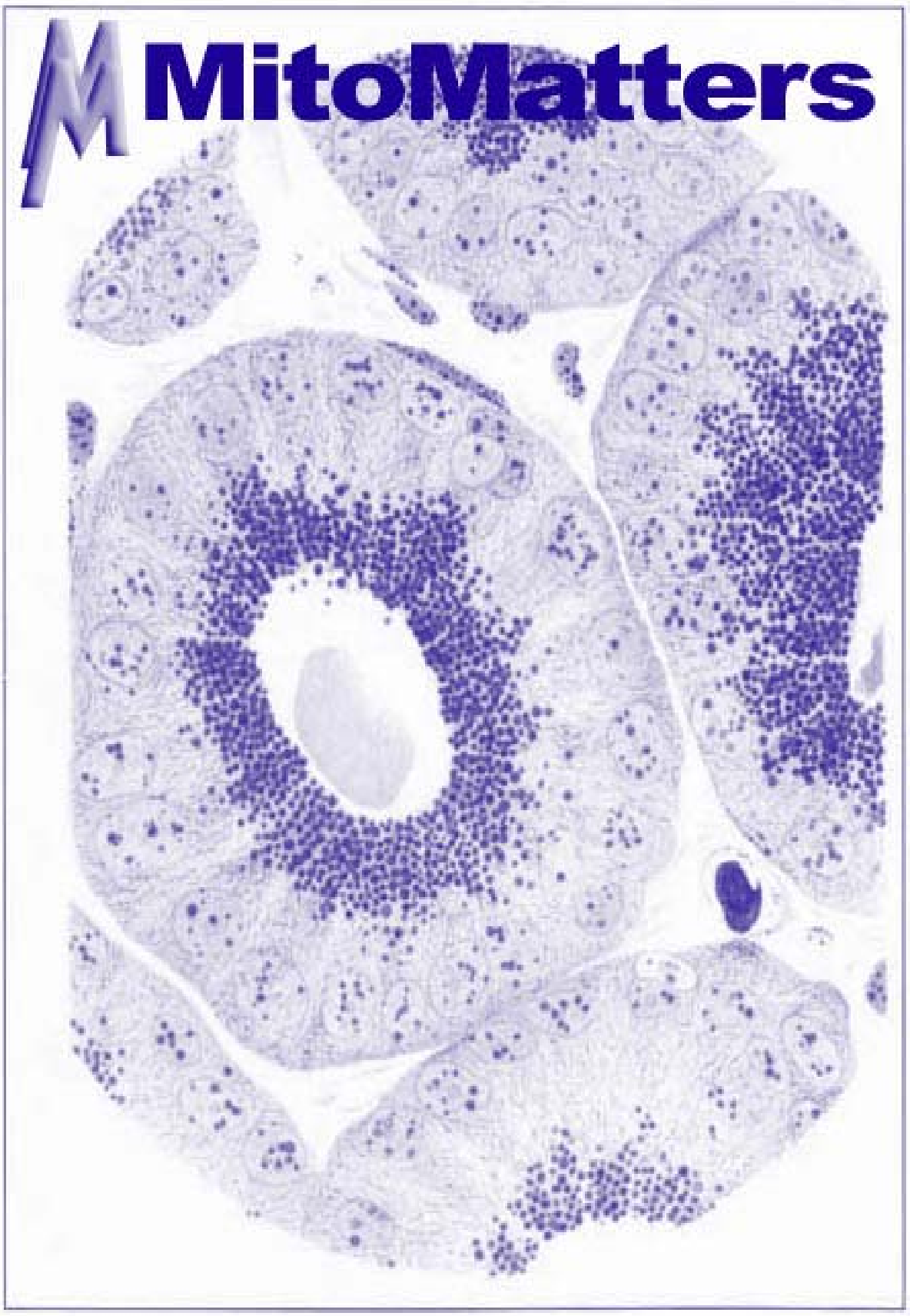
MitoMatters



The Official Newsletter of the Mitochondria Research Society,
Volume 1, Issue 3, 2002

Founder's note

Dear MRS member:

With this newsletter you will find the Ballot for The Mitochondria Research Society officers. The nominating committee headed by Mariana Gerschenson, Ph.D., has selected two candidates for the president and two for the secretary treasurer. Please choose one candidate from each category by crossing (X) the box next to the candidate's short biography and mail in the envelope provided or fax (410-502-7244) your ballot by November 30, 2002.

