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Effects of functional electrical stimulation in denervated thigh muscles of paraplegic patients mapped with T_2 imaging

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Abstract

Object—Functional electrical stimulation (FES) for paraplegic patients, with the long-term goal of ultimately restoring muscle function, is associated with several positive effects: improvement of blood circulation, skin condition, peripheral trophism and metabolism, prophylaxis against decubitus ulcer and better physical fitness. Since fibres of denervated muscles (lacking a supplying nerve) need to be activated directly, the fraction of elicited muscle tissue follows the geometric distribution of the electrical field which can be simulated using electro-physiological computer models. Experimental validation of these results, however, has not yet been established.

Materials and Methods—We acquired T_2 parameter images using a multi-slice multi-spin-echo MR sequence before and immediately after FES in 9 denervated paraplegic patients and 3 healthy subjects in order to visualise the geometric distribution of activation by electrically induced muscle stimulation in denervated vs. innervated (healthy) thigh muscle.

Results and Conclusion—After realigning and normalisation, maps of relative T_2 increase were calculated. The results demonstrate that the spatial distribution of short term effects of FES of denervated muscle tissue of paraplegic patients who regularly perform FES can be visualised by T_2 parameter images. This may be used to refine models of the electrical field of FES in muscle and fibre activation in the future.

Keywords

high field MRI; T_2 mapping; human muscle; denervated muscle; functional electrical stimulation; paraplegia; FES

Introduction

Functional electrical stimulation (FES) for paraplegic patients has long been subject of research with the long-term goal of restoring lost muscle functions, as e.g. standing up or hand grasp additionally has several positive therapeutic effects and can further be seen as a sports activity [1,2,3]. The benefits include improvement of local blood circulation and skin

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condition, improvement of peripheral trophism and metabolism, prophylaxis against decubitus ulcer, better overall physical fitness and cosmetic effects. In contrast to healthy muscle, where all regions supplied by an excited nerve are activated, fibres of denervated muscles need to be activated directly by the externally applied electrical field, which is generated via two large surface electrodes. The activation pattern and, therefore, the effect of training follows the geometric distribution of the electrical field which can be simulated using electro-physiological computer models [4, 5, 6]. The results of the computer simulations are activation patterns showing where activation would occur at locations of fibre endings as well as along the length of a fibre. Experimental validation of these results, however, has not yet been established.

MR signal change in skeletal muscle following denervation was subject of research as early as 1988 by Polak [7]. Studying rat muscles, they reported an increase of both T_1 and T_2 relaxation times in denervated muscle. Subsequent research on denervation of muscle focused on the time course and underlying physiological mechanisms of signal abnormalities (including prolonged T_2 relaxation times) following denervation as well as the effect's potential as a non-invasive diagnostic tool [8]. Increased T_2 relaxation times were found in denervated human skeletal muscle [9], as well as in chronically denervated rat muscle [10, 11, 12]. Knee torque measurements [13, 14] and determination of the increase in muscle cross sectional area in CT slices [13, 14] are gross measurements of the overall effects of FES, performed regularly in a period of several months. On the other hand, histological analysis of muscle biopsies [13, 15, 16] may provide detailed information about the stimulation's long-term effects, although at specific locations only. In contrast, magnetic resonance imaging can be used to study the short-term effects and their spatial distribution. Muscle activation can be mapped using MRI by exploiting the effect of prolonged T_2 relaxation times after exercise. This has long been known and used in healthy subjects [17] and athletes [18] e.g. to visualise inter-individual differences in muscle recruitment, and can be used for diagnostic purposes in metabolic diseases and myopathies [19].

The scope of this work is to investigate the applicability of T_2 parameter imaging used for visualising the effect of functional electrical stimulation (FES) in denervated muscles. In this pathological condition, electrical stimulation is governed by a different mechanism of activation than in healthy muscle, i.e. by directly activating muscle fibres, instead of eliciting contractions via a supplying nerve, as is the case for healthy muscle tissue. These different mechanisms necessitate the use of very different stimulation parameters – mainly voltages, FES pulse lengths and timings. As a consequence, the patient's FES protocol is not suitable for use with healthy subjects (intolerably high voltages) and vice versa (i.e. would be too weak). Nevertheless, we applied the same method of T_2 parameter imaging to a group of healthy subjects before and after functional electrical stimulation. This has the only purpose of illustrating that the method of T_2 parameter imaging – which is well established in literature for monitoring voluntary and electrically induced contraction of healthy muscle tissue – yields the expected results at our system, when using the same MRI sequence and post processing methods as applied to paraplegic patients.

