

ORIGINAL**Which muscle shows fasciculations by ultrasound in patients with ALS?**

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Abstract : The purpose of the present study was to elucidate the relative frequencies of fasciculations assessed by sonography in a large number of muscles in patients with amyotrophic lateral sclerosis (ALS). The patients diagnosed as having ALS were retrospectively assessed by muscle sonography. The frequencies of having fasciculations were compared among the 15 muscles and the subtypes according to the initially affected body region. Overall, approximately half of the muscles had fasciculations (48.8%), in the average of 11.4 muscles per patient. The frequency of fasciculations tended to be lower in the patients with longer disease durations upon testing. Biceps brachii had the highest frequency, followed by extensor digitorum communis, whereas sternocleidomastoid and rectus abdominis had the lowest frequencies. The frequencies of fasciculations were similar among the clinical subtypes. In conclusion, in patients with ALS, fasciculations were detected most frequently in proximal arm muscles by sonography, whereas truncal muscles had lower frequencies. Fasciculations tended to be less evident in the advanced disease stage, possibly reflecting muscle degeneration. Appropriate selection of muscles to observe fasciculations is important for diagnosis of ALS. *J. Med. Invest.* 63 : 49-53, February, 2016

Keywords : ALS, fasciculation, sonography, muscle

INTRODUCTION :

Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disease that predominantly affects the upper and lower motor neurons. Fasciculations, that are localized and temporary muscle twitching, have been recognized as a hallmark of this disease entity, suggesting hyperexcitability of motoneurons and motor axons. Given the high sensitivity to detect fasciculations in comparison with clinical examination, needle electromyography has been utilized for the detection. Because of the diagnostic importance, Lambert proposed an electrodiagnostic criteria of ALS ("Lambert criteria") that required the presence of fasciculation potentials (FPs) in the multiple body segments (1). The diagnostic weight of FPs in electrodiagnosis of ALS had been slightly downgraded by the subsequent diagnostic criteria (e.g., the revised El Escorial criteria (2, 3)). Given the low diagnostic sensitivity of the electrodiagnostic criteria, new electrodiagnostic criteria were awaited to achieve high diagnostic sensitivity, especially in the early disease stage (4). To take advantage of fasciculations as a highly characteristic, albeit non-specific, finding of ALS, widespread FPs in the presence of reinnervation were recently included in the Awaji ALS electrodiagnostic criteria and validated by a number of studies (5, 6). Detection of FPs is of clinical significance, because FPs tend to appear earlier than other signs of reinnervation (e.g., increased durations, amplitudes, and phases of motor unit potentials) and application of the Awaji criteria was reported to diagnose ALS earlier than the revised El Escorial criteria (7, 8).

Ultrasonography (US) has also been recently utilized as a

sensitive measure of muscle movements, including fasciculations. Because of its ability to observe a wide area of muscle tissues non-invasively, detection of fasciculations by US was reported to be superior to that by needle electromyography (9). However, there has no detailed analysis as to the distribution of fasciculations by US in order to set up an efficient and sensitive recording protocol. Thus, the aim of the study was to elucidate the distribution of fasciculations by US in patients with ALS and its possible association with clinical information.

MATERIALS AND METHODS :*Inclusion criteria of the ALS patients*

In this retrospective study the patients with ALS were assessed from April, 2013 to April, 2015. The patients who fulfilled the criteria for definite or probable ALS according to the revised El Escorial ALS criteria (2) combined with the Awaji electrodiagnostic criteria (5). Briefly, Awaji electrodiagnostic criteria recognizes fasciculation potential as a sign of active denervation along with fibrillation and positive sharp wave, provided that concurrent evidence of reinnervation is present in the same muscle (i.e., long-duration, high-amplitude motor units, late recruitment, polyphasia). Given the potential disagreement of how the Awaji criteria and the revised El Escorial criteria are to be combined (10), both clinically probable and laboratory-supported probable criteria were enrolled as originally stated in the revised El Escorial criteria (2). The patients were classified into four subtypes based on the initial