With this newsletter you will also find the membership application/renewal form. If you have not already renewed your membership, please do so as soon as possible. With your renewal you will continue to receive an uninterrupted subscription to the *Mitochondrion* journal, this newsletter as well as other membership benefits.

We hope you enjoy this issue of *MitoMatters*.

Sincerely,

Keshav K. Singh, Ph.D
Founder, The Mitochondria Research Society

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Practical Issues in the Design of Studies Evaluating Therapy in Mitochondrial Disorders

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Mitochondrial disorders represent a heterogenous group of conditions which have altered electron transport chain flux as a common cellular consequence. Under the general term of "mitochondrial disorders" there are a number of specific genetic mutations that are responsible including both nuclear and mitochondrial DNA encoded mutations [1]. For example, point mutations in mitochondrial DNA, alterations in thymidine metabolism (MNGIE syndrome), alterations in nuclear encoded sub units of electron transport chain (i.e. SURF and NDUF mutations), mitochondrial deletion and depletion syndrome (nuclear defects) and defects in subunit assembly proteins (SCO2) [2]. The converse is also true in that for a given point mutation, such as MELAS 3243, there may be a variety of phenotypic presentations ranging from deafness to strokes to short stature to diabetes or a combination of all or none of the above.

Perhaps because of difficulties in establishing the diagnosis in some centers the identification and recognition of mitochondrial disorders as a specific disease entity is probably underestimated. Recent estimates show that mitochondrial disorders are more common than ALS and myasthenia gravis and probably as common as some of the common muscular dystrophies (ie. Duchenne and Myotonic Muscular Dystrophy). In spite of this high prevalence there have been few studies evaluating therapy and most of these are single case reports or small case series. There are a number of issues which issues in addition to the lack of appreciation of these conditions as causes of neurological disease that have likely contributed to the paucity of clinical investigations into experimental therapeutics. [3]

Issues Relevant to study design

Patient selection and recruitment: The first issue to consider is whether to evaluate individuals with a common phenotype or genotype. For example, a reduction in seizure frequency may be an appropriate outcome variable in a child with severe MELAS 3243, whereas this would be inappropriate in a patient with the same MELAS 3243 mutation who only manifests with hearing loss and type 2 diabetes. Conversely, if patients are selected based on a given symptom, such as ptosis and external opthalmoplegia, the underlying genetic and biochemical basis for this may be highly variable (i.e., CPEO vs MELAS vs MNGIE). Ideally, it will be important to study a given genotype (i.e., MELAS 3243) with similar phenotypic characteristics, however, from a practical stand-point, we will likely have to have studies of more heterogeneous groups and later on narrow down the sub-groups.

Selection of the outcome variables: This is probably one of the most critical factors to consider in the development of studies in experimental therapeutics. The outcome variable(s) must reflect the fundamental biochemical process that is targeted by the intervention. For example, phosphocreatine recovery kinetics by ^{31}P-MRS [4] would be an appropriate outcome variable in a 2 week study of multiple co-factor and creatine intervention in MELAS syndrome whereas IQ scores, incidence of stroke, and quality of life would be poor choices for the would not be capable of detecting differences even if ultimately the intervention could influence the latter outcome variables. Other important issues such as the test-retest reliability and sensitivity of the outcome measures will influence the sample size required to detect an effect of treatment. For example, quality of life scores require a very large number of research subjects, whereas tests with high test-retest reliability, such as customized strength and muscle endurance testing equipment, will detect significant differences with far fewer participants.

