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**Original Article**

# Analysis of the crosstalk in mechanomyographic signals along the longitudinal, lateral and transverse axes of elbow flexor muscles during sustained isometric forearm flexion, supination and pronation exercises

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**Abstract**

**Objective**: To analyse the influence of muscle fibre axis on the degree of crosstalk in mechanomyographic (MMG) signals during sustained isometric forearm flexion, pronation and supination exercises performed at 80% maximum voluntary contraction (MVC) at an elbow joint angle of 90°. **Methods**: MMG signals in longitudinal, lateral and transverse directions of muscle fibres were recorded from the elbow flexors of twenty-five male subjects using triaxial accelerometers. Cross-correlation coefficients were used to quantify the degree of crosstalk in all nine possible pairs of fibre axes, all muscle pairs and all exercises. **Results**: MMG root mean square (RMS) was statistically significant among the fibre axes ($p<0.05$, $\eta^2=0.17$-$0.34$) except for biceps brachii and brachioradialis in supination and brachialis in flexion. Overall mean crosstalk values in the three muscle pairs (biceps brachii & brachialis, brachialis & brachioradialis and brachioradialis & biceps brachii) were found to be 6.09-52.17%, 4.01-61.42% and 2.16-51.85%, respectively. Crosstalk values showed statistical significance among all nine axes pairs ($p<0.05$, $\eta^2=0.16$-$0.51$) except for biceps brachii & brachialis during pronation. The transverse axes pair generated the lowest mean crosstalk values (2.16-9.14%). **Conclusion**: MMG signals recorded using accelerometers from the transverse axes of muscle fibres in the elbow flexors are unique and yield the least amount of crosstalk.

**Keywords:** Crosstalk, Elbow Flexors, Mechanomyography, Muscle Fibre Axis, Signal Propagation

## Introduction

Mechanomyography (MMG) has been used for the assessment of skeletal muscle activity over the last three decades[1]. MMG is an effective alternative to electromyography (EMG) in providing global information on muscle function[2] since MMG signals detect more of the muscle during activation and hence precise placement of the sensor over the muscle is not critical. Additionally, MMG yields more promising results than EMG specifically during fatiguing contractions and is thus a suitable alternative to EMG for the assessment of muscle fatigue[2-4] and the contractile properties of muscle[5]. Despite other related benefits of this technique, such as its ease of operation and decreased signal processing cost[6], crosstalk is a drawback that hinders the application of MMG in clinical applications[7]. Specifically, clinical applications, such as prosthetic control, require the separate identification of the functions of individual muscles, but crosstalk might result in misinterpretation of individual muscle functions. Crosstalk can be defined as contamination of the signals from the observed muscle by the signals from muscle(s) in close proximity[8]. Theoretically, MMG signals from two separate muscles should not have a high common signal percentage as these signals are generated by two unique sources and are thus less likely to show an identical waveform shape. Unfortunately, the literature does not include many studies on crosstalk in MMG signals[9-12] suggesting further investigation on issues related to crosstalk in MMG signals in order to precisely define the use of MMG as a muscle assessment tool.

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The authors have no conflict of interest.

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**Figure 1.** Illustration of postural settings for flexion exercise.

**Figure 2.** Illustration of the postural settings for the pronation and supination exercises.

As detailed in the literature, the MMG signals originating from a muscle spread in all directions[13] and are filtered by the surrounding objects, including bones, tendons, fat and skin[14]. A few studies performed using multiaxial accelerometers have revealed interesting observations on the amplitudes of the MMG signals in different directions. MMG root mean square (RMS) provides useful information on the number of motor units recruited which provides better insights to understand the physiological process of muscle activation[15]. Previous studies have also shown that the MMG signals measured along the x, y and z axes of an accelerometer might correspond to the muscle vibrations produced in the longitudinal, lateral and transverse directions to muscle fibres.

Mean MMG RMS values along the lateral axis of biceps brachii (BB) muscle fibres have been found greater than the mean MMG RMS values along the longitudinal axis during isometric contractions at different levels of effort[16]. MMG signals are generated mainly through the lateral expansion of BB muscle fibres, thus the signals generated along the lateral axis of muscle fibres were considered for analysis[17]. A grid of 13 × 12 accelerometers were used to analyse MMG amplitude of tibialis anterior muscle during isometric contractions[18,19]. Both these studies concluded that, for a single motor unit, the MMG amplitude is mainly affected in lateral direction only.

A few other studies found that the MMG signals along different axes of muscle fibres are frequency band specific. It was observed that MMG signals propagate mainly along the longitudinal axis to the muscle fibres if the frequencies are greater than 25 Hz and along lateral axis if the frequencies are less than 25 Hz during submaximal isometric contractions of BB muscle having a grid of 15 accelerometers on it[20]. A triaxial accelerometer was used over the rectus femoris muscle, which was activated through external stimulation, and it was observed that the MMG signals along the longitudinal fibre axis showed a higher concentration in the 16-34 Hz band, whereas the MMG signals along the lateral and transverse fibre axes showed a higher concentration in the 22-29 Hz band[21]. A strong association was revealed between the MMG signals along the longitudinal and transverse axes of the accelerometer[22]. A common observation in these studies reveal that the lateral axis of muscle fibres provides higher MMG amplitude.

However, when taken together, none of these studies, have analysed MMG signals and its crosstalk from the elbow flexors along the longitudinal, lateral and transverse directions to muscle fibres. Such an analysis which involves the elbow joint could be useful in clinical applications, like the development of prosthetic control[23]. Previous studies on crosstalk have analysed the MMG signals from forearm[10-12] and leg muscles[24]. The proximity, number and size of muscles contribute to the resulting crosstalk[24]. For example, the leg can contain a smaller number of large muscles than the forearm, and thus, the fraction of crosstalk among forearm muscles is greater than that reported for leg muscles. To this effect, the accessible location and specific architecture of the BB[25] makes the arm a preferred location for the quantification and analysis of crosstalk in MMG signals[26].

