents the addition of another hospital actively preparing and submitting data.

Dr. Chance also addressed progress on regulatory matters. The department is currently navigating two sets of regulations. Initially, a newly appointed, recently graduated attorney undertook a comprehensive rewrite of the regulatory language. However, following a review by the lead regulations attorney, specific revisions were requested to revert aspects of the changes. The regulations are presently being redrafted and are anticipated to proceed to a public hearing shortly. These regulations pertain to CCHD screening incorporating the new algorithm, as well as the addition of one metabolic panel condition to the screening panel. The department aims to have these changes finalized by the end of 2025.

Furthermore, the department is pursuing a mandate requiring all CCHD testing data, including the total number of tests conducted, referral status, and echocardiogram outcomes, to be reported to the State of Nebraska in any format that hospitals are capable of providing. This initiative aims to transition birth defects data collection from a passive to an active surveillance model, ensuring that confirmed case data and comprehensive screening outcomes are available for ongoing monitoring. Despite adjustments to the project timeline, these developments remain promising and are expected to enhance program capabilities significantly.

Other Business

It was announced that this meeting marked Dr. James Harper's final appearance as a committee member. In recognition of his dedicated service, he was presented with a plaque and an admiralship. To honor Dr. Harper's significant contributions, Children's Nebraska established the Dr. James Harper Hematology Philanthropy Fund, which will provide support for children with sickle cell disease.

Additionally, 20-year service awards were presented to Dr. Khalid Awad, Kathryn Heldt, Dr. Richard Lutz, Dr. William Rizzo, and Dr. Jill Skrabal.

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Data

A closed session of members and staff was held for review of data. Information regarding Nebraska births, including births screened, unscreened births, out-of-hospital births, out-of-hospital births screened, unsatisfactory specimens, confirmed positive cases, confirmed negative cases, repeated normal cases, and pending cases, was provided.

Adjourned

Dr. Khalid Awad adjourned the meeting at 11:38 a.m.

Respectfully Submitted:

Jillian Chance
Michaela Howard
Yousif Ibrahim
Tyra Mills
Derek Ross
Krizia Sanoy
Sarah Ward

Executive Summary: Inclusion of Krabbe Disease in the Nebraska Newborn Screening Panel

Introduction

Krabbe Disease (KD), particularly the infantile form (IKD), is a rapidly progressive neurodegenerative disorder with significant morbidity and early mortality if untreated. Following a robust evidence review and recent inclusion of IKD in the U.S. Recommended Uniform Screening Panel (RUSP) in 2024, Nebraska is evaluating its readiness to include IKD in its newborn screening (NBS) panel. Early detection through NBS enables timely intervention via hematopoietic stem cell transplantation (HSCT), ideally administered within 30 to 45 days of life. However, successful implementation will require Nebraska to develop a rapid diagnostic and referral infrastructure.

Key Justifications for Inclusion

- Krabbe Disease meets RUSP criteria: early identification improves outcomes.

- Second-tier psychosine testing improves specificity and reduces false positives.

- Without screening, diagnosis is delayed and treatment opportunities are missed.

- Nebraska's lab partner Revvity Omics is equipped to perform required tests.

Actionable Recommendations

1. 1. Adopt IKD screening in the Nebraska NBS panel using a two-tier test: GALC enzyme activity followed by DBS psychosine quantification.
2. 2. Develop a statewide rapid diagnosis and referral system to ensure infants are evaluated and prepared for HSCT by day 30–45 of life.
3. 3. Establish partnerships with experienced out-of-state transplant centers and negotiate expedited care and insurance coordination.
4. 4. Engage Revvity Omics for both primary and secondary laboratory testing to ensure timely and accurate results.
5. 5. Train follow-up and clinical staff using ACMG ACT Sheets, clinical algorithms, and video materials to prepare for rare but urgent cases.
6. 6. Build a long-term follow-up system to track health outcomes of identified cases and ensure care continuity.

7. 7. Prepare the public health infrastructure for rare positive screens (~1 IKD case every 13–14 years in Nebraska).
8. 8. Ensure health equity in access, family education, and out-of-state treatment navigation.

Conclusion

While the incidence of IKD in Nebraska is low, the potential benefit of early diagnosis and intervention is profound. Implementing IKD screening will require focused efforts to ensure infrastructure, staffing, and inter-state collaboration are in place. This investment will position Nebraska as a national leader in rare disease detection and childhood health equity.

Considerations

- Detection of later-onset phenotypes remains uncertain and may not yield actionable interventions.
- Psychosocial and logistical burdens on families, including travel and insurance authorization, must be addressed.
- Implementation requires investment in clinical coordination, training, and interstate partnerships.
- Rapid diagnostic confirmation and family decision-making are critical within the newborn's first weeks.
- Nebraska currently lacks an in-state pediatric transplant center, necessitating out-of-state referrals.
- HSCT must occur within 30–45 days of life to be effective, requiring urgent coordination and care infrastructure.
- Low frequency of IKD in Nebraska (estimated 1 case every 13–14 years) raises cost-benefit concerns.

Evidence Review: Krabbe Disease

Amy Brower, PhD
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Clinical Professor
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INTRODUCTION

Newborn screening (NBS) is a cornerstone of public health that enables the early identification and treatment of severe, often life-threatening, genetic disorders. The inclusion of Krabbe Disease (KD), specifically the infantile form (IKD), in the Nebraska NBS panel requires a thorough review of clinical evidence, screening technology, treatment efficacy, and implementation readiness. Krabbe Disease is a rare autosomal recessive lysosomal storage disorder caused by mutations in the *GALC* gene, resulting in rapid and irreversible neurological deterioration if untreated.

Recent advances in screening—particularly the use of second-tier psychosine testing—and new clinical data demonstrating improved outcomes with early hematopoietic stem cell transplantation (HSCT) led to the addition of IKD to the U.S. Recommended Uniform Screening Panel (RUSP) in July 2024. Successful treatment depends on rapid diagnosis and referral: ideally, infants must receive HSCT within the first 30 to 45 days of life. However, this level of urgency poses a significant challenge for Nebraska, which does not currently have a pediatric transplant center or a formalized infrastructure for expedited referral and diagnostic coordination.

This review evaluates the clinical and operational justification for adding infantile Krabbe disease (IKD) to Nebraska's newborn screening (NBS) panel and offers evidence-based recommendations to support public health decision-making. In addition to analyzing programmatic and laboratory readiness, the review incorporated national guidance, including recommendations from the Advisory Committee on Heritable Disorders in Newborns and Children (ACHDNC), and drew upon recent publications that became available following the inclusion of IKD in the U.S. Recommended Uniform Screening Panel (RUSP) in July 2024. The focused literature review prioritized studies that addressed screening performance, treatment outcomes, and implementation models relevant to the post-RUSP landscape. While Nebraska's current NBS partner laboratory, Revvity Omics, has the capacity to perform both first-tier GALC enzyme activity screening and second-tier psychosine testing and molecular diagnostics, effective implementation will also require the establishment of a rapid, coordinated response system for diagnosis and timely referral to out-of-state transplant centers with experience in treating IKD.

BACKGROUND AND DISEASE OVERVIEW

Krabbe disease (KD), which affects 0.3–2.6 per 100 000 live births, is a rare, autosomal recessive lysosomal disorder caused by variants in the GALC gene (chromosome 14q31; OMIM: #245200) that reduce galactosylceramidase (GALC) activity.[1] GALC plays a critical role in the catabolism of galactolipids, including galactosylceramide and psychosine, which are essential components of myelin. Deficient GALC activity leads to the pathological accumulation of psychosine, a highly cytotoxic metabolite that causes the dysfunction and death of myelin-producing oligodendrocytes in the central nervous system and Schwann cells in the peripheral nervous system. This results in progressive, global demyelination and severe neurodegeneration.[2]

Clinically, KD can be divided into two subtypes based on age at symptom onset with the most common form, infantile KD (IKD), having onset by 12 months with extreme irritability, hypertonia of limbs, hypersensitivity to external stimuli, and episodic fever. Progression is rapid, with a plateau of motor and cognitive development followed by regression and death in early childhood.[3] Late-onset KD (LOKD) generally presents with developmental regression, spastic paraplegia, ataxia, wide gait, and visual loss.[4] Disease presentation at less than 1 year of age generally correlates with a more severe, rapid disease course than later presentation. The estimate of the proportion of children who present in the first 6 months after birth ranges from 40% to 70%, and when untreated, these infants have a median survival of 1.5 years.[5]

LOKD accounts for approximately 15% to 20% of known cases. LOKD has a broader clinical spectrum, with symptom onset typically occurring between 12 and 36 months of age, although some individuals may present even later in childhood, adolescence, or adulthood. Hallmark features of LOKD include developmental regression, spastic paraplegia, gait abnormalities, cerebellar ataxia, vision loss, and sometimes psychiatric or cognitive decline. Compared to IKD, disease progression in LOKD is generally slower, and untreated individuals may survive for several years; median survival has been estimated at 9.5 years. However, the clinical trajectory can vary widely depending on the specific genotype and psychosine levels.[5]

Currently, the only disease-modifying therapy for KD is hematopoietic stem cell transplantation (HSCT), which aims to provide functional GALC enzyme from engrafted donor-

derived microglia.[6] For IKD, the timing of HSCT is critical; it must be administered before irreversible neurological injury occurs. The optimal window for HSCT is typically within the first 30 to 45 days of life, ideally while the child is still presymptomatic. Early HSCT can stabilize disease progression and improve long-term survival. However, the procedure is associated with significant risks, including a 10% mortality rate within 100 days of transplant and potential for long-term morbidity, including motor, cognitive, and visual impairment.[7] Nevertheless, survival and neurodevelopmental outcomes are significantly better for children who undergo HSCT early compared to those who are diagnosed after symptom onset.

Given the rapid and devastating progression of IKD, especially in the absence of early intervention, newborn screening (NBS) offers a critical opportunity for timely diagnosis and referral to treatment. The recent addition of IKD to the U.S. Recommended Uniform Screening Panel (RUSP) reflects growing consensus on the utility of early detection in altering disease course and improving outcomes for affected infants.