Abbreviations :

ALS=amyotrophic lateral sclerosis
EDC=extensor digitorum communis
FCU=flexor carpi ulnaris
FDI=first dorsal interosseous
FDP=flexor digitorum profundus
FDS=flexor digitorum superficialis
FP=fasciculation potential
US=Ultrasonography

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region of clinical involvement (arm, leg, bulbar, and bibrachial), according to the clinical history and initial neurological examination by treating neurologists. The diagnosis of the bibrachial subtype was based on the classification by Wijesekera, *et al.* and those eventually met the above-mentioned ALS criteria (11). This study was approved by the Institutional Review Board of Vihara Hananoso Hospital and Tokushima University. The subjects or their caregivers gave written informed consent at the time of the testing.

Sonography

The sonographic examinations were performed using LOGIQ7 (GE) with an 11-MHz linear-array transducer. A single technician (N.T.) who was not aware of the clinical information performed the sonography. The participants were tested in the supine position. The room temperature was maintained at 23-25 degrees Centigrade. The skin temperatures at the neck and the limbs were measured and maintained >32 degrees Centigrade by covering a blanket. Similar to the previous study, all scans were made in the transverse plane with standard transducer locations corresponding to muscle bellies (9). We observed the muscle for 20 seconds to determine the presence or absence of fasciculations. The definition of fasciculations was made as involuntary twitching of small parts of the muscle, at least two of which were required to be classified as being present. We tested unilaterally on the side of the initial onset. In case of symmetric onset, the right side was selected.

Statistical analysis

The chi-square test and ANOVA with a Games-Howell post-hoc test were used for statistical analysis [SPSS software (version 22.0) (Tokyo, Japan)]. A statistically significant *P* value was set at 0.05.

RESULTS :

Typical presentation of fasciculations is shown in Figure 1, that shows brief twitching of a part of the muscle. The clinical characteristics of the patients are shown in Table 1. Sonographic evaluation for fasciculations were performed in total of 640 muscles, averaging 11.4 muscles per patient [11.4 ± 2.2 (range 7-5)]. Overall, approximately a half (48.8%) of the tested muscles showed fasciculations, ranging from 0 to 100% in each patient. Table 2 shows the frequencies of fasciculations in the 15 tested muscles. In comparison to the grand average of all the patients (48.8%), four

muscles showed significantly different frequencies : biceps brachii and extensor digitorum communis had higher frequencies [66% and 68%, *P*=0.013 and 0.011, respectively (the chi-square test)], whereas sternocleidomastoid and rectus abdominis had lower frequencies (25% and 26%, *P*=0.002 and 0.0078, respectively (the chi-square test)). In addition, the medial head of gastrocnemius tended to have lower frequency than the grand average [36%, *P*=0.07 (the chi-square test)].

Table 1 : patient characteristics and overall sonographic findings

	ALS
Number	56
Age (mean \pm SD ; range)	67.3 \pm 10.5 (38-88)
Gender (men/women)	31/25
Disease duration (months)	27.5 \pm 29.6 (1-110)
Height (cm)	159 \pm 9.8 (134-187)
Weight (kg)	53.4 \pm 11.1 (31-84)
Body Mass Index	21.2 \pm 3.5 (14.5-30.9)
Region of onset	Arm 19 (34%) Leg 17 (30%) Bulbar 12 (22%) Bibrachial 8 (14%)
sonography	
Fasciculations present [individual patient]	312/640 (48.8%) [range : 0-100%, 48.8 \pm 22.1%]
Number of muscles tested/ patient	11.4 \pm 2.2 (7-15)

To study possible associations, multivariate analysis was performed between the overall frequency of fasciculations and the patients' descriptive parameters. There was an inverse correlation between the disease duration and the frequency (Figure 2). Otherwise, no correlation was present between the frequency and the following : age, gender, height, weight, and the body mass index.