To our knowledge T_2 mapping of the effect of electro stimulation has not yet been studied previously, in neither denervated human skeletal muscle in vivo nor animal models thereof. Here, we present data on the spatial distribution of the activation in long-term denervated skeletal muscle and (innervated) healthy subjects after functional electrical stimulation, measured with T_2 parameter MR imaging.

Materials and Methods

Subjects

Nine paraplegic patients with conus cauda lesion and resulting denervation of the muscles in the lower limbs were studied. The patients, who had participated in an FES program comprising regular stimulation as a daily routine for several years, were scanned before and after performing their individual regular FES training, consisting of leg extensions in an upright sitting position. All patients were characterised by a complete lower motor neuron lesion resulting in flaccid paraplegia and total denervation of all leg muscles. For comparison, three healthy subjects, performing FES exercise, were scanned before and after stimulation. However, different electrodes and stimulation protocols had to be used (see below), due to inherent differences between denervated and healthy muscles. Note also that due to the functionally different pathway of muscle stimulation, i.e. via the electric field distribution (denervated muscle) or via motor neurons, healthy subjects are not used as controls but to demonstrate the sensitivity and specificity of the method presented. Patients were divided into two groups, based on their response to functional electrical stimulation, in terms of knee extension torque, which is interrelated with cross sectional area of the thigh muscles and time of denervation until they had started regular FES training. The six well responding patients (group A) achieved a knee torque ranging from 16 to 31 Nm (21 ± 6 Nm), while three patients (group B) achieved only 0 to 7.5 Nm (3.6 ± 3.8 Nm). Knee torque of the leg studied, denervation time before starting regular FES and time of regular FES training are listed in Tab. 1. Typically, each patient's right leg was studied, if this was not prevented by injury or recent operation on this limb.

As denervated muscle fibres have to be activated directly by the electrical field and thus relatively high voltages have to be applied, large electrodes are used [20]. Empirically, best results are achieved using electrodes made of soft flexible conductive rubber attached via a wet sponge sheet or directly using electrode gel as contact medium [21]. The size of the electrodes was 12×18 cm for denervated muscle as opposed to 10×5 cm, used for stimulation of healthy subjects. Electrodes were placed on the anterior side of the right thigh, over the distal and proximal end of the quadriceps femoris. Functional electrical stimulation of the patients was achieved with biphasic rectangular impulses (40 ms total pulse duration, pulse pause of 10 ms, resulting in a stimulation frequency of 20 Hz. Burst length was 2 s with 2 s pause) and a maximum intensity of $160 V_{pp}$, which was suitable to elicit tetanic contractions, after an initial training phase of several months of daily FES with different stimulation parameters [13]. Patients who achieved full extension of the knee, i.e. patients in group A, used additional weights (3.2 ± 1.2 kg) at the ankle to increase the training intensity. The leg was extended in 3 to 6 series of 12 to 15 repetitions, with 2 to 3 min pause between series [14]. Stimulation parameters for healthy subjects were 800 μ s total width of biphasic impulse, stimulation frequency of 30 Hz, bursts of 1 s and 2.5 s pause, 3 series with 2 min pause between the series. Voltage was adjusted individually to achieve full knee extension throughout the FES experiment, with a maximum of $40 V_{pp}$. Seven kilogram of additional weight was attached to the ankle of the stimulated leg. The differences in stimulation techniques are an unavoidable compromise, as parameters that efficiently elicit muscle contractions in a denervated extremity causes intolerable pain in healthy subjects. It is evident that after full contraction of a single muscle group (or two, if adjacent) the pain threshold has been reached in healthy subjects.