SCREENING METHODOLOGY AND LABORATORY CONSIDERATIONS

Newborn screening (NBS) for IKD, as defined by low GALC enzyme activity and psychosine ≥ 10 nM, was added to the Recommended Uniform Screening Panel (RUSP) in July 2024.[8] New York implemented NBS for KD in August 2006 based on low GALC levels in dried-blood spots using synthetic substrate and analyzed by multiplex tandem mass spectrometry (MS/MS) assay for five lysosomal storage disorders (LSDs) (KD, Gaucher, Pompe, Niemann-Pick, and Fabry disease).[9] Infants with low GALC activity were then screened for GALC variants by complete DNA sequencing and deletion analysis. In 2016, New York reported a positive predictive value of only 1.4% for IKD and an unexpectedly high number of variants of unknown significance after 8 years of screening. Based on these findings, and the experience of ten other NBS programs with IKD NBS, second-tier DBS testing to assess whether an elevated psychosine concentration is present in newborns with low GALC enzyme activity was recommended.[10] Most NBS programs have added second-tier tests, including DBS psychosine concentration to improve screening specificity and GALC gene molecular analysis to help predict phenotype, including a

30-kb deletion, the most common pathogenic variant found in European studies of IKD.[11] A summary of state NBS programs and IDK NBS, where applicable, follows below.

IKD NBS Summary of State Programs (May 20, 2025)

- 12 states with status of "universally screened"
  - Georgia, Illinois, Indiana, Kentucky, Minnesota, Missouri, New Jersey, New York, Ohio, Pennsylvania, South Carolina, Tennessee
- 11 of the 12 states reported total NBS fees
  - Range $41.72 (PA) to $235 (MN)
  - Average $130.16
- All 12 states provided a LIMS System Report for all screened conditions
  - LIMS System
    - 8/12 reported Revvity
    - 3/12 reported Neometrics/Natus
    - 1/12 reported Other
  - Follow-up System
    - 4/12 reported Revvity
    - 2/12 reported Neometrics/Natus
    - 4/12 reported Internally Developed
    - 1/12 reported Other
    - 1/12 reported OZ
- 7 of the 12 states provide details on KD screening method
  - 6/7 report 1st tier as MS/MS of GALC
  - 1/7 report 1st tier as fluorometry of GALC
- 6/7 report 2nd tier
  - 3/6 report 2nd tier psychosine
  - 3/6 report 2nd tier gene sequencing
  - 1/6 report 2nd tier 30-kb deletion testing
- 6 of the 12 states provide details on KD equipment

- 2/6 report RevityQSight 225 MSMS screening system
- 1/6 report RevvityOmics for 2nd tier 30-kb deletion
- Other equipment: Fluorimeter, Waters TQD, XEVO TQS Micro, ABI 3730, Sciex LDT

NBS programs are essential public health programs that perform laboratory screening, conduct follow-up on actionable results, and refer infants to clinical care for diagnosis and treatment as necessary. Increasingly, NBS programs are implementing long-term follow-up of NBS identified individuals to support screening, counseling, and service delivery. Components of IKD NBS include a first-tier screen to identify low GALC enzyme activity, a second-tier screen to identify psychosine $\geq$ 10 nM, a diagnostic evaluation including sequencing and deletion analysis of the *GALC* gene, urgent referral to specialists and a transplant center, and long-term follow-up of IKD and LOKD. Second-tier psychosine testing is "...virtually essential...before calling out screening results..." and has reduced the false-positive rate of the GALC screen.[12]

The factors influencing screening methodology and laboratory readiness to screen for any NBS condition can vary from state to state but readiness for IKD NBS may be enhanced if the state already screens for other LSDs, such as Mucopolysaccharidosis type I (MPS I). While MPS I is screened in Nebraska, the first-tier screen is performed by MS/MS, and the second-tier is sequencing of the alpha-L-iduronidase (IDUA) gene, it does not involve a second-tier MS/MS screen and the recommendation to treat IKD with HSCT by 30 to 45 days of life are not requirements for MPS I screening.[13]

Since 2018, Revvity Omics laboratory located in Pittsburgh, PA performs the DBS NBS for newborns born in Nebraska. Revvity Omics has the following accreditations: (1) College of American Pathologists (CAP), (2) Clinical Laboratory Improvement Amendments (CLIA), (3) Commission on Laboratory Accreditation (COLA), and (4) Joint Commission (JC). First-Tier Screen: Revvity offers the NeoLSD MSMS Kit, an in vitro diagnostic (IVD) mass spectrometry based kit for NBS of Pompe, MPS-I, Fabry, Gaucher, Niemann-Pick A/B, and KD.[14] At least two states appear to utilize Revvity for IKD screening and this is expected to ensure testing cutoffs and decision schemes will meet specificity/sensitivity and other performance targets.

Second-Tier Screen: Revvity also offers a test that includes both sequencing and deletion/duplication (CNV) analysis of all coding regions of *GALC* using next-generation

sequencing (NGS) technology.[15] Revvity offers a psychosine biochemical assay and lists DBS as a sample source but it is unclear if any NBS programs utilize Revvity for this testing. The strategy outlined in the RUSP recommends screening for IKD characterized by low GALC levels and psychosine $\geq$ 10nM, so additional information from the Revvity team would be helpful.

Short-term Follow-up: Follow-up is essential to communicate screen-positive results to primary care physicians (PCPs), coordination of confirmatory testing, and referral of identified newborns to appropriate specialists and/or treatment centers. Nebraska utilizes Newborn Screening ACT Sheets and Algorithms developed by the American College of Medical Genetics and Genomics (ACMG). IKD and LOKD ACT Sheets and Algorithms are available and were published in 2022, and are accompanied by short animated videos produced in 2023 are also available.[16] For IKD, follow-up staff will need to work closely with specialists and treatment centers to ensure the newborn is rapidly referred to a transplant center to increase the chance that the newborn is transplanted as close to 30 days of age as possible. There are only a few centers in the US with the experience and capability to transplant young children with Krabbe disease, so a consideration if the potential need to assist the family to quickly travel out-of-state for their treatment and care.

Long-term Follow-up: The longitudinal follow-up of NBS identified newborns is important to achieve the best possible outcomes by expanding the ability of state public health agencies to provide screening, counseling and services to these newborns and children and to collaborate with clinicians, public health agencies and families to create a system of care that can assess and coordinate follow-up and treatment of newborn screening conditions. Long-term follow-up efforts facilitate understanding of the condition, improve the delivery of NBS, help to ensure the best possible health outcomes, and track indicators for infants who may be at risk for a disorder. This is especially the case for LOKD and may include care coordination to assure access to both care and treatment as well as periodic assessments of outcomes. Long-term follow-up requires collaboration between clinicians, public health agencies, and families.

CLINICAL UTILITY AND JUSTIFICATION FOR INCLUSION

Until it was terminated in April 2025, the Advisory Committee on Heritable Disorders in Newborns and Children (ACHDNC) conducted evidence reviews of conditions that were nominated for inclusion on the RUSP and makes recommendations for RUSP inclusion to the US Secretary of Health and Human Services (HHS). KD was nominated and rejected for the RUSP in 2010. Improvements in the screening methodology, advancements in the understanding of the natural history of KD, and evidence of benefit from transplantation in the first months of life for IKD led to a renomination in 2022 and eventual RUSP inclusion in 2024. The addition of psychosine as a second-tier screen improved the specificity of screening for IKD and this improvement, coupled with the potential benefit of allowing families to choose disease-modifying therapy for IKD in early infancy, led the ACHDNC in 2024 to recommend adding IKD to the RUSP.[6] The ACHDNC noted that without newborn screening, diagnosis of IKD is generally made after significant clinical symptoms develop, past when HSCT can be effective. The committee also stated for IKD NBS to be as effective as possible, it is important to have systems in place to support families in making challenging decisions soon after diagnosis about whether to pursue HSCT and to ensure rapid access to HSCT if chosen. The committee stated that the benefit of newborn detection of later-onset phenotypes of KD is uncertain.

The ACHDNC Evidence Review Group cited a retrospective study of 6 infants (4 males, 2 females) with IKD identified through NBS between January 2016 and February 2019 and treated with HSCT at 4 pediatric centers: (1) Duke Children's Hospital, (2) Ann & Robert H. Lurie Children's Hospital, (3) Nationwide Children's Hospital, and (4) St. Louis Children's Hospital.[13] All had negative family histories. Families were notified of the abnormal NBS result at a median of 6 days of age (range 5-16, Figure 1).

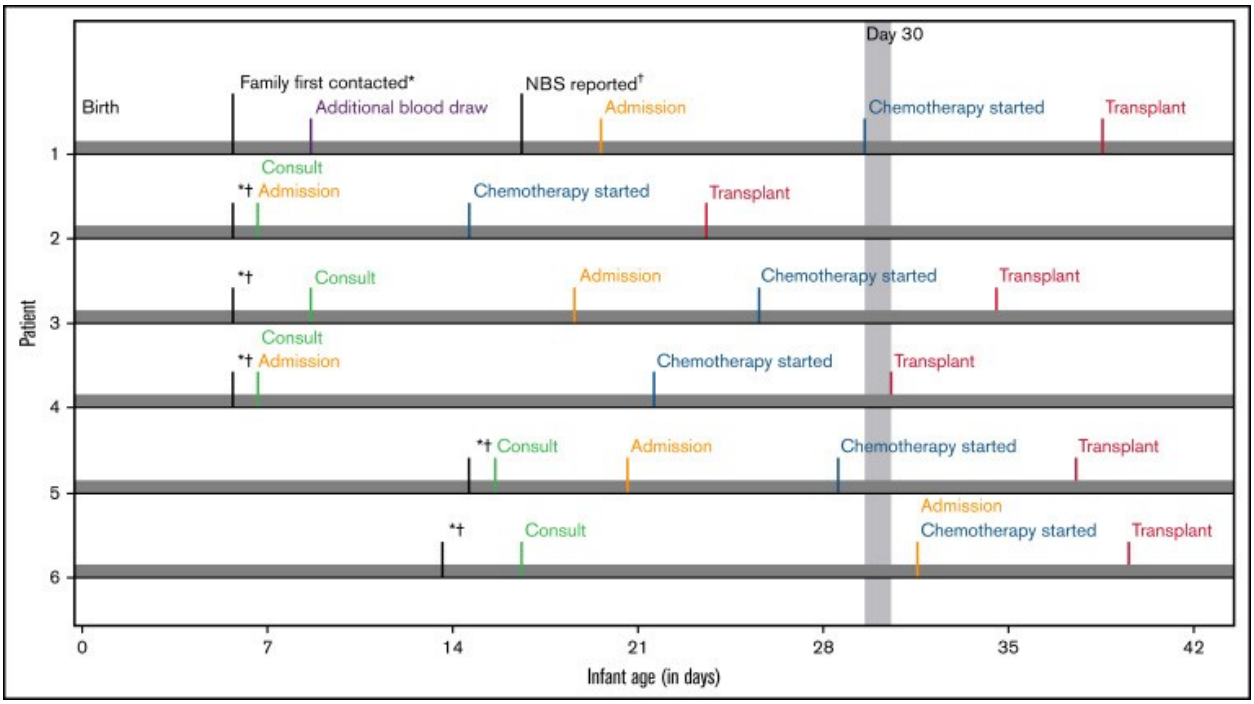


Figure 1: Timelines for patient diagnosis, referral and transplant. Timelines are presented for each infant showing when (1) families were first alerted to abnormal NBS (date indicated by *), (2) NBS results were finalized (indicated by †), (3) the initial consultation occurred, (4) the infant was admitted to the hospital, (5) chemotherapy was started, and (6) transplant occurred. The timeline is shown in infant age or "days old," where day of birth = 0 days old. The goal of transplant occurring prior to 30 days is highlighted by gray shading beyond this time point.