Table 3 shows the frequencies of fasciculations according to the region of onset. There was no difference in the frequencies of the all tested muscles, the arm muscles, and the leg muscles. However, in the patients who were tested within one year since disease onset, the frequency of fasciculations in the arm tended

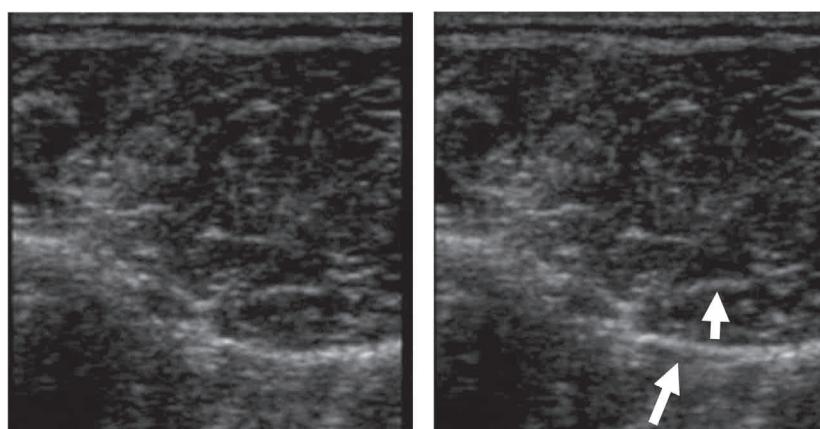


Figure 1 : serial recording of muscle sonography showing typical fasciculations in a patient with amyotrophic lateral sclerosis. In comparison to the baseline image of the biceps brachii (left), there was brief twitching of a part of the muscle causing upward deflection (right).

Table 2 : frequency of fasciculations by sonography

	Number of patients with present/absent fasciculations (% positive)	P value (vs. grand average : 48.8%)
SCM	11/33 (25%)	0.002
Tongue	24/26 (48%)	0.9
Deltoid	22/28 (44%)	0.5
Biceps brachii	37/19 (66%)	0.013
Triceps brachii	30/25 (55%)	0.4
EDC	32/15 (68%)	0.011
FDS	12/11 (52%)	0.7
FCU	12/11 (52%)	0.7
FDP	11/11 (50%)	0.9
FDI	28/27 (51%)	0.8
Quadriceps	30/23 (57%)	0.3
Tibialis anterior	25/27 (48%)	0.9
Gastrocnemius, medial head	19/34 (36%)	0.07
Soleus	10/12 (45%)	0.8
Rectus abdominis	9/26 (26%)	0.0078

SCM, sternocleidomastoid ; EDC, extensor digitorum communis ; FDS, flexor digitorum superficialis ; FCU, flexor carpi ulnaris ; FDP, flexor digitorum profundus ; FDI, first dorsal interosseous

to be lower in the bulbar-onset patients than the limb-onset patients [$P=0.065$ (ANOVA with the Games-Howell post-hoc test)].

DISCUSSION :

In this retrospective study we have performed sonographic evaluation up to 15 muscle groups to determine the relative frequencies of fasciculations. Biceps brachii and extensor digitorum communis had higher frequencies than the grand average, whereas sternocleidomastoid and rectus abdominis had lower frequencies.

Sonographic evaluation of fasciculations in ALS

Fasciculations are observed with a number of underlying conditions, including normal individuals, cramp-fasciculation syndrome, metabolic disorders (e.g., hyperparathyroidism, hyperthyroidism, hypomagnesemia), drug-induced conditions (e.g., caffeine, lithium, terbutaline, anti-cholinesterase, theophylline), motor neuropathies and motor neuron diseases (12). In ALS, fasciculations appear to originate from motor axons and neuronal bodies (13). Because the presence of fasciculations in progressive weakness and muscle atrophy highly indicates motor neuron disorders, fasciculations became an integral part of diagnosis of ALS.

