Challenges and opportunities in clinical trials for spinal muscular atrophy

D. Hirtz, MD; S. Iannaccone, MD, FAAN; J. Heemskerk, PhD; K. Gwinn-Hardy, MD; R. Moxley III, MD; and L.P. Rowland, MD

Abstract—Spinal muscular atrophy (SMA) is the most common fatal neuromuscular disease of infancy. SMA type I is the most severe and mortality is usually due to respiratory failure. In type II the disability is of later onset and less severe, and prognosis has improved primarily due to supportive care. Type III is the mildest form with onset usually of weakness in adolescence or young adulthood. SMA is an autosomal recessive disorder with deletions or mutations of the gene at the 5 q11 locus. There is no specific prevention or treatment, but current progress toward potential therapies has been substantial and several candidates including histone deacetylase (HDAC) inhibitors are under consideration for further evaluation. The authors sought to address the challenges and opportunities for testing new therapies for SMA.

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Spinal muscular atrophy (SMA) is a severe and often devastating neurologic disorder of infants and children. The clinical spectrum extends from the most severely affected (SMA I) to those who have relatively preserved strength and a normal life expectancy (SMA III). There is no known treatment for this disorder, but recent findings have led to biologically plausible candidates for therapeutic intervention. Clinical trials for this disease are being planned and executed. We sought to address the challenges of and opportunities for effectively organizing trials of potential new therapeutic agents.

Incidence and prevalence. Although rare, SMA is the most common fatal neuromuscular disease of infancy and the third most common diagnosis of neuromuscular diseases seen in clinics for children <18 years.[1] One in 50 people carries this autosomal recessive gene.[2-4] SMA I has the highest incidence of the three types, but because these children usually do not survive past age 2 years, SMA II and III are more prevalent than SMA I.[5] Estimates based on a national voluntary registry and a few prospective studies give a frequency of 8 to 11 per 100,000 live births. There have been no epidemiology studies in North America.

SMA type I. SMA type I is called Werdnig-Hoffmann disease or infantile onset SMA. Weakness and profound hypotonia are noticeable in the first few months of life. There is a striking discrepancy between the infant's normal social awareness and interaction and motor development. Spontaneous movements are very limited except in the hands and feet, and the infant lies on his or her back in a frog-leg position. Deep tendon reflexes are absent but sphincter tone and sensation are intact. Polyminimyoclonus, or muscle trembling, can be seen in the fingers, and fasciculations are often present in the tongue. Because intercostal muscles are weak, the diaphragm is used to breathe and pectus excavatum and flaring of the lower ribs result. The babies tire easily when feeding and may lose weight and fail to thrive. Both weaknesses from malnutrition and respiratory insufficiency cause susceptibility to aspiration. The most common cause of death is respiratory failure; children rarely survive beyond age 2 years.

SMA type II. In type 2 disease, intermediate or juvenile SMA, milestones are usually normal until onset of weakness between 6 and 18 months of age. The legs tend to be weaker than the arms, so children often come to medical attention because of failure to walk. The pattern of deep tendon reflexes may be variable. Children can sit without support when placed, sometimes walk with bracing, and are now surviving into adolescence and beyond. Good pulmo-

From the National Institute of Neurological Disorders and Stroke (Drs. Hirtz, Heemskerk, and Gwinn-Hardy), NIH, Bethesda, MD; Children's Medical Center, University of Texas Southwest (Dr. Iannaccone), Dallas; University of Rochester Medical Center (Dr. Moxley), NY; and Columbia University (Dr. Rowland), New York, NY.

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Address correspondence and reprint requests to Dr. Deborah Hirtz, 6001 Executive Blvd., Room 2212, Bethesda, MD 20892; e-mail: hirtzd@ninds.nih.gov

nary function and care seems to be a key to prolonged survival.