The objective of this study was to analyse the crosstalk in MMG signals along the longitudinal, lateral and transverse axes of elbow flexor muscles during sustained isometric forearm flexion, pronation and supination exercises. Specifically, we tested the hypothesis for the existence of a particular axis of fibres in elbow flexor muscles that produces minimal common signal. Three muscles, namely the BB, brachialis (BRA) and brachioradialis (BRD), were selected due to their intermediate size and inter-muscular distance because crosstalk has been found to be a function of the distance between neighbouring muscles[6]. Three exercises, namely, sustained isometric forearm flexion, pronation and supination at an elbow joint angle of 90°, were performed for the identification and quantification

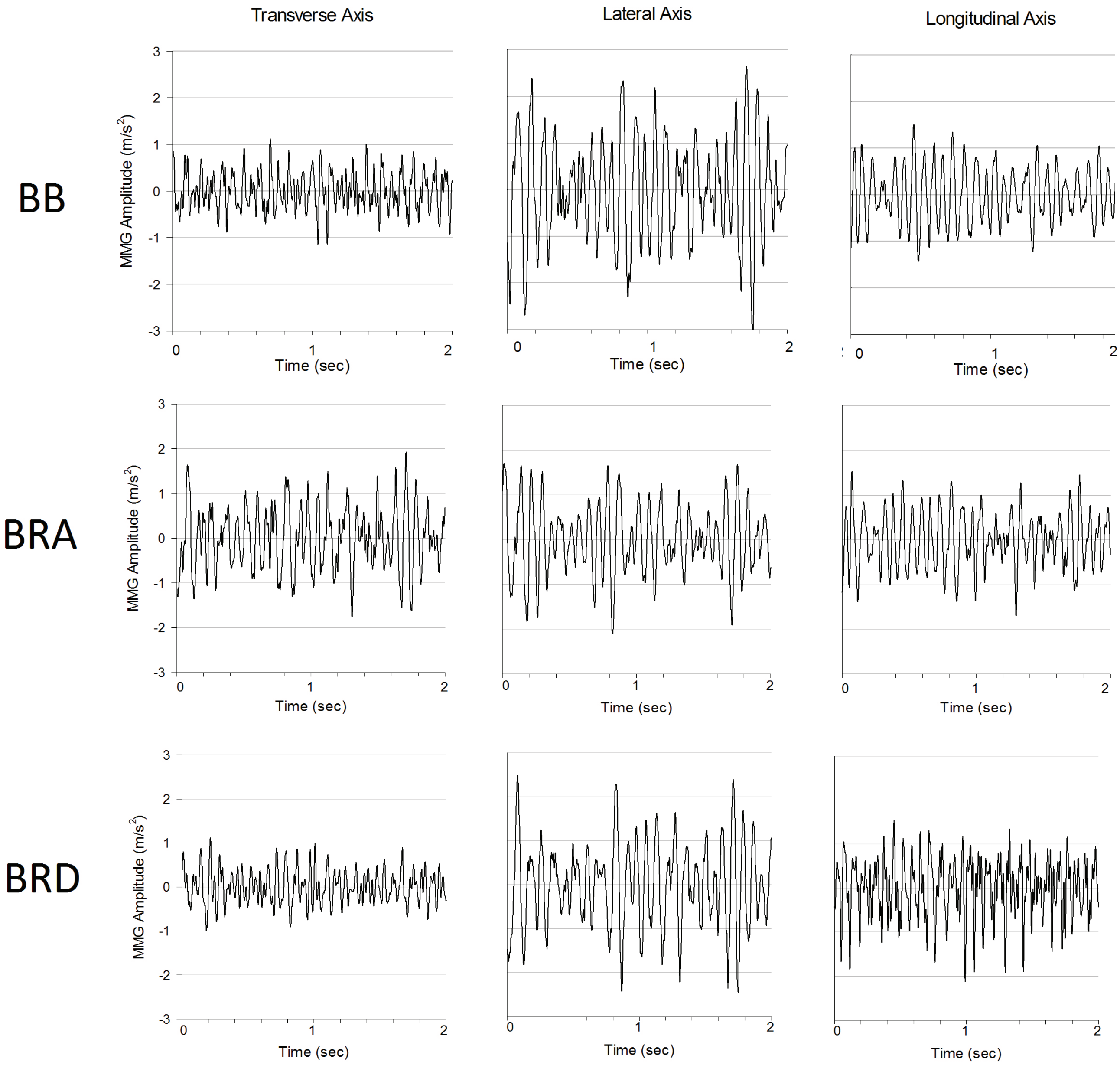


**Figure 3(a).** MMG signals from the BB, BRA and BRD muscles of a subject during the FLEXION excercise. These signals were used for the quantification of crosstalk in the transverse, lateral and longitudinal axes of muscle fibres.

of crosstalk from BB, BRD and BRA muscles. Isometric muscle actions which ensures no change in muscle length during activation were deemed suitable for the analysis of crosstalk in MMG signals as the % of common signal might be affected by the variation in muscle length[6]. Each of the three considered muscles has its own distinct biomechanical function during sustained isometric exercises. The BRA inserts into the ulna and is therefore exclusively responsible for elbow flexion, whereas the BB inserts into the radius and therefore participates in elbow flexion and forearm supination[27]. In contrast, the BRD passes through the elbow joint and is responsible for both elbow joint flexion and forearm pronation[28]. Crosstalk in MMG signals from triaxial accelerometers, that were anatomically aligned to the longitudinal, lateral and transverse axes of elbow flexor muscles, was quantified in all nine possible pairs of muscle fibre axes in each of the three tested muscle pairs. To avoid the occurrence of physiological tremors at maximal effort[6], MMG signals at 80% maximum voluntary contraction (MVC) were considered for the analyses performed in this study.

## Methods

### *Subjects*

Twenty-five young, healthy and untrained male subjects [(age=25.52(4.54) years, weight=68.21(9.65) kg, height=167.52(3.17) cm; all data are shown as the mean(SD)] with no history of neuromuscular injury participated in the experiment. Written informed consent was obtained from all the subjects. This study was approved by the local Medical Research & Ethics Committee (MREC), Ministry of Health, Malaysia and was performed according to the guidelines established by the Declaration of Helsinki.

