

Importance of Sample Size for the Estimation of Repeater F Waves in Amyotrophic Lateral Sclerosis

Jia Fang, Ming-Sheng Liu, Yu-Zhou Guan, Bo Cui, Li-Ying Cui

Department of Neurology, Peking Union Medical College Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100730, China

Abstract

Background: In amyotrophic lateral sclerosis (ALS), repeater F waves are increased. Accurate assessment of repeater F waves requires an adequate sample size.

Methods: We studied the F waves of left ulnar nerves in ALS patients. Based on the presence or absence of pyramidal signs in the left upper limb, the ALS patients were divided into two groups: One group with pyramidal signs designated as P group and the other without pyramidal signs designated as NP group. The Index repeating neurons (RN) and Index repeater F waves (Freps) were compared among the P, NP and control groups following 20 and 100 stimuli respectively. For each group, the Index RN and Index Freps obtained from 20 and 100 stimuli were compared.

Results: In the P group, the Index RN ($P = 0.004$) and Index Freps ($P = 0.001$) obtained from 100 stimuli were significantly higher than from 20 stimuli. For F waves obtained from 20 stimuli, no significant differences were identified between the P and NP groups for Index RN ($P = 0.052$) and Index Freps ($P = 0.079$); The Index RN ($P < 0.001$) and Index Freps ($P < 0.001$) of the P group were significantly higher than the control group; The Index RN ($P = 0.002$) of the NP group was significantly higher than the control group. For F waves obtained from 100 stimuli, the Index RN ($P < 0.001$) and Index Freps ($P < 0.001$) of the P group were significantly higher than the NP group; The Index RN ($P < 0.001$) and Index Freps ($P < 0.001$) of the P and NP groups were significantly higher than the control group.

Conclusions: Increased repeater F waves reflect increased excitability of motor neuron pool and indicate upper motor neuron dysfunction in ALS. For an accurate evaluation of repeater F waves in ALS patients especially those with moderate to severe muscle atrophy, 100 stimuli would be required.

Key words: Amyotrophic Lateral Sclerosis; Repeater F waves; Upper Motor Neuron

INTRODUCTION

Amyotrophic lateral sclerosis (ALS) is an idiopathic, fatal neurodegenerative disease of human motor system, and it includes the participation of other systems, such as sensory system, autonomic system, and basal ganglia.^[1-3] There is no highly effective disease-modifying treatment. The median survival is 3–4 years from symptom onset and there is considerable variation in progression rate overall.^[3,4] An electromyography (EMG) examination can be used to detect subclinical involvement of lower motor neurons (LMNs), which is diagnostically most useful in the presence of upper motor neuron (UMN) signs within the same territory.^[5,6] During the early course of the disease, patients may lack overt clinical signs of an UMN lesion. In the later stages, severe LMN involvement may mask previously existent UMN signs.^[7] Early diagnosis of

the disease is important for proper management of ALS patients.

The F wave is a late muscle response which results from antidromic activation of one or a small number of motor neurons following peripheral nerve electrical stimulation of their axons.^[8] In routine EMG examinations, 10–20 F waves are commonly used. Consecutively recorded F waves vary in latency and amplitude. F waves can be useful for the evaluation of motor neuron pool integrity and changes in alpha motor neuron excitability in central motor disorders.^[9,10] One electrophysiological expression of the excitatory state of the motor neurons could theoretically be their tendency to produce repeated backfirings.^[11]

In neurogenic abnormalities such as motor neuron disease, an increased number of repeater F waves have been demonstrated.^[12] However, in ALS, the F wave persistence was low due to reduction of the number of motor neurons, sometimes, no F response was recorded in a routine series of

Access this article online

Quick Response Code:



Website:
www.cmj.org

DOI:
10.4103/0366-6999.151107

Address for correspondence:

Prof. Li-Ying Cui,
Department of Neurology, Peking Union Medical College Hospital,
Neuroscience Center, Chinese Academy of Medical Sciences and Peking
Union Medical College, Beijing 100730, China
E-Mail: pumchcly@sina.com

20 F responses even in some apparently normal muscles. The clinical significance of repeater F waves as a sign of UMN dysfunction has not been fully evaluated so far. The aim of this study was to assess the clinical significance of repeater F waves in ALS and investigate the effects of different F wave sample sizes on the accuracy of the information obtained from ALS patients.