SMA type III. In patients with the mildest form, type 3 (Kugelberg-Welander disease), independent ambulation is achieved and survival is usually normal.[5-8] Onset of apparent weakness may be anytime after age 18 months, but is often in late childhood or adolescence. It may be confused with limb-girdle muscular dystrophy. The gait is typically waddling with lumbar lordosis, genu recurvatum, and a protuberant abdomen. Deep tendon reflexes may be present or absent. If onset of weakness is later than age 2 years, it is highly likely that ambulation will be possible into the fifth decade or beyond.[9] Although the clinical picture may not be typical of a neurodegenerative disease, a decrease in motor units over time has been documented.[10]

The classification for SMA into three types is useful for understanding prognosis but they are biologically not distinct. With fewer copies of survival motor neuron gene (SMN) 2 (or SMN copy count), there is increase in severity of weakness. However, all patients must have at least one copy of the *SMN2* gene, because complete loss of SMN would be an embryonic lethal condition.[11,12]

Genetics. SMA is an autosomal recessive disorder caused by homozygous deletions or mutations of the *SMN1* gene at the 5 q11 locus[13,14] which result in reduction of full length (fl-SMN) protein necessary for lower motor neuron function.[15-17] There are two copies of the SMN gene on chromosome 5q that code for SMN protein: *SMN1* and *SMN2*. *SMN1* encodes fl-SMN protein, while *SMN2* mostly encodes a protein that is lacking in exon 7 (Δ7-SMN, a less stable protein). The severity of the disease can be modified by extra copies of the *SMN2* gene.[17] All patients have reduced levels of fl-SMN protein, but those with the phenotype of SMA type 1 have as little as 9% of the normal amount of fl-SMN, those with SMA type 2 have 14%, and with SMA type 3, about 18%. Once fl-protein levels approach 23% of normal levels, motor neuron function appears normal, and carriers usually have 45 to 55% fl-SMN protein.

Potential for therapies. There is no cure for SMA, but there has been substantial progress toward possible therapies due to advances in understanding the molecular genetic mechanisms. Since the disease phenotype is proportional to the amount of fl-SMN present,[18] one strategy for pharmacologic intervention in SMA is to enhance production of fl-SMN. Mechanisms for potential specific therapies include enhanced expression of the *SMN2* gene, altering *SMN2* transcript splicing to increase the level of fl-SMN RNA, and other strategies to increase the level or activity of SMN. Active agents including histone deacetylase (HDAC) inhibitors have been identified that can increase the level of fl-SMN.[19-23] These compounds cause deacetylation of histone as well as nonhistone proteins and transcription factors, and thereby can upregulate SMN mRNA ratios.[24] HDAC inhibitors used in humans for other clinical conditions are phenylbutyrate and hydroxyurea.[17] Studies of sodium butyrate in a mouse model and lymphoid cell lines from patients with SMA showed promise,[21] as have cell culture studies of valproic acid.[22] Acyclorubicin has shown ability to raise SMN levels in fibroblasts from patients with SMA I, but has a high toxicity profile.[25] Safety and toxicity for all of these compounds needs to be better defined, and all are under consideration for further evaluation.

Existing resources and current therapeutic trials. There are projects funded by the National Institute of Neurologic Disorders and Stroke (NINDS) and by private foundations working on generating candidate therapies for SMA. The NINDS SMA Project is a preclinical pilot program to develop therapeutics for SMA, using guidance from industry, academia, and the FDA (www.smaproject.org). Candidate compounds will be identified and will undergo extensive screening to see if they enhance fl-SMN expression, have pharmaceutical suitability, and are active in animal models of SMA. This program supports facilities to chemically optimize active compounds, to perform a standardized battery of cellular assays, and to test drugs in animal models of SMA. In addition, private foundations aggressively support therapeutics development efforts that generate candidates for clinical testing. These organizations fund additional resources for clinical research in SMA such as the Indiana University School of Medicine International SMA Patient Registry, funded by Families of SMA since 1986. The registry has compiled information on over 1,100 families including type of SMA, tests used for diagnosis, age at diagnosis, living status, and cause of death. The registry is a source of statistical data and serves as a database of patients for clinical trials (http://www.fsma.org/registry2002.shtmal).