NMR protocol

T_2 parameter images were calculated from transversal multi-slice multi-echo (MSME) images of the thigh using a 3T whole body scanner (Bruker Medspec S300, Ettlingen, Germany) with a quadrature receive/transmit birdcage RF coil ($d = 27$ cm, $I = 27$ cm)

provided by the manufacturer.. Six echoes per slice were acquired with a Carr Purcell Meiboom Gill (CPMG) scheme, allowing equidistant echo spacing. To cover a sufficient T_E range for the expected T_2 of skeletal muscle, spin echoes were acquired at 25 – 150 ms, in steps of 25 ms. 14 slices with a slice thickness of $d = 8$ mm and a slice gap of 5 mm (4 patients) or 18 slices with $d = 10$ mm and 9 mm gap (5 patients) were acquired using a T_R of 3 s, while safely remaining below SAR limits, as monitored by the MR system. Acquisition matrix size was constant at 128×96 and reconstructed to 128^2 . The field of view was varied between 10×16 cm² and 19×23 cm², depending on the cross section of each patient's thigh.

Gaussian shaped slice excitation and refocusing pulses with 3.2 and 2.4 ms pulse duration, respectively, were used with a slice selection gradient of 2.51 mT/m. T_2 parameter images were calculated by fitting mono exponential curves to the even echoes, to exploit the advantage of the CPMG scheme of correctly refocusing the even echos in the xy -plane, even for non-ideal 180° -pulses, in order to allow reproducible T_2 measurements and to improve inter-subject comparability of T_2 values [22, 23].

Re-positioning was done using markers on the patient's skin and the patient table. MR-visible markers were placed at the position of the stimulation electrodes. The first scan after exercise was acquired immediately after re-positioning the patients and subjects as quickly as possible, which was accomplished in 6.1 ± 1.8 min after the last FES pulse. Within this time, electrodes were removed, the marker was attached at the electrode position, the patient was carefully padded and re-positioned in the scanner and finally, slice positions were checked on an updated localiser image. As a quality control, two vials filled with 25 ml of gadolinium doped agarose gel [24] (taken from the Eurospin Test Object 5 phantom (Diagnostic Sonar, Scotland, UK)) with transverse relaxation times of $T_2 = 58 \pm 2$ ms and $T_2 = 47 \pm 2$ ms were placed next to the patient's thigh during all measurements. The tubes were placed within the field of view, separated from the thigh by about 2 cm using sponge rubber, to avoid temperature induced T_2 changes in the phantoms during the measurement.

Data processing

Data acquired before and after exercise were realigned and normalised using minc tools [25] (<http://www.bic.mni.mcgill.ca/ServicesSoftware/MINC>), to enable comparison of data on a per-pixel basis. The data set acquired after exercise was used as template image. All geometric operations were calculated for the first echo images and subsequently applied to the remaining echoes. Calculation of T_2 parameter images was done with the free tool ImageJ [26, 27] (<http://rsb.info.nih.gov/ij/>) and the MRI Analysis Calculator plug-in, performing a nonlinear least squares fit of a mono exponential T_2 dephasing curve, $S(T_E) = S_0 e^{-T_E/T_2}$, employing a simplex algorithm.

The background surrounding the thigh cross sections in the images was masked by fully automatic segmentation. To discriminate signal from the background, signal intensity was calculated in a large circular region of interest (ROI) fully within the leg (diameter = half of FOV, centred in the image) in the centre slice, first echo image, assuming that background intensity (noise and ghost signal) was less than 25% of the mean value in that ROI. To reduce bias in T_2 fitting originating from noise distributed across each image, background noise was estimated as standard deviation of signal in a region of interest placed outside the muscle, not affected by ghost artifacts in phase encoding direction.

Activation images were calculated pixel-by-pixel as relative T_2 increase by subtraction of pre exercise T_2 maps from post exercise data, and dividing by pre exercise T_2 s (i.e. “activation” is defined as $\Delta T_2 / T_2$). Anatomically matched regions of interest (ROI) were placed well within in the m. vastus lateralis (VL), m. vastus medialis (VM), m. vastus

intermedialis (VI), m. rectus femoris (RF), adductor muscles (AD), m. semitendinosus and semimembranosus (SM) and m. biceps femoris (BF) to evaluate activation and absolute T_2 pre and post exercise. ROIs were drawn manually on the first echo images of the post exercise scans and applied to the pre and post exercise activation images, respectively. In case it was evident that a muscle was only activated partially (i.e. in healthy tissue, when excitation is elicited via a stimulated nerve), the ROI was restricted to the fraction of activated tissue to maintain comparability to muscles exhibiting more uniform activation.