METHODS

Subjects

Fifty patients, 27 males and 23 females with mean age 49 ± 7 years (range 36–64), diagnosed as having definite, probable, or laboratory-supported probable ALS using the revised EI Escorial criteria,^[13] were studied. The median duration of ALS patients from symptom onset was 14 months (range 3–72). Twenty-five age- and gender-matched healthy subjects, 13 male and 12 female recruited from the family members of the patients and faculty members were used as controls. Their mean age was 51 ± 7 years (range 41–62). Subjects with diseases such as cervical myelopathy, multifocal motor neuropathy and entrapment neuropathy etc., were excluded by appropriate examinations. Patients with diabetes mellitus, alcohol abuse and other systemic or neurological diseases were excluded from the study. At the time of the investigation, none of the ALS patients were taking riluzole or antispasticity drugs.

We defined UMN signs in the upper limb by the presence of spasticity, increased tendon reflexes, reserved tendon reflexes in weak limbs or a positive Hoffman sign. The investigation was focused around left abductor digiti minimi (ADM), because of split-hand syndrome in ALS.^[14] The patients were divided into two groups according to the presence or absence of pyramidal signs in the left upper limbs. The group with pyramidal signs in the left upper limbs was designated as the P group and the group without pyramidal signs in the left upper limbs was designated as the NP group.

Nerve conduction studies

A Viking IV EMG machine (Nicolet Biomedical, Madison, Wisconsin, USA) performed the electrophysiological studies. Routine nerve conduction studies included motor and sensory conduction studies of the median, ulnar, tibial and peroneal nerves. Particular emphasis was paid to exclude the possibility of conduction block by applying proximal stimulation of median and ulnar nerves in axilla and Erb's point and by comparing amplitudes of compound muscle action potential (CMAP) responses between different stimulation sites. Motor conduction parameters included measurements of peak to peak amplitude of CMAP, distal motor latency (DML) and motor conduction velocity (MCV). Measurements of peak to peak amplitude of sensory action potential and sensory conduction velocities were recorded in sensory conduction studies. Nerve conduction studies were compared with normal values of the neurophysiology laboratory of Peking Union Medical College Hospital.^[15]

F wave studies

The examination was performed in a warm room. All subjects were examined in a supine position as relaxed as possible to avoid voluntary activity. The skin temperature of the examined limb was measured and maintained $>32^{\circ}\text{C}$ by covering them with a blanket. A hundred consecutive stimuli with a frequency of 1 Hz and duration of 0.1 ms were delivered to ulnar nerves using supramaximal surface stimulation. A belly tendon, silver electrode, surface montage was used to record ADM responses. Ulnar nerves were stimulated at the wrist, 6–7 cm from the proximal active recording electrode on the ADM. Filter settings were 20 Hz to 2 kHz, amplifier gain was 0.5 mV for F waves and 5 mV for M responses. In order to differentiate responses from background noises, only deflections that had amplitudes of at least $40 \mu\text{V}$ were accepted as F waves. The late components of M responses and A waves were excluded from F wave measures. F waves similar in latency, amplitude and configuration, were defined as repeater F waves from a given neuron, called a repeating neuron (RN). We analyzed the number of individual repeater F waves using Index RN and the number of F waves that repeat using Index repeater F waves (Freps). Index RN = $100 \times \text{number of RN} / \text{number of traces with different F wave shapes in a series of 100 stimuli}$. Index Freps = $100 \times \text{number of repeater F waves} / \text{total numbers of traces with F waves in the same nerve}$.^[11] The number of repeater F waves were evaluated from the series of 20 and 100 consecutive sweeps respectively. To simulate the procedure in routine recordings, F waves following 20 stimuli were not randomly selected but were taken from the first 20 samples of each series of 100 F waves. The numbers of repeater F waves following 20 and 100 stimuli were compared.