Currently in the United States, there are three collaborative clinical trial groups focused on SMA. AmSMART (American Spinal Muscular Atrophy Randomized Trials, http://acsresearch.swmed.edu/amsmart/) is a clinical consortium that has established a framework for clinical trials, has investigated and validated outcome measures, has performed a pilot trial of treatment with creatine, and plans future pilot trials with other drugs. Project Cure SMA (http://www.fsma.org/clinicaltrials/shtml) has also established a clinical network to do natural history studies, refine clinical outcome measures, and collect data on safety and tolerability of valproate and other drugs. The third network, the Pediatric Neuromuscular Clinical Research Network, has recently been established to provide a regional network in the Northeast for conducting natural history studies and finding and testing effective treatments for SMA (http://www.unmc.edu/sma/). In addition, there are active clinical trials consortia in Europe

as well (European Neuromuscular Center, http://www.enmc.org/trials/trial.cfm).

Lessons from other disorders. Investigators from outside the field of SMA provide useful models for collaborative clinical trial networks; these included pediatric oncology, muscular dystrophy, amyotrophic lateral sclerosis (ALS), Huntington disease, and Parkinson disease (PD). The Children's Oncology Group (COG) is a multicenter collaboration that has transformed pediatric cancer from a usually fatal disease to one that is curable in over 75% of patients (http://www.childrensoncologygroup.org). It began with the unification of four North American pediatric clinical trials groups. It now encompasses 238 pediatric cancer programs in North America, Australia, New Zealand, Switzerland, and the Netherlands, and consolidates administrative and regulatory activities within a single group operations center. Accomplishments include consolidation of data management operations, transfer of data sets, expanded web-based remote data entry systems, and developing criteria and infrastructure for performance monitoring and regulatory compliance. It provides access to state-of-the-art treatment protocols for the vast majority of pediatric cancer patients in the United States.[26]

ALS is an example of an adult onset motor neuron disease for which large scale clinical trials are now underway. Prior to 1995, clinical research in ALS consisted of small trials performed at single sites with a few multicenter trials funded primarily by pharmaceutical companies. These were largely unsuccessful and funding interest by pharmaceutical companies waned. Since then, four regional academic consortia, including first the Western ALS group (WALS) and then the northeastern ALS group (NEALS), have formed and begun treatment trials.[27-29] This model of regional consortia allows access to treatment trials for a large number of patients. A larger umbrella ALS research group is in the process of being formed with goals that include the following: 1) to develop a comprehensive ALS database that defines natural history, 2) to enhance communication, and 3) to foster development of new clinical investigators.

Duchenne muscular dystrophy (DMD) is an example of an x-linked disorder for which effective treatment has been developed as a result of multicenter clinical trials. Those trials were only possible through the formation of a collaborative group called the CIDD group (Clinical Investigators in Duchenne Dystrophy). This group began with four centers in the early 1980s. The principal investigators, clinical coordinators, and biostatisticians participated equally to define the disease, establish criteria for treatment, develop trial treatment protocols, and establish validation of outcome measures. Their early therapeutic trials were negative, but in 1987 they found that prednisone was a beneficial treatment for DMD. Thus, information gained in small scale studies performed by separate groups was used to plan larger longer term studies.[30-33]

Efforts in collaborative research by the Parkinson Study Group (http://www.parkinson-study-group.org) and the Huntington Study Group (http://www.huntington-study-group.org), both progressive degenerative disorders of the CNS, were described. Benefits of collaborative research included diversity of patients enrolled, generalizability, and sufficient sample size and statistical power. The group developed a consensus about a stepwise research plan, interacts with sponsors and regulatory bodies, communicates with the advocacy community and the public, and generates research and training standards and codes of conduct. This organizational structure maintains a strong emphasis on individual investigator-initiated work, which has great value for generating new ideas and hypotheses. Both groups have links to numerous publications on their websites.