The primary study endpoint was the evaluation performed 1 year after HSCT and descriptive endpoints included clinical, neurodiagnostic, and neurodevelopmental outcomes with follow-up ranging from 30 to 58 months. Prior to transplant, the diagnosis was confirmed through low GALC activity in leukocytes, elevated psychosine levels, and/or *GALC* genotyping. All infants had extremely elevated pyschosine levels (range 24-73 nmol/L; normal <2 nmol/L) and genotyping found known pathogenic variants in 5 infants and the 30-kb deletion in 3 infants. Infants underwent standard evaluations to assess health, infectious disease screening, organ function, neurologic evaluations including magnetic resonance imaging (MRI) and nerve conduction studies (NCS), neurodevelopmental testing, and clinical status of IKD. Chemotherapy started at a median of 27 days of age (range 14-32). All infants tolerated myeloablative chemotherapy well, engrafted donor cells, and were alive with normal GALC levels with a median of 47.5 months ʾfollow-up (range 30-58). The children (30-58 months old) were reported to have varying degrees of developmental delay. Five infants underwent neurodevelopmental testing 1

year after HSCT. All scores were >1 SD below the mean using aged-based norms (mean: 100; SD: 15; range 45-160; Figure 2).

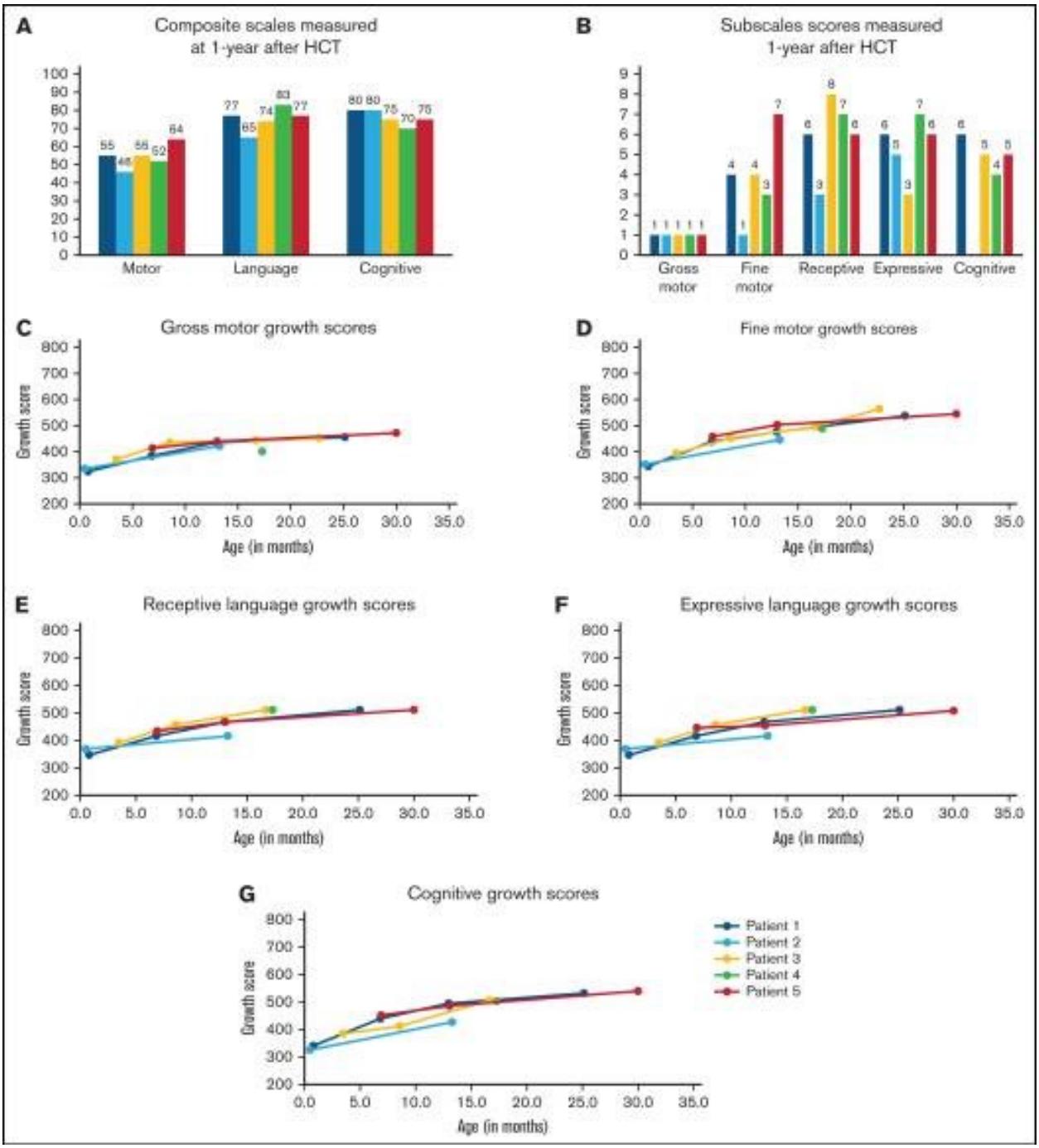


Figure 2: Neurodevelopmental outcomes assessed 1 year after transplant and over time. Infants underwent developmental testing using the BSID-III at 1-year posttransplant (A-B) and at additional time points after transplant (C-G). Composite scores were assigned for motor, language, and cognitive development (A). Composite scores are classified as extremely low (≤69), borderline (70-79), low average (80-89), average (90-109), high average (110-119), superior (120-129), and very superior (≥130). Subscales were also assigned for gross motor, fine

motor, receptive language, expressive language, and cognitive development (B). Growth scores (mean: 500; SD: 100; range, 200-800) were calculated using raw scores to reflect longitudinal growth independent of age14 and were helpful in showing ongoing development in infants/children with low age-based scores. Growth scores are presented for individual patients (Patients 1-5) over time for the subscales: gross motor (C), fine motor (D), receptive language (E), expressive language (F), and cognitive (G).

In this retrospective review, only 1 infant received a transplant prior to 30 days of age (24 days old), and the other babies were 31 to 40 days of age at HSCT. All children are continuing to achieve developmental milestones, although more slowly than unaffected peers. Due to the small numbers, no conclusion could be made regarding the impact of timing of HSCT on outcomes. However, in contrast to these 6 NBS identified newborns, untreated infants develop quadriparesis, severe motor delay (equivalent to 1 month old), autonomic instability by 1 year of age, and early death.[5]

The ACHDNC Expedited Evidence-Based Review Final Report (Final Report) in February 2024 presented a survival curve comparing 7 NBS identified IKD cases to 51 individuals with IKD born in states that do not offer screening and who were detected based on signs and symptoms in their first year of life and who did not qualify for HSCT based on their disease status (Figure 3).[17]

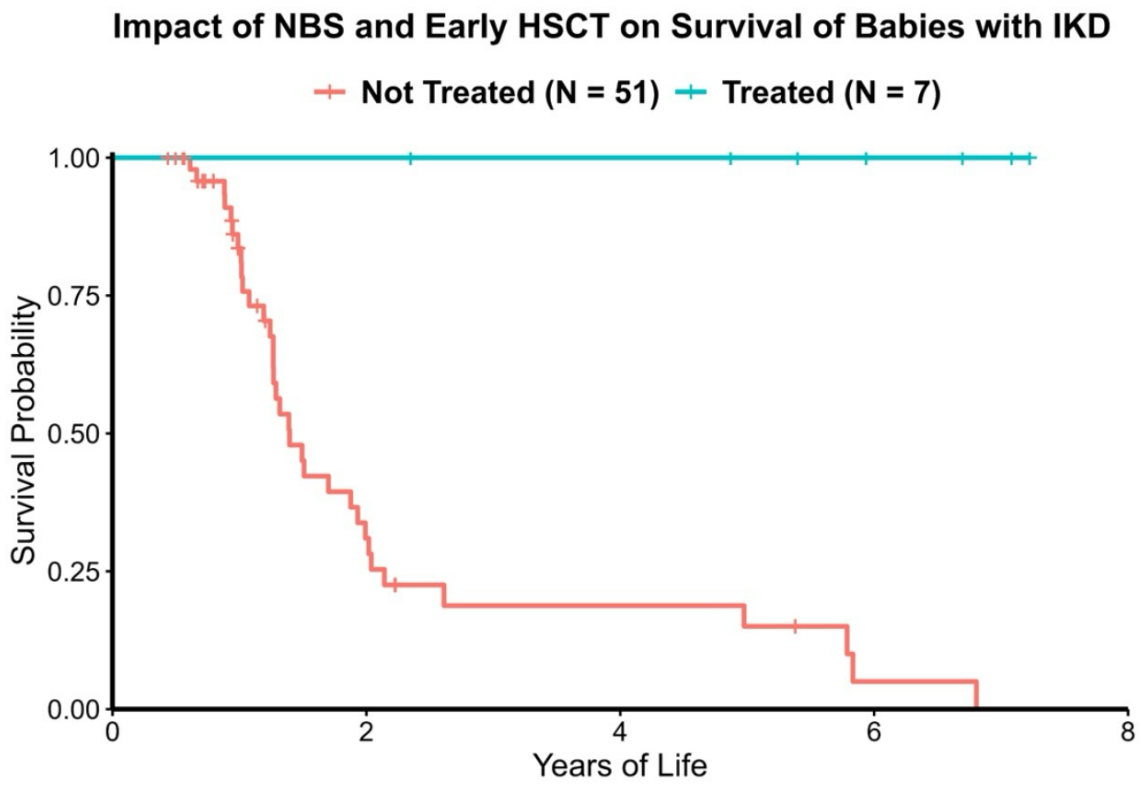


Figure 3: Survival curve comparing 7 NBS identified IKD with 51 clinically identified IKD.