### *Experimental procedure*

The experiment was conducted in two separate sessions with a gap of at least twelve hours between the two sessions for each participant. In the first session, the subject became familiarized with the experimental protocol and signed the consent form. Personal and anthropometric details of the subject were then noted. The subject was asked to perform a set of warmup exercises, which included arm stretching, elbow joint flexion and five to six repetitions of biceps curls with low weights. At the end of the warmup session, the subject was ready for MVC determination. MVCs were then determined for the sustained isometric forearm flexion, supination and pronation exercises through three trials with a 2 minutes rest period between any two consecutive trials and a 10 minutes rest period between any two consecutive exercises. During each exercise, the subject sat comfortably on a chair with a straight back, shoulders slightly abducted and 90° angle between arm and forearm. The angle between the subject's arm and forearm was measured using a digital goniometer and the subject's wrist flexion was restricted using a wrist-hand orthosis. The subject's MVC for the flexion exercise was determined as the maximum weight (dumbbell) during a sustained isometric hold for 3-4 seconds with the forearm in a supinated position (Figure 1). Subsequently, for the pronation exercise with postural settings maintained (Figure 2), the weights were replaced by a shovel handle of a wrist dynamometer (Baseline™ Evaluation Instruments, Fabrication Enterprises Inc., NY, USA) that was held with the wrist in neutral position. MVC for this exercise was considered as the maximum effort the subject could produce for 3-4 seconds during forearm pronation. Similarly, for the supination exercise using the same dynamometer, MVC was considered as the maximum effort the subject could produce for 3-4 seconds during forearm supination. In all the trials, proper posture was ensured by a dedicated observer who measured any off-axis variations periodically using a digital goniometer. Large variations (>±5%) in a trial were not allowed and considered unsuccessful. In addition, strong verbal encouragement was given to the subject to exert maximum effort with MVC being determined as the maximum from three successful trials.

In the second session of the experiment, sensors were placed over the skin by a physician who was present on site. The subject's skin was appropriately cleaned and shaved prior to placement. It was assumed that the relative movement between the sensor and skin beneath is negligible. The subject followed the same postural settings as adopted during the determination of MVC. The subject was then asked to perform sustained isometric forearm flexion, pronation and supination exercises at 80% MVC for 6 seconds with a rest period of 10 minutes between exercises. For all the exercises, proper posture, off-axis precautionary measures, maximum ±5% variations, announcement of time elapsed and verbal encouragement were observed. The generated signals were recorded in a personal computer for further analysis.

### *Recording of MMG signals*

The MMG signals were collected using three triaxial accelerometers (ADXL335, Analogue Devices, USA; full-scale range =±3 g; typical frequency response=0.5-500 Hz; senstivity=330 mVg$^{-1}$; size=15 mm × 15 mm × 1.5 mm; weight <1.5 grams). The accelerometers were attached to the skin surface over the muscle bellies with 3M Micropore and double-sided adhesive tape while the subject maintained a neutral arm position. The anatomical position of each muscle belly was determined according to a previous study[29]: BB – into the bulk of the muscle in the middle of the arm; BRA – two finger breadths proximal to the elbow crease along and just lateral to the tendon and bulk of the biceps; and BRD – midway between the biceps tendon and lateral epicondyle along the flexor crease. The x, y and z axes of each of the accelerometers were positioned along the estimated longitudinal, lateral and transverse directions of the muscle fibres, respectively. All three muscles considered in the present study are parallel fascicle muscles[30], and thus, the muscle fibre direction for each of the three muscles was estimated by considering the straight line between the muscle origin and muscle insertion as the longitudinal axis. In contrast, the direction perpendicular to the longitudinal axis to the skin/accelerometer from the muscle was taken as the transverse axis, and the direction perpendicular to the longitudinal and transverse axes was considered the lateral axis. The origin and insertion of each muscle were detected according to a previous study[29]: BB, origin=supraglenoid tuberosity (long head) and coracoid process of scapula (short head), insertion=bicipital tuberosity of radius; BRA, origin=volar surface of the distal half of the humerus, insertion=tuberosity of the ulna and volar surface of the coronoid process; BRD, origin=supracondylar area of the lateral aspect of the humerus, insertion=lateral aspect of the radius, just above the styloid process. The output obtained at each sensor was connected to the data acquisition unit (NI cDAQ 9191 with the NI 9205 module, National Instruments, Austin, TX, USA), which was connected to a personal computer over Wi-Fi. The sampling rate used was 1 kHz, which is largely higher than the minimum required to capture MMG signals that have been reported to be between 5-100 Hz[31]. MMG signals were recorded during all the trials, and the signals were recorded for 6 seconds during each trial.