Issues addressed. Three central questions were examined: 1) prioritizing candidate drug therapies for use in clinical trials; 2) issues in optimizing clinical trial design including choice of outcome measures, use of placebo controls, and impact of natural history; 3) organization of collaborative research; structure, support services, data management, data sharing, and roles of advocacy groups.

Prioritizing candidate drug therapies. Five drugs met the following two criteria: 1) ability to increase expression of *SMN2* in cultured cells and 2) previous use in humans for any indication. These were aclarubicin, hydroxyurea, indoprofen, phenylbutyrate, and valproate. NINDS compiled reviews of pharmacologic, clinical, and SMA-relevant data, forming the basis for recommendations for proceeding with clinical testing or for gathering additional preclinical data. Drugs requiring additional modification before clinical testing, for example to improve the ability to cross the blood–brain barrier, will be considered by the NINDS SMA Project for preclinical therapeutics development. Drugs that can increase *SMN2* expression in cell culture at clinically achievable concentrations in the CNS are good candidates for clinical studies in SMA.

The Food and Drug Administration recommends that an IND application be submitted when a new indication is being considered, even for drugs approved for human use in other indications. This gives the FDA an early opportunity to comment on the study design and help avoid problems that may interfere with later approval for new use. In addition, obtaining orphan drug status from the FDA provides major incentives for eventual marketing of a drug for a rare disease, including extended marketing exclusivity and substantial tax credits for expenses related to clinical trials.

Issues in clinical trial design. Determining the best outcome measures to use in trials and standardizing them across different trials and centers is crit-

ical. Outcome measures were organized according to survival time (or time to event), nutrition and growth measures, strength and motor function, physiologic measures such as motor unit number estimate (MUNE) and compound muscle action potential (CMAP), biochemical measures including SMN RNA, and quality of life (QOL) measures. The infant outcome measures are especially needed; i.e., standardization of motor function using the test of infant motor performance (TIMP) and infant pulmonary function tests. A validation study is currently under way at one center but enrollment is behind schedule; it was affirmed that the research community should support the completion of this study by referring patients when possible. Establishing reliability of the MUNE/CMAP is crucial because these tests appear to relate very closely to the disease process, but require different levels of patient compliance. The challenges to standardizing this sensitive outcome measure include the time and expense required for training and reliability testing. It may be necessary to identify and appropriately compensate specific key EMG investigators to participate in trials.

Survival or time to ventilatory support are clearly primary outcome measures for children with type I disease. Motor function should be considered for the primary outcome of intervention trials; the TIMP, gross motor function measure (GMFM), and the Children's Hospital of Philadelphia Test of Strength in Spinal Muscular Atrophy should be validated and the GMFM compared to the Hammersmith Scale for toddlers and children for assessing motor function. Studies must incorporate inter-rater reliability monitoring.

Muscle strength testing as an outcome measure can presently be determined by two methods of quantitative strength testing: hand-held myometry and fixed myometry connected to a computer. A study to compare hand held myometry (generally used in Europe) with fixed myometry (used in the United States) would be helpful. Pulmonary function tests (PFTs) should be considered as a strength measure, although these are not yet reliable in infants. Other secondary outcome measures include growth and quality of life. If trials are instituted in a presymptomatic clinical phase, deterioration, for example in the MUNE, would become an important outcome.

Placebo controls, possibly for some phase II safety studies but particularly for phase III studies, are recommended though understandably difficult to accept for parents. Without these, efficacy and toxicity cannot be determined. Patients should be stratified within the study according to motor function; i.e., non-sitters, sitters, and walkers. Approaches to design include crossover as well as placebo-controlled studies. Investigators should be consistent in their approach to patients considering clinical trials, and should not promote one experimental therapy over another, but should educate families about equipoise.