The Final Report assessed four key questions and applied their decision analysis to IKD NBS. A summary of the four questions and analysis is included below.

- Key Question 1: Clinical Validity of IKD NBS
  - The RUSP nomination for infantile Krabbe disease newborn screening specifies first-tier screening with low GALC enzyme activity and second-tier dried-blood spot screening with psychosine, with diagnostic referral for psychosine ≥10 nM.
  - Psychosine is highly specific for Krabbe disease. Increasing the threshold for a positive second-tier psychosine screening test would decrease the number of less severe phenotypes that would be detected.
  - The risk of missing a case of infantile Krabbe disease with newborn dried blood spot psychosine < 10 nM is nearly zero (i.e., the sensitivity of psychosine ≥10 nM for IKD is nearly 100%).

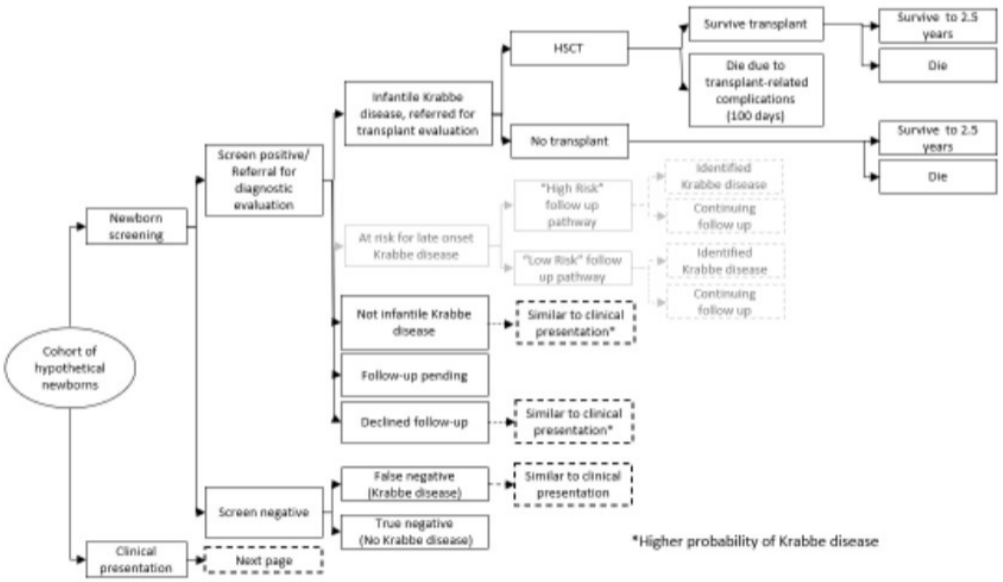
- Based on the information from 9 state newborn screening programs from 2016 to 2023, using psychosine ≥10 nM as the only second-tier test, the number of cases of IKD ranged from 0 in Indiana, New York, and South Carolina, which collectively screened about 874,000 infants, to 5 cases of IKD in Illinois, which reported screening 848,000 infants. Based on 3.55 million screened newborns described for this report, the overall IKD case detection rate with psychosine ≥10 nM is about 3.1 per million infants screened. No program reported that they would miss a case of infantile Krabbe disease with a psychosine ≥10 nM.
- The rationale for second-tier psychosine testing with a threshold of 10 nM is to decrease the identification of LOKD (i.e., any phenotype other than IKD ranging from onset later in childhood or in adulthood) relative to current practice. No cases of LOKD would have been identified by the newborn screening programs with psychosine ≥10 nM among the 3.55 million newborns screened as described in this report. This threshold also eliminates the detection of LOKD across the total population of infants screened of about 9.29 per million infants screened.

- Key Question 2: Impact of IKD NBS Compared with Usual Case Detection
  - HSCT around the first month of life for infantile Krabbe disease is associated with decreased risk of mortality.
- Key Question 3: Benefits and Negative Consequences for Families of IKD NBS
  - Family Experiences and Perspectives - One study addressing family attitudes about Krabbe disease newborn screening based on 170 respondents to an online survey from December 2019 to February 2020. All respondents were impacted by having a family member with Krabbe disease. Overall, nearly all respondents (97%) “feel that KD NS should be implemented in every state.”
  - Health Disparities - Public health newborn screening can potentially reduce disparities in care because all infants have access to screening. One study found that the proportion of Krabbe disease among Black or Asian individuals identified through newborn screening was twice as high as in the hospital dataset. It also reported that within the hospital dataset, non-Hispanic White patients had a lower average time gap

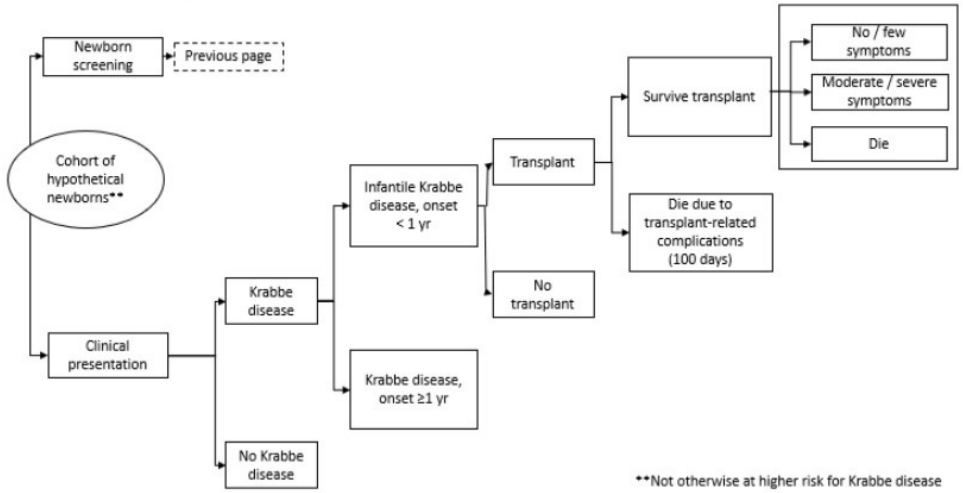
from first presentation to diagnosis (64 days) compared with non-Hispanic Black children (101 days), Asian children (136 days), and Hispanic children (120 days).

- Key Question 4: Potential Population-Level Outcomes of IKD NBS
  - The decision-analytic model was updated to reflect screening for infantile Krabbe disease only, not later-onset phenotypes, using a screening algorithm in which only infants with dried-blood spot psychosine ≥10 nM would be referred for diagnostic evaluation.
- Decision Analysis of IKD NBS
  - For this expedited review, the Final Report used updated data from state newborn screening programs to revise the screening parameters included in the simulation model. A schematic of the IKD NBS decision model is included below based on the annual newborn cohort in the US of 3.65M newborns (Figure 4).

a. Newborn screening using the revised nomination screening algorithm†



b. Clinical presentation



*Only newborns with low GALC enzyme activity and psychosine ≥10 nM referred for diagnostic evaluation. Health states shown in gray were included in the previous modeling analysis and are not included in the analysis of the revised nomination.

- Under a policy of infantile Krabbe disease newborn screening, 11.3 infants (range: 5.6 – 20.2) are estimated for referral for diagnostic testing due to a positive screen annually assuming an annual newborn cohort of 3.65 million newborns. Each of these infants are projected to be identified with likely IKD and referred for HSCT, of whom 9.9 (range: 3.5 – 19.9) would be expected to receive HSCT and 1.4 (range: 0.3 – 2.2) would not receive HSCT, either due to ineligibility based on disease status or declined by

family. Of those infants receiving HSCT, 1.0 (range: 0.3 – 1.2) would be expected to die from complications within 100 days of transplant. By 30 months of age, 9.2 infants would be alive and 2.1 (range: 0.5 – 4.0) would be expected to have died (1.0 from HSCT and 1.1 from KD).

- Under a policy of clinical presentation in the absence of NBS, 24.2 infants (range 8.6–43.3) would be identified with KD. Of these, 11.3 (range: 4.0 – 20.2) would be expected to present with symptoms in the first year of life and 12.9 (range: 4.6 – 23.0) would be expected to present later. 1.1 clinically identified infants (range: 0.0–4.0) would be expected to receive HSCT. By 30 months of age, 7.9 (range 3.5 – 10.6) would be expected to have died either from HSCT complications or KD progression
- Summary - With universal infantile Krabbe disease newborn screening, this analysis predicts that 11.3 infants (range: 5.6 – 20.2) annually would be diagnosed with IKD and be referred for evaluation for HSCT. Of these, 9.9 (range: 3.5 – 19.9) would receive HSCT. Of the infants who received HSCT, 1.0 (range: 0.3 – 1.2) would die from complications of HSCT within 100 days and all others would be alive at 2.5 years. Without universal KD newborn screening, relying on clinical presentation, 11.3 (range: 4.0 – 23.0) infants would present before age 1 year, of whom 1.1 (range: 0 – 4.0) would be eligible for and receive HSCT. Of the remaining 10.2, 7.8 (range: 3.5 – 10.6) infants would be expected to die from KD by age 2.5 years.

ESTIMATED IMPACT ON NEBRASKA'S NBS PANEL

Testing newborns for IKD presents unique challenges compared with other diseases identified through NBS. For infants to access HSCT, complex care coordination is immediately needed and may include referral to an out-of-state transplant center. Prior reports established HSCT before 30 days of age as a benchmark among presymptomatic infants. The retrospective study authors proposed a timeline of first-tier screen (GALC activity), second-tier screen (psychosine), results between 2 and 4 days of life, diagnostic and treatment evaluations beginning between 3 and 5 days of life, and HSCT when the infant is between 17 to 20 days of age. They also suggested some strategies for NBS programs and transplant centers to mitigate delays including: (1)

establishing a relationship between the NBS program and transplant centers; (2) performing transplant-related processes and disease evaluations in parallel, (3) partnering with Medicaid and private health care payers to minimize third-party payer approval time, and (4) initiating chemotherapy before final donor selection, assuming that candidate donors have been selected (not customary for HCT centers).[13] HSCT is currently the treatment of choice for IKD and experts anticipate that the gene therapies in development will also require treatment in the first few weeks of life to optimize outcomes.

