Using wavelet analysis to reveal the muscle functional recovery following nerve reinnervation in a rat model

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***Abstract*— Targeted muscle reinnervation (TMR) technique has been successful in many amputees for providing sufficient electromyography (EMG) signal to control advanced prosthetics. However, it seems to lack further understanding of the recovery progress of muscle functions after targeted muscle reinneveration surgery. In this study, a rat TMR model was developed to investigate intramuscular EMG activity changes after reinnervation. Using the discrete wavelet decomposition and average rectified algorithm, the recorded EMG showed a gradual improvement in the reinnervated muscle within four weeks. Future work will be performed to further assess the efficiency of reinnveration therapy after the surgery.**

I. INTRODUCTION

Targeted muscle reinnervation (TMR) is a new technique developed by Dr. Todd Kuiken et. al, which transfers the remaining limb nerves to alternative muscles (targeted muscles) to produce sufficient electromyography (EMG) information for real time control of advanced prosthetic arms [1-4]. Although TMR has achieved great successes in the control of multifunctional myoelectric prostheses for many above-elbow amputees, it is imperative to further study the muscle recovery process after reinnervation. Obviously, it is unseemly to conduct these fundamental investigations on human subjects [5].

In this study, we developed a TMR model on rats. To investigate muscle recovery process, implanted electrode was used to record intramuscular EMG signal. Then the TMR group, denervation group and control group rats were compared to show the EMG activity changes after innervation. However, it contained plenty of noise in the recorded EMG signal. To analyze the recorded EMG signal, discrete wavelet transform (DWT) was used to reveal muscle activity progress.

II. MATERIALS AND METODS

A. Animal Preparation

The experimental procedures were approved by the Ethics Committee for Animal Research, the Shenzhen Institutes of Advanced Technology, Chinese Academy of Sciences. 18 Sprague-Dawley rats (200-250g, SPF level, provided by the Guangdong Medical Laboratory Animal center) were used in the experiment. All rats were housed in a temperature controlled room during a 12-hour day/night cycle with accessing to food and water ad libitum. Then 18 rats were randomly divided into the TMR group (n=6), denervation group (n=6), and control group (n=6). In TMR group, the pectoralis major muscle of the right side chest was selected as targeted (reinnervated) muscle, and the left side pectoralis major muscle was intact for comparison. The original median nerve was transected and transferred into the pectoralis major muscle. In denervation group, the original nerves of the right side pectoralis major muscle were transected, and the left side pectoralis major muscle remained intact. In control group, the left and right side pectoralis major muscles were both intact. The detailed TMR surgical procedures were described before in our previously paper [5].

B. Implantation of EMG recording electrode

The surgical implantation of intramuscular EMG recording electrodes were performed following the protocol in [6]. Briefly, a 5 pin connector (Omnetic, Minneapolis, MN) was secured to the skull and the Teflon coated stainless wires (Cat No. 793500, A-M system Inc) were passed subcutaneously either to the back or the pectoralis muscle.

C. EMG data collection and treadmill locomotion

The treadmill locomotion and EMG recording program was performed once per week within 4 weeks after the implantation of the electrodes in all rats. The treadmill locomotion program consisted of a running session (30 s) and a rest session (30 s), and repeated 3 times. Furthermore, the treadmill was set at a 15 degree incline, with a belt speed of 9 m/min at running session. To record EMG data, a biological signal acquisition system was used in the experiment (MedLab-U8C502, Nanjing MedEase Science and Technology Co., Ltd, China). During the locomotion, the rat EMG signal was recorded at a sample rate of 2000 Hz and a amplifier gain of 1000.

D. Power spectral density distribution of raw EMG signal

To characterize the frequency contents of the raw EMG signal, the power spectral density (PSD) was estimated using Welch method. Fig. 1 shows an representative PSD distribution of raw EMG signal from both sides pectoralis major muscle of a TMR group rat at running and rest session. It can be observed that the frequencies below about 100 Hz are the main noise source.

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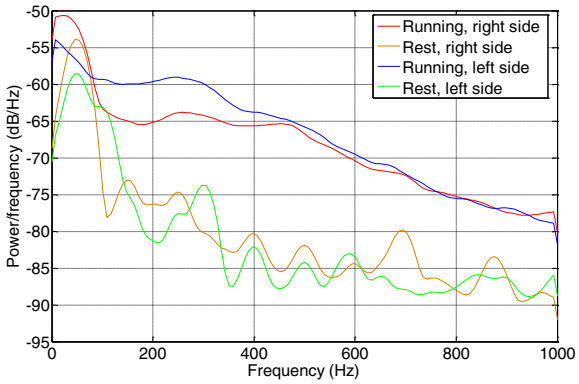

Figure 1. The power spectral density distribution of a TMR rat during running and rest session. The EMG of right side pectoralis major muscle was recorded in channel 1, and the left side pectoralis major muscle EMG was recorded in channel 2. The rat EMG signal at the rest session can be treated as background noise.