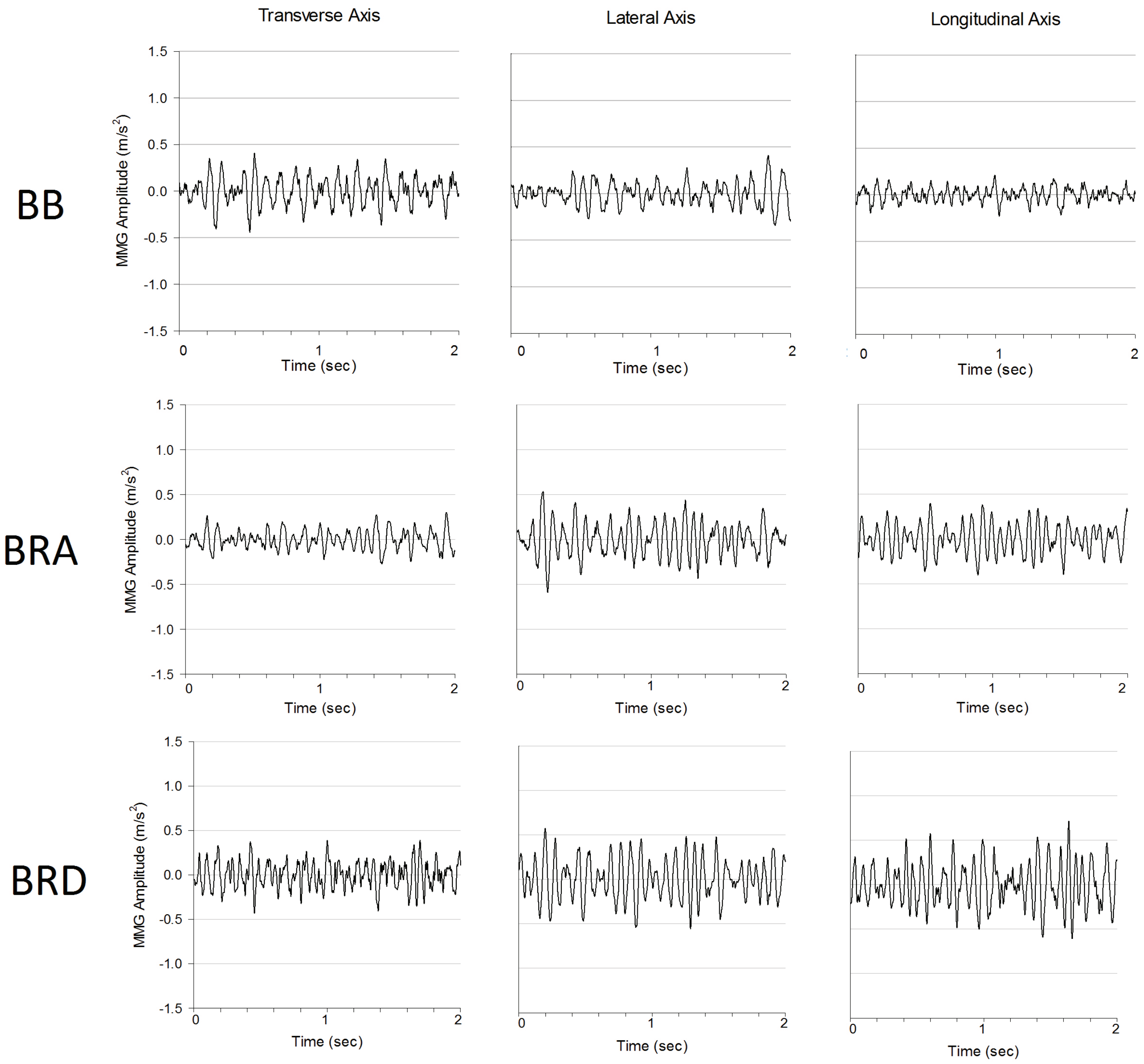


**Figure 3(b).** MMG signals from the BB, BRA and BRD muscles of a subject during the PRONATION excercise. These signals were used for the quantification of crosstalk in the transverse, lateral and longitudinal axes of muscle fibres.

Data acquisition and storage were performed using custom-written programmes in LabVIEW™ (version 2016, National Instruments, Austin, TX, USA).

### *Data analysis*

The data detected by the sensors along the x, y and z directions to the muscle fibres, which were stored on a personal computer, were digitally bandpass filtered (fourth-order Butterworth filter) at 5-100 Hz to obtain the MMG signals[32]. The MMG signals for a period of 2 seconds corresponding to the middle 33% of each 6 seconds isometric contraction were then extracted to remove the effect of the signal transition period, as recommended by a previous study[24]. The MMG RMS was determined by taking the absolute RMS of the signal from each muscle in the three directions of the accelerometer. Cross-correlation coefficients, which were calculated using equation (1), were determined to quantify crosstalk in the MMG signals from two associated muscles[33], and the coefficients were obtained for all nine possible pairs of the accelerometer axes.

