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Pompe Disease in Infants: Improving the Prognosis by Newborn Screening and Early Treatment



WHAT'S KNOWN ON THIS SUBJECT: Newborn screening program is now technically feasible in ultrarare diseases and can successfully identify patients with Pompe disease before symptoms have progressed to the point where they would be likely to be identified through routine pediatric care.



WHAT THIS STUDY ADDS: This is the first report of improved clinical outcomes in patients with Pompe disease who were identified by a national newborn-screening program and were treated before the onset of clinically recognizable symptoms of Pompe disease.

abstract



OBJECTIVE: Pompe disease causes progressive, debilitating, and often life-threatening musculoskeletal, respiratory, and cardiac symptoms. Favorable outcomes with early intravenous enzyme-replacement therapy and alglucosidase alfa have been reported, but early clinical diagnosis before the development of severe symptoms has rarely been possible in infants.

METHODS: We recently conducted a newborn screening pilot program in Taiwan to improve the early detection of Pompe disease. Six of 206 088 newborns screened tested positive and were treated for Pompe disease. Five had the rapidly progressive form of Pompe disease, characterized by cardiac and motor involvement, and were treated soon after diagnosis. The sixth patient was started on treatment at 14 months of age because of progressive muscle weakness. Outcomes were compared with treated patients whose disease was diagnosed clinically and with untreated historical control subjects.

RESULTS: At the time of this report, patients had been treated for 14 to 32 months. The 5 infants who had early cardiac involvement demonstrated normalization of cardiac size and muscle pathology with normal physical growth and age-appropriate gains in motor development. The infant without cardiac involvement also achieved normal motor development with treatment. Survival in patients who had newborn screening was significantly improved compared with those in the untreated reference cohort ($P = .001$). Survival in the treated clinical comparators was reduced but not statistically different from that in the newborn screening group ($P = .48$).

CONCLUSIONS: Results from this study indicate that early treatment can benefit infants with Pompe disease and highlight the advantages of early diagnosis, which can be achieved by newborn screening. *Pediatrics* 2009;124:e1116–e1125

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KEY WORDS

inborn errors of metabolism, neuromuscular disorders, newborn screening, outcome, treatment

ABBREVIATIONS

GAA—acid α -glucosidase
rhGAA—recombinant human acid α -glucosidase
CRIM—cross-reactive immunologic material
DBS—dried blood spot
NTUH—National Taiwan University Hospital
NAG—neutral glucosidase
CXR—chest radiograph
ECG—electrocardiogram
CK—creatinine kinase
BNP—b-type natriuretic peptide
LVMI—left ventricular mass index
PAS—periodic acid-Schiff
H&E—hematoxylin and eosin
AIMS—Alberta Infant Motor Scales
PDMS-II—Peabody Developmental Motor Scales, Second Edition

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Pompe disease (glycogen storage disease type II, acid maltase deficiency) was first described by Dr Johannes Pompe in 1932 in an infant who had rapidly progressive cardiomyopathy and was found at autopsy to have massive deposition of glycogen in the heart and skeletal muscle.¹ Pompe disease was classified as a lysosomal storage disorder when it was determined that the primary pathology resulted from a deficit of the lysosomal enzyme acid α -glucosidase (GAA) (EC 3.2.1.20).² Worldwide incidence of Pompe disease is estimated to be 1 in 40 000² and may be higher in certain regions (Israel³ and Taiwan⁴).

Pompe disease has been classified as infantile onset or late onset, although a continuum in disease severity, indicative of a single disease with a variable age at onset and rate of progression, is found. The most severe form is typically diagnosed in infants between 3 and 5 months of age during the assessment of a respiratory infection, cardiomegaly, or hypotonia. In natural history studies in infants, the gap between diagnosis and ventilator use or death was 1 to 2 months; nearly all infants died by the age of 18 months.^{5,6}

A Phase 2/3 trial of enzyme-replacement therapy with recombinant human GAA (rhGAA) (alglucosidase alfa [Genzyme Corporation, Cambridge, MA]) in 18 patients who were ≤ 6 months of age at enrollment (corrected for gestational age)⁷ showed that all patients survived after 1 year of treatment, and 15 infants reached 18 months of age; however, 3 patients required invasive ventilatory assistance, 5 were tube-fed, and 5 made no gains in motor development.⁷ In a separate clinical trial of infants and children who were between 3 and 43 months of age at enrollment and had more advanced disease progression, 15 of 21 patients survived to the end of the study (median treatment duration: ~ 2 years), 7 of 16 patients

who were free of invasive ventilation at baseline remained so, and 13 of the total 21 patients demonstrated improvements in motor development.⁸ These results suggest that earlier therapeutic intervention, before extensive muscle damage, could result in better outcomes.

Cross-reactive immunologic material (CRIM) status may also affect treatment outcomes in infantile-onset Pompe disease. CRIM-negative infants have shown poorer outcomes, even with early initiation of treatment.^{7,9,10} CRIM-positive patients have mutations that result in the production of endogenous GAA protein with little, if any, enzyme activity. The presence of endogenous GAA protein may confer some immunologic protection or tolerance against the development of anti-rhGAA antibodies, because the exogenous enzyme is not perceived as foreign protein by the immune system. CRIM-negative infants have no detectable endogenous enzyme and tend to develop high and sustained levels of antibodies to exogenously administered enzyme; therefore, any evaluation of the impact of treatment in infants with Pompe disease must consider CRIM status.

We recently conducted a large-scale newborn screening pilot program to evaluate the impact of early detection in and treatment of infants with Pompe disease.^{11,12} Between October 1, 2005, and December 31, 2007, $\sim 45\%$ of all newborns in Taiwan had GAA activity measured in dried blood spots (DBSs). Of 206 088 newborns screened, 6 cases of Pompe disease were diagnosed. Here we report outcomes in those 6 patients.