Results

Cross section images of the thigh of three paraplegic patients and one healthy subject at approximately 25 cm below the trochanter major are shown in Fig. 1. Images are reconstructed from the first echo of the MSME sequence. The regions of interest were drawn manually in all slices, to enable quantification of activation along each muscle. In the images, the large variation in size and anatomy, i.e. distribution of fat and relative size and shape of the muscles between paraplegic patients as well as compared to the healthy subject is evident. Especially for patients in group B (e.g. patient #8 in Fig. 1), the thigh's diameter is reduced, and the proportion of fat and connective tissue is higher than in normal subjects.

T_2 of reference test objects was very reproducible and varied only within $0.7 \pm 3.2\%$ (mean $\pm$ SD) between pre- and post exercise scans.

Activation images ($\Delta T_2/T_2$) of thigh muscles of a paraplegic patient and a healthy subject after FES are shown in Fig. 2, a profile along the thigh is shown in Fig. 3. This sagittal image was reconstructed from the set of axial slices, interpolated to 61 slices. A median filter with a radius of one pixel was applied to the sagittal slice. Generally, activation is distributed more evenly in thigh muscles of paraplegic patients than in the healthy subjects, where distinct muscles, mainly RF, VM and VL, are activated (Figs. 2 (a) and (b)). Using T_2 increase as an index, muscles of well trained patients (group A) are activated to a larger extent than healthy muscles. In patient group B, relative T_2 change is generally much lower than in well trained patients or in activated muscles of healthy subjects (cf. Figs. 4 and 5). Both patient groups, however, show activation in the posterior muscles (adductors, m. semimembranosus and biceps femoris), where healthy subjects do not show any T_2 increase after functional electrical stimulation, with parameters suitable and tolerable to normal innervated muscles. This is also shown quantitatively as average over each patient and subject group, respectively, in Figs. 4 and 5. Figure 5 shows profiles of relative T_2 change along the axis of the leg as group averages for patient groups A, B and healthy subjects. Data from two anterior ROIs (RF and VM muscles) and two posterior ROIs (SM and BF muscles) are shown. It can be clearly seen in Fig. 5 that activation in patient group A is limited to anterior muscles, while muscles in group B are activated more homogeneously, however at a smaller magnitude. In healthy subjects, activation is also limited to anterior muscles, but less homogeneous than in healthy subjects (Fig. 5 c).

In patients responding well to FES (group A), average activation in the anterior muscle groups is $16 \pm 1\%$, and only $7 \pm 1\%$ in posterior groups, measured as mean $\pm$ standard error of the mean across the regions under the distal electrode and between the electrodes, as given in Fig. 4. In patients of group B, who's lower limbs have a smaller cross section, and the electrodes thus cover a larger fraction of the thigh, average activation is distributed almost equally, with $3.9 \pm 0.8\%$ in anterior and $4.0 \pm 0.9\%$ in posterior muscles. In the activated anterior muscles of healthy subjects activation varies in a similar range as seen in well trained patients and there is even a slight deactivation with a relative T_2 change of $-3.3 \pm 0.7\%$ in the passive adductor muscle groups (see Figs. 2 and 5).

Discussion and Conclusion

Our results demonstrate that activation after FES of denervated muscles of patients with a conus cauda lesion can be mapped by T_2 parameter images acquired with a multi-slice multi-spin-echo MR sequence. In appropriate test objects the variability of the measurement method applied using a 3T scanner was sufficiently low, i.e. below 1 %.

Our general hypothesis for this work is that FES leads to T_2 changes also in degenerated denervated muscles, as it is known for healthy muscle tissue. This was confirmed successfully. The second hypothesis was that the distribution of activation using the FES protocol applied to patients with degenerated denervated muscle is different to the distribution of activation achieved in healthy subjects performing FES, of course with a different stimulation protocol. We were able to show that FES induced T_2 changes in those patients extends across the entire thigh including the hamstrings, as was expected due to the training status of the thigh muscles: In contrast to weak responders (or patients not performing FES), the degeneration of the anterior muscles of well trained patients is far less progressed, although they never performed FES applying the electrodes to the anterior side of the thigh.