Another challenge in IKD NBS has been distinguishing between affected and unaffected babies due to the lack of sensitivity of the screen for low GALC enzyme activity. Psychosine as a second-tier screen can rapidly discriminate between IKD ($\geq$10 nmol/L), LOKD (>2 and < 10 nmol/L), pseudodeficiencies, and carriers (<2 nmol/L).[18] Nebraska's screening laboratory Revvity has established protocols for GALC MS/MS, psychosine MS/MS, and *GALC* molecular analysis. At least 2 states currently use Revvity for IKD NBS.

In 2023, 24,111 babies were reported born in Nebraska.[19] A positive first-tier screen (low GALC) of 53 per 100,000 screened (range 6.6-172.3 per 100,000) applied to 24,111 newborns estimates 12.78 newborns would be reflexed to second-tier psychosine screen each year.[17] Using a screen positive defined as low GALC and $\geq$ 10 nmol/L psychosine of 0.30977 per 100,000 (range 0.15464-0.55426 per 100,000), this rate applied to 24,111 newborns estimates the identification of 1 IKD every 13-14 years.[17]

RECOMMENDATIONS

Krabbe Disease (KD), particularly the infantile form (IKD), is a rapidly progressive neurodegenerative disorder with significant morbidity and early mortality if untreated. Following a robust evidence review and recent inclusion of IKD in the U.S. Recommended Uniform Screening Panel (RUSP) in 2024, Nebraska is evaluating its readiness to include IKD in its newborn screening (NBS) panel. Early detection through NBS enables timely intervention via hematopoietic stem cell transplantation (HSCT), ideally administered within 30 to 45 days of life. However, successful implementation will require Nebraska to develop a rapid diagnostic and referral infrastructure.

Key Justifications for Inclusion

- Krabbe Disease meets RUSP criteria: early identification improves outcomes.
- Second-tier psychosine testing improves specificity and reduces false positives.
- Without screening, diagnosis is delayed and treatment opportunities are missed.
- Nebraska's lab partner Revvity Omics is equipped to perform required screening tests.

Key Considerations for Inclusion

- Detection of later-onset phenotypes remains uncertain and may not yield actionable interventions.
- Psychosocial and logistical burdens on families, including travel and insurance authorization, must be addressed.
- Implementation requires investment in clinical coordination, training, and interstate partnerships.
- Rapid diagnostic confirmation and family decision-making are critical within the newborn's first weeks.
- Nebraska currently lacks an in-state pediatric transplant center, necessitating out-of-state referrals.
- HSCT must occur within 30–45 days of life to be effective, requiring urgent coordination and care infrastructure.
- Low frequency of IKD in Nebraska (estimated 1 case every 13–14 years) raises cost-benefit concerns.

Actionable Recommendations

1. Adopt IKD screening in the Nebraska NBS panel using a two-tier test: GALC enzyme activity followed by DBS psychosine quantification.
2. Develop a statewide rapid diagnosis and referral system to ensure infants are evaluated and prepared for HSCT by day 30–45 of life.

3. Establish partnerships with experienced out-of-state transplant centers and negotiate expedited care and insurance coordination.

4. Engage Revvity Omics for both primary and secondary laboratory testing to ensure timely and accurate results.

5. Train follow-up and clinical staff using ACMG ACT Sheets, clinical algorithms, and video materials to prepare for rare but urgent cases.

6. Build a long-term follow-up system to track health outcomes of identified cases and ensure care continuity.

7. Prepare the public health infrastructure for rare positive screens (~1 IKD case every 13–14 years in Nebraska).

8. Ensure health equity in access, family education, and out-of-state treatment navigation.

Conclusion

While the incidence of IKD in Nebraska is low, the potential benefit of early diagnosis and intervention is profound. Implementing IKD screening will require focused efforts to ensure infrastructure, staffing, and inter-state collaboration are in place. This investment will position Nebraska as a national leader in rare disease detection and childhood health equity, and facilitate future panel expansions.

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Executive Summary: Inclusion of GAMT Deficiency in the Nebraska Newborn Screening Panel

Introduction

Guanidinoacetate Methyltransferase Deficiency (GAMT-D) is a rare but serious disorder of creatine biosynthesis that can lead to profound intellectual disability, epilepsy, and behavioral disturbances if left untreated. Following its inclusion in the U.S. Recommended Uniform Screening Panel (RUSP) in January 2023, Nebraska is evaluating the feasibility and public health impact of adding GAMT-D to its newborn screening (NBS) panel. Early detection through NBS enables prompt treatment with creatine supplementation, ornithine, and dietary modifications, which can prevent neurological symptoms entirely if started presymptomatically. Implementation in Nebraska will require laboratory validation, provider education, and systems for confirmatory testing and long-term follow-up.

Key Justifications for Inclusion

- GAMT-D meets RUSP criteria: early identification and treatment lead to normal developmental outcomes.
- First-tier mass spectrometry testing is low-cost and can be multiplexed with other disorders.
- Without screening, diagnosis is often delayed for years, missing the window for effective intervention.
- Revvity Omics can support second-tier sequencing and could expand to first-tier biochemical screening.

Key Considerations for Inclusion

- Extremely low incidence (estimated 1 case every 20 years in Nebraska) may challenge perceived cost-benefit.
- Confirmatory testing requires access to specialized metabolic providers and follow-up care.
- Psychosocial, logistical, and insurance barriers may limit timely treatment access.
- Laboratory validation and personnel training are needed for first-tier test implementation.

- Statewide education is required for primary care and follow-up teams to recognize and manage cases.

Actionable Recommendations

1. Add GAMT-D to the Nebraska NBS panel using a first-tier tandem mass spectrometry test for GAUC and creatine.
2. Validate laboratory protocols and cutoffs using existing dried blood spot quality assurance materials.
3. Establish a confirmatory testing protocol with metabolic genetics and biochemical follow-up.
4. Coordinate with Revvity Omics and/or partner labs to offer GAMT gene sequencing as a second-tier test.
5. Educate clinical and NBS staff using ACMG ACT Sheets, diagnostic algorithms, and patient education tools.
6. Build a long-term follow-up system to track development and treatment adherence in diagnosed infants.
7. Prepare for rare positive screens (~1 case per 20 years) with a rapid-response care coordination plan.
8. Promote health equity in education, diagnosis, and care coordination for families receiving results.

Conclusion

Although GAMT-D is an ultra-rare condition, its inclusion in the Nebraska NBS panel is justified by the transformative impact of early diagnosis and treatment. With minimal laboratory burden and the availability of effective therapy, implementation would align Nebraska with national standards while protecting infants from otherwise preventable neurologic injury. Proactive preparation will ensure a sustainable model for rare disease screening and future panel expansion.

Evidence Review: GAMT Deficiency

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INTRODUCTION

Newborn screening (NBS) is a cornerstone of public health that enables the early identification and treatment of severe, often life-altering, genetic disorders. The inclusion of Guanidinoacetate Methyltransferase Deficiency (GAMT-D) in the Nebraska NBS panel requires a thorough review of clinical evidence, screening technology, treatment efficacy, and implementation feasibility. GAMT-D is a rare autosomal recessive disorder of creatine biosynthesis caused by mutations in the *GAMT* gene, leading to toxic accumulation of guanidinoacetate (GAUC) and creatine deficiency, which together cause progressive neurological impairment if left untreated.

Recent clinical studies—including those published after GAMT-D was added to the U.S. Recommended Uniform Screening Panel (RUSP) in January 2023—demonstrate that early identification through NBS followed by timely treatment with creatine supplementation, ornithine, and dietary modification can prevent the onset of epilepsy, intellectual disability, and movement disorders. In several sibling case series, individuals treated in the neonatal period achieved normal developmental milestones. This reinforces the value of presymptomatic diagnosis and underscores the importance of universal screening.

This review evaluates the clinical and operational justification for adding GAMT-D to Nebraska's newborn screening (NBS) panel and offers evidence-based recommendations to support public health decision-making. In addition to analyzing programmatic and laboratory readiness, the review incorporated national guidance, including recommendations from the Advisory Committee on Heritable Disorders in Newborns and Children (ACHDNC), and drew upon publications made available following the RUSP designation. The focused literature review prioritized studies that addressed diagnostic accuracy, treatment efficacy, long-term developmental outcomes, and state-level implementation strategies. While Nebraska's current NBS partner laboratory, Revvity Omics, offers second-tier sequencing for *GAMT*, effective implementation will require validation and deployment of a first-tier GAUC assay using tandem

mass spectrometry, along with provider education, confirmatory testing protocols, and follow-up infrastructure to ensure timely diagnosis and access to metabolic treatment.

BACKGROUND AND DISEASE OVERVIEW

Guanidinoacetate methyltransferase deficiency (GAMT-D), which likely affects between ~0.038 and 0.4 per 100 000 live births (eg. 0.5 and 2 per million live births and 1 per 250,000 live births) is an autosomal recessive disorder of creatine (CRE) biosynthesis due to pathogenic variants in the *GAMT* gene (chromosome 19p13.3; OMIM: #601240) that primarily affects the brain and muscles.[1,2] GAMT-D leads to low plasma and brain CRE levels and elevated concentrations of guanidinoacetate (GAUC) in the brain, cerebrospinal fluid, blood, and urine. A shortage of creatine—essential for cellular energy metabolism—combined with the accumulation of neurotoxic levels of GAUC, results in severe and progressive neurological impairment.