METHODS

Screening Program

The Pompe disease newborn screening program was approved by the institutional review board of the National Taiwan University Hospital (NTUH). It was integrated into the preexisting

newborn screening program at the NTUH Screening Center. Taiwan does not require informed consent for newborn screening; however, this pilot screening program was reviewed with parents prenatally, and written informed consent for screening from 1 parent per infant was required. The routine newborn screening DBS specimen was used for Pompe disease screening via enzyme assay, which was conducted as previously described.¹¹ Briefly, GAA enzyme activity of $<55\%$ of the population mean triggered retesting of GAA activity, as well as total neutral glucosidase (NAG) activity in the original DBS. When the NAG/GAA ratio was >100 , the newborn was recalled immediately for confirmatory testing in whole blood; otherwise, when GAA activity was $<25\%$ and NAG/GAA ratio was >25 , a second DBS was obtained and tested. When in the second DBS GAA activity was $<8\%$, NAG/GAA ratio was >60 , and percentage of total GAA inhibition was $>80\%$, the infant was referred to NTUH for diagnostic confirmation via GAA activity assay in whole blood and clinical evaluation.

Clinical Evaluation

For diagnostic confirmation, physical examination, chest radiographs (CXRs), and electrocardiograms (ECGs) were performed. When cardiac involvement was suspected by CXR or ECG, an echocardiogram was performed. When hypertrophic cardiomyopathy was confirmed, the infant was admitted to the hospital for additional baseline evaluations and initiation of treatment with alglucosidase alfa. When no cardiac involvement was found, the patient was followed closely but no treatment was initiated.

Baseline Ancillary Testing

Blood chemistry included creatinine kinase (CK) and B-type natriuretic peptide (BNP). Ejection fraction, thickness of the ventricular walls and interventricular septum, left ventricle end-systolic di-

mension, left ventricle end-diastolic dimension, and calculated left ventricular mass index (LVMI) were measured with echocardiography. Fibroblasts were obtained by skin biopsy for additional confirmation of GAA deficiency by enzyme assay. Quadriceps muscle biopsy specimens were obtained after informed consent. Muscle specimens were fixed in glutaraldehyde, processed for high-resolution light microscopy, and stained with periodic acid-Schiff (PAS) as previously described.¹³ Additional tissue was fixed in formalin, paraffin processed, and stained with hematoxylin and eosin (H&E) stain. Informed consent was obtained for DNA extraction for mutation analysis and for data collection.

Treatment Protocol

Patients with confirmed cardiomyopathy at diagnosis were treated with alglucosidase alfa (manufactured at 2000-L bioreactor capacity); asymptomatic patients were treated when Pompe-associated symptoms appeared. Treatment costs were reimbursed by the Taiwan Bureau of National Health Insurance. After baseline evaluations, intravenous alglucosidase alfa (20 mg/kg) was given every other week, infused according to the product insert. Safety, including infusion-associated reactions, was monitored during the entire treatment period. All patients underwent a physical therapy regimen, including extremity and trunk muscle power training; exercises to enhance head support from prone and supine positions; and exercises to assist with sitting, standing, and solid food intake.

GAA Activity, Genotyping, and Western Blot Analysis

GAA activity in mononuclear cells and/or fibroblasts was measured by using the artificial substrate 4-methylumbelliferyl- α -D-glucopyroside (Fluka Chemical Corp, Ronkonkoma, NY). Genomic DNA from peripheral blood cells was used for muta-

tion analysis of the GAA gene.¹⁴ CRIM status was determined by Western blot analysis.

Follow-up Clinical and Ancillary Testing

Blood chemistry tests were performed monthly. Motor and gastrointestinal evaluations were repeated every 3 months. Quadriceps muscle biopsy, from the opposite leg used for the baseline biopsy, was repeated 6 months after the first infusion.

The Alberta Infant Motor Scale (AIMS) and the Peabody Developmental Motor Scale, Second Edition (PDMS-II), were used. The AIMS is an observational measure of infant motor performance that can be administered from birth through the age of independent walking. It assesses the sequential development of motor milestones.¹⁵ The PDMS-II is a skill-based measure of gross and fine motor development for infants and children from 6 months to 6 years of age and consists of 4 gross motor and 2 fine motor subtests.¹⁶

Clinical Comparators

Ten infants who had Pompe disease diagnosed on the basis of clinical manifestations rather than NBS and were treated with rhGAA at NTUH were included as clinical comparators. Of these, 5 received a diagnosis after clinical presentation in populations that were not covered by the screening program (CLN1–CLN5), and the others were identified and treated between December 2002 and the start of the screening program (CLN6–CLN10). Eleven patients from NTUH who died before the availability of enzyme-replacement therapy and who were described in a previous natural history survey⁵ were used as untreated historical comparators. Survival was compared among the 3 cohorts by using Kaplan-Meier analysis.¹⁷

RESULTS

Cases Detected by the Screening Program

Six of 206 088 newborns screened tested positive for Pompe disease (referred to as NBS1–NBS6) by DBS screen and confirmatory enzyme activity assay in whole blood. The infants were aged 7 days to 40 days at diagnosis; baseline characteristics are summarized in Table 1. All were born to healthy, nonconsanguineous parents; none had obvious clinical symptoms at birth; and all were CRIM-positive (Fig 5, which is published as supporting information at www.pediatrics.org/content/full/124/6/e1116). The 2 cases identified since the publication of the initial results of the screening program,¹¹ NBS5 and NBS6, both were recalled for confirmatory testing in whole blood immediately after the results of the first DBS test were known. Five patients (NBS2–NBS6) had cardiomyopathy at the time of diagnosis; NBS1, who did not, is presented separately.