Enrollment in trials may be difficult because of small numbers of patients. Regionalization of study groups would help enrollment. Better epidemiologic data would also help by providing information regarding distribution of patients. Families should be referred to enroll in clinical trials as soon as they are diagnosed. Thus, support for a registry of all patients with newly diagnosed SMA, initially national with the potential to expand to international, was considered top priority, as was setting goals for the registry. Perhaps a statement about how to seek information about the registry and clinical trials should be sent to the referring physician with any positive diagnostic laboratory DNA report. The institution of routine neonatal screening for SMA would be of substantial benefit to early diagnosis and referral for clinical trials. Then, innovative designs involving presymptomatic patients could be considered.

Organization of collaboration. Few patients are newly diagnosed each year in any one single center, making enrollment for Phase III studies a challenge. Coordination on a national and international level is desirable and there is need to maximize resources and recruitment of patients and families, and to move as quickly as possible toward evaluation of safety and effectiveness of potential treatments. Enlisting the widespread support of families and their physicians will be crucial.[34] A number of existing resources can be identified that will further these goals.

We recommend that an organized collaborative infrastructure be developed. A central element should be enhancement of the existing registry. This would help identify the maximum number of potentially eligible clinical trial subjects, and would serve to assess the impact and economic burden of SMA, quality of life, natural history, epidemiology, and family history. There would be potential for biologic specimen collection as a shared resource. Methods for recruitment include websites, support groups, newborn screening, and DNA testing centers. Goals for a minimum database should be set that would be determined by the proposed use of the registry.

SMA patient and family organizations exist throughout the world. Advocacy groups can help greatly with public education, patient recruitment, participation on committees, infrastructure support, funding trials and preclinical studies, working with industry to develop therapies and fund clinical research, helping with clinical and patient safety issues, and incorporating families' needs into research planning.

Within a collaborative structure, regional groups could be organized and a central steering body could be developed that would be new, inclusive, and transparent. Such a steering body or administrative core would be initially comprised of patient representatives and scientific leaders of existing projects. This organization would initially be project driven, but could lead to a permanent infrastructure with a coordinating statistical center, common databases and data elements, and widespread accessibility.

Final recommendations. *Establish an international SMA study group.* Enhance cooperation of clinical researchers, family/advocacy groups, and government representatives to maximize the participation of families and patients, to develop consensus on validated, widely accepted outcome measures, to broaden the content and use of a resource registry, and to establish standards for clinical care. The formation of this group would initially center around these specific projects. International participation would be encouraged.

Expedite drug development for clinical trials in SMA. Encourage and promote the development of new therapies, review candidate compounds using established criteria, and ensure consistency and uniformity in the assessment of future compounds.

Form a working group on outcome measures for clinical trials. Goals include comparison and assessment of outcome measures for clinical trials, especially for the youngest age group, standardization and validation of outcome measures, and development of methods to ensure reliability.

Expand and enhance the existing SMA patient registry. Create a common database and data elements, ascertain natural history, facilitate communication among families, eliminate of duplication of resources, and provide assistance in organizing and contacting possible subjects for clinical trials. The possible additional function as a biologic repository, with phenotypic characterization available for samples, should be considered. Data should be accessible to all investigators through a central coordinating center. Ideally, the registry could include data on burden of disease and quality of life. Support and input from parents and advocacy groups would be essential.

Develop guidelines for clinical care. Variations in management, nutritional, orthopedic, and pulmonary care affect the design of clinical trials because they modify the clinical course, particularly in terms of mortality and pulmonary function. Therefore, standardization of optimal care would be desirable both for patient benefit and for understanding the effect of treatment in clinical trials. Review and classification of the literature to develop evidence-based guidelines for clinical care should be attempted; if this is not possible, consensus-based guideline should be developed and studies to support evidence-based recommendations should be undertaken.

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Appendix

Workshop Chairs: Richard T. Moxley III, MD, University of Rochester Medical Center, Rochester, NY; Lewis P. Rowland, MD, Columbia University, New York, NY.

Organizing Committee: Deborah Hirtz, MD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD; Susan T. Iannaccone, MD, FAAN, Texas Scottish Rite Hospital for Children, Dallas, TX; Katrina Gwinn-Hardy, MD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD; Jill Heemskerk, PhD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD.