All procedures were done according to the protocols approved by the Ethical Committee of Clinical Research of Peking Union Medical College Hospital and adhered to the principles of the declaration of Helsinki. All ALS patients and control subjects provided their written informed consent to participate in the study.

Statistical analysis

Descriptive statistics were generated for all variables. The Shapiro–Wilk test was used to estimate the probability that the data being analyzed present a normal distribution. The homogeneity was tested using a Levene test. For data that were not normally distributed even after data transformation, nonparametric statistical comparisons were performed. The Kruskal–Wallis test was used for multiple group comparisons and the Mann–Whitney test was used to examine comparisons between two groups. The Bonferroni method was used, and the *P* value was then corrected for type 1 error due to multiple comparisons. A statistically significant *P* value was set at 0.05. All statistical analyses were performed using SPSS for windows, version 21 (SPSS, Inc., Chicago, Illinois, USA).

RESULTS

For the 75 ADM muscles examined, there were 25 ADM muscles in the P, NP and control group respectively. Motor conduction studies were consistent with anterior horn diseases in all patients. The DML of the ALS patients were significantly prolonged compared with the controls ($P < 0.001$). The mean ulnar CMAP values of the ALS patients were significantly lower compared with the controls ($P < 0.001$). The mean MCV of the ALS patients were significantly slower than the controls ($P = 0.002$). There were no significant differences between the P and NP groups for DML ($P = 0.64$), CMAP ($P = 0.093$) and MCV ($P = 0.091$) [Table 1]. There was no conduction block or CMAP temporal dispersion in the 75 ulnar nerves examined. The sensory nerve conduction results were normal.

In the P group, the Index RN ($P = 0.004$) and Index Freps ($P = 0.001$) obtained from 100 stimuli were significantly higher than from 20 stimuli. In the NP group, there were no significant differences between the Index RN ($P = 0.244$) or Index Freps ($P = 0.183$) obtained from 20 and 100 stimuli. In the control group, the Index RN ($P = 0.02$) and Index Freps ($P = 0.018$) obtained from 100 stimuli were significantly higher than from 20 stimuli. For F waves obtained from 20 stimuli, no significant differences were identified between the P and NP groups for Index RN ($P = 0.052$) and Index Freps ($P = 0.079$); The Index RN ($P < 0.001$) and Index Freps ($P < 0.001$) of the P group were significantly higher than the control group; The Index RN ($P = 0.002$) of the NP group was significantly higher than the control group; There was no significant difference between the NP and control groups for the Index Freps ($P = 0.023$). For F waves obtained from 100 stimuli, the Index RN ($P < 0.001$) and Index Freps ($P < 0.001$) of the P group were significantly higher than the NP group; The Index RN ($P < 0.001$) and Index Freps ($P < 0.001$) of the P group were significantly higher than the control group; The Index RN ($P < 0.001$) and Index Freps ($P < 0.001$) of the NP group were significantly higher than the control group [Figure 1]. The maximum number of

Table 1: Demographic characteristics and motor nerve conduction measurements of study groups

Items	P	NP	C
Age (years)*	48 ± 6	51 ± 7	51 ± 7
Gender (male : female)*	13:12	14:11	13:12
Duration (months)†	14 (14)	12 (12)	-
DML (ms)*	2.58 ± 0.31‡	2.53 ± 0.48§	2.15 ± 0.28‡§
CMAP (mV)*	6.84 ± 4.16‡	8.61 ± 4.07§	16.72 ± 3.11‡§
MCV (m/s)*	55.19 ± 8.03‡	53.96 ± 9.27§	61.08 ± 6.86‡§

*For values that were normally distributed, variables were expressed as mean ± SD; †For values that were not normally distributed, variables were expressed as median (interquartile range); ‡Parameters in the P group which were significantly different from the control group ($P < 0.05$); §Parameters in the NP group which were significantly different from the control group ($P < 0.05$). P: Limbs with pyramidal signs; NP: Limbs without pyramidal signs; C: Control group; DML: Distal motor latency; CMAP: Compound muscle action potential; MCV: Motor conduction velocity; SD: Standard deviation; -: Nil.

individual repeater F waves have required at least 80 stimuli in 90% ALS patients of the P group, 67% ALS patients of the NP group and 82% subjects of the control group.