Possible mechanisms of variable frequencies of fasciculations in ALS

The present study showed non-uniform frequencies of fasciculations in ALS ; more specifically, of the 15 tested muscles biceps brachii and extensor digitorum communis had greater detection sensitivities, whereas sternocleidomastoid and rectus abdominis were lower than the ground average. This tendency (i.e., limb > truncal, upper extremity> lower extremity, possibly proximal limb> distal limb) was somewhat consistent with previous studies and will be discussed in detail.

Sonographically, Misawa, *et al.* reported sensitivities of fasciculations in six muscles (i.e., tongue, biceps brachii, first dorsal interosseous (FDI), low-thoracic paraspinalis, vastus lateralis, and tibialis anterior), the biceps being the most sensitive muscle and the paraspinalis the lowest (9). Overall, the sensitivities of their study were higher than ours (48-88%), possibly reflecting the different recording durations (20 seconds in the present study and at least 30 seconds by Misawa, *et al.*).

Electrophysiologically using needle electromyography, Misawa, *et al.* in the same study claimed FDI as the most sensitive among them, followed by biceps brachii and the tibialis anterior being the least sensitive, but the reason for inconsistency of the orders between the needle electromyography and sonography was unknown. Higashihara, *et al.* examined 11 muscles (3 in the cranial, 5 in the cervical, 1 thoracic (paraspinalis), and 2 in the lumbosacral segments), showing triceps brachii being the highest, followed by trapezius, biceps brachii, and vastus medialis (10). Sekiguchi, *et al.* reported the frequencies of FPs in 8 muscles (1 in the cervical, 1

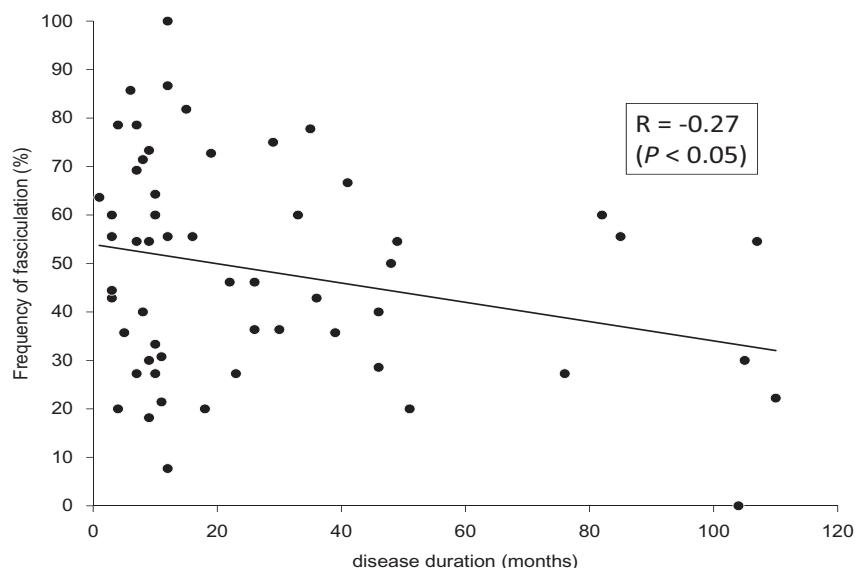


Figure 2 : there was inverse relation between the disease duration and the frequency of the muscles that showed fasciculations.

Table 3 : frequencies of fasciculations based on the region of onset.

