Electrical impedance myography for the assessment of children with muscular dystrophy: a preliminary study.

S B Rutkove and B T Darras

Department of Neurology, Beth Israel Deaconess Medical Center, and Department of Neurology, Children’s Hospital Boston, Harvard Medical School, Boston, MA, USA

Email: srutkove@bidmc.harvard.edu

Abstract. Electrical impedance myography (EIM) provides a non-invasive approach for quantifying the severity of neuromuscular disease. Here we determine how well EIM data correlates to functional and ultrasound (US) measures of disease in children with Duchenne muscular dystrophy (DMD) and healthy subjects. Thirteen healthy boys, aged 2-12 years and 14 boys with DMD aged 4-12 years underwent both EIM and US measurements of deltoid, biceps, wrist flexors, quadriceps, tibialis anterior, and medial gastrocnemius. EIM measurements were performed with a custom-designed probe using a commercial multifrequency bioimpedance device. US luminosity data were quantified using a gray-scale analysis approach. Children also underwent the 6-minute walk test, timed tests and strength measurements. EIM and US data were combined across muscles. EIM 50 kHz phase was able to discriminate DMD children from healthy subjects with 98% accuracy. In the DMD patients, average EIM phase measurements also correlated well with standard functional measures. For example the 50 kHz phase correlated with the Northstar Ambulatory Assessment test ($R = 0.83$, $p = 0.02$). EIM 50 kHz phase and US correlated as well, with $R = -0.79$ ($p < 0.001$). These results show that EIM provides valuable objective measures Duchenne muscular dystrophy severity.

1. Introduction

Duchenne muscular dystrophy (DMD) is a progressive neuromuscular disease that affects approximately 1 in 3000 male births [1]. Caused by mutations in the dystrophin gene that encodes a membrane structural support protein, DMD begins impacting boys at just 2-4 years of age [2]. Initially subtle weakness affecting the proximal lower extremities progresses until the child has increasing difficulty walking. By 12 years, most boys are wheelchair-bound and have limited upper extremity function; nearly all die from a combination of respiratory and heart failure before age of 30 [1]. Currently, only corticosteroids have been shown to slow the loss of function in this disease [3]. However, since the genetic basis of the disease is now well-understood, a variety of new potential treatments are being tested in clinical trials [4]. One major limitation to this testing is that the outcome measures or biomarkers used to assess drug efficacy are limited. Functional measures, such as the distance a boy can walk in 6 minutes, the 6-minute walk test, is one major measure used, but it is limited by a variety of factors [5]. New, non-invasive, methods for assessing disease status are needed. One potential method is the use of localized impedance measurements of muscle, or electrical impedance myography (EIM) [6]. Studies in other neuromuscular diseases, including amyotrophic...
lateral sclerosis [7] and spinal muscular atrophy [8], have shown that EIM is sensitive to the severity of disease and can serve as a useful biomarker of disease change over time. In this study, we preliminarily evaluate the potential use of EIM in the assessment of DMD by comparing EIM data to standard functional measures in a group of DMD boys and a group of healthy boys while also assessing the relationship between EIM and ultrasonography.

2. Methods
A total of 19 boys, aged 2 to 12 years, mean age 7.3 years with genetically confirmed DMD and a total of 20 healthy boys, ages 2 to 12 years, mean age 7.4 years, were recruited for study at Children's Hospital Boston. The study was approved by the hospital's institutional review board; parents provided written informed consent and children verbal assent. All boys underwent functional testing, including the 6MWT, the North Star Ambulatory Assessment, and quantitative strength testing.

2.1. EIM Testing
All subjects had EIM testing of unilateral deltoid, biceps, wrist flexors, quadriceps (rectus femoris), tibialis anterior, and medial gastrocsemi. EIM measurements were performed with the Imp SFB7 (Impedimed, Inc, Sydney Australia), configured with a custom-designed hand-held electrode array, based on a design that has been previously described [9] with current flow parallel to the muscles. Three different array sizes were used depending on the size of the child. Although multifrequency data were collected from each muscle, only the 50 kHz data were analyzed here for convenience sake. Additional analysis of the multifrequency data will be performed separately.

2.2. Ultrasound Measurements
Ultrasound was also performed with the Terason t3000 system (Teracorp, Inc, Burlington, MA) using a 10 MHz probe. Measurements were performed on the identical set of muscles on which EIM was performed in the identical locations were the EIM probe was placed. The ultrasound probe was placed transversely on the muscle. Single images were obtained from each muscle. The images were then downloaded into Photoshop ® (Adobe Inc, San Jose, CA) and a gray-scale analysis was performed [10]. In this analysis, a selected area of muscle was chosen from the image and a mean luminosity value measured; in disease, the luminosity increases [10].