Participants: Enrico S. Bertini, MD, Bambino Gesú Children's Research Hospital, Rome, Italy; Christina B. Brahe, PhD, Universita Cattolica del Sacro Cuore, Rome, Italy; Wilson W. Bryan, MD, US Food and Drug Administration, Rockville, MD; Arthur Burghes, PhD, Ohio State University, Columbus, OH; Kate Bushby, MD, FRCP, University of Newcastle upon Tyne, United Kingdom; Avital Cnaan, PhD, Children's Hospital of Philadelphia, Philadelphia, PA; P. Michael Conneally, PhD, Indiana University School of Medicine, Indianapolis, IN; Anne M. Connolly, MD, Washington University School of Medicine, St. Louis, MO; Robin Conwit, MD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD; Thomas Crawford, MD, Johns Hopkins University, Baltimore, MD; Merit E. Cudkowicz, MD, Massachusetts General Hospital, Charlestown, MA; Valerie A. Cwik, MD, Muscular Dystrophy Association, Tucson, AZ; Basil T. Darras, MD, Children's Hospital, Harvard Medical School, Boston, MA; Marianne de Visser, MD, PhD, University of Amsterdam, Amsterdam, The Netherlands; Darryl C. DeVivo, MD, Columbia University, New York, NY; Stephen J. Durako, Westat, Rockville, MD; Diana M. Escolar, MD, Children's National Medical Center, Washington, DC; John J. Feeney III, MD, US Food and Drug Administration, Rockville, MD; Richard S. Finkel, MD, The Children's Hospital of Philadelphia, Philadelphia, PA; Sharron E. Gargosky, PhD, Ucycld Pharma, Inc., Scottsdale, AZ; Rene Gonin, PhD, Westat, Rockville, MD; Jacqueline M. Gray, BS, Indiana University School of Medicine, Indianapolis, IN; Sharon E. Hesterlee, PhD, Muscular Dystrophy Association, Tucson, AZ; Linda S. Hynan, PhD, University of Texas Southwestern Medical Center, Dallas, TX; Cynthia Joyce, SMA Foundation, New York, NY; Petra Kaufmann, MD, MSc, Columbia University, New York, NY; Cheryl Kitt, PhD, National Institute of Arthritis and Musculoskeletal and Skin Diseases, NIH, Bethesda, MD; Story C. Landis, PhD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD; Robert T. Leshner, MD, Children's National Medical Center, Washington, DC; Audrey Lewis, Families of SMA, Libertyville, IL; Christian L. Lorson, PhD, University of Missouri, Columbia, MO; Leslie A. Morrison, MD, University of New Mexico School of Medicine, Albuquerque, NM; Tan T. Nguyen, MD, PhD, Food and Drug Administration, Rockville, MD; Robert E. Pacifici, PhD, Eli Lilly and Company, Research Triangle Park, NC; Shree Pandya, MS, PT, University of Rochester Medical Center, Rochester, NY; Marc C. Patterson, MD, Columbia University Medical Center, New York, NY; Audrey S. Penn, MD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD; Thomas Prior, PhD, Ohio State University, Columbus, OH; Karen R. Rabb, RN, CCRC, CCRA, Texas Scottish Rite Hospital for Children, Dallas, TX; Bernard Ravina, MD, National Institute of Neurologic Disorders and Stroke, NIH, Rockville, MD; Sandra P. Reyna, MD, University of Utah School of Medicine, Salt Lake City, UT; Sabina I. Robinson, PhD, Science Applications International Corp., Pittsburgh, PA; Albert T. Roy, CCRP, Children's Oncology Group, Bethesda, MD; Lee L. Rubin, PhD, Curis, Inc., Cambridge, MA; Jeremy M. Shefner, MD, PhD, Upstate Medical University, Syracuse, NY; Neil Shneider, MD, PhD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD; Ira Shoulson, MD, University of Rochester Medical Center, Rochester, NY; Louise R. Simard, PhD, Sainte-Justine Hospital Research Center, Montreal, QC Canada; Christopher Spancake, PhD, Families of SMA, Libertyville, IL; Giovanna M. Spinella, MD, Office of Rare Diseases, NIH, Bethesda, MD; Charlotte J. Sumner, MD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD; Kathryn J. Swoboda, MD, PhD, University of Utah School of Medicine, Salt Lake City, UT; Elizabeth A. Thom, PhD, George Washington University Biostatistics Center, Rockville, MD; Eduardo F. Tizzano, MD, PhD, Hospital Sant Pau, Barcelona, Spain; Andoni J. Urtizberea, MD, Association Française contre les Myopathies, Evry, France; Jiri Vajsar, MD, MSc, FRCP-C., The Hospital for Sick Children, Toronto, ON Canada; Louis Viollet, MD, PhD, Association Francaise contre les Myopathies, Evry, France; Ching H. Wang, MD, PhD, Stanford University Medical Center, Stanford, CA; Brunhilde Wirth, PhD, University Clinic of Cologne, Cologne, Germany; Katherine Woodbury-Harris, PhD, National Institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD; Brenda L. Wong, MD, MBBS, MRCP, Cincinnati Children's Hospital Medical Center, Cincinnati, OH; Robert A. Zalutsky, PhD, National institute of Neurologic Disorders and Stroke, NIH, Bethesda, MD.

