CHINESE HAMSTER OVARY CELL-DERIVED RECOMBINANT HUMAN ACID α-GLUCOSIDASE IN INFANTILE-ONSET POMPE DISEASE

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Objective To conduct an open-label, multinational, multicenter study examining the safety and efficacy of recombinant human acid α-glucosidase (rhGAA) in treatment of infantile-onset Pompe disease.

Study design We enrolled 8 infant patients who had Pompe disease with GAA activity <1% of normal, cardiomyopathy, and hypotonia. In the 52-week initial phase, rhGAA was infused intravenously at 10 mg/kg weekly; an extension phase continued survivors' treatment with 10 to 20 mg/kg of rhGAA weekly or 20 mg/kg every 2 weeks for as long as 153 weeks. Safety measurements included adverse events, laboratory tests, and anti-rhGAA antibody titers. Efficacy evaluations included survival, ventilator use, echocardiograms, growth, and motor and cognitive function.

Result After 52 weeks of treatment, 6 of 8 patients were alive, and 5 patients were free of invasive ventilator support. Clinical improvements included ameliorated cardiomyopathy and improved growth and cognition. Five patients acquired new motor milestones; 3 patients walked independently. Four patients died after the initial study phase; the median age at death or treatment withdrawal for all patients was 21.7 months, significantly later than expected for patients who were not treated. Treatment was safe and well tolerated; no death was drug-related.

Conclusion rhGAA improved ventilator-free survival, cardiomyopathy, growth, and motor function in patients with infantile-onset Pompe disease compared with outcomes expected for patients without treatment. *(J Pediatr 2006;149:89-97)*

Pompe disease (also known as glycogen storage disease type II, acid maltase deficiency, or glycogenosis type II) is a rare autosomal recessive metabolic muscle disease. Deficiency in acid α-glucosidase (GAA) results in lysosomal accumulation of glycogen in many body tissues and ultimately leads to multisystemic pathology.

Historically, Pompe disease has been classified into different subtypes on the basis of the age at symptom onset, extent of organ involvement, and rate of progression to death.[1] The clinical spectrum ranges from a rapidly progressive form (infantile-onset) to a more slowly progressive form (late-onset), with considerable variability and overlap in these extremes.[2-4] The combined incidence of all forms of Pompe disease is approximately 1:40,000.[5,6]

Patients with infantile-onset Pompe disease typically present before 12 months of age with progressive, hypertrophic cardiomyopathy that may obstruct left ventricular outflow; profound muscle weakness and hypotonia; non-attainment or loss of motor milestones; difficulty feeding; and failure to thrive. These patients have a dramatically

AE	Adverse event	CRIM	Cross-reacting immunological material
AIMS	Alberta Infant Motor Scale	ERT	Enzyme replacement therapy
BAER	Brainstem auditory evoked response	IAR	Infusion-associated reaction
BSID-II	Bayley Scale of Infant Development, second edition	Ig	Immunoglobulin
		LVMI	Left ventricular mass index
CHO	Chinese hamster ovary	OAE	Otoacoustic emission
CNS	Central nervous system	(rh)GAA	(recombinant human) α-glucosidase

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Conflict of Interest: P.S.K., A.A., and Y.T.C. have received research/grant support from Genzyme Corporation. P.S.K. and M.N. are members of the Pompe Disease Advisory Board for Genzyme Corporation. Y.T.C. has served as a consultant for Genzyme Corporation. If therapy for Pompe disease proves successful commercially, Duke University and the inventors of the cell line used to generate the enzyme used in this clinical trial (CHO-1 in this manuscript) may benefit financially pursuant to the Duke University's Policy on Inventions, Patents, and Technology Transfer.

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shortened life span. In patients who are not treated, the median age of death ranges from 6.0 to 8.7 months.[4,7] In the most rapidly progressive form, also termed "classic" infantile Pompe disease, the mortality rate is as high as 92% to 95% in the first year of life.[7] In an historical cohort of patients manifesting Pompe disease in the first year of life, irrespective of phenotype, 74% died by 1 year of age, 91% by 2 years of age, and 93% by 3 years of age.[7] Death generally results from cardiac and respiratory failure.[1,3,7]

No approved specific treatment for Pompe disease currently exists. However, recombinant human GAA (rhGAA) has shown physiological activity both in animal disease models and in early clinical trials.[8-15] In 3 pilot studies in severely affected infants, rhGAA (purified from transgenic rabbit milk [11-14] or from Chinese hamster ovary [CHO] cell cultures[8]) markedly ameliorated cardiomyopathy and prolonged all patients' survival beyond 1 year. One of 6 patients given rhGAA from rabbit milk (a preparation that is no longer available) and 1 of 3 patients given CHO cell-derived rhGAA walked independently and remained ventilator-free. The remaining 7 patients from these 3 studies showed lesser degrees of motor improvement and eventually required ventilation. As of January 2006, 3 of the 9 patients in these pilot studies had died and 6 remained alive (unpublished data). The 3 patients who died ranged in age from 1.2 to 4.3 years at the time of death, and the 6 patients who were living ranged in age from 3.7 to 6.5 years as of January 2006 (unpublished data).