Retention/Duration of studies: Subject retention is an issue for several of the mitochondrial cytopathies result in significant medical complications which can arise during the course of the study. For example, in some of our studies with MNGIE syndrome we were unable to complete the protocol in any of the 3 patients due to frequent nutritional and electrolyte acute episodes which occurred during the course of the study that confounded the outcome measurements. A second confounding issue is that of dementia and/or psychiatric issues which compromise compliance and accuracy of the outcome variables. As the duration of the study increases, these issues of retention become more apparent due to subject fatigue with the protocol.

Funding Issues: A lack of recognition of mitochondrial cytopathies as being an important cause of disease is one factor that has lead to difficulty obtaining funding for clinical trials. The lack of a coordinated multi-centered clinical trials collaborative effort has also been a limitation, however, there are steps currently being taken to rectify this with a North American Collaborative being established. In spite of the increased awareness and funding of this area of research, funding is still extremely limited. Until further clinical research is conducted using smaller trials that take into account some of the aforementioned design issues, the probability of successful funding for large randomized double-blind, multi-centered trials will be very limited. It is important for clinicians to realize that the "ideal" study will not be funded until more supportive studies are completed in order to develop optimal compounds and combinations of compounds that may be efficacious in mitochondrial cytopathy.

Suggestions for future research

One way of overcoming some of the issues of clinical and genetic heterogeneity is to take a "final common pathway" approach. By this I mean that consideration should be first given to evaluating compounds that target the final common pathways of mitochondrial dysfunction and use outcome measurements that evaluate these processes. Although not invariably present, an increase in oxidative stress, a decrease in electron transport chain flux with a resultant in decrease in oxygen consumption, a depletion of alternative energy pathways with a reduction in cellular energy charge (elevated lactate and reduced phosphocreatine) are cellular features of many mitochondrial disorders [5]. There have been several interventions that have been tried in the past with limited success including,

- Reduction in lactate production (i.e. dichloroacetate)[6;7].
- Reduction in oxidative stress (i.e. Vitamin C, coenzyme Q10, Vitamin E)[4;8-10].
- Bypass of electron transport chain defect (i.e. coenzyme Q10, succinate, Vitamin K3)[4;9;10].
- Alternative energy source (creatine monohydrate)[11-14].
- Combination therapy (various combinations of vitamins and co-factors)[4;10;15].
- Increased mitochondrial efficiency (aerobic exercise)[16;17].
- Gene shifting therapies to reduce DNA mutational burden (resistance exercise, myo-toxins injection)[18-20].

A first step would be to evaluate a combination of compounds that target two or more of the final common pathways of energy dysfunction in mitochondrial disorders with and without superimposed exercise. Such a study should utilize multiple sites and use clinical (endurance, functional tasks, strength) and biochemical markers (oxidative stress markers, PCr recovery, muscle oxygenation (near infra-red spectroscopy), lactate, etc.) of efficacy.

Once the initial trials are completed, it should be possible to have sufficient experience and sample size estimates to design large trials with functional outcomes. When one appreciates the huge advances in chemotherapy treatments for children with acute lymphoblastic leukemia over the past few decades, the potential for such combination therapies could be a significant advance for those with mitochondrial cytopathies.

References

1. Simon DK, Johns DR. Mitochondrial disorders: clinical and genetic features. Annu Rev Med 1999;50:111-27.

2. Servidei S. Mitochondrial encephalomyopathies: gene mutation. Neuromuscul Disord 2002;12:334-9.

3. Chinnery PF, Turnbull DM. Epidemiology and treatment of mitochondrial disorders. Am J Med Genet 2001;106:94-101.

4. Eleff S, Kennaway NG, Buist NR, et al. 31P NMR study of improvement in oxidative phosphorylation by vitamins K3 and C in a patient with a defect in electron transport at complex III in skeletal muscle. Proc Natl Acad Sci U S A 1984;81:3529-33.