E. EMG signal decomposition

The discrete wavelet transform analyzed the signal at different frequency bands with different resolutions by decomposing the signal into a coarse approximation and detail information [7]. The number of decomposition levels was selected based on the frequency content of the signal. Table 1 describes the frequency ranges corresponding to different wavelet decomposition levels. The wavelet coefficients were computed with db4 wavelet function, since Daubechies' s function is recommended for analyzing EMG signal [8]. Figs. 2 and 3 show approximation and detailed coefficients of four level db4 wavelet decomposition on EMG signals from right and left side of pectoralis major muscle of a TMR group rat at 30s running session. It can be seen that the noise were mainly in the frequency range of cd4 and ca4. It's probably mainly due to motion artifacts and power line interference (50 Hz and corresponding harmonics). Thus the coefficients of cd4 and ca4 were not used in the evaluation of muscle function recovery using EMG.

TABLE I. FREQUENCY RANGES CORRESPONDING TO DIFFERENT LEVELS OF DISCRETE WAVLETE DECOMPOSITION

Decomposed signal	**Frequency range (Hz)**
cd1	500-1000
cd2	250-500
cd3	125-250
cd4	62.5-125
ca4	0-62.5

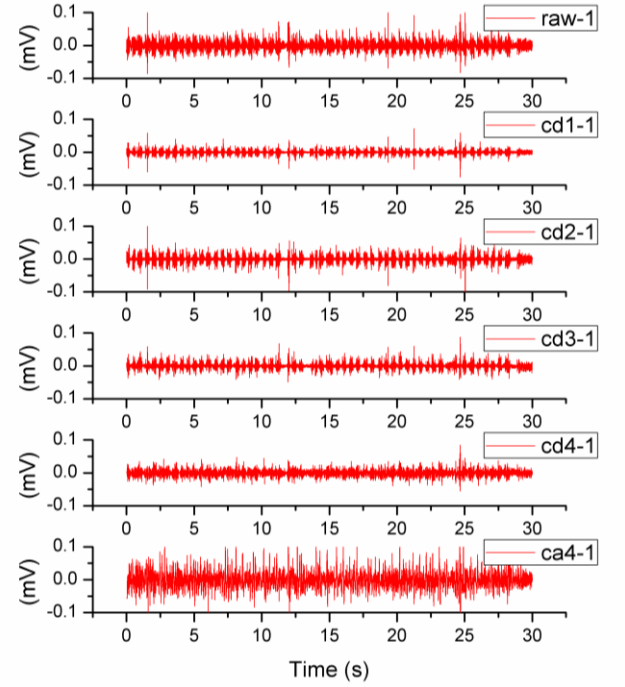

Figure 2. The approximation and detailed coefficients of four level discrete wavelet decomposition from reinnervated pectoralis major muscle in a TMR rat during running session. It can be observed that noise signal is mainly in the cd4 and ca4 coefficients. Raw-1 stands for raw EMG signal recorded from channel 1 (right side pectoralis major muscle).

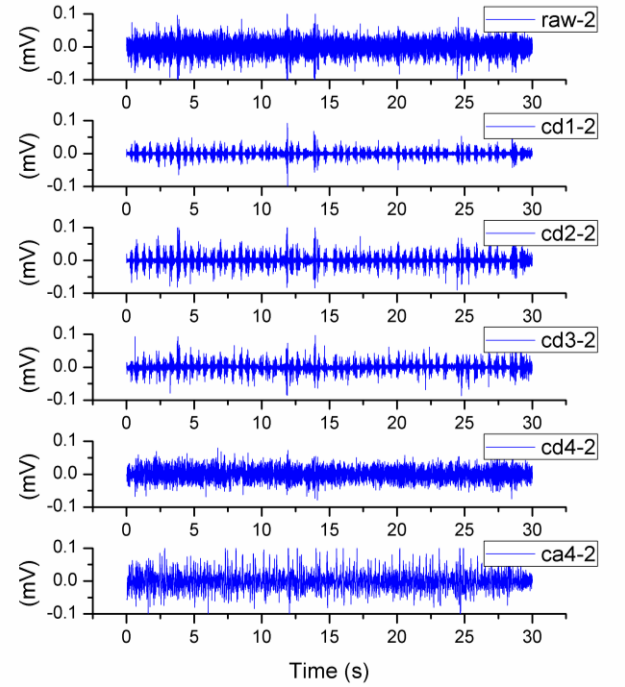

Figure 3. The approximation and detailed coefficients of four level discrete wavelet decomposition from normal pectoralis major muscle in a TMR rat during running session. It can be observed that noise signal mainly lies in the cd4 and ca4 coefficients. Raw-2 stands for raw EMG signal recorded from channel 2 (left side pectoralis major muscle).