Some patients with Pompe disease have a small amount of natural, but inactive, GAA enzyme. This material is called cross-reacting immunologic material (CRIM) because it, like rhGAA, is recognized by anti-GAA antibodies. In the Amalfitano et al CHO-cell rhGAA study,[8] patients who did not become ambulatory lacked CRIM (as determined by means of Western blotting) and produced high anti-rhGAA antibody titers.[8] In the Van den Hout et al study,[11] the patient with the best motor response to rhGAA from transgenic rabbit milk was also CRIM-positive, although these authors did not find a correlation between CRIM status and antibody formation and did not consider that antibodies had an effect on clinical outcomes.

To confirm the promising preliminary results of these studies in a larger group of patients, investigators in the United States and Europe conducted this phase II, open-label, multicenter, multinational trial of enzyme replacement therapy (ERT) with CHO-derived rhGAA in 8 patients with infantile-onset Pompe disease. Results from the initial 52 weeks of therapy and an extension study are presented here, along with the current status of surviving patients.

METHODS

This trial was initially conducted at centers in the United States and Europe and was approved by their institutional review boards and ethics committees. Written informed consent was obtained from parents or guardians. Eight patients with skin fibroblast GAA activity <1% of the normal mean (assayed against 4-methylumbelliferyl-α-d-glucopyranoside), cardiomegaly (cardiothoracic ratio >0.5 by means of chest radiography), and left ventricular hypertrophy (left ventricular mass index [LVMI] $\geq$65 g/m^2, $\geq$2 SD higher than the age-appropriate normal mean[16]) were enrolled. Exclusion criteria included respiratory insufficiency (O_2 saturation in room air <90%, arterial pCO2 >50 mm Hg, or any ventilator use), signs and symptoms of cardiac failure and a cardiac ejection fraction <40%, or a major congenital abnormality or significant organic disease (unrelated to Pompe disease and likely to decrease survival rate). Patients still living after the 52-week initial phase were enrolled into an extension protocol.

Study Methods and Treatments administered

Two preparations (CHO-1 and CHO-2) of the investigational product, rhGAA, were purified from 2 different CHO cell lines transfected with the cDNA for human GAA. The protein sequences of the rhGAAs produced from each preparation are identical to each other and to a commonly occurring form of human GAA, with a calculated protein mass of 99.4 kDa. The recombinant proteins also contain 7 asparagine-linked glycosylation sites and 13 cysteine residues, 12 of which are involved in disulfide linkages. Both enzymes have similar specific activities toward a synthetic substrate and were prepared as 110-kD precursor proteins in a frozen liquid or lyophilized form.

All patients underwent baseline assessments within the 7 days before their first infusion of rhGAA. During the initial 52-week phase, all patients started receiving weekly intravenous infusions of CHO-1 at a dose of 10 mg/kg. In an attempt to improve overall clinical response, the doses were increased for patients H, C, and F. The dose for patient H was increased to 20 mg/kg weekly after week 43 of the initial phase and maintained at that level for 26 further doses. Patient C received 2 doses at 20 mg/kg, starting after week 90, and patient F received 10 doses, starting after week 70. All doses of rhGAA were administered in 3-hour infusions (2 mg/kg for 30 minutes, and the remaining dose for 2.5 hours). An independent safety monitoring board reviewed safety data. Exposure to CHO-1 ranged from 17 to 100 weeks. When a more robust manufacturing process was developed, patients who survived (patients A, C, E, and F) were transitioned to the second rhGAA preparation (CHO-2). These 4 patients received CHO-2 for periods ranging from 17 to 54 weeks. Total exposure to rhGAA, including the extension phase and both preparations, ranged from 17 to 153 weeks. At the time this study was initiated, results from preclinical studies in animal models[9,10] and results from previous clinical trials[8,11,14] suggested that a dose higher than that required for ERT for other lysosomal storage disorders was necessary to have a clinical effect in Pompe disease.

Study Assessments

Safety evaluations included assessment of adverse events (AE); routine physical examinations; vital signs; routine blood