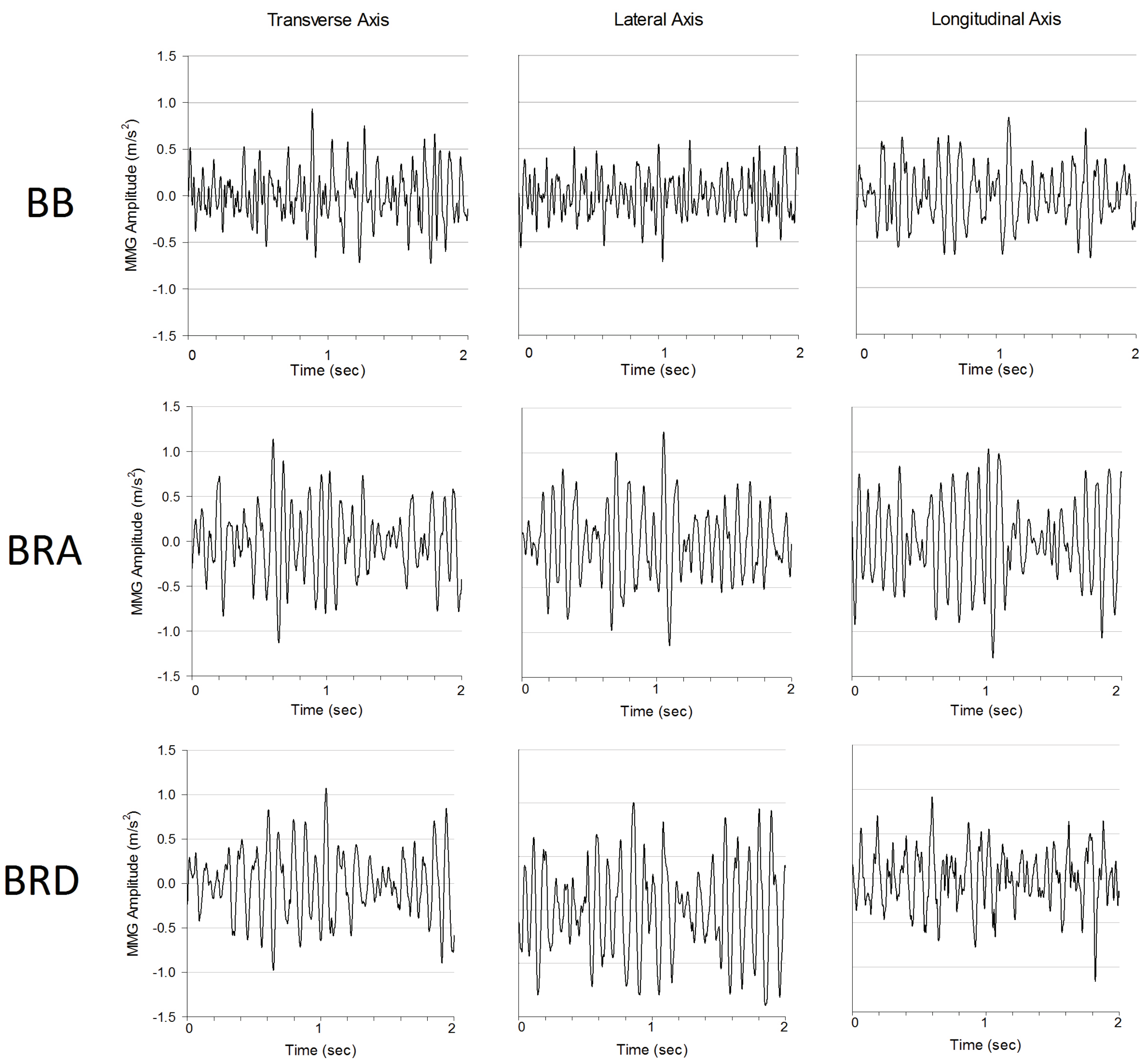


**Figure 3(c).** MMG signals from the BB, BRA and BRD muscles of a subject during the SUPINATION excercise. These signals were used for the quantification of crosstalk in the transverse, lateral and longitudinal axes of muscle fibres.

$$R_{p,q}(\tau) = \frac{1}{a \times b \times \omega(\tau)} \sum_{n=0}^{N-1} P_t(n) Q_t(n+\tau);\ 1-N<\tau<M \qquad (1)$$

where $P_t$ and $Q_t$ are the MMG signals from the investigated muscle pair, $a=\sqrt{\sum_{n=0}^{N-1} P_t^2(n)}$, $b=\sqrt{\sum_{n=0}^{M-1} Q_t^2(n)}$, $N$ and $M$ are the lengths of $P_t$ and $Q_t$, $\tau$ represents the time lag between the signals taken from 1-$N$ to $M$ and $\omega$ is the weighting factor given by equation (2).

$$\omega(\tau) = \begin{cases} \dfrac{\max(M,N)+\tau}{\max(M,N)}, & -N<\tau<0 \\ 1, & \tau=0 \\ \dfrac{\max(M,N)-\tau}{\max(M,N)}, & 0<\tau<M \end{cases} \qquad (2)$$

The peak cross correlation coefficient, $R_{p,q}$ was squared to quantify common variance (% crosstalk) using equation (3) in the three muscle pairs, namely, BB & BRA (MP1), BRA & BRD (MP2) and BB & BRD (MP3), with BB, BRA and BRD designated as muscle 1, 2 and 3 respectively.

$$Crosstalk(\%) = [\max(R_{p,q})]^2 \times 100\% \qquad (3)$$

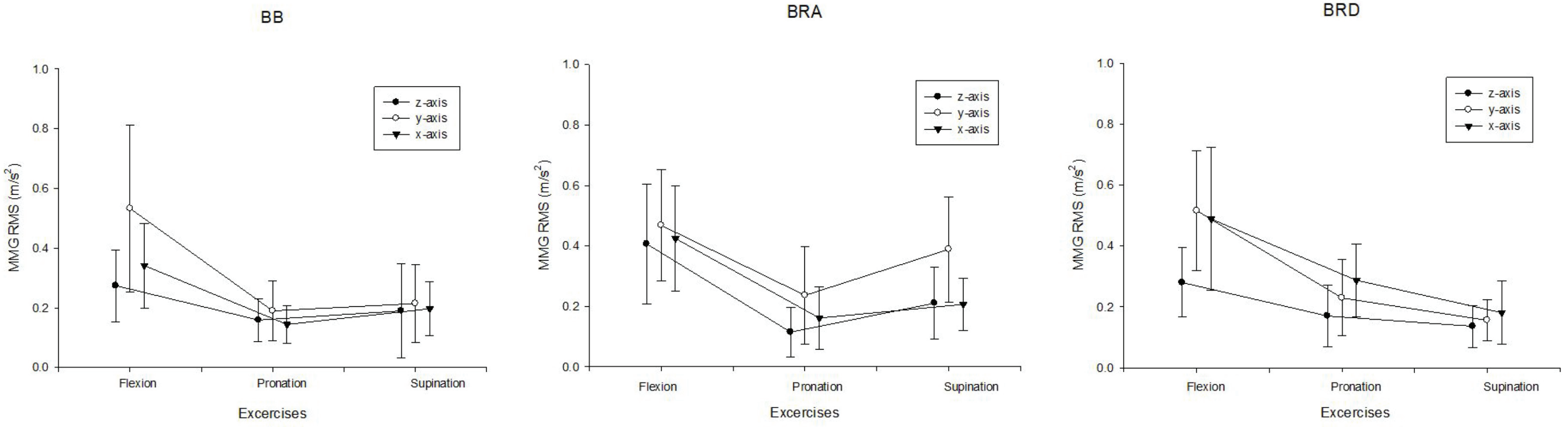


**Figure 4.** Mean MMG RMS values plotted for the three exercises in BB, BRA and BRD muscles where z, y and x-axes represent the transverse, lateral and longitudinal axes to muscle fibres.