Infants With Cardiac Involvement

Clinical Presentation

CXR in the 5 patients with hypertrophic cardiomyopathy (NBS2–NBS6) revealed prominent heart shadows (Fig 1). ECG showed biventricular hypertrophy, although the PR intervals were not significantly shortened. BNP and CK levels were markedly elevated (Fig 2).¹⁸ Despite the presence of significant cardiac disease, these infants showed no signs of cardiac disease at rest and had normal routine physical examinations and normal weight gain. Three infants (NBS3–NBS5) demonstrated cyanosis with crying or feeding. Echocardiography revealed thickened ventricular walls and interventricular septum in all 5 infants. LVMI was significantly increased (108.9–186.0 g/m²; reference range¹⁹: 47.4 ± 6.2 g/m²), although the left ventricular

TABLE 1 Characteristics of Patients Whose Pompe Disease Was Detected by Screening

Characteristic	NBS1	NBS2	NBS3	NBS4	NBS5	NBS6
Gestational age, wk	38	39	40	39	37	38
Gender	Female	Female	Male	Male	Female	Male
Birth weight, kg	3.3	3.0	2.9	3.9	3.3	3.4
Age at diagnosis, d	40	19	22	9	33	7
Body weight at diagnosis, kg	4.4	3.3	3.2	3.5	4.6	3.3
Symptoms at diagnosis	—	—	Crying cyanosis	Feeding cyanosis	Feeding cyanosis	—
Age at first infusion	14 mo	26 d	29 d	17 d	34 d	12 d
GAA activity, nmol/mg per h ^a						
Lymphocyte	1.83	0.34	1.58	0.45	3.82	0.83
Fibroblast	0.64	0.06	—	0.11	0.14	0.06
CRIM status	+	+	+	+	+	+
GAA gene mutation						
Allele 1	c.424_440del (p.S142LfsX29) ^b	c.1935C→A (p.D645E)	c.1935C→A (p.D645E)	c.1935C→A (p.D645E)	c.1935C→A (p.D645E)	c.1935C→A (p.D645E)
Allele 2	c.811A→G (p.T271A) ^b	c.1411_1414delGAGA (p.E471fsX5)	c.2842insT (p.L948LfsX70) ^b	c.784G→A (p.E262K)	c.1935C→A (p.D645E)	c.1062C→G (p.Y354X)
Current age, mo	40	34	33	26	20	15
Gross motor status, walk at n mo	15	14	20	14	12.5	13
CK, U/L	101	922	1126	597	565	752
Echocardiography						
LVEF, %	70	56	70	66	73	77
LVMI, g/m ²	42.7	120.0	170.1	186.0	120.3	108.9

LVEF indicates left ventricular ejection fraction.

^a Normal range for GAA activity in lymphocytes and fibroblasts is >60 nmol/mg per h.

^b Novel mutation.

ejection fraction was not changed significantly.

Baseline Muscle Histology

In patients NBS2 to NBS6, histology revealed significant involvement of the muscle fibers at baseline (Fig 3A); the degree of muscle fiber damage indicated advanced disease despite the clinical findings of normal muscle tone and muscle strength in these infants. On H&E staining, most sections showed severe vacuolization of myocytes. Electron microscopy revealed lysosomal glycogen in most cells. Many myocytes had stage 2 (mild) disease.²⁰ Other myocytes were affected by late stage 4 disease and were devoid of myofibrils; myofibrils were completely replaced by cytoplasmic glycogen. Mitochondria remained floating in the cytoplasmic glycogen. Cytoplasmic glycogen was occasionally associated with cell debris in the form of electron-dense membrane whorls. Analysis of PAS-stained sections revealed variable amounts of glycogen in most skeletal myocytes.

Treatment Responses

Patients' ages at diagnosis, defined as the time the patients were brought to the NTUH clinic after positive DBS en-

zyme assay, were between 7 and 33 days, and first infusion of alglucosidase alfa was given between 1 and 7 days after diagnosis (Table 1). Dura-

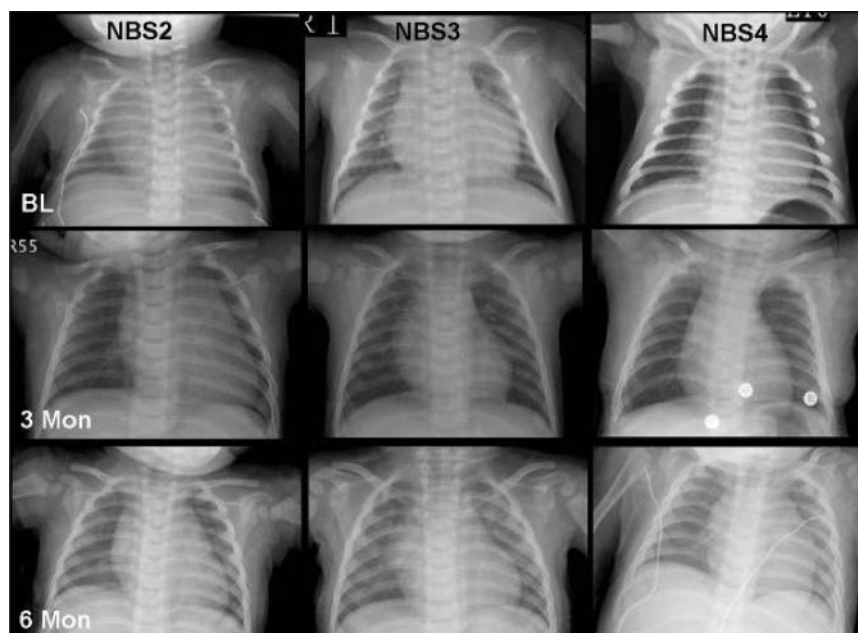


FIGURE 1

CXR for 3 patients who were detected by screening for infantile-onset Pompe disease. Top, Pictures at diagnosis; middle, 3 months after treatment; bottom, 6 months after treatment. The pictures revealed continuous decrease in heart sizes compared with the baseline studies.