The absence of F waves in a series of 20 stimuli was noted in the left ulnar nerves of 7 ALS patients with the relative preservation of CMAP. For the 7 patients, the strength of the distal upper limb with moderate to severe muscle atrophy ranged from 2 to 4 grades according to the Medical Research Council scale. The CMAP amplitude of the 7 ALS patients were significantly lower than the other 43 ALS patients ($P = 0.002$). Four patients belonged to the P group while the other 3 patients belonged to the NP group. There

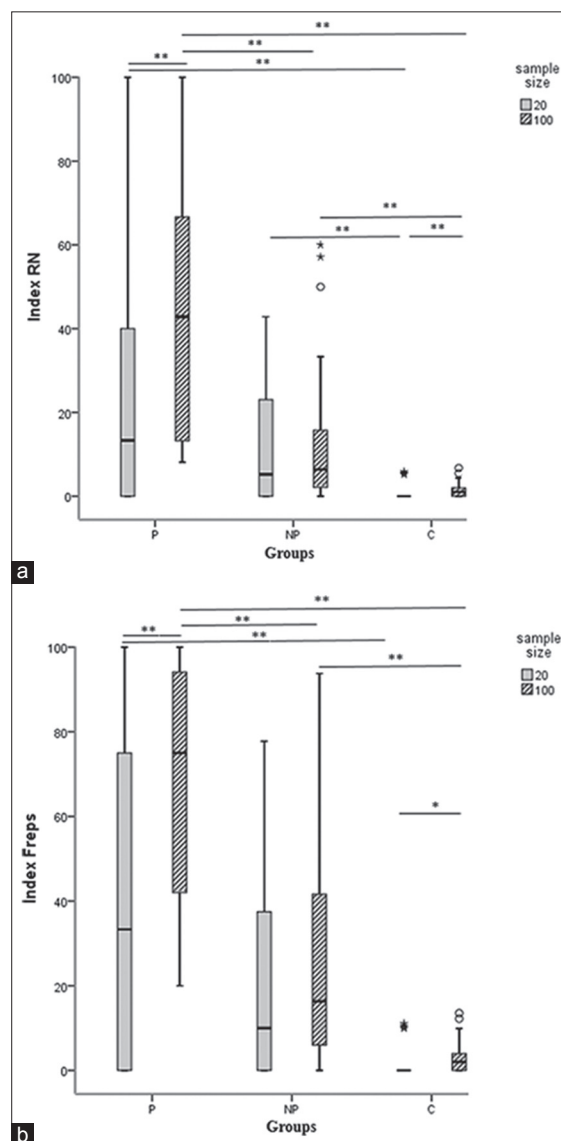


Figure 1: The comparison of repeater F waves among the P, NP and control groups and the comparison of repeater F waves obtained from 20 with those from 100 stimuli for each group. The edges indicate the 25th and 75th percentile. The open circle (o) designates values below or above 1.5 box-lengths from the box-edges, and the asterisk (*) designate values below or above 3.0 box-lengths from the box-edges. (a) Index RN; (b) Index Freps (* $P < 0.05$, ** $P < 0.01$). P: Limbs with pyramidal signs; NP: Limbs without pyramidal signs; C: Control group; RN: Repeating neuron.

was no significant difference between the 4 patients in the P group and the 3 patients in the NP group for disease duration and CMAP amplitude. In the following 80 stimuli, a number of F waves were recorded with increased repeater F waves in left ulnar nerves of the 4 patients in the P group, while none or just 1 F wave was recorded in the left ulnar nerves of the 3 patients in the NP group [Table 2, Figures 2 and 3].