Without early intervention, GAMT-D typically leads to profound neurological impairment. Affected individuals often present with global developmental delay, severe intellectual disability, and limited expressive language, typically restricted to only a few words. Epilepsy is common and frequently refractory. Behavioral manifestations may include features of autism spectrum disorder and self-injurious behaviors, such as head-banging. Additional findings include extrapyramidal symptoms such as tremors, dystonia, or facial tics. Hypotonia and significant delays in gross motor milestones (e.g., sitting, walking) are also observed. In more severe cases, there may be neuroregression with loss of previously acquired motor functions, including head control and independent sitting. To date, more than 50 cases of GAMT-D have been documented in the peer-reviewed literature. In the absence of early treatment, affected individuals commonly present with hypotonia, moderate to severe intellectual disability, minimal speech development, epilepsy, significant behavioral disturbances, and movement disorders such as ataxia and dystonia.[3-6] Although neonates with GAMT-D are typically asymptomatic due to transplacental creatine transfer, clinical symptoms begin to emerge in infancy or early childhood. Without a known family history or the benefit of newborn screening, diagnosis is often delayed due to the nonspecific nature of early symptoms. In one cohort study

of 22 patients, developmental concerns such as delay, seizures, and motor abnormalities were noted at a mean age of 14 months (range: 3–24 months), yet the average age at diagnosis was substantially later at 8.5 years (range: 9 months to 25 years).[4]

Current standard of care is oral supplementation with creatine and ornithine, sodium benzoate to facilitate alternative nitrogen excretion, and a protein-restricted diet aimed at reducing arginine intake.[7] Routine monitoring of plasma guanidinoacetate (GUAC), creatine levels, and comprehensive amino acid profiles is recommended to guide treatment. Management of GAMT-D requires lifelong treatment. During the first 6–12 months of life, laboratory assessments are typically performed every 1–2 months, with monitoring intervals extended as the metabolic profile stabilizes. Given that therapeutic decisions are primarily guided by biochemical markers, affected individuals may be managed remotely by metabolic specialists and dietitians. Case series indicate that presymptomatic treatment substantially reduces the risk of developing neurological complications such as intellectual disability, epilepsy, behavioral abnormalities, and movement disorders. However, the existing evidence base is limited by small sample sizes, heterogeneous outcome measures, and variability in follow-up duration. A mixed-method cohort study published in 2025 involved 4 sibling pairs with GAMT-D and 8 age-matched healthy sibling pairs demonstrated that younger siblings who were diagnosed and treated earlier exhibited markedly better outcomes across all assessed domains, including cognition, development, motor skills, adaptive function, behavior, and seizure control.[8] Remarkably, children treated during the neonatal period had outcomes comparable to healthy controls, and those treated during infancy showed significantly improved outcomes relative to siblings treated after age two. These findings underscore the critical importance of presymptomatic diagnosis and early initiation of treatment, providing strong support for the inclusion of GAMT-D in universal newborn screening programs.

SCREENING METHODOLOGY AND LABORATORY CONSIDERATIONS

Newborn screening (NBS) for GAMT-D IKD, as defined by low CRE and elevated GAUC, was added to the Recommended Uniform Screening Panel (RUSP) in January 2023.[9] GAMT normally catalyzes the conversion of GAUC to CRE, and in GAMT-D, impaired enzyme activity leads to the accumulation of GAUC. NBS for GAMT-D measures both GAUC and CRE concentrations in dried blood spots (DBS) using flow injection tandem mass spectrometry (MSMS), which can be multiplexed with routine assays for acylcarnitines and amino acids, utilizing either derivatized or non-derivatized extracts.[10] A positive screen is characterized by elevated GAUC levels and a high GAUC-to-CRE ratio. While sequencing of the GAMT gene—known to have over 50 pathogenic or likely pathogenic variants—can support the diagnosis, confirmation is based on low plasma CRE and elevated plasma GAUC levels. Magnetic resonance spectroscopy may assist in evaluating brain CRE and GAUC levels but is typically reserved for research and is not required for diagnosis. Arginase deficiency, a condition that can also elevate GAUC, should be ruled out in ambiguous cases. Most diagnoses can be confirmed within two months of a positive screen using available biochemical assays. Because GAMT-D lacks a late-onset phenotype, no further follow-up is needed for infants with normal CRE and GAUC levels.

NBS for GAMT-D began in Australia in 2002, and by April 2022, approximately 1.4 million newborns had been screened with one confirmed case. In British Columbia, a 2012–2018 pilot screened nearly 250,000 newborns without identifying a case. Several U.S. states and international programs have implemented or piloted newborn screening (NBS) for guanidinoacetate methyltransferase (GAMT) deficiency using various methodologies and instrumentation tailored to their laboratory infrastructure and goals.[11] Michigan initiated GAMT screening in September 2022 using a commercially available non-derivatized kit for amino acids and acylcarnitines on Waters TQD instruments, enhanced by the addition of internal standards for guanidinoacetate (GUAC) and creatine. Challenges related to instrument sensitivity were addressed by emphasizing instrument cleanliness and adding three preventative maintenance visits per year. Michigan uses both GUAC levels and the GUAC/creatine ratio (GUAC×1000÷CRE) to determine screen-positives, with age-specific cutoffs for infants ≤71 hours and >72 hours of age, and refers positive cases to a metabolic coordinating center. In contrast, New York began screening in 2018 with a three-tiered method, later optimized to a two-tiered approach by

modifying the ion of interest to eliminate the need for second-tier HPLC testing. This change significantly reduced the number of follow-up tests and referrals. New York considers a GUAC×1000÷CRE ratio ≥12.0 and GUAC ≥5.00 as referral-level thresholds and follows screen-positive samples with GAMT gene sequencing. Utah has been screening since 2015, initially using a derivatized MS/MS method and transitioning to a non-derivatized method in 2019. The state mandates two screens—one in the first 48 hours and another between days 7–16—reporting stable GUAC levels across timepoints but observing a marked drop in creatine levels in later specimens. Molecular sequencing of the *GAMT* gene, often performed on dried blood spots via PCR amplification and Sanger sequencing of all six coding exons, is used either as a second-tier test or as part of confirmatory diagnostics depending on program goals. While sequencing provides valuable genotype information for clinical decision-making, referral for diagnostic evaluation is typically based on biochemical findings regardless of molecular results.[11] A summary of state NBS programs and GAMT-D NBS, where applicable, follows below.

GAMT-D NBS Summary of State Programs (June 1, 2025)

- 13 states with status of "universally screened"
  - Arizona, California, Connecticut, Delaware, Florida, Kentucky, Maryland, Massachusetts, Michigan, Minnesota, New York, Pennsylvania, Utah
- 13 of the 13 states reported total NBS fees
  - Range $0 (NY, FL) to $235 (MN)
  - Average $134.63
- All 13 states provided a LIMS Follow-up System all screened conditions
  - 4/13 reported Revvity
  - 3/13 reported Neometrics/Natus
  - 3/13 reported Other
  - 2/13 reported Internally Developed
  - 1/13 reported StarLims
- 9 of the 13 states provide details on GAMT-D screening method

- 2/9 report 1st tier as MS/MS of GUAC, CRE, GUAC/CRE
- 3/9 report 1st tier as MS/MS of GAUC, GUAC/CRE
- 3/9 report 1st tier as GAUC
- 1/9 report 1st tier as MS/MS with no details

- 5 of the 13 states provide details on 2nd tier
  - 3/5 report 2nd tier *GAMT* gene sequencing
  - 1/5 report 2nd tier GAUC MSMS
  - 1/5 report 2nd tier as “target"
- 5 of the 13 states provide details on GAMT-D equipment
  - 2/5 report Waters MS/MS
  - 1/5 report Scion LDT
  - 1/5 report XEVO TQ MS, TQS Micro, ABI 3730
  - 1/5 report NeoBase2 Kit

NBS programs are essential public health programs that perform laboratory screening, conduct follow-up on actionable results, and refer infants to clinical care for diagnosis and treatment as necessary. Increasingly, NBS programs are implementing long-term follow-up of NBS identified individuals to support screening, counseling, and service delivery. Components of GAMT-D NBS include a first-tier screen to identify low CRE and elevated GAUC levels, a second-tier screen (some NBS program only) to sequence the *GAMT* gene, referral to genetics or metabolic specialist, and long-term follow-up.

The factors influencing screening methodology and laboratory readiness to screen for any NBS condition can vary from state to state. Implementation of GAMT-D NBS presents several challenges, including test validation, securing adequate funding, staffing limitations, and managing competing programmatic priorities. GAMT deficiency can be screened alongside acylcarnitine and amino acid disorders using either derivatized or non-derivatized MS/MS methods. A single-tier test is sufficient, though non-derivatized methods may require more cleaning or be affected by older equipment. Adding GAMT to existing panels may require re-validation of current assays. 35 of 53 NBS programs (66%) responded to a survey from the

ACHDNC Evidence Review Group and 45% of them estimated it would take between 2 and 3 years to implement GAMT-D NBS, and noted that an FDA-approved testing kit would facilitate the implementation. 47% did not think a second-tier test would be necessary. Based on interviews with two state NBS programs (Utah and New York) that have successfully implemented GAMT screening, the estimated additional cost per infant—beyond existing NBS program operating expenses—is likely well under $1. However, both programs utilized laboratory-developed tests (LDTs) made possible by in-house technical expertise and infrastructure. As a result, this cost estimate may not be generalizable to other states, particularly those lacking the resources to independently develop and validate their own assays.

Since 2018, Revvity Omics laboratory located in Pittsburgh, PA performs the DBS NBS for newborns born in Nebraska. Revvity Omics has the following accreditations: (1) College of American Pathologists (CAP), (2) Clinical Laboratory Improvement Amendments (CLIA), (3) Commission on Laboratory Accreditation (COLA), and (4) Joint Commission (JC).

First-Tier Screen: No information could be found regarding Revvity capability to screen for GAMT-D.

Second-Tier Screen: Revvity offers a "follow-up" test to sequence the *GAMT* gene.[12]

Short-term Follow-up: Follow-up is essential to communicate screen-positive results to primary care physicians (PCPs), coordination of confirmatory testing, and referral of identified newborns to appropriate specialists and/or treatment centers. Nebraska utilizes Newborn Screening ACT Sheets and Algorithms developed by the American College of Medical Genetics and Genomics (ACMG). GAMT-D ACT Sheets and Algorithms are available and were published in 2022, and are accompanied by short animated videos produced in 2023 are also available.[13] For GAMT-D, follow-up staff will need to work closely with genetic or metabolic specialists. A clinician will perform confirmatory testing for GAMT deficiency by evaluating levels of creatine and GUAC in plasma and urine. Prematurity and total parental nutrition (TPN) can affect these biomarkers so guidance around when to best test may be warranted for these groups. Arginase deficiency can also cause elevated GUAC. The diagnostic method and criteria include: (1) Low plasma/urine creatine; (2) Elevated plasma/urine GUAC; (3) Creatine depletion in brain MR

spectroscopy; (4) GAMT gene variant may be found; (5) Clinical findings; and (6) Family history. Development of action plan templates and fact sheets for primary care physicians (PCP) and families, including any confirmatory testing, is needed.[11]

Long-term Follow-up: The longitudinal follow-up of NBS identified newborns is important to achieve the best possible outcomes by expanding the ability of state public health agencies to provide screening, counseling and services to these newborns and children and to collaborate with clinicians, public health agencies and families to create a system of care that can assess and coordinate follow-up and treatment of newborn screening conditions. Long-term follow-up efforts facilitate understanding of the condition, improve the delivery of NBS, help to ensure the best possible health outcomes, and track indicators for infants who may be at risk for a disorder. Long-term follow-up requires collaboration between clinicians, public health agencies, and families.