*Statistical analysis*

The statistical analyses were performed using SPSS (version 20, IBM SPSS Statistics, NY, USA). Results [mean(SD)] of the Shapiro-Wilks's test indicated that MMG RMS [$p<0.05$, skewness=0.96(0.46)-3.60(0.46), kurtosis=0.08(0.90)-15.67(0.90)] and crosstalk [$p<0.05$, skewness=0.87(0.46)-2.67(0.46), kurtosis=1.52(0.90)-7.08(0.90)] were found to be not normally distributed, and thus non-parametric statistical tests were used for further analysis of the data. The Kruskal-Wallis test was used to statistically analyse the MMG RMS and crosstalk values. All subsequent post hoc analysis was performed using the Mann-Whitney test. A significance level of $\alpha=0.05$ (95% confidence interval) was used, hence $p<0.05$ was considered statistically significant. The effect size ($\eta^2$) used in this study was according to Cohen's interpretation[34]:<0.2 (small), >0.2-0.5 (medium) and >0.5 (high).

## Results

Figures 3(a), 3(b) and 3(c) shows the MMG signals used for quantification of crosstalk, which were obtained from the BB, BRA and BRD muscles of a subject along the longitudinal, lateral and transverse directions of the muscle fibres during sustained isometric forearm flexion, pronation and supination exercises. The mean MMG RMS values along the three muscle fibre axes during the sustained isometric forearm flexion, pronation and supination exercises were plotted (Figure 4). For each of the three muscles in each of the three exercises, MMG RMS was found to be statistically significant among the fibre axes ($p<0.05$, $\eta^2=0.17-0.34$) except for BB and BRD in supination, and BRA in flexion. Similarly, for each of the three fibre axes in each of the three muscles, MMG RMS was found to be statistically significant among the exercises ($p<0.05$, $\eta^2=0.12-0.42$). Likewise, for each of the three fibre axes in each of the three exercises, MMG RMS was found to be statistically significant among the muscles ($p<0.05$, $\eta^2=0.11-0.26$). For each of the three muscles per exercise, the transverse axis showed the lowest MMG RMS mean(SD) values [0.11(0.08)-0.40(0.19)] against the longitudinal [0.14(0.06)-0.48(0.23)] and lateral [0.15(0.06)-0.53(0.27)] axes. Furthermore, for each of the three muscles, the MMG RMS mean(SD) values obtained during sustained isometric forearm flexion [0.27(0.11)-0.53(0.27)] were higher than those obtained during pronation [0.11(0.08)-0.28(0.11)] and supination [0.13(0.06)-0.38(0.17)] exercises. The analysis of MMG RMS mean(SD) values of the individual muscles found that BB [0.14(0.06)-0.53(0.27)] yielded higher values than BRA [0.11(0.08)-0.46(0.18)] and BRD [0.13(0.06)-0.51(0.19)].

In all the cross-correlation analyses, most of the peak coefficients were observed at time lag ($\tau$) of approximately 0 second (Figure 5). The peak cross-correlation

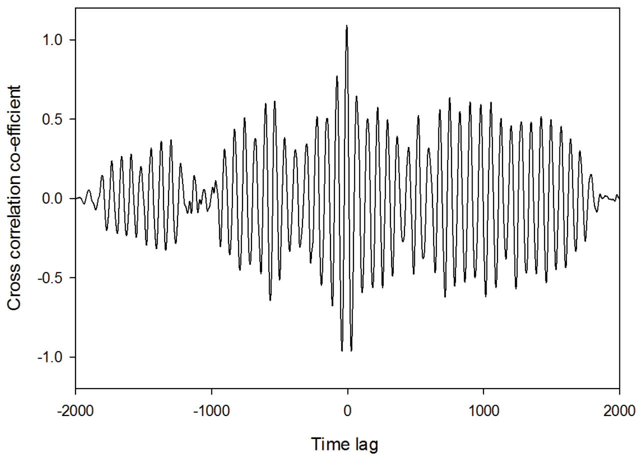


**Figure 5.** Correlogram of MMG signals recorded from longitudinal axes of MP1 (BB & BRA) during sustained isometric forearm flexion task performed by a subject. The figure is plotted for entire range of time lags and the peak correlation is nearly at 0-time lag.

coefficient values obtained at τ=0 from equation (1) ranged from 0.14-0.78 (mean crosstalk ranged from 2.16-61.42%). Specifically, the mean crosstalk values during the sustained isometric forearm flexion, pronation and supination exercises ranged from 2.74-61.42%, 4.48-42.90% and 2.16-59.03%, respectively. The range of mean crosstalk values obtained for muscle pairs MP1, MP2 and MP3 were the following: 4.81-52.17%, 4.01-61.42% and 2.74-51.85% (flexion), 4.93-48.38%, 5.06-42.90% and 4.48-23.86% (pronation) and 6.09-28.93%, 8.44-59.03% and 2.16-21.97% (supination). Crosstalk values showed statistical significance among all nine axes pairs in the three muscle pairs during sustained isometric forearm flexion, pronation and supination exercises with mostly medium effect sizes ($p<0.05$, $\eta^2=0.16-0.51$) except for MP1 during the pronation exercise (Table 1). Figure 6 shows the bar chart representation of the mean crosstalk values obtained for all nine axes pairs in the three muscle pairs during the sustained isometric forearm flexion, pronation and supination exercises. The transverse axes pair recorded the MMG signals with the lowest mean crosstalk values (2.16-9.14%) in all exercises and muscle pairs. The post hoc analysis of the crosstalk values revealed 58% of the comparisons between the transverse axes and remaining eight axes pairs, from all exercises and muscle pairs, were statistically significant ($p<0.05$, $\eta^2=0.11-0.71$), as shown in Table 2.

## Discussion

The study aimed to analyse the crosstalk in the MMG signals along the longitudinal, lateral and transverse axes of elbow flexor muscles. Specifically, the study investigated the hypothesis that the level of crosstalk in MMG signals would show differences among all possible pairs of fibre axes. In addition, the potential existence of an axis of muscle fibres that would yield the lowest level of crosstalk in MMG signals during the sustained isometric forearm flexion, pronation and supination exercises was also investigated. To test these hypotheses, MMG signals were recorded using triaxial accelerometers that were anatomically aligned to the longitudinal, lateral and transverse axes of the elbow flexor muscles, and these signals were employed to quantify the MMG RMS values for the three axes and the degree of crosstalk in the nine axes pairs.