5. Tarnopolsky MA, Beal MF. Potential for creatine and other therapies targeting cellular energy dysfunction in neurological disorders. Ann Neurol 2001;49:561-74.

6. Taivassalo T, Matthews PM, De Stefano N, et al. Combined aerobic training and dichloroacetate improve exercise capacity and indices of aerobic metabolism in muscle cytochrome oxidase deficiency. Neurology 1996;47:529 34.

7. De Stefano N, Matthews PM, Ford B, Genge A, Karpati G, Arnold DL. Short-term dichloroacetate treatment improves indices of cerebral metabolism in patients with mitochondrial disorders. Neurology 1995;45:1193-8.

8. Bresolin N, Bet L, Binda A, et al. Clinical and biochemical correlations in mitochondrial myopathies treated with coenzyme Q10. Neurology 1988;38:892-9.

9. Barbiroli B, Frassineti C, Martinelli P, et al. Coenzyme Q10 improves mitochondrial respiration in patients with mitochondrial cytopathies. An in vivo study on brain and skeletal muscle by phosphorous magnetic resonance spectroscopy. Cell Mol Biol (Noisy-le-grand) 1997;43:741-9.

10. Argov Z, Bank WJ, Maris J, et al. Treatment of mitochondrial myopathy due to complex III deficiency with vitamins K3 and C: A 31P-NMR follow-up study. Ann Neurol 1986;19:598-602.

11. Tarnopolsky M, Martin J. Creatine monohydrate increases strength in patients with neuromuscular disease. Neurology 1999;52:854-7.

12. Tarnopolsky MA, Roy BD, MacDonald JR. A randomized, controlled trial of creatine monohydrate in patients with mitochondrial cytopathies. Muscle Nerve 1997;20:1502-9.

13. Klopstock T, Querner V, Schmidt F, et al. A placebo-controlled crossover trial of creatine in mitochondrial diseases. Neurology 2000;55:1748-51.

14. Borchert A, Wilichowski E, Hanefeld F. Supplementation with creatine monohydrate in children with mitochondrial encephalomyopathies. Muscle Nerve 1999;22:1299-300.

15. Matthews PM, Ford B, Dandurand RJ, et al. Coenzyme Q10 with multiple vitamins is generally ineffective in treatment of mitochondrial disease. Neurology 1993;43:884-90.

16. Taivassalo T, Shoubridge EA, Chen J, et al. Aerobic conditioning in patients with mitochondrial myopathies: physiological, biochemical, and genetic effects. Ann Neurol 2001;50:133-41.

17. Taivassalo T, De Stefano N, Argov Z, et al. Effects of aerobic training in patients with mitochondrial myopathies. Neurology 1998;50:1055-60.

18. Taivassalo T, Fu K, Johns T, Arnold D, Karpati G, Shoubridge EA. Gene shifting: a novel therapy for mitochondrial myopathy. Hum Mol Genet 1999;8:1047-52.

19. Clark KM, Bindoff LA, Lightowlers RN, et al. Reversal of a mitochondrial DNA defect in human skeletal muscle. Nat Genet 1997;16:222-4.

20. Andrews RM, Griffiths PG, Chinnery PF, Turnbull DM. Evaluation of bupivacaine-induced muscle regeneration in the treatment of ptosis in patients with chronic progressive external ophthalmoplegia and Kearns-Sayre syndrome. Eye 1999;13 (Pt 6):769-72.

Mitochondria in the Nucleus

In 1958 Australian electron microscopists H. Hoffman and G.W. Grigg reported the presence of mitochondria in the nucleus of the mouse lymph nodes. Since then there had been reports of mitochondrial presence in the nucleus of cancer cells. Recently, Bakeeva et al (2001) provide evidence that mitochondria are found in the nucleus of rats that were given alcohol. They claim that this phenomenon is reproducible. For more details see:

Bakeeva et al (2001) Mitochondria enter the nucleus, Biochemistry 66:133-1336.