References

1. Iannaccone ST, Khatri IA. Childhood spinal muscular atrophy. In: Younger DS, ed. Motor disorders. Philadelphia: Lippincott Williams & Wilkins, 2005.
2. Emery AEH. Population frequencies of inherited neuromuscular diseases—a world survey. Neuromuscul Disord 1991;1:19–29.
3. Merlini L, Stagni SB, Marri E, Granata C. Epidemiology of neuromuscular disorders in the under-20 population in Bologna Province, Italy. Neuromuscul Disord 1992;2:197–200.
4. Pearn J. Incidence, prevalence and gene frequency studies of chronic childhood spinal muscular atrophy. J Med Genet 1978;15:409–413.
5. Byers RK, Banker BQ. Infantile muscular atrophy. Arch Neurol 1961;5: 140–164.
6. Dubowitz V. Chaos in the classification of SMA: a possible resolution. Neuromuscul Disorders 1995;5:3–5.
7. Zerres K, Rudnik-Schoneborn S, Forrest E, Lusakowska A, Borkowska J, Hausmanowa-Petrusewicz I. A collaborative study on the natural

history of childhood and juvenile onset proximal spinal muscular atrophy (type II and III SMA): 569 patients. J Neurol Sci 1997;146:67–72.
8. Zerres K, Davies KE. 59th ENMC International Workshop: Spinal muscular atrophies: recent progress and revised diagnostic criteria. Neuromuscul Disord 1999;9:272–278.
9. Russman BS, Buncher CR, White M, et al. Function changes in spinal muscular atrophy II and III. The DSN/SMA Group. Neurology 1996;47: 973–976.
10. Bromberg MB, Swoboda KJ, Lawson VH. Counting motor units in chronic motor neuropathies. Exp Neurol 2003;Suppl 1:53–57.
11. Monani UR, Coovert DD, Burghes AH. Animal models of spinal muscular atrophy. Hum Mol Genet 2000;9:2451–2457.
12. DiDonato CJ, Brun T, Simard LR. Complete nucleotide sequence, genomic organization, and promoter analysis of the murine survival motor neuron gene (SMN). Mamm Genome 1999;10:638–641.
13. Brzustowicz LM, Lehner T, Castilla LH, et al. Genetic mapping of chronic childhood-onset spinal muscular atrophy to chromosome 5 q11.2-13.3. Nature 1990;344:540–541.
14. Melki J, Abdelhak S, Sheth P, et al. Gene for chronic proximal spinal muscular atrophies maps to chromosome 5q. Nature 1990;344:767–768.
15. Gilliam TC, Brzustowicz LM, Castilla LH, et al. Genetic homogeneity between acute (SMA I) and chronic (SMA II & III) forms of spinal muscular atrophy. Nature 1990;345:823–825.
16. Lefebvre S, Burlet P, Liu Q, et al. Correlation between severity and SMN protein level in spinal muscular atrophy. Nat Genet 1997;16:265–269.
17. Iannaccone ST, Smith SA, Simard LR. Spinal muscular atrophy. Curr Neurol Neurosci Rep 2004;4:74–80.
18. Gavrilov DK, Shi X, Das K, Gilliam TC, Wang CH. Differential SMN2 expression associated with SMA severity. Nat Genet 1998;20:230–231.