F. EMG activity evaluation and statistical analysis

$$ARV = \frac{1}{N}\sum_{n=1}^{n=N}\left|X(n)\right| \quad (1)$$

The energy in the EMG signal for right and left side pectral major muscles was estimated using average rectified value (ARV) method. ARV EMG is defined as a time windowed mean of the absolute value of the EMG signal, see equation 1 [9]. It is similar to integrated EMG features, and can be used to quantify muscle activity over time [10-11].

The EMG signal of 30 s period at running session was selected for analysis in all groups. ARV EMG data of right and left side was compared at different frequency bands (raw signal, cd1, cd2, cd3). The statistical results were expressed as mean ± SEM. Paired t-test was used to determine the statistically significant differences. The criterion of significance was set at $P<0.05$ for all computations.

III. RESULTS

A. Average rectified value of raw EMG signal

ARV EMG of the right and side pectroralis major muscle in all groups is shown in Fig. 4. ARV EMG in denervation group was minimal, followed by TMR group and control group. However, it shows no statistically significant differences between the right and left side muscle in all groups during the first week after surgery. In the fourth week post surgery, only denervation group shows statistically difference between the right and left side muscle. The ARV of raw EMG signal did not provide useful information to characterize muscle activity changes after reinnervation.

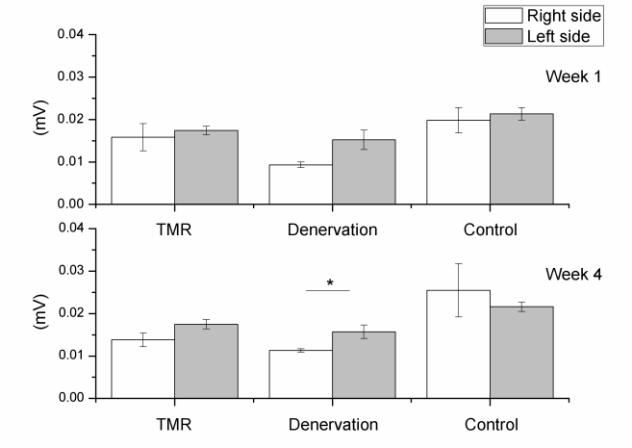

Figure 4. ARV EMG during treadmill locomotion in the first and fourth week in all groups. Values are mean±SEM for 6 rats. *, significantly different between the right side and the left side at $P< 0.05$.

B. Average rectified value of detailed wavelet coefficients

ARV of wavelet coefficients cd1, cd2, and cd3 were used to compare the right and left side muscle in all groups.

ARV of the detailed coefficients during treadmill locomotion in TMR group is shown in Fig. 5. It was statistically significant different between the right and left side muscle at coefficients of cd1, cd2, and cd3 in the first week after surgery. Then there was an improvement in mean EMG amplitude of the right side muscle within four weeks. At the fourth week, the ARV of the detailed coefficients of the right side and the side were similar, no significant difference in EMG activity was found between the reinnervated muscle and normal muscle.

ARV of the detailed coefficients during treadmill locomotion in denervation group was shown in Fig. 6. It was significant different between right and left side muscle at the first week after denervation surgery. No improvements in the muscle activity of the denervated muscle was found. At the fourth week, the EMG activity of the denervated muscle remained significantly less than the normal muscle at coefficients of cd1 and cd2.

ARV of the detailed coefficients during treadmill locomotion in control group was shown in Fig. 7. No statistical difference was found between the right and left side muscle.

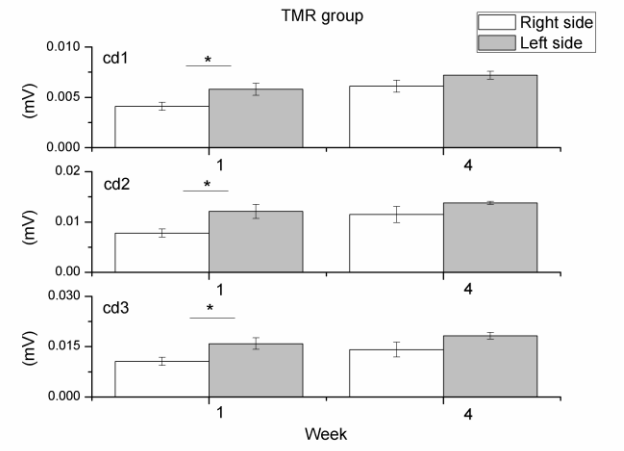

Figure 5. ARV of detailed coefficients cd1, cd2, and cd3 during treadmill locomotion in the first and fourth week in TMR group. Values are mean± SEM for 6 rats. *, significantly different between the right side and the left side at $P<0.05$.