DISCUSSION

The role of repeater F waves in UMN lesions has not been fully evaluated because previous studies have compared patients with normal subjects and have mostly employed 10–20 supramaximal stimuli to obtain F waves so far.^[11,12] In the present study, an adequate number of F waves were employed to investigate relatively accurate F wave properties. The results of the nerve conduction study identified significantly decreased CMAP, prolonged DML and slowed MCV of the ALS patients compared with the control group, which were consistent with previous studies.^[9,16]

In ALS, despite the fall in overall F wave frequency, there are significantly increased numbers of RNs and repeater F waves. These features probably reflect a heightened degree of excitability within spinal motor neuron pool as a result of UMN involvement.^[17] Our study exhibits that in ALS, an

increased number of repeater F waves have been attributed to a decreased number of motor neurons that are capable of responding to antidromic stimulations and to an increased level of excitability of anterior horn cells.

The present study demonstrated that in a series of 20 stimuli, there was no significant difference between repeater F waves in the P and NP group, while in a series of 100 stimuli, there were significantly increased repeater F waves in the P group than the NP group. What's more, the repeater F waves obtained from 100 stimuli were significantly increased than from 20 stimuli in the P group and the controls. Different sample sizes are needed for evaluation of different F wave parameters and for normal subjects and neuropathic patients.^[18] More stimuli would increase the number of individual repeater F waves, and accurate evaluation of repeater F waves would require at least 100 stimuli.^[19] The required number may be higher in patients with diseases causing a depletion of motor axons or motor neurons. In patients with neuropathies, larger sample size of F waves may be required than normal subjects.^[18] There is a potential risk of producing false negative or false positive results if an inadequate sample size is employed.^[18]

In our study, F response was absent in a routine series of 20 F responses in some patients, however, in the following 80 stimuli, a number of F waves were recorded with increased repeater F waves in the 4 ALS patients of the P group. The F wave persistence may be influenced by many factors such as nerve conduction block, reduction in the number of motor neurons and axons as well as transient impairment of motor neuron excitability.^[20] ALS is pathologically characterized by proximal neurofilamentous swelling within the axons of motor neuron.^[21-23] Some ALS patients showed transitory conduction blocks, increased temporal CMAP dispersion, or prolonged DML and F wave latencies which mimic a demyelinating neuropathy.^[24] Previous studies reported “pseudo-conduction block” due to ongoing Wallerian degeneration in ALS.^[24] Larger sample size can help clarify the pathophysiology of F wave disappearance in the routine 20 stimuli in ALS and differentiate ALS from some diseases with symptoms similar to ALS, such as multifocal motor neuropathy, endocrinopathies, paraneoplastic syndromes and cervical spondylosis.^[17,25]

Table 2: Numbers of RNs and repeater F waves recorded following 100 stimuli of the ALS patients with absence of F waves in the first 20 stimuli

Subject	RNs		Repeater F waves	
	20 stimuli	100 stimuli	20 stimuli	100 stimuli
1 (P group)	0	2	0	6
2 (P group)	0	1	0	9
3 (P group)	0	3	0	8
4 (P group)	0	1	0	3
5 (NP group)	0	0	0	0
6 (NP group)	0	0	0	0
7 (NP group)	0	0	0	0

P: Limbs with pyramidal signs; NP: Limbs without pyramidal signs; ALS: Amyotrophic lateral sclerosis; RN: Repeating neuron.

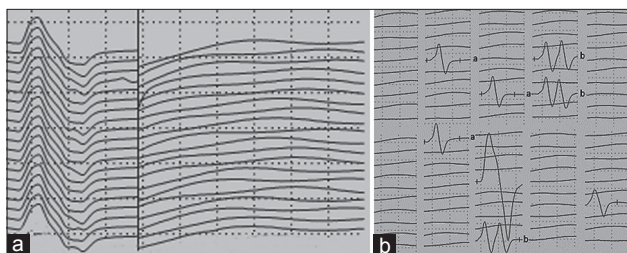


Figure 2: Twenty consecutive traces exhibiting absence of F wave during a train of 20 supramaximal stimuli to the left ulnar nerve at the wrist in a 50-year-old male patient of the P group. A number of repeater F waves were obtained during the following 80 supramaximal stimuli. Letters to the right of record identify waves on the basis of waveform and latency. (a) F waves recorded from 20 stimuli; (b) F waves recorded from 100 stimuli. Calibration: M response: 5 mV/division, 5 ms/division; F waves: 500 μ V/division, 5 ms/division.