	Arm-onset	Leg-onset	Bulbar-onset	Bibrachial	P values
Number (M/F)	19 (11/8)	17 (9/8)	12 (8/4)	8 (3/5)	0.9 (Chi-square)
Number of tested muscles	12.1±2.1	10.4±2.0	12.8±2.3	13.0±6.8	0.15
Overall frequency of fasciculations (% of muscles)	46.7±22.1	51.9±21.5	46.3±22.1	48.7±21.8	0.8
Fasciculations in arm (% of muscles)	58.9±29.9	49.7±34.4	56.0±23.0	58.4±32.5	0.8
Fasciculations in leg (% of muscles)	33.7±34.9	57.2±32.2	47.2±40.0	48.9±32.6	0.4
Number (M/F) : Disease duration ≤1 year	10 (6/4)	5 (2/3)	11 (7/4)	0	0.9 (Chi-square)
Overall frequency of fasciculations (% of muscles)	53.8±21.4	57.5±13.2	41.5±20.9	N/A	0.24
Fasciculations in arm (% of muscles)	69.8±20.8	72.0±10.9	52.1±19.3	N/A	0.065
Fasciculations in leg (% of muscles)	37.5±35.2	45.8±41.7	42.4±38.3	N/A	0.9

thoracic (paraspinalis), 6 in the lumbosacral segments (including 1 paraspinalis) , showing that FDI and gastrocnemius being the highest and the lumbar paraspinalis being the lowest in sensitivity (14). These findings are in accordance with previous reports studied by electromyography (Cappellari *et al.*, 1999, Krarup, 2011). Of note, the localization of FPs is disease-stage-dependent and tends to show proximally in the early disease stage, and to shift distally later in the disease course (15).

The pathogenesis of preferential presence of fasciculations in proximal arm muscles and low frequency in truncal muscles has not been fully identified, but there have been important observations as potential explanations. First, the origin of FPs has been controversial, and is likely to be multifactorial, including distal axons, anterior horn cells, and supraspinal sites (16). As spontaneous firing, FPs imply axonal and/or neuronal hyperexcitability. Indeed, motor neurons and axons in the peripheral and central nervous systems have been reported to have dysfunctions in ionic channels, namely impaired “slow” potassium currents and upregulated persistent Na⁺ currents (17-20). Although regional difference of axonal excitability has been reported in axons innervating a limb muscle as evidenced by *in vivo* “threshold tracking” (21-23), its application for a short paraspinal nerve is technically challenging, thus it is uncertain whether there is different excitability or channel properties between the axons innervating the limb muscles and paraspinalis.

Clinical significance

The present data suggest several points that are relevant to clinical practice. First, the present data would guide the sonographic evaluation to detect fasciculations in a patient with suspected ALS in terms of selection of muscles. Although the position of sonography in the diagnosis of ALS has not been established in the recent criteria (2, 5), sonographic detection of fasciculations would anticipate the presence of FPs in the respective muscle and thus would reduce the total number of invasive needle examination as “echo-guided needle electromyography” (24). Second, our results showing lower frequencies of fasciculations with longer disease durations are consistent with electrophysiological observation (8), in that FPs could be the initial electrophysiological change by needle electromyography that precedes fibrillations and positive waves. It is possible that chronically denervated and degenerated muscles loses firing properties. Third, the present data have shown diffuse fasciculations beyond initially involved regions, even in patients with short disease durations. In an early disease stage when only focal muscular atrophy is evident in a single limb, sonographic evaluation of other, often strong, muscle groups might suggest diffuse neuron loss and axonal dysfunction.

Limitation of the study

This study has limitations. First, the numbers of the tested muscles were not uniform in each patient because of the retrospective methodology. Second, sonographic evaluation of fasciculations is operator-dependent, however, a recent report claimed high inter-rater agreement and ability to distinguish fasciculations from motion artifact (25). Third, there has been no uniformly accepted protocol on sonographic evaluation of fasciculations, such as a duration for observation and selection of a transducer and frequency ranges. In needle electromyography, Mills recommended 60 seconds of observation for fasciculation potentials (26). However, theoretically shorter time of observation could detect fasciculations effectively by sonography because sonography observes wider area of muscle tissues than needle electromyography. A prospective, large-scale study to compare electrodiagnostic and sonographic features would clarify the issues.

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CONFLICT OF INTEREST STATEMENT :

None of the authors has conflict of interest.

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