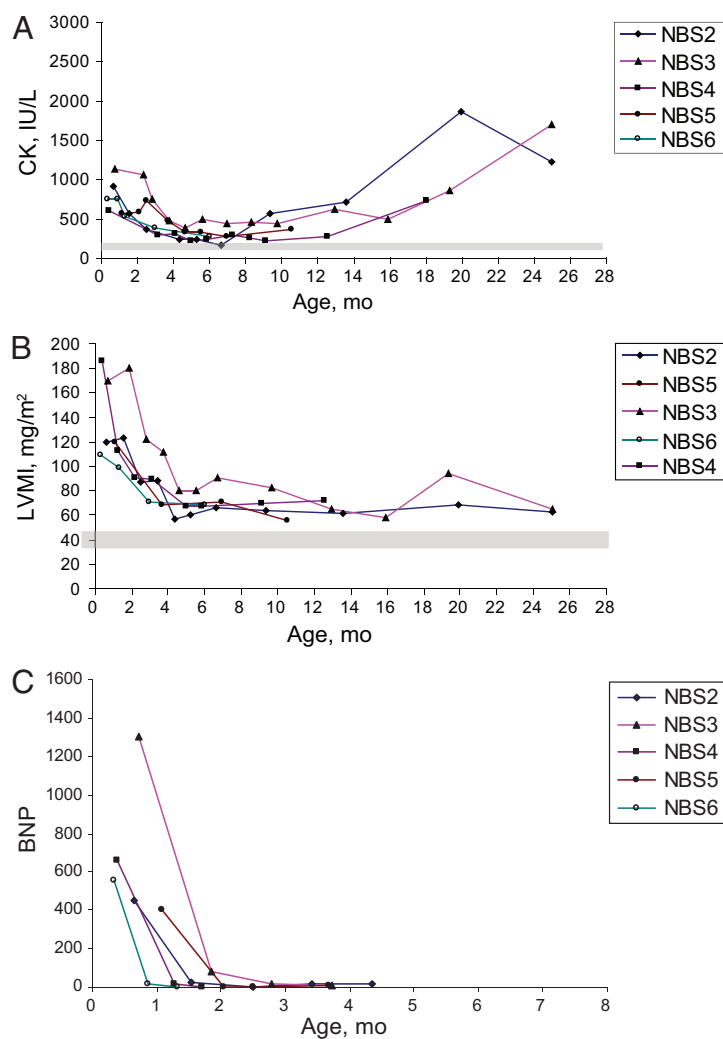


FIGURE 2

Cardiac parameters from baseline through the present for NBS2 to NBS6. A, CK levels; normal range (indicated by gray bar): 38 to 160 U/L. B, LVMI; normal range (gray bar): 47.4 ± 6.2 g/m². C, BNP, normal for infants <1 month of age: median: 28.61 pg/mL; 97.5th percentile: 169.10 pg/mL ($n = 23$).

tion of treatment was between 14 months and 33 months for these patients. All patients survived, remained free of mechanical ventilation, and had normal growth for the duration of the study. Heights were within normal Taiwanese ranges, but NBS3's weight was below the 10th percentile at all time points.

Cardiac Parameters

After treatment with alglucosidase alfa, NBS2 to NBS6 demonstrated a gradual decrease in heart size, as revealed in serial CXR (Fig 1), ECG, and LVMI (Fig 2). CK levels decreased rap-

idly but increased in the older patients after 18 months of age (Fig 2). This elevation of CK might be caused by increased activity (walking) and was not related to antibody titers (data not shown). BNP decreased to normal 1 month after start of treatment (Fig 2).

Motor and Cognitive Development

AIMS assessments showed that NBS2 to NBS6 had mild early motor development delay in the first few months of life (Fig 6, which is published as supporting information at www.pediatrics.org/content/full/124/

6/e1116). Delay in head control was the most common sign, followed by trunk weakness. Results from the PDMS-II revealed that the motor delay was more prominent in gross motor development than in fine motor development (Fig 6). During therapy with alglucosidase alfa, all patients had improvements in motor skills that resulted in attainment of normal AIMS scores and motor development by approximately 1 year of age (Fig 6).

Muscle Histology

On H&E stain of follow-up quadriceps muscle biopsies for patients NBS2 to NBS6 (Fig 3B), most myocytes were intact, although small vacuolization could be seen in some. Electron microscopy showed generally healthy myocytes with intact myofibrils. There were occasional small collections of cytoplasmic glycogen between myofibrils and small lysosomes containing cell debris only (data not shown). In high-resolution light microscopy PAS-stained sections, glycogen had been cleared from most skeletal myocytes; the sarcomeric banding pattern of healthy myofibrils could be clearly seen. Scattered among the healthy myocytes were a few end-stage (stage 4) cells filled with cytoplasmic glycogen, stained purple.

Infant Without Cardiac Involvement

NBS1 was followed closely after diagnosis of Pompe disease at age 40 days. Diagnostic confirmation for this patient was delayed because the family was reluctant to bring the infant in for confirmatory testing (by whole-blood enzyme assay). No cardiac abnormality was revealed by CXR, ECG, or echocardiography; CK remained normal. Head lag and truncal hypotonia were noted at 4 months of age. At 10 months of age, the patient sat only with support from both arms, had prominent hypotonia in the lower extremities,