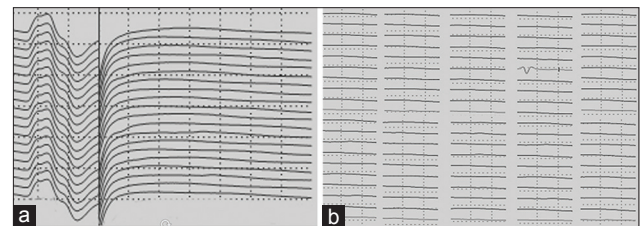


Figure 3: Twenty consecutive traces exhibiting absence of F wave during a train of 20 supramaximal stimuli to the ulnar nerve at the wrist in a 61-year-old male patient of the NP group. Only one F wave was recorded during the following 80 supramaximal stimuli. (a) F waves recorded from 20 stimuli; (b) F waves recorded from 100 stimuli. Calibration: M response: 5 mV/division, 5 ms/division; F waves: 500 μ V/division, 5 ms/division.

F wave is suitable as a probe for changes in the spinal cord excitatory state.^[26] In moderate to severe atrophy, UMN signs can be masked by LMN signs. Increasing the number of stimuli to obtain repeater F waves can give hints for additional central pathology in individual cases and provide information for differential diagnosis especially when F waves are absent in the routine 20 stimuli. For our study, patients with pure LMN dysfunction such as Kennedy's disease, spinal muscular atrophy, paralytic poliomyelitis etc., may be more qualified as the controls. However, such patients with a matched age and disease duration are difficult to find in our study.

On the basis of our study, the usually recommended sample size of 10–20 was inadequate for the assessment of ALS patients, if full potentials of F wave parameters especially repeater F waves were to be explored. An adequate number of F waves can ensure a better and more reliable exploration of motor neuron pool by F waves in ALS.