CLINICAL UTILITY AND JUSTIFICATION FOR INCLUSION

Until it was terminated in April 2025, the Advisory Committee on Heritable Disorders in Newborns and Children (ACHDNC) conducted evidence reviews of conditions that were nominated for inclusion on the RUSP and makes recommendations for RUSP inclusion to the US Secretary of Health and Human Services (HHS). GAMT-D was nominated and rejected for evidence review in 2016 because no case had been identified prospectively through NBS. Improvements in the screening methodology, advancements in the understanding of the natural history of GAMT-D, and evidence from states screening for GAMT-D led to a renomination in 2021 and eventual RUSP inclusion in 2023.[14] The ACHCNC noted that although GAMT deficiency is much rarer than other newborn screening conditions—such as phenylketonuria (~6 per 100,000) or maple syrup urine disease (~0.5 per 100,000)—it can be screened at low additional cost with high accuracy and few diagnostic referrals. In addition, the diagnostic process is well established, relies on readily available biochemical tests, and can be completed in early infancy. They stated that since GAMT-D has no late-onset form, infants with normal results require no further follow-up, treatment involves accessible dietary interventions, and early initiation significantly improves neurologic outcomes. Without

screening, diagnosis is often delayed. Therefore, the ACHDNC concluded that, despite its rarity, GAMT deficiency meets key criteria for newborn screening due to the clarity of diagnosis and the proven benefits of early treatment.

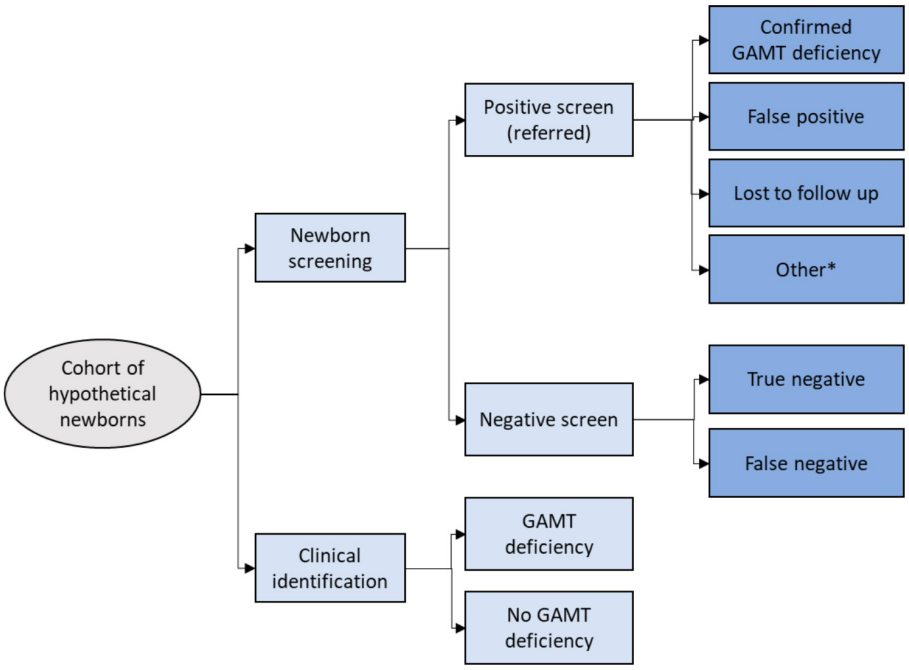
The ACHDNC GAMT Deficiency Final Report assessed four key questions and applied their decision analysis to GAMT-D. A summary of the four questions, findings, and decision analysis is included below.

- Key Question 1: What is the natural history and epidemiology of GAMT deficiency?
  - GAMT deficiency is rare and some affected individuals might not ever be diagnosed, subsequently there are gaps in the evidence related to the epidemiology, including the birth prevalence and whether there are higher risk populations.
  - The full range of pathogenic alleles has not been characterized, therefore assessment of the frequency of specific variants in existing databases could underestimate the expected birth prevalence of GAMT deficiency.
- Key Question 2: What is the analytic or clinical validity of newborn screening or GAMT deficiency?
  - High-throughput NBS with MS/MS has been incorporated into two U.S. state NBS programs using a laboratory developed test and each program has identified one case each.
  - Including programs in Australia and Canada as well as the US, 3 cases have been detected in about 2.9 million infants screened, or about 0.1 case per 100,000 newborns screened (i.e., about 1 case per 970,000 newborns screened).
  - The number of infants each year requiring diagnostic evaluation is low compared to other conditions included in the RUSP.
  - Diagnostic evaluation can be completed in <1 month following a positive screen.
- Key Question 3: What are the harms associated with newborn screening for GAMT deficiency?
  - No harms were identified.
- Key Question 4: What are the benefits and harms of pre-symptomatic or early treatment of GAMT deficiency compared to when GAMT deficiency is usually identified?

- Case series suggest that pre-symptomatic or earlier initiation of treatment of GAMT deficiency is associated with improved neurological outcomes, including reduced risk of intellectual disability and less frequent seizures.
- None of the reports provide developmental results based on a standardized quantitative measure.

- Decision Analysis of GAMT-D NBS
- The Final Report used a decision analysis model and a schematic of the model is shown in Figure 1.

Figure 1. GAMT Deficiency Model Schematic



*Includes diagnosis of non-targeted conditions and unknown determination due to death before confirmatory testing

- Based on data from the Utah and New York newborn screening programs, it is projected that screening a U.S. birth cohort of 3.6 million infants annually would result in approximately 93 referrals for follow-up testing (range: 62–135) for suspected GAMT deficiency. Of these, an estimated 7 infants (range: 1–22) would be confirmed to have GAMT deficiency, while approximately 77 (range: 59–88) would be false positives. Additionally, around 10 infants per year (range: 2–26) may receive a diagnosis of a non-targeted condition or remain undiagnosed due to death prior to confirmatory testing.

These projections highlight the relatively low false-positive rate and the potential for early diagnosis through newborn screening compared to clinical identification alone.

- Summary - Modeling projections estimate 7 cases of GAMT deficiency (range: 1-22) would be identified annually through national NBS. There is insufficient evidence to compare to the estimated cases detected in the absence of NBS. There is insufficient evidence to model any clinical outcomes beyond case identification to quantify the potential benefits of screening.

Since the RUSP recommendation in January 2023, a mixed-method cohort of four sibling pairs with GAMT-D and eight age-matched healthy sibling pairs from Canada and published in May 2025 provides useful information on health outcomes.[15] The study aimed to determine the effect of early treatment in GAMT-D by comparing sibling pairs with 1 sibling diagnosed and treated symptomatically, while the other was diagnosed and treated earlier. Interviews and surveys of the sibling pairs provided the assessment data about stages of development, motor and cognitive skills, self-care skills, coordination, behavior, learning abilities/school performance, social skills, and medical history. The mean age of treatment start for the younger sibling was 1.28 years and 5.33 years for the older sibling.

In this report, younger siblings diagnosed and treated earlier consistently showed superior outcomes across all domains, including development, cognition, motor skills, adaptive functioning, behavior, and seizure control. Remarkably, children treated in the neonatal period demonstrated outcomes comparable to healthy controls, while those treated in infancy still outperformed siblings treated after age two. These results highlight the critical importance of presymptomatic treatment and provide strong support for the inclusion of GAMT-D in newborn

screening programs to enable early diagnosis and improve long-term outcomes. Figures 2

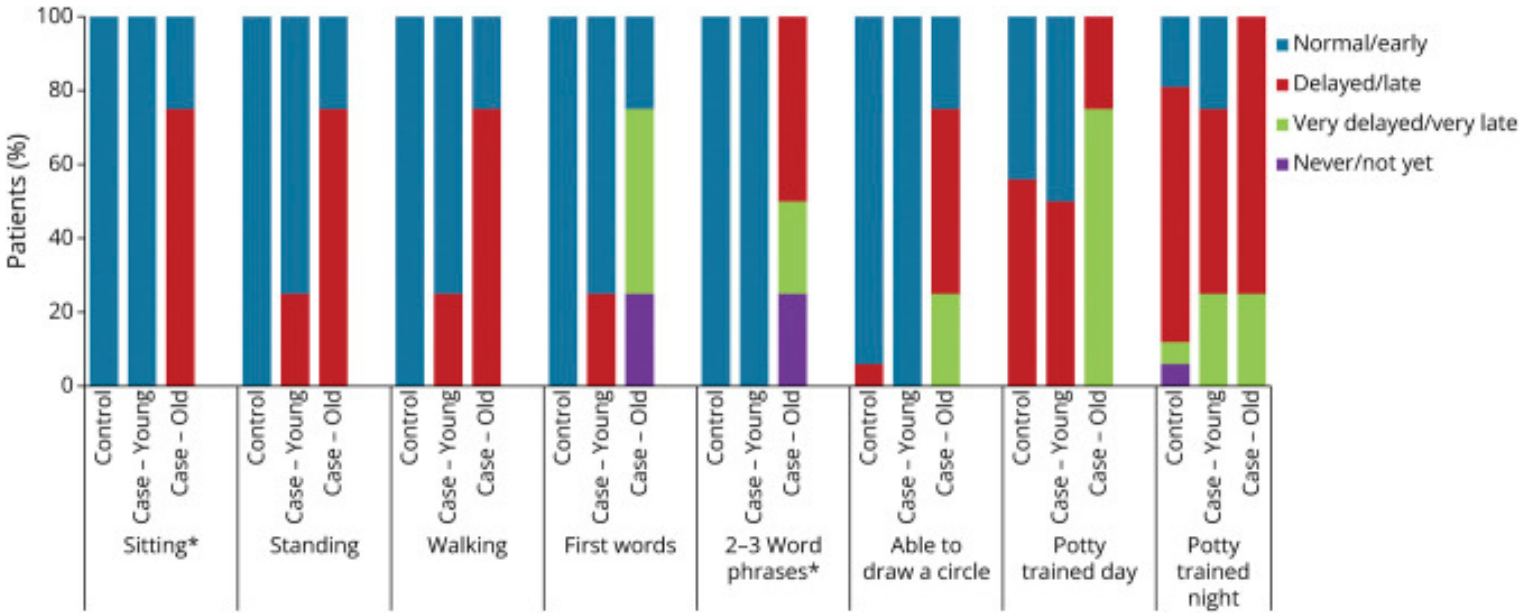


Figure 2: Developmental Milestones. The percentage of participants who achieved early developmental milestones within the normal age range (normal/early), achieved them at an older age (delayed/late, very delayed/very late), or never/not yet achieved them.

through 4 from the publication demonstrate the findings.