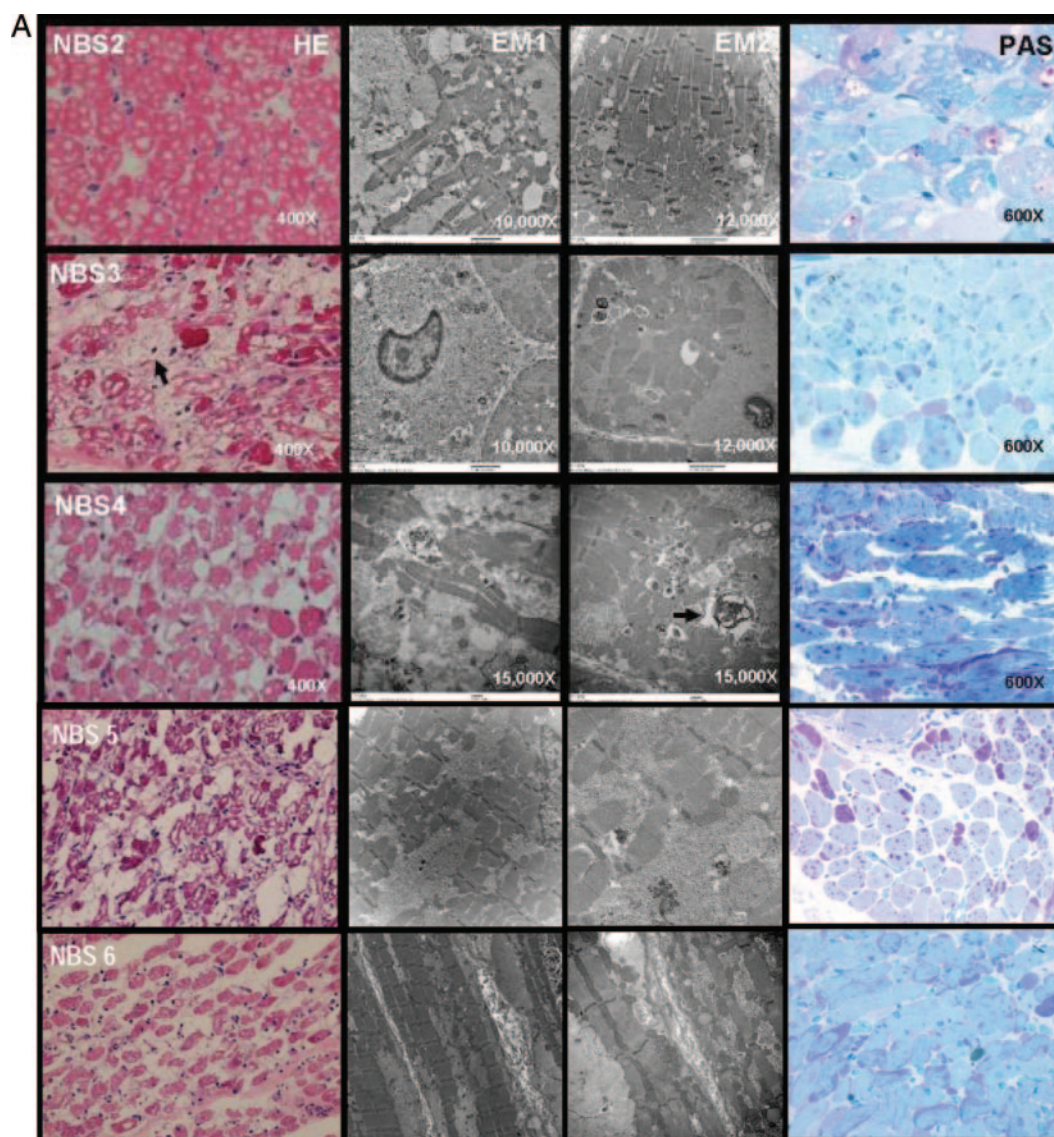


FIGURE 3

Muscle histology (H&E stain, electron microscopy, and PAS stain) at the time of diagnosis (A) and 6 months after treatment (B) for NBS2 to NBS6. A, An arrow in the H&E picture for NBS3 indicates a myocyte filled with storage materials. An arrow in EM2 for NBS4 indicates a vacuole containing cell debris in the form of electron-dense membrane whorls. B, In the H&E stain, arrows indicate small vacuolization in some of the myocytes. In EM, the arrow shows occasional small collections of cytoplasmic glycogen in between myofibrils. PAS stains show rare end-stage cells (arrows) that are filled with glycogen, staining purple, in between normal cells.

could barely support her weight when pulled to stand, and vomited frequently. Videofluoroscopic swallow study showed oropharyngeal dysphagia without aspiration. An upper gastrointestinal tract barium study revealed delayed gastric emptying. Muscle biopsy at 11 months of age revealed multiple, small, punctuate vacuoles in most myocytes; electron microscopy and special stain suggested

that the vacuoles contained lipid (Fig 7, which is published as supporting information at www.pediatrics.org/content/full/124/6/e1116).

At 14 months of age, treatment with alglucosidase alfa was initiated in response to the development of motor symptoms that indicated Pompe disease. The patient's motor development improved quickly, including walking at

15 months, running at 19 months, and climbing stairs at 21 months. Pretreatment CK levels remained in the normal range, 136 and 108 U/L at 11 and 14 months of age, respectively, but decreased to 46 and 19 U/L after 3 and 6 months of treatment. No repeat muscle biopsy was obtained.