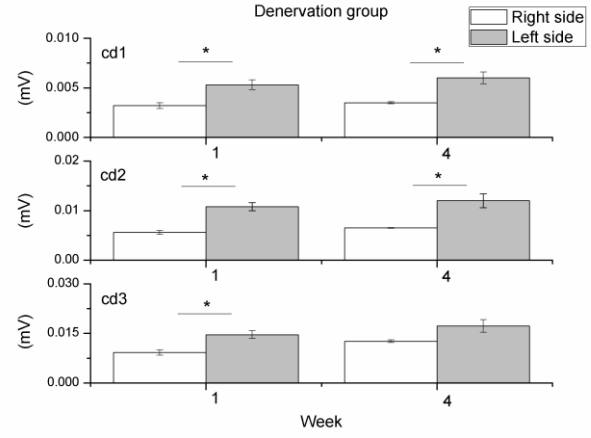

Figure 6. ARV of detailed coefficients cd1, cd2, and cd3 during treadmill locomotion in the first and fourth week in denervation group. Values are mean±SEM for 6 rats. * indicates significantly different between the right side and the left side at $P<0.05$.

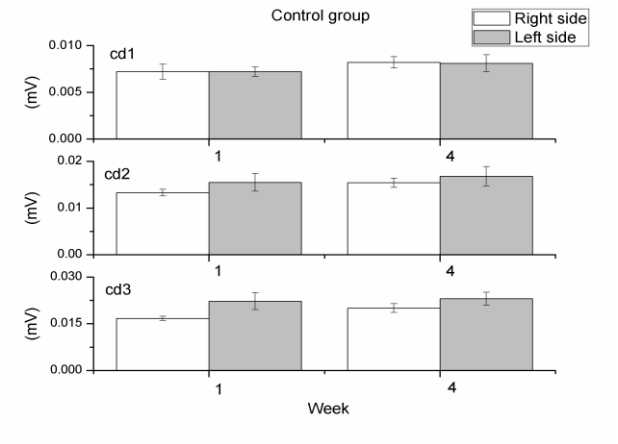

Figure 7. ARV of detailed coefficients cd1, cd2, and cd3 during treadmill locomotion in the first and fourth week in control group. Values are mean ± SEM for 6 rats. No significant difference was found between the right side and the left side at $P<0.05$.

IV. Discussion

In this study, we developed a rat model to investigate the recovery process after targeted muscle reinnervation using intramuscular EMG electrode. However, the recorded EMG signal was contaminated by noises such as power line interference and motion artifacts. The discrete wavelet transform was used to reduce the influence of noise. The ARV of DWT coefficients reveal the gradual increase in muscle activity after TMR within four weeks. Compared with the normal muscle, the EMG activity of the targeted muscle may suggest an innervation between the transferred nerve and the targeted muscle within four weeks after TMR surgery. For amputees after TMR surgery, the muscles need time a long time to reinnervate and heal [12]. Moreover, it was reported in animal experiments that cell therapy or Chondroitinase ABC can reduce muscle innervation time and improves functional recovery [13-14]. Since the success of the TMR technique depends on the sufficient EMG information produced by the reinnverated muscle, these advances in neuroscience may benefit TMR surgery candidates. In future, the developed rat TMR model will be used to investigate and assess the therapy of enhancement reinnervation between transferred nerve and targeted muscles to guide clinical practice.

Discrete wavelet decomposition and average rectified algorithm have been used in this study to distinguish difference in the EMG activity between the normal muscle and the reinnervated muscle. Discrete wavelet transform is used to obtain the EMG information at specific frequency bands. Average rectified is a popular method to quantify the EMG activity over a time period. Besides ARV, there are a lot of EMG feature extraction methods such as peak rectified value, root mean square, median frequency, zero crossing, etc. Future work will compare the performance of these methods, and select the optimal feature to characterize EMG recovery process.

V. Conclusion

This paper investigated reinnvervated muscle EMG activity changes after by comparing with normal and denervated muscle in a rat model with implanted recording electrode. Combined with discrete wavelet decomposition and average rectified algorithm, the results suggest a gradual improvement in EMG activity in the reinnvervated muscle within four weeks. The developed rat TMR model and EMG analysis algorithm could be used for fundamental research of reinnervation and optimal training protocol after TMR to guide clinical practice.