REFERENCES

1. Su XW, Broach JR, Connor JR, Gerhard GS, Simmons Z. Genetic heterogeneity of amyotrophic lateral sclerosis: Implications for clinical practice and research. *Muscle Nerve* 2014;49:786-803.
2. Kiernan MC, Vucic S, Cheah BC, Turner MR, Eisen A, Hardiman O, *et al.* Amyotrophic lateral sclerosis. *Lancet* 2011;377:942-55.
3. Menke RA, Körner S, Filippini N, Douaud G, Knight S, Talbot K, *et al.* Widespread grey matter pathology dominates the longitudinal cerebral MRI and clinical landscape of amyotrophic lateral sclerosis. *Brain* 2014;137:2546-55.
4. Bäumer D, Talbot K, Turner MR. Advances in motor neurone disease. *J R Soc Med* 2014;107:14-21.
5. Liu MS, Cui LY. Electrophysiological, pathological, and molecular study in diagnosis of neuromuscular disease. *Chin Med J* 2012;125:3203-6.
6. de Carvalho M, Dengler R, Eisen A, England JD, Kaji R, Kimura J, *et al.* Electrodiagnostic criteria for diagnosis of ALS. *Clin Neurophysiol* 2008;119:497-503.
7. Drory VE, Kovach I, Groozman GB. Electrophysiologic evaluation of upper motor neuron involvement in amyotrophic lateral sclerosis. *Amyotroph Lateral Scler Other Motor Neuron Disord* 2001;2:147-52.
8. Fisher MA. AAEM Minimonograph #13: H reflexes and F waves: Physiology and clinical indications. *Muscle Nerve* 1992;15:1223-33.
9. Argyriou AA, Polychronopoulos P, Talelli P, Chroni E. F wave study in amyotrophic lateral sclerosis: Assessment of balance between upper and lower motor neuron involvement. *Clin Neurophysiol* 2006;117:1260-5.
10. Argyriou AA, Karanasios P, Makridou A, Makris N. F-wave characteristics as surrogate markers of spasticity in patients with secondary progressive multiple sclerosis. *J Clin Neurophysiol* 2010;27:120-5.
11. Chroni E, Tendero IS, Punga AR, Stålberg E. Usefulness of assessing repeater F-waves in routine studies. *Muscle Nerve* 2012;45:477-85.
12. Petajan JH. F-waves in neurogenic atrophy. *Muscle Nerve* 1985;8:690-6.
13. Brooks BR, Miller RG, Swash M, Munsat TL, World Federation of Neurology Research Group on Motor Neuron Diseases. El Escorial revisited: Revised criteria for the diagnosis of amyotrophic lateral sclerosis. *Amyotroph Lateral Scler Other Motor Neuron Disord* 2000;1:293-9.
14. Shibuya K, Misawa S, Nasu S, Sekiguchi Y, Mitsuma S, Beppu M, *et al.* Split hand syndrome in amyotrophic lateral sclerosis: Different excitability changes in the thenar and hypothenar motor axons. *J Neurol Neurosurg Psychiatry* 2013;84:969-72.
15. Tang XF, Yang T, Yang BX, Liu XZ, Rong ZP. Electromyographic findings in normal Chinese. Analysis of 310 subjects. *Chin Med J* 1984;97:613-22.
16. de Carvalho M, Chio A, Dengler R, Hecht M, Weber M, Swash M. Neurophysiological measures in amyotrophic lateral sclerosis: Markers of progression in clinical trials. *Amyotroph Lateral Scler Other Motor Neuron Disord* 2005;6:17-28.
17. Peioglou-Harmoussi S, Fawcett PR, Howel D, Barwick DD. F-response frequency in motor neuron disease and cervical spondylosis. *J Neurol Neurosurg Psychiatry* 1987;50:593-9.
18. Chroni E, Taub N, Panayiotopoulos CP. The importance of sample size for the estimation of F wave latency parameters in the ulnar nerve. *Muscle Nerve* 1994;17:1480-3.
19. Fisher MA, Hoffen B, Hultman C. Normative F wave values and the number of recorded F waves. *Muscle Nerve* 1994;17:1185-9.
20. Yokota T, Inaba A, Yuki N, Ichikawa T, Tanaka H, Saito Y, *et al.* The F wave disappears due to impaired excitability of motor neurons or proximal axons in inflammatory demyelinating neuropathies. *J Neurol Neurosurg Psychiatry* 1996;60:650-4.
21. Delio DA, Fiori MG, Lowndes HE. Motor unit function during evolution of proximal axonal swellings. *J Neurol Sci* 1992;109:30-40.
22. King AE, Dickson TC, Blizzard CA, Woodhouse A, Foster SS, Chung RS, *et al.* Neuron-glia interactions underlie ALS-like axonal cytoskeletal pathology. *Neurobiol Aging* 2011;32:459-69.
23. Rao MV, Nixon RA. Defective neurofilament transport in mouse models of amyotrophic lateral sclerosis: A review. *Neurochem Res* 2003;28:1041-7.
24. Ahdab R, Créange A, Saint-Val C, Farhat WH, Lefaucheur JP. Rapidly progressive amyotrophic lateral sclerosis initially masquerading as a demyelinating neuropathy. *Neurophysiol Clin* 2013;43:181-7.
25. Cui LY, Liu MS, Tang XF. Single fiber electromyography in 78 patients with amyotrophic lateral sclerosis. *Chin Med J* 2004;117:1830-3.
26. Ibrahim IK, el-Abd MA. Giant repeater F-wave in patients with anterior horn cell disorders. Role of motor unit size. *Am J Phys Med Rehabil* 1997;76:281-7.

Received: 07-10-2014 **Edited by:** Li-Min Chen

How to cite this article: Fang J, Liu MS, Guan YZ, Cui B, Cui LY. Importance of Sample Size for the Estimation of Repeater F Waves in Amyotrophic Lateral Sclerosis. *Chin Med J* 2015;128:515-9.

Source of Support: Nil. **Conflict of Interest:** None declared.