2.3. Data Analysis
Data was evaluated both for individual muscles and averaged over the 6 muscle examined. Unpaired t-tests and Spearman correlations were performed; significance was set at p < 0.05, two-tailed.

3. Results

3.1. Overall EIM Data
Figure 1A provides a histogram of 6-muscle averaged EIM 50 kHz phase values for 20 healthy boys and 19 DMD boys. Figure 1B provides the same data for quadriceps alone. As can be seen there is a clear differentiation between the groups. A receiver operating characteristic analysis demonstrated EIM’s ability to discriminate healthy and sick muscle with an area under the curve, or accuracy, of 98% for the 6-muscle mean EIM values. Figure 2 shows phase correlated with age with DMD and controls, respectively, revealing a gradual increase in healthy children and stability or a slight decrease with age in the DMD boys. For DMD, R = -0.30, p = 0.21; for control R = 0.59, p = 0.008.

3.2. Relationship between EIM data and function measures
Figure 3 shows the correlation between the mean EIM values and the North Star Ambulatory Assessment for the mean EIM data for only the DMD boys (R = 0.83, p = 0.02).
Figure 1. Comparison of EIM 50 kHz phase data for DMD and healthy boys for A. Six-muscle average and B. Quadriceps alone

Figure 2. Average 6-muscle EIM phase data versus age. A. DMD boys and B. Normal subjects. Note the difference in overall trajectories

Figure 3. Correlation between North Star Ambulatory Assessment (NSAA) and EIM phase in 8 DMD boys in whom this assessment was obtained.

3.3. Relationship between EIM and ultrasound luminosity
Figure 4A shows the correlation between the 6-muscle mean EIM data and 6-muscle mean luminosity (R = -0.79, p < 0.001); Figure 4B shows data for quadriceps alone (R = -0.67, p = < 0.001).
4. Discussion
These results show that EIM is sensitive to disease and can readily differentiate normal children from those with DMD. In addition, there is an age dependence to the values in that the data suggest progressively lower values children with DMD but progressively higher values in healthy controls. Moreover, EIM is clinically meaningful in that there is a strong correlation between EIM values and a standard functional assessment tool. Finally, this study confirms that the changes in EIM actually relate directly to tissue pathology. The increasing fibrosis and fatty infiltration, observed on muscle ultrasound, correlate strongly with the impedance data.

There are several limitations to this work. First, we are only looking at a relatively small group of children. Second, the impedance device and handheld array have considerable limitations in that we have to use different size devices for different children. This could be affecting our analysis to some extent and a separate study comparing the characteristics of the data obtained from the 3 different sized arrays will also need to be performed. Third, we have only evaluated one characteristic of the impedance data: the longitudinal 50 kHz phase. We have also acquired multifrequency data in both the longitudinal and transverse directions and thus much data still needs to be analyzed. It is possible that other frequencies may provide more powerful markers of disease status than these 50 kHz values. Similarly the reactance and resistance may also be useful to assess and will be analyzed as well.

In conclusion, EIM appears to be a promising marker of disease in boys with Duchenne muscular dystrophy. Further study utilizing this simple approach to disease assessment are warranted.

Acknowledgments
This work was funded by NIH grant AR060850

References
[1] Bushby K, Finkel R, Birnkrant DJ, Case LE, Clemens PR, et al C 2010 Lancet Neurol 9 77-93.
[2] Prior TW and Bridgeman SJ 2005 J Mol Diagn 7 317-26.
[3] Manzur AY, Kuntzer T, Pike M and Swan A 2008 Cochrane Database Syst Rev CD003725.
[4] Nelson SF, Crosbie RH, Miceli MC and Spencer MJ 2009 Curr Opin Neurol 22 532-8.
[5] Bushby K and Connor E 2011 Clinical investigation 1 1217-1235.
[6] Rutkove SB 2009 Muscle Nerve 40 936-946.
[7] Rutkove SB, Caress JB, Cartwright MS, Burns TM, Warder J, David WS, et al 2012 Amyotrophic Lateral Sclerosis
 [8] Rutkove SB, Gregas MC and Darras BT 2012 Muscle & nerve 45 642-7.
 [9] Narayanaswami P, Speiker AJ, Mongiovì P, Keel JC, Muzin SC and Rutkove SB 2012 Muscle Nerve 46 257-63.
[10] Jansen M, van Alfen N, Nijhuis van der Sanden MW, van Dijk JP, Pillen S and de Groot J 2012 Neuromuscular disorders 22 306-17.