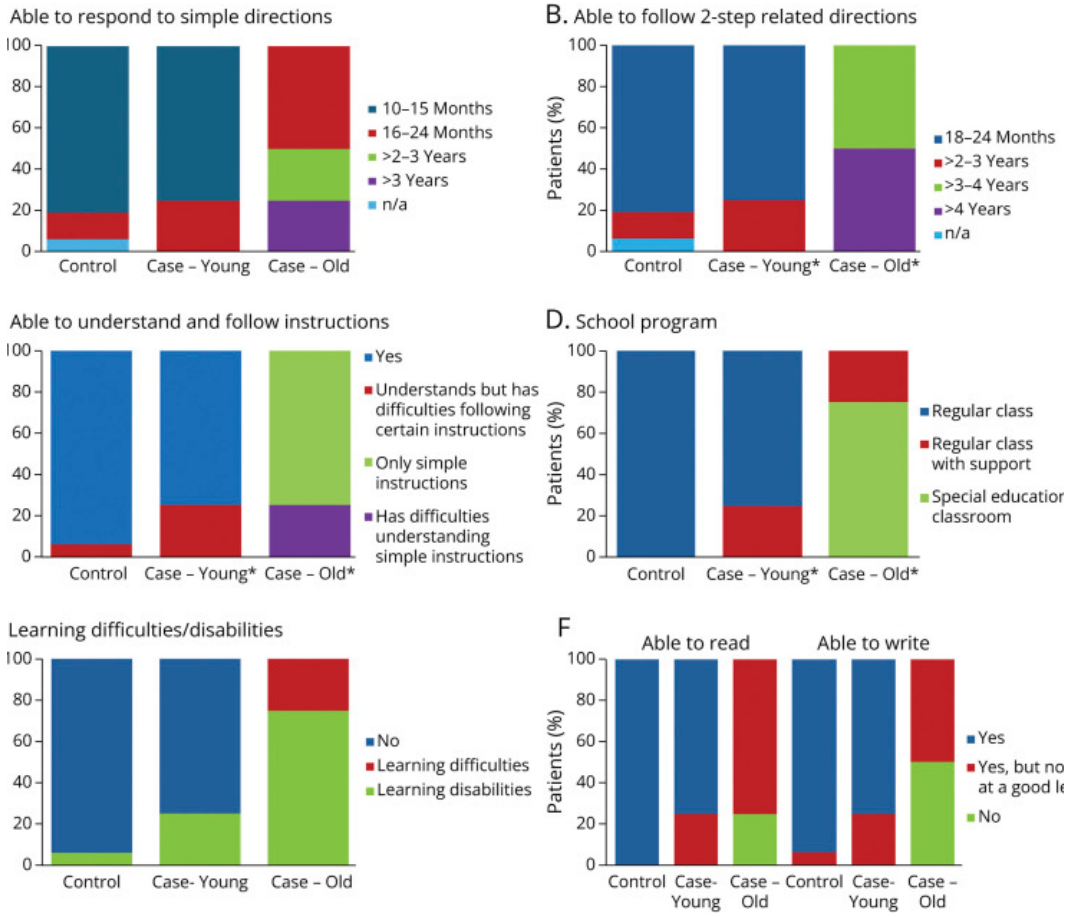


Figure 3. The age in which participants were able (A) to respond to simple directions and (B) to follow 2-step–related directions. (C) The ability to understand and follow complex and simple instructions. The percentage (D)of participants who attend regular class, regular class with support, or special education classroom; (E) of participants with learning difficulties/disabilities; and (F) of participants who can read and write. * p value <0.05, case-young vs case-old.

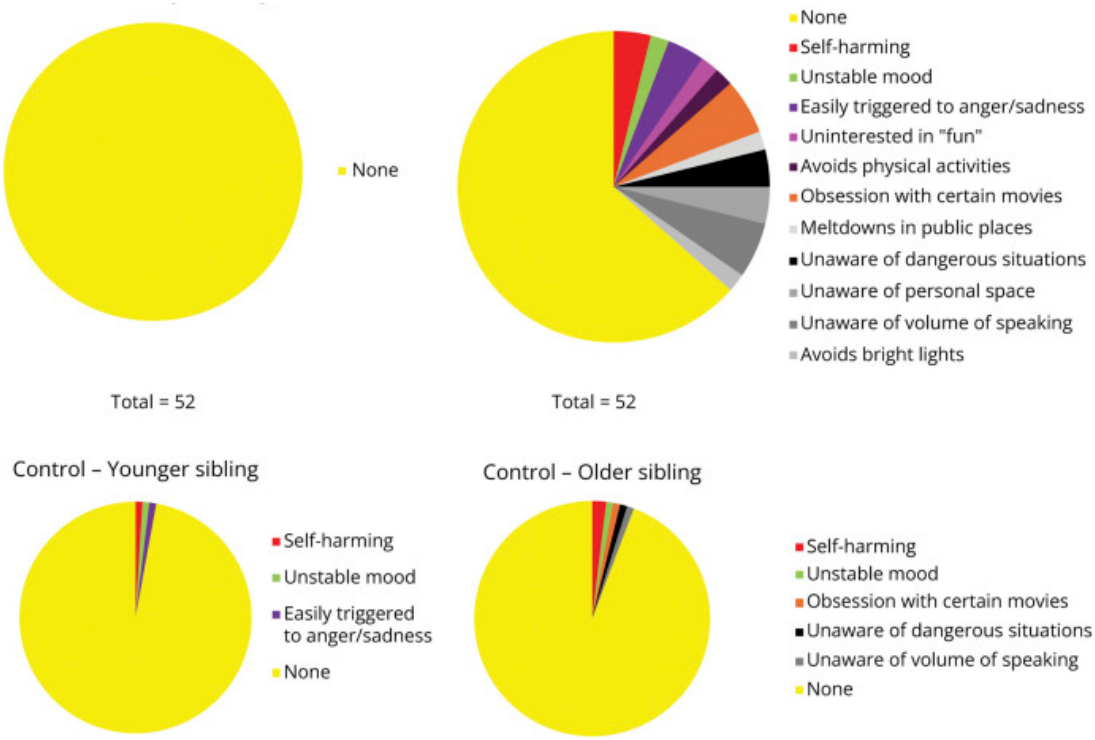


Figure 4. Of 13 different behavioral abnormalities listed in the questionnaire, the parents reported this spectrum of abnormalities in their children. The pie chart represents the number of observations for each behavioral abnormality. The total number reflects the product of the 13 listed abnormalities and the number of participants.

ESTIMATED IMPACT ON NEBRASKA'S NBS PANEL

In 2023, 24,111 babies were reported born in Nebraska.[16] Assuming the screening algorithm includes CRE and GAUC, a positive first-tier screen of 2.6 per 100,000 screened (range 1.7-3.8 per 100,000) applied to 24,111 newborns estimates approximately 1 newborn every 1.5 to 2 years referred for second-tier screening.[14] Using a screen positive of 0.2 per 100,000 (range 0.02-0.6 per 100,000), this rate applied to 24,111 newborns estimates the identification of 1 GAMT-D every 20 years.[14]

RECOMMENDATIONS

Guanidinoacetate Methyltransferase Deficiency (GAMT-D) is a rare but serious disorder of creatine biosynthesis that can lead to profound intellectual disability, epilepsy, and behavioral disturbances if left untreated. Following its inclusion in the U.S. Recommended Uniform Screening Panel (RUSP) in January 2023, Nebraska is evaluating the feasibility and public health

impact of adding GAMT-D to its newborn screening (NBS) panel. Early detection through NBS enables prompt treatment with creatine supplementation, ornithine, and dietary modifications, which can prevent neurological symptoms entirely if started presymptomatically. Implementation in Nebraska will require laboratory validation, provider education, and systems for confirmatory testing and long-term follow-up.

Key Justifications for Inclusion

- GAMT-D meets RUSP criteria: early identification and treatment lead to normal developmental outcomes.
- First-tier mass spectrometry testing is low-cost and can be multiplexed with other disorders.
- Without screening, diagnosis is often delayed for years, missing the window for effective intervention.
- Revvity Omics can support second-tier sequencing and could expand to first-tier biochemical screening.

Key Considerations for Inclusion

- Extremely low incidence (estimated 1 case every 20 years in Nebraska) may challenge perceived cost-benefit.
- Confirmatory testing requires access to specialized metabolic providers and follow-up care.
- Psychosocial, logistical, and insurance barriers may limit timely treatment access.
- Laboratory validation and personnel training are needed for first-tier test implementation.
- Statewide education is required for primary care and follow-up teams to recognize and manage cases.

Actionable Recommendations

1. Add GAMT-D to the Nebraska NBS panel using a first-tier tandem mass spectrometry test for GAUC and creatine.
2. Validate laboratory protocols and cutoffs using existing dried blood spot quality assurance materials.
3. Establish a confirmatory testing protocol with metabolic genetics and biochemical follow-up.
4. Coordinate with Revvity Omics and/or partner labs to offer GAMT gene sequencing as a second-tier test.
5. Educate clinical and NBS staff using ACMG ACT Sheets, diagnostic algorithms, and patient education tools.
6. Build a long-term follow-up system to track development and treatment adherence in diagnosed infants.
7. Prepare for rare positive screens (~1 case per 20 years) with a rapid-response care coordination plan.
8. Promote health equity in education, diagnosis, and care coordination for families receiving results.

Conclusion

Although GAMT-D is an ultra-rare condition, its inclusion in the Nebraska NBS panel is justified by the transformative impact of early diagnosis and treatment. With minimal laboratory burden and the availability of effective therapy, implementation would align Nebraska with national standards while protecting infants from otherwise preventable neurologic injury. Proactive preparation will ensure a sustainable model for rare disease screening and future panel expansion.

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