19. Andreassi C, Jarecki J, Zhou J, et al. Aclarubicin treatment restores SMN levels to cells derived from type 1 spinal muscular atrophy patients. Hum Molec Genet 2001;10:2841–2849.
20. McCaffrey PG, Newsome DA, Fibach E, Yoshida M, Su MS. Induction of gamma-globin by histone deacetylase inhibitors. Blood 1997;90:2075–2083.
21. Chang JG, Hsieh-Li HM, Jong YJ, Wang NM, Tsai CH, Li H. Treatment of spinal muscular atrophy by sodium butyrate. Proc Natl Acad Sci USA 2001;98:9808–9813.
22. Sumner CJ, Huynh TN, Markowitz JA, et al. Valproic acid increases SMN levels in spinal muscular atrophy patient cells. Ann Neurol 2003; 54:647–654.
23. Brichta L, Hofmann Y, Hahnen E, et al. Valproic acid increases the SMN2 protein level: a well-known drug as a potential therapy for spinal muscular atrophy. Hum Mol Genet 2003;12:2481–2489.
24. Jarecki J, Chen J, Whitney M, et al. Identification and profiling of compounds that increase full-length SMN2 nRNA levels. Presented at the Sixth Annual International Spinal Muscular Atrophy Research Group Meeting; Chicago, IL; 2002. Abstract.
25. Andreassi C, Jarecki J, Zhou J, et al. Aclarubicin treatment restores patients. Hum Molec Genet 2001;10:2841–2849.
26. Ross JA, Severson RK, Pollock BH, et al. Childhood cancer in the United States. A geographical analysis of cases from the Pediatric Cooperative Clinical Trials groups. Cancer 1996;77:201–207.
27. Traynor BJ, Zhang H, Shefner J, et al. Functional outcome measures as clinical trial endpoints in ALS. Neurology 2004;63:1933–1935.
28. Shefner JM, Cudkowicz ME, Schoenfeld D, et al. A clinical trial of creatine in ALS. Neurology 2004;63:1656–1661.
29. Cudkowicz ME, Shefner JM, Schoenfeld DA, et al. A randomized, placebo-controlled trial of topiramate in amyotrophic lateral sclerosis. Neurology 2003;61:456–464.
30. Brooke MH, Fenichel GM, Griggs RC, et al. Clinical investigation in Duchenne dystrophy: 2. Determination of the "power" of therapeutic trials based on the natural history. Muscle Nerve 1983;6:91–103.
31. Brooke MH, Fenichel GM, Griggs RC, et al. Clinical investigation of Duchenne muscular dystrophy: interesting results in a trial of prednisone. Arch Neurol 1987;44:812–817.
32. Brooke MH, Griggs RC, Mendell JR, et al. Clinical trial in Duchenne dystrophy. 1. The design of the protocol. Muscle Nerve 1981;4:186–197.
33. Mendell JR, Moxley RT, Griggs RC, et al. Randomized, double-blind six-month trial of prednisone in Duchenne's muscular dystrophy. N Engl J Med 1989;320:1592–1597.
34. Caldwell PHY, Butow PN, Craig JC. Pediatricians' attitudes towards randomized controlled trials involving children. J Pediatr 2002;141: 798–803.