In 6 infants who were treated with alglucosidase alfa, only 1 infusion-

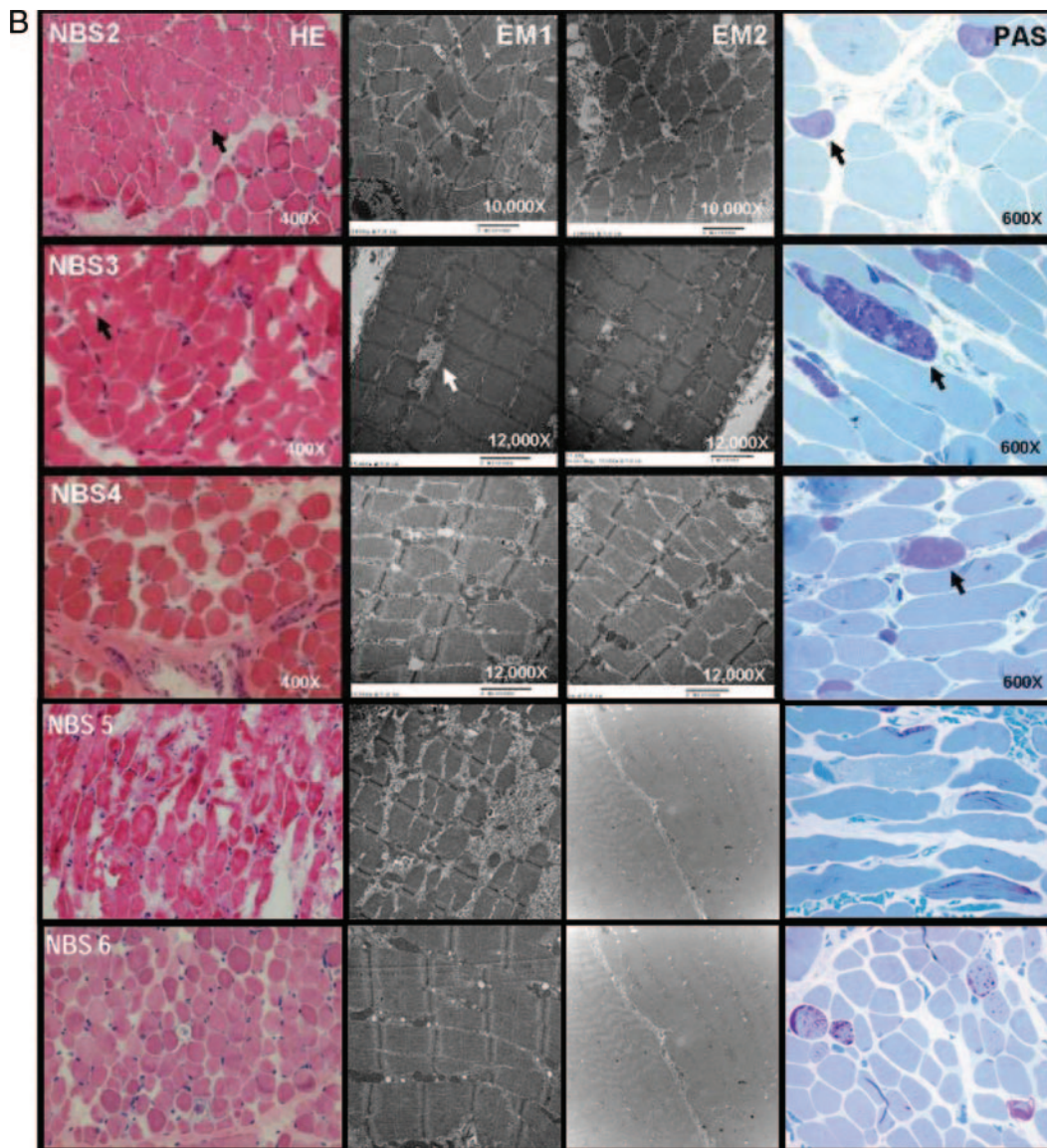


FIGURE 3
Continued.

associated reaction occurred, in patient NBS5 6 months after treatment initiation. The reaction was mild and manifested as transient skin rash. Several episodes of bronchiolitis occurred in NBS1, and 1 episode of gastroenteritis and fever occurred in NBS4, which was assessed to be unrelated to enzyme administration.

Clinical Comparators

All 10 clinical comparators were symptomatic at diagnosis and were CRIM-positive. These patients have been

treated for between 14 months and 76 months. Two were treated at 6 months of age and died at ages 20 months and 48 months with no motor development gains (Table 2, which is published as supporting information at www.pediatrics.org/content/full/124/6/e1116). Another 2 patients, who were treated at 6 months of age, were dependent on respiratory support through tracheostomy 24 hours a day by ages 64 and 73 months; they achieved no motor development. One patient's disease was diagnosed and treated at the age of 2

months because of an audible heart murmur. He had near-normal motor milestones; however, another patient whose disease was diagnosed and treated at the age of 2 months because of elevated liver enzymes (alanine transaminase and aspartate transaminase) had delayed development. The other 4 patients, whose disease was diagnosed between the ages of 4 months and 5 months and began treatment between the ages of 4 months and 6 months, all had delays in motor development.