Make More Mitochondria

Always wondered about how do cells regulate mitochondrial number? In a study published recently, Williams and his colleagues report the development of transgenic mice over-expressing a signaling protein called calmodulin-dependent protein kinase (CaMK). When CaMK is activated, it and another protein called calcineurin, trigger a pathway that leads to production of more mitochondria. Since levels of mitochondrial proteins decrease with normal aging, this study may help develop therapies to increase physical endurance in the aged individuals.

Wu H et al (2002) Regulation of mitochondrial biogenesis in skeletal muscle by CaMK. Science 296:349-352.

Paternal Mitochondrial Inheritance

Every mitochondriac knows that human mitochondrial DNA is inherited from the mother. Not true any more! A Danish group has documented the first known exception to the rule. This group describes a man whose muscle cells contain mitochondria that came mainly from his father.

Schwartz M and Vissing J (2002) Paternal Inheritance of Mitochondrial DNA New England J of Med 347:576-580.

Mighty Mouse

Bruce Spiegelman and his colleagues has identified PGC1, a protein that functions as a molecular switch. This protein converts a "fast-twitching" muscle (which tires quickly), into high-endurance "slow-twitch" muscle in mice. This discovery may in the future lead to new treatments of degenerative muscle disease, and could also help in developing drugs for endurance athletes like marathon runners.

Lin et al (2002) Transcriptional co-activator PGC-1 alpha drives the formation of slow-twitch muscle fibres. Nature. 418:797-801.

Upcoming Meetings

Japanese Society for Mitochondria Research and Medicine meeting
18- 22nd Dec. 2002, Contact: Hideyuki J. Majima, Ph.D. Department of Radiology, Kagoshima Univ. School of Dentistry, 8-35-1 Sakuragaoka, Kagoshima 890-8544, Japan, E-mail: hmajima@denta.hal.kagoshima-u.ac.jp, Tel +81-99-275-6270, 6272, Fax +81-99-275-6278

Asian Society for Miotochondrial Research and Medicine meeting,
5-6th Feb, 2003. Contact: Hong Kyu Lee, M.D. Department of Internal Medicine, Seoul National University College of Mediicne, 28 Yongondong, Chongno-ku, Seoul, Korea, hkleemd@plaza.snu.ac.kr. Tel: 822-760-2266, Fax: 822-765-7966.

3rd European Metabolic Course: The Department of Metabolic and Endocrine Disorders and the Laboratory of Paediatrics and Neurology at the University Children's Hospital Nijmegen are organizing this meeting in collaboration with the Orphan Europe Academy on October 29 - November 2, 2003.The course is designed for Paediatricians with 2 to 5 years clinical experience in the metabolic field. It is pitched at a high level and restricted to 33 participants. To receive the full programme, please contact: Guilaine Arduin, Manager Orphan Europe Academy, Orphan Europe Immeuble le Wilson - Cedex 70, 92046 Paris la Défense – France, E-mail: garduin@orphan-europe.fr, Tel: 33.1.47.73.94.20, Fax: 33.1.49.00.18.00

6th Euromit meeting, June 30- July 4th, 2004.
Contact Jan Smeitink, MD, PhD. Nijmegen Center for Mitochondrial Disorders, Department of Pediatrics, University Medical Center Nijmegen. Geert Grooteplein 10, PO BOX, 9101, 6500 HB Nijmegen, The Netherlands. E-mail j.smeitink@cukz.umcn.nl, Tel: 0031-24-3614430, Fax: 0031-24-3616428

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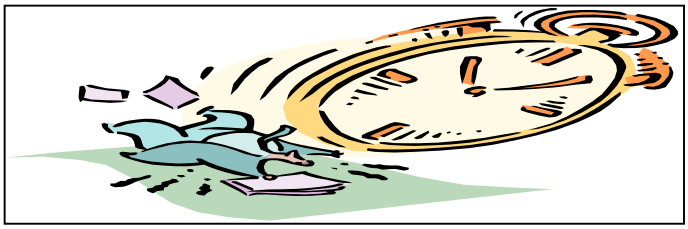

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