The range of peak cross-correlation coefficient values (0.14-0.78) observed in the present study is comparable to the common variances (0.37-0.51) reported in [35], whereas the authors measured lateral oscillations from the three superficial quadriceps femoris muscles using a uniaxial MMG signal during concentric and eccentric isokinetic muscle actions. The crosstalk values obtained among all nine axes pairs of the three muscle pairs were statistically significant ($p<0.05$) during the sustained isometric forearm flexion, pronation and supination exercises, except for MP1 during pronation, as shown in Table 1. This observation is very similar to that obtained in a previous study[12], which found that crosstalk in MMG signals along the three axes of the muscle fibres of forearm muscles showed statistical significance ($p<0.05$) during four different isometric wrist postures.

The transverse axes pair exhibited the lowest mean crosstalk values ($R_{p,q}$=0.14-0.30, crosstalk=2.16-9.14%) among all nine axes pairs in almost all the exercises and muscle pairs investigated in the current study. This observation concurs with the results obtained in a previous study[12], which revealed that the transverse axes pair yielded the lowest degree of crosstalk in MMG signals (11.38-25.55%) recorded from forearm muscles during isometric wrist postures. However, these researchers only considered three accelerometer aligned axes pairs (xx, yy & zz), whereas

**Table 1.** Statistical significance of the crosstalk values obtained for the nine axes pairs of the three muscle pairs during the flexion, pronation and supination exercises.

| **Muscle Pair** | **MP1** | | | **MP2** | | | **MP3** | | |
|---|---|---|---|---|---|---|---|---|---|
| **Exercise** | **Flexion** | **Pronation** | **Supination** | **Flexion** | **Pronation** | **Supination** | **Flexion** | **Pronation** | **Supination** |
| $\eta^2$ | 0.48 | 0.18 | 0.20 | 0.51 | 0.30 | 0.36 | 0.46 | 0.16 | 0.23 |
| ***p-value*** | **<0.05** | 0.10 | **<0.05** | **<0.05** | **<0.05** | **<0.05** | **<0.05** | **<0.05** | **<0.05** |

*Bold font indicates the statistical significance ($\eta^2$: effect size). MP1: BB & BRA, MP2: BRA & BRD and MP3: BRD & BB.*

**Table 2.** Post hoc analysis of crosstalk values between transverse and other axes pairs.

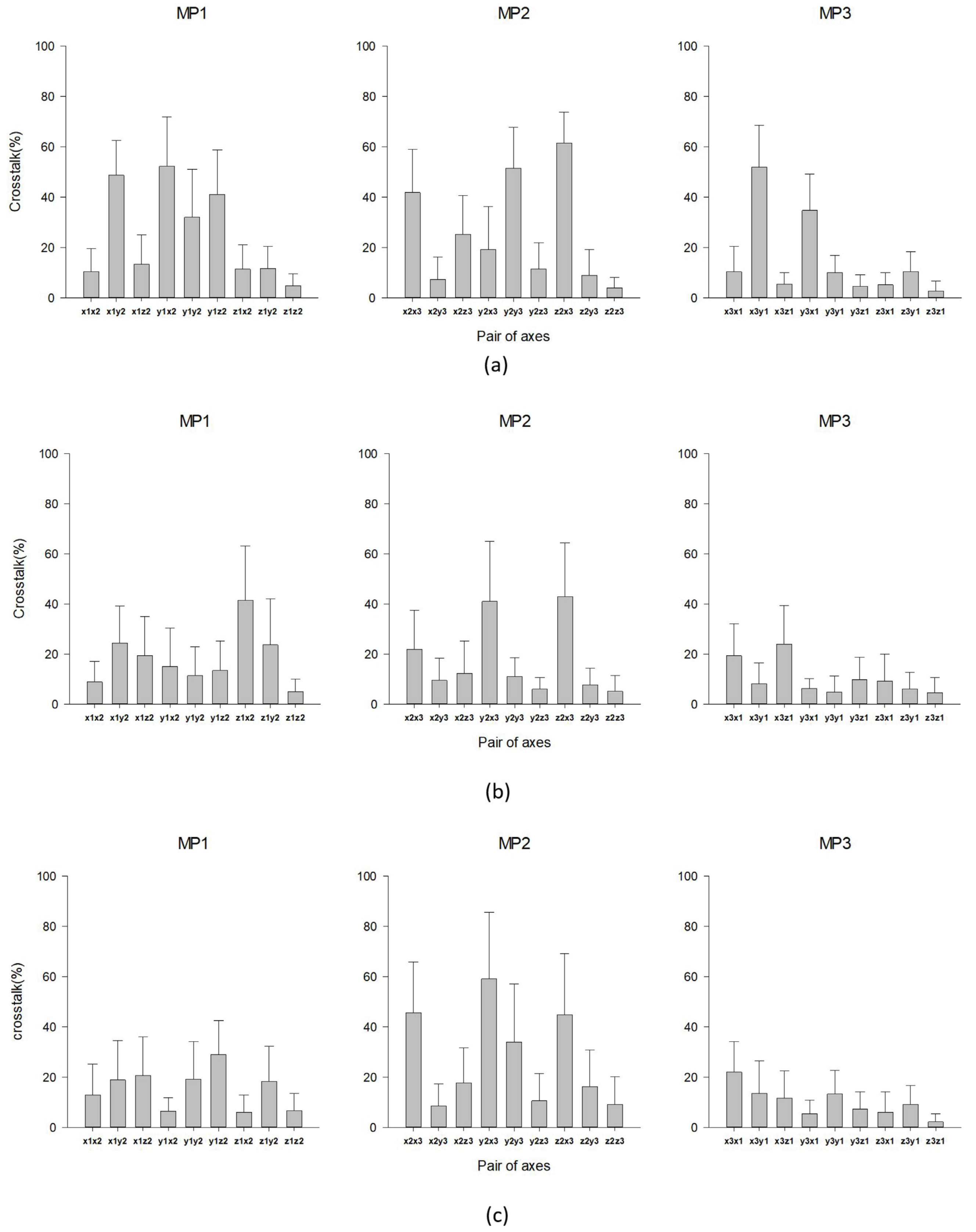
| MP1 | | | | MP2 | | | | MP3 | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Axes Pair** | **Flexion** | **Pronation** | **Supination** | **Axes Pair** | **Flexion** | **Pronation** | **Supination** | **Axes Pair** | **Flexion** | **Pronation** | **Supination** |
| x1x2 & z1z2 | **<0.05** | 0.10 | 0.20 | x2x3 & z2z3 | **<0.05** | **<0.05** | **<0.05** | x3x1 & z3z1 | **<0.05** | **<0.05** | **<0.05** |
| x1y2 & z1z2 | **<0.05** | **<0.05** | 0.20 | x2y3 & z2z3 | **<0.05** | 0.80 | 0.50 | x3y1 & z3z1 | **<0.05** | 0.80 | **<0.05** |
| x1z2 & z1z2 | **<0.05** | **<0.05** | **<0.05** | x2z3 & z2z3 | **<0.05** | 0.60 | **<0.05** | x3z1 & z3z1 | 0.90 | 0.60 | **<0.05** |
| y1x2 & z1z2 | **<0.05** | 0.40 | **<0.05** | y2x3 & z2z3 | **<0.05** | **<0.05** | **<0.05** | y3x1 & z3z1 | **<0.05** | 0.80 | 0.50 |
| y1y2 & z1z2 | **<0.05** | 0.70 | 0.20 | y2y3 & z2z3 | **<0.05** | 0.20 | **<0.05** | y3y1 & z3z1 | **<0.05** | 0.40 | **<0.05** |
| y1z2 & z1z2 | **<0.05** | 0.40 | 0.10 | y2z3 & z2z3 | **<0.05** | 0.50 | **<0.05** | y3z1 & z3z1 | 0.60 | 0.50 | 0.70 |
| z1x2 & z1z2 | **<0.05** | **<0.05** | 0.10 | z2x3 & z2z3 | **<0.05** | **<0.05** | **<0.05** | z3x1 & z3z1 | 0.20 | 0.10 | **<0.05** |
| z1y1 & z1z2 | **<0.05** | **<0.05** | 0.30 | z2y3 & z2z3 | **<0.05** | 0.20 | **<0.05** | z3y1 & z3z1 | **<0.05** | 0.50 | 0.20 |