The activated areas in the thighs of well trained patients were roughly delineated by muscle boundaries and activation was relatively homogeneous across the muscle cross section, although the examined muscles of all paraplegic patients were denervated and thus are activated by direct stimulation of the muscle fibres, rather than by distributing activation across the muscle via motor neurons. The reason for this finding is likely to be the difference in electrical properties of muscles, fat between the muscles, and fasciae. It was therefore considered appropriate to select regions of interest matched to the muscles, rather than e.g. evaluating activation in regions only defined by position relative to the electrodes or distances from reference points in the thigh. Activation maps were calculated as relative T_2 changes, because the high inter-subject variability even between healthy subjects, as was found here and reported in literature [28], suggests that comparison of relative changes in signal intensity within subjects is most appropriate. Variation in geometry of the individual muscles and the entire thigh between patients was substantial, depending on training status and denervation time before FES training.

The different distribution of activation in the thigh of denervated patients compared to healthy subjects is a consequence of the different activation mechanisms, the relatively large size of the electrodes and the high electrical field strength applied, which is necessary for direct excitation of the muscle fibres. As expected, activation was found predominantly in anterior muscles, but also its extension into posterior muscle groups is consistent with the long-term training effects of FES in denervated muscles: Were the hamstrings not activated at all, they would degenerate further, i.e., cross sectional areas would decrease more and contractile muscle tissue would be replaced by fat and connective tissue. A reduction in total knee extension torque in patients having reached a high training level has been observed before [14] and is attributable to co-contractions of the quadriceps' antagonists, an unwanted effect [20] that leads to a non-perfect correlation between knee torque and muscle tension in the extensors and to biased force measurements. It is one goal of this study, by mapping the effect of FES, to improve the understanding of functional electrical stimulation, refine models and thus to help optimise FES protocols.

In healthy subjects, muscle activation is mediated by motor neurons. As soon as a muscle or a group of similarly excited muscles are activated, the subject would not tolerate further increase of stimulation amplitude, as electrical stimulation of normal muscle can be very unpleasant. Thus, only distinct muscles or fractions of muscles can be activated. Activation

is very heterogeneous between subjects, as it depends on distribution of the electric field and anatomy. In this study, we found only the anterior muscles to be stimulated in healthy subjects. Averaging activation in all anterior muscles, as given for the patients (groups A and B) in the results section, is therefore not meaningful. The antagonists are not activated at all (see Figs. 2 and 5). This is expected when electrically stimulating healthy muscles, which are elicited via stimulated motor neurons, by using surface electrodes placed on the anterior side of the thigh.

We are well aware that comparing patient results with those of subjects is not straight forward, due to the differences in the stimulation parameters, electrode sizes, exercise protocol details and, obviously, the mode of activation of muscle cells. However, the main focus of this work is to demonstrate that studying the spatial distribution of activation in *denervated* and successfully FES-trained human thigh muscle is feasible with T_2 mapping. So far, this is only possible via simulation in electro-physiological models and could be inferred from the long-term effects of FES. We show data of healthy muscle, activated using a stimulation protocol more suitable for healthy subjects, merely for a qualitative comparison and to demonstrate the sensitivity ($\Delta T_2/T_2$) and specificity (muscle groups activated) of the method.

The long T_2 of lipids and the fact that the lipid's relaxation time is not prolonged by muscle activation introduces an offset to the basal T_2 of tissue containing fat, which results in a reduced relative T_2 change. The T_2 mapping method therefore becomes less sensitive to muscle activation with increasing lipid content. This effect manifests as a partial volume effect in the pixel-by-pixel T_2 fit, as well as a bias in the ROI analysis, when pixels only containing fat or connective tissue are included in the ROI. This effect may be modelled by including knowledge about the fat content of the tissue (measured in a separate experiment) or with an approach based on elevated basal absolute T_2 values compared to normal muscle tissue. Here, we report gross T_2 changes without correcting for this bias, to avoid the uncertainty introduced by such a high order fit.