References

[1] T. A. Kuiken, G. Li, B. A. Lock, R. D. Lipschutz, L. A. Miller, K. A. Stubblefield, and K. B. Englehart, "Targeted muscle reinnervation for real-time myoelectric control of multifunction artificial arms," *Jama*, vol. 301, no. 6, pp. 619–628, 2009.

[2] T. A. Kuiken, L. A. Miller, R. D. Lipschutz, B. A. Lock, K. Stubblefield, P. D. Marasco, P. Zhou, and G. A. Dumanian, "Targeted reinnervation for enhanced prosthetic arm function in a woman with a proximal amputation: a case study," *The Lancet*, vol. 369, no. 9559, pp. 371–380, Feb. 2007.

[3] L. A. Miller, K. A. Stubblefield, R. D. Lipschutz, B. A. Lock, and T. A. Kuiken, "Improved myoelectric prosthesis control using targeted reinnervation surgery: a case series," *IEEE Trans. Neural Syst. Rehabil. Eng.*, vol. 16, no. 1, pp. 46–50, 2008.

[4] G. Li, A. E. Schultz, and T. A. Kuiken, "Quantifying pattern recognition-based myoelectric control of multifunctional transradial prostheses," *IEEE Trans. Neural Syst. Rehabil. Eng. Publ. IEEE Eng. Med. Biol. Soc.*, vol. 18, no. 2, pp. 185–192, Apr. 2010.

[5] H. Zhou, F. Wu, L. Yang, L. Zhang, H. Guo, and G. Li, "A preliminary analysis of reconstructed nerve function using targeted muscle reinnervation in a rat model," in *Neural Engineering (NER), 2013 6th International IEEE/EMBS Conference on*, 2013, pp. 1602–1605.

[6] R. R. Roy, D. L. Hutchison, D. J. Pierotti, J. A. Hodgson, and V. R. Edgerton, "EMG patterns of rat ankle extensors and flexors during treadmill locomotion and swimming," *J. Appl. Physiol. Bethesda Md 1985*, vol. 70, no. 6, pp. 2522–2529, Jun. 1991.

[7] Q. Xu, H. Zhou, Y. Wang, and J. Huang, "Fuzzy support vector machine for classification of EEG signals using wavelet-based features," *Med. Eng. Phys.*, vol. 31, no. 7, pp. 858–865, Sep. 2009.

[8] R. Chowdhury, M. Reaz, M. Ali, A. Bakar, K. Chellappan, and T. Chang, "Surface Electromyography Signal Processing and Classification Techniques," *Sensors*, vol. 13, no. 9, pp. 12431–12466, Sep. 2013.

[9] R. Merletti and P. Di Torino, "Standards for reporting EMG data," *J Electromyogr Kinesiol*, vol. 9, no. 1, pp. 3–4, 1999.

[10] A. Phinyomark, P. Phukpattaranont, and C. Limsakul, "Feature reduction and selection for EMG signal classification," *Expert Syst. Appl.*, vol. 39, no. 8, pp. 7420–7431, Jun. 2012.

[11] A. E. Hibbs, K. G. Thompson, D. N. French, D. Hodgson, and I. R. Spears, "Peak and average rectified EMG measures: which method of data reduction should be used for assessing core training exercises?," *J. Electromyogr. Kinesiol. Off. J. Int. Soc. Electrophysiol. Kinesiol.*, vol. 21, no. 1, pp. 102–111, Feb. 2011.

[12] K. A. Stubblefield, L. A. Miller, R. D. Lipschutz, and T. A. Kuiken, "Occupational therapy protocol for amputees with targeted muscle reinnervation," *J. Rehabil. Res. Dev.*, vol. 46, no. 4, pp. 481–488, 2009.

[13] C. Lazerges, P.-A. Daussin, B. Coulet, R. Boubaker el Andalousi, J.-P. Micallef, M. Chammas, Y. Reyne, and F. Bacou, "Transplantation of primary satellite cells improves properties of reinnervated skeletal muscles," *Muscle Nerve*, vol. 29, no. 2, pp. 218–226, Feb. 2004.

[14] M. J. Sabatier, B. N. To, S. Rose, J. Nicolini, and A. W. English, "Chondroitinase ABC reduces time to muscle reinnervation and improves functional recovery after sciatic nerve transection in rats," *J. Neurophysiol.*, vol. 107, no. 3, pp. 747–757, Nov. 2011.