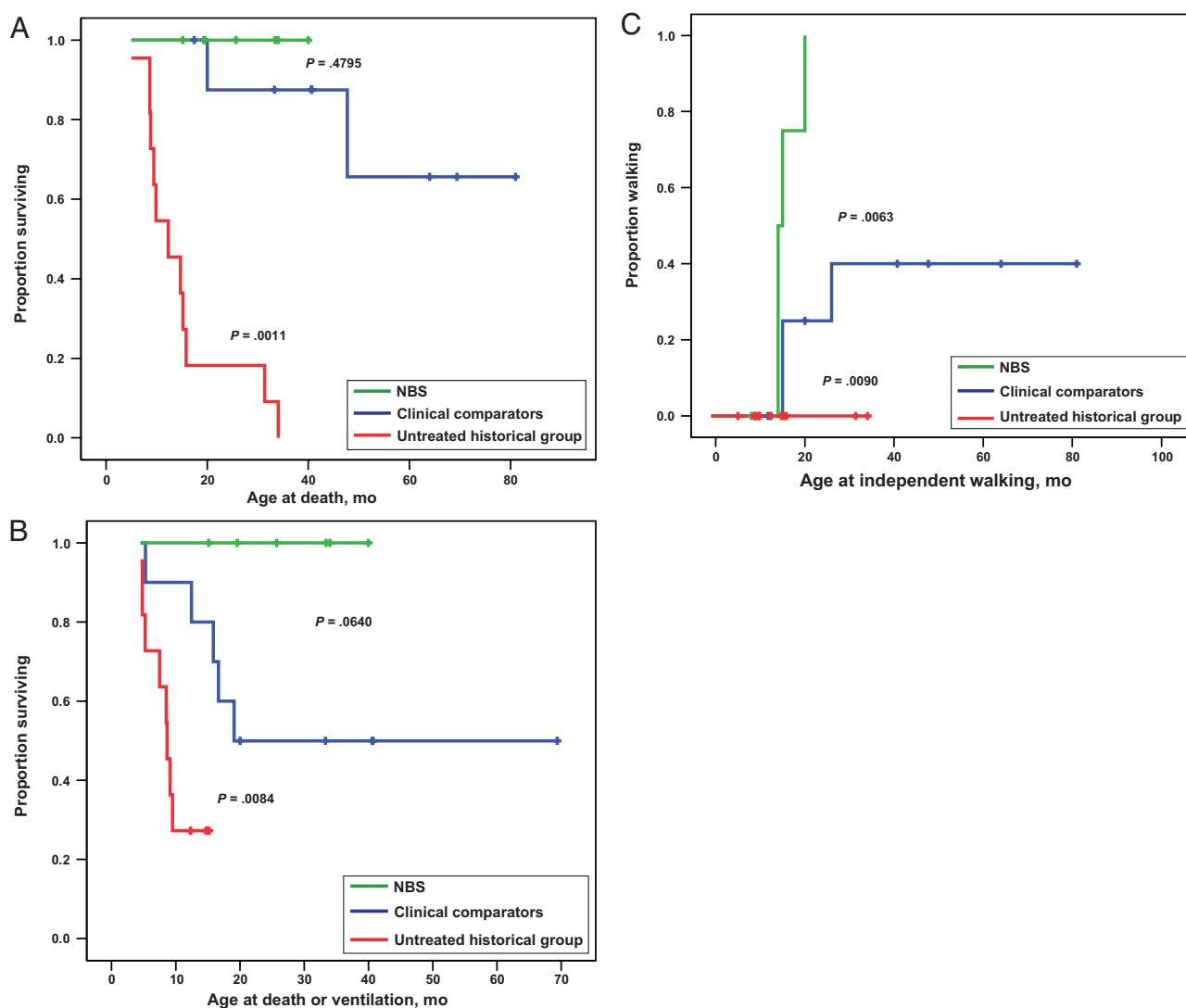


FIGURE 4

Survival and motor outcomes for patients whose disease was detected by newborn screening compared with those whose disease was diagnosed clinically. Green represents the screened cases (NBS); blue represents the clinical cases (clinical comparators); red represents cases from the natural history survey (untreated historical group). A, Kaplan-Meier survival curve for age at death; the difference between the NBS cases and the untreated cases was significant ($P = .001$). B, Kaplan-Meier ventilator-free survival curve for age at death or ventilation; the differences between the NBS cases and the untreated cases was significant ($P = .008$). C, Kaplan-Meier 1-minus-survival plot illustrates the time from birth to independent walking. The differences between the NBS cases and both the clinical cases and the untreated cases were significant ($P = .006$ and $P = .009$).

Outcomes Comparison Among Newborn Screening, Clinical Comparators, and Untreated Reference Cohorts

All patients in the newborn screening group survived; at the end of the study, their ages ranged from 15 months to 40 months. Kaplan-Meier analysis found that survival in these patients was significantly improved as compared with the untreated reference cohort ($P = .001$; Fig 4). Survival in the

treated clinical comparators was reduced but was not statistically different from the newborn screening group ($P = .48$). Survival free of ventilation was also compared and was markedly improved in the newborn screening group compared with both the untreated cohort ($P = .008$) and the clinical comparators ($P = .06$; Fig 4). Kaplan-Meier freedom-from-event analysis identified earlier independent walking in the newborn screening group than the un-

treated reference cohort ($P = .009$; Fig 4) or the clinical comparators ($P = .006$); there was no difference in time to walking between the treated clinical comparators and the untreated reference cohort ($P = .22$).

DISCUSSION

The 6 infants with Pompe disease identified by our newborn screening pilot program had uniformly positive responses to treatment with alglucosi-

dase alfa. This response is more consistent than has been seen in other published studies of infants who were treated with rhGAA,^{8,21} despite that 5 of these 6 patients had very early cardiac involvement, indicating severe, rapidly progressive Pompe disease. All 6 patients had very low GAA activity, comparable to our clinical cases and data from other studies.⁶ The infants seemed to benefit greatly from early detection because treatment was initiated before the onset of obvious symptoms and before significant irreversible muscle damage had occurred. The patients demonstrated reduction of LVMI, age-appropriate motor gains, and improvement in muscle morphology. The small number of end-stage cells in their follow-up muscle biopsies did not seem to affect adversely muscle function, as demonstrated by the achievement of normal motor development.

The infant without cardiac involvement, NBS1, also seems to have benefited from early treatment and may not have been detected until much later without screening. The muscle biopsy result, although different from that seen in the other patients, is not inconsistent with a Pompe disease diagnosis; in late-onset Pompe disease, some muscle fibers may show little to no glycogen,²² and lipid storage has been reported in Pompe disease.^{23,24} This case is likely comparable to the patients included in a trial of older infants and children between 3 and 43 months of age previously described as “nontypical infantile type.”²⁵ Such patients who show signs of disease in the first years of life, even without cardiac manifestation, tend to have very poor outcomes when not treated.²⁶ The patient also may represent a juvenile-onset case, because some patients juvenile-onset Pompe disease have been reported to have had developmental delay in childhood, motor symptoms in the first

years of life, and CK levels within the normal range.^{27,28} Because this patient may have had a course of Pompe disease that would not necessarily be fatal in infancy, we removed NBS1 from the survival analysis and recalculated the *P* value, which remained nonsignificant when compared with the clinical comparators (*P* = .54) and significant when compared with the historical control subjects (*P* = .005).

The outcomes of the treated comparator infants who were detected after clinical manifestations in this study varied. One infant who began treatment at 2 months of age had outcomes similar to the NBS infants who were treated early, whereas another patient who began treatment at 2 months of age had a similar response to that seen in cases of Pompe disease treated at ≥ 6 months of age^{7,21}; however, their outcomes are markedly better than the known natural history of patients with untreated infantile Pompe disease.⁵

It is important to note that all of the infants reported here were CRIM-positive. The Taiwanese population has a high incidence of the missense mutation p.D645E, which is consistent with CRIM-positive status; 5 (83%) of the 6 patients identified in this study had this mutation. Other world populations may have a higher incidence of CRIM-negative mutations; therefore, the clinical results obtained in this study may not be entirely predictive of expected results in other populations.

This study has several potential limitations. The sample of treated patients is small and the length of follow-up is still limited; although our results are promising, it is possible that even with very early identification and treatment, not all infants with Pompe disease will show the type of treatment response seen here. Questions about long-term clinical status in patients with infantile-onset Pompe disease re-

main; the children in this study continue to develop normally, but there is a shortage of published data on the duration of improvement in muscle and cardiac function in patients who have infantile-onset Pompe disease and are treated with alglucosidase alfa. In addition, although we found significant differences between the patients who were identified through newborn screening and the comparator groups of untreated patients and treated patients whose diagnosis was made after clinical symptoms became evident, the comparison is limited by its retrospective nature.

The cumulative clinical experience from multiple trials performed to date and the availability of a high-throughput testing by using DBS samples^{29,30} have set the stage for the implementation of newborn screening for Pompe disease. Results from this study will help to guide the development of appropriate confirmatory diagnostic testing to be performed after an abnormal newborn screening result, as well as clinical protocols that can be used to determine the optimal time to initiate treatment.

CONCLUSIONS

In comparison with findings from previous treated cohorts, the results from this study indicate that early treatment with alglucosidase alfa is critical in the treatment of infants with Pompe disease and highlight the need for early diagnosis, which can be achieved by newborn screening.