The approach of measuring T_2 using a MSME method with CPMG phase cycling scheme was extensively tested on phantoms: T_2 times measured with this method were comparable to the results of much more time consuming single-spin-echo sequences, consecutively varying T_E in 6 steps. Note also that T_2 results of the 3-point fit were still accurate when deliberately misadjusting the refocusing pulses by $\pm 45^\circ$, to test the robustness of the protocol.

Re-positioning of the thigh is an important issue, because not only position and orientation but also shape of the thigh muscles may vary before and after exercise. This requires realignment and normalisation of pre- and post exercise data for a pixel-by-pixel analysis. The high stability of T_2 measurements in the vials placed next to the thigh indicates that the variability of the T_2 measurements in vivo is due to variations in tissue T_2 change and inter-subject variations.

In conclusion, our results show that electrically induced activation of muscle tissue of denervated paraplegic patients, who regularly perform FES, can be visualised by T_2 parameter imaging. T_2 changes after FES were found in denervated muscles as well as in healthy subjects, with characteristic differences in distribution and intensity of activation, caused by the different techniques necessary for functional electrical stimulation of denervated and healthy muscles. The findings advance present knowledge on the geometric distribution of activation in denervated human musculature in vivo and may be used to refine models of the electrical field of FES in muscle and fibre activation in the future.

Results may further support the optimisation of parameters for clinical therapy and allow regular follow-up examination.

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Fig. 1.

Cross sections of the thigh of three paraplegic patients and one healthy subject 25 cm below trochanter major; first echo of MSME sequence (parameters see methods section). ROIs were placed manually across the whole volume covered, in the m. vastus lateralis (VL), m. vastus medialis (VM), m. vastus intermedius (VI), m. rectus femoris (RF), adductor muscles (AD) m. semimembranosus and semimembranosus (SM) and m. biceps femoris (BF) to evaluate activation and absolute T_2 pre and post exercise.

Fig. 2.

Representative slice showing muscle activation after functional electrical stimulation (a) in a paraplegic patient (#1, group A) and (b) in a healthy subject (#1). The image shows relative T_2 changes, i.e. post exercise minus pre exercise T_2 , normalised to pre exercise T_2 in %. Electrode size and position is indicated by grey lines. The red arrow indicates the position of the intersecting sagittal slice from patient #1 shown in Fig. 3.

Fig. 3.

A sagittal slice showing the profile of the effect of FES parallel to the thigh in a paraplegic patient's muscles. The slice intersects the axial slice shown in Fig. 2 from the same patient at the position is indicated by the red arrow.

Fig. 4.

Activation after FES in thigh muscle of paraplegic patients and healthy subjects, at a position (a) beneath the distal electrode and (b) between the electrodes, 10 cm above the centre of the distal electrode. Columns represent mean activation in the ROIs, averaged across three slices before group averaging. Error bars indicate standard error of the mean.

Fig. 5.

Profiles of relative T_2 change along the long axis of the leg as group averages for (a) six well trained paraplegic patients, (b) three patients with a weak response to FES and (c) three healthy subjects. Data from two anterior ROIs (RF and VM muscles) and two posterior ROIs (SM and BF muscles) are shown. Distal direction is towards positive values on the axis of abscissa.

Table 1

Knee torques, denervation times and time of regular FES training of the paraplegic patients prior to MRI.

Patient number	knee torque [Nm]	denervation time [yrs] pre FES	FES training time [yrs]	total denervation time [yrs]
<i>Group A</i>				
A1	30.6	1.7	2.2	3.9
A2 *	21.1	1.2	1.9	3.1
A3	15.7	0.8	2.6	3.4
A4	17.6	0.8	2.5	3.3
A5	21.5	2.0	7.8	9.8
A6	17.1	1.8	11.2	13.0
<i>Group B</i>				
B7 **	7.5	0.7	2.3	3.0
B8	3.3	7.6	2.5	10.1
B9 *	0	8.7	3.1	11.8

* FES exercise and T_2 imaging was done on left leg

** Very low compliance to FES training.