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REFERENCES

- Pompe JC. Concerning idiopathic hypertrophy of the heart [in Dutch]. *Ned Tijdschr Geneeskd*. 1932;76:304–311
- Hirschhorn R. Glycogen storage disease type II: acid alpha-glucosidase (acid maltose) deficiency. In: Scriver CR, ed. *The Metabolic and Molecular Bases of Inherited Disease*. 8th ed. New York, NY: McGraw-Hill; 2001:3389–3420
- Bashan N, Potashnik R, Barash V, Gutman A, Moses SW. Glycogen storage disease type II in Israel. *Isr J Med Sci*. 1988;24(4–5):224–227
- Lin CY, Shieh JJ. Molecular study on the infantile form of Pompe disease in Chinese in Taiwan. *Zhonghua Min Guo Xiao Er Ke Yi Xue Hui Za Zhi*. 1996;37(2):115–121
- Kishnani PS, Hwu WL, Mandel H, Nicolino M, Yong F, Corzo D. A retrospective, multinational, multicenter study on the natural history of infantile-onset Pompe disease. *J Pediatr*. 2006;148(5):671–676
- van den Hout HM, Hop W, van Diggelen OP, et al. The natural course of infantile Pompe's disease: 20 original cases compared with 133 cases from the literature. *Pediatrics*. 2003;112(2):332–340
- Kishnani PS, Corzo D, Nicolino M, et al. Recombinant human acid alpha-glucosidase: major clinical benefits in infantile-onset Pompe disease. *Neurology*. 2007;68(2):99–109
- Nicolino M, Byrne B, Wraith JE, et al. Clinical outcomes after long-term treatment with alpha-glucosidase alfa in infants and children with advanced Pompe disease. *Genet Med*. 2009;11(3):210–219
- Amalfitano A, Bengur AR, Morse RP, et al. Recombinant human acid alpha-glucosidase enzyme therapy for infantile glycogen storage disease type II: results of a phase I/II clinical trial. *Genet Med*. 2001;3(2):132–138
- van den Hout JM, Kamphoven JH, Winkel LP, et al. Long-term intravenous treatment of Pompe disease with recombinant human alpha-glucosidase from milk. *Pediatrics*. 2004;113(5). Available at: www.pediatrics.org/cgi/content/full/113/5/e448
- Chien YH, Chiang SC, Zhang XK, et al. Early detection of Pompe disease by newborn screening is feasible: results from the Taiwan screening program. *Pediatrics*. 2008;122(1). Available at: www.pediatrics.org/cgi/content/full/122/1/e39
- Kemper AR, Hwu WL, Lloyd-Puryear M, Kishnani PS. Newborn screening for Pompe disease: synthesis of the evidence and development of screening recommendations. *Pediatrics*. 2007;120(5). Available at: www.pediatrics.org/cgi/content/full/120/5/e1327
- Lynch CM, Johnson J, Vaccaro C, Thurberg BL. High-resolution light microscopy (HRLM) and digital analysis of Pompe disease pathology. *J Histochem Cytochem*. 2005;53(1):63–73
- Ko TM, Hwu WL, Lin YW, et al. Molecular genetic study of Pompe disease in Chinese patients in Taiwan. *Hum Mutat*. 1999;13(5):380–384
- Piper MC, Pinnell LE, Darrah J, Maguire T, Byrne PJ. Construction and validation of the Alberta Infant Motor Scale (AIMS). *Can J Public Health*. 1992;83(suppl 2):S46–S50
- Folio MR, Fewell RR. *Peabody Developmental Motor Scales, 2nd ed. (PDMS-2)*. Austin, TX: PRO-ED, Inc; 2000
- Kaplan EL, Meier P. Nonparametric estimation from incomplete observations. *J Am Stat Assoc*. 1958;53:457–481
- Hwu WL, Chien YH, Lee NC, Chen CA, Wu MH. Elevation of BNP in newborns with hypertrophic cardiomyopathy due to Pompe disease. *Heart*. 2008. Available at: <http://heart.bmj.com/cgi/eletters/94/10/1307#8129>. Accessed March 30, 2008
- Vogel M, Staller W, Buhlmeyer K. Left ventricular myocardial mass determined by cross-sectional echocardiography in normal newborns, infants, and children. *Pediatr Cardiol*. 1991;12(3):143–149
- Thurberg BL, Lynch Maloney C, Vaccaro C, et al. Characterization of pre- and post-treatment pathology after enzyme replacement therapy for Pompe disease. *Lab Invest*. 2006;86(12):1208–1220
- Kishnani PS, Nicolino M, Voit T, et al. Chinese hamster ovary cell-derived recombinant human acid alpha-glucosidase in infantile-onset Pompe disease. *J Pediatr*. 2006;149(1):89–97
- Schooser BG, Muller-Hocker J, Horvath R, et al. Adult-onset glycogen storage disease type 2: clinico-pathological phenotype revisited. *Neuropathol Appl Neurobiol*. 2007;33(5):544–559
- Sarnat HB, Roth SI, Carroll JE, Brown BI, Dungan WT. Lipid storage myopathy in infantile Pompe's disease. *Arch Neurol*. 1982;39(3):180–183
- Verity MA. Infantile Pompe's disease, lipid storage, and partial carnitine deficiency. *Muscle Nerve*. 1991;14(5):435–440
- Slonim AE, Bulone L, Ritz S, Goldberg T, Chen A, Martiniuk F. Identification of two subtypes of infantile acid maltase deficiency. *J Pediatr*. 2000;137(2):283–285
- Hagemans ML, Winkel LP, Hop WC, Reuser AJ, Van Doorn PA, Van der Ploeg AT. Disease severity in children and adults with Pompe disease related to age and disease duration. *Neurology*. 2005;64(12):2139–2141
- Winkel LP, Hagemans ML, van Doorn PA, et al. The natural course of non-classic Pompe's disease: a review of 225 published cases. *J Neurol*. 2005;252(8):875–884
- Van der Beek NA, Hagemans ML, Reuser AJ, et al. Rate of disease progression during long-term follow-up of patients with late-onset Pompe disease. *Neuromuscul Disord*. 2009;19(2):113–117
- Li Y, Scott CR, Chamoles NA, et al. Direct multiplex assay of lysosomal enzymes in dried blood spots for newborn screening. *Clin Chem*. 2004;50(10):1785–1796
- Zhang XK, Elbin CS, Chuang WL, et al. Multiplex enzyme assay screening of dried blood spots for lysosomal storage disorders by using tandem mass spectrometry. *Clin Chem*. 2008;54(10):1725–1728

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