*Bold font indicates the statistical significance. MP1: BB & BRA, MP2: BRA & BRD and MP3: BRD & BB. (x1, x2, x3: longitudinal axis of BB, BRA, BRD; y1, y2, y3: lateral axis of BB, BRA and BRD; z1, z2, z3: transverse axis of BB, BRA and BRD).*

the present study included all nine possible (six additional) non-aligned axes pairs for the quantification of crosstalk. Thus, our findings provide further evidence to support the hypothesis on the existence of an axis pair (transverse axes) which generate unique signals (lowest crosstalk). Incidentally, we also observe the transverse and lateral axes to exhibit the lowest and highest MMG RMS values respectively, among the three muscle fibre axes, as shown in Figure 4. This observation on the lateral axis concurs with the results of [16], in which the authors observed higher MMG RMS values along the lateral axis as compared to the longitudinal axis, recorded using a biaxial accelerometer from the BB muscle during submaximal to maximal sustained isometric muscle actions. Taken together, these findings suggest that the lateral axis of muscle fibre appears to be the preferred mode of direction of MMG signals and the levels of crosstalk increases with levels of MMG signals[6].

The overall crosstalk values in MMG signals from the elbow flexor muscles ranged from 2.16-61.42% in the current study. This range of crosstalk is comparable to those obtained in two previous studies[10,24], which yielded crosstalk from the superficial quadriceps femoris muscles during isometric muscle actions and forearm muscles during isometric muscle actions with ranges of 1.53-50.97% and 1.69-64.05%, respectively. Muscle size, number of proximity muscles and inter-muscular distance contribute to crosstalk in MMG signals[24]. Given these parameters, arm muscles are projected to have mean crosstalk values between those observed for leg muscles and those found for forearm muscles. Our findings for the mean crosstalk value in the elbow flexors (2.16-61.42%) support this assumption, as arm muscles are medium sized, rather isolated and with an inter-muscular distance that is less than the quadriceps femoris but greater than the forearm muscles. In addition, muscle fat, tendons and tissues between muscles act as low-pass filters[14]. Thus, the filter architecture of synergistic arm muscles differs from that of agonist/antagonist forearm muscles. Crosstalk between the synergistic muscle pairs has been observed to be greater than the crosstalk in agonist/antagonist muscle pairs[6]. Nevertheless, the range of mean crosstalk values observed in the current study for synergistic arm muscles was lower than the range of mean crosstalk values obtained in a previous study[10] for agonist/antagonist forearm muscles during isometric wrist postures. This difference in findings might be due to the variation in the fascicle organization of muscle fibres and other issues related to the accessibility of the concerned muscles in these studies.

These findings on crosstalk in MMG signals along the three different axes of elbow flexor muscles revealed significant observations that could be useful in many areas related to the application of MMG as a reliable technique for muscle function assessment, such as the development of standards for myographic signal acquisition[31], control techniques of arm prosthetics[23], rehabilitation monitoring systems[36] and for the assessment of limb functions in the fields of sports and athletics[37]. The present study nevertheless could benefit from a few potential limitations in this work, which could improve the robustness of the methodology used. The use of ultrasound imaging of the concerned muscles during exercises can provide a better estimation of minor changes in the orientation



(a)

(b)

(c)

**Figure 6.** Crosstalk in nine axes pairs (x1, x2, x3: longitudinal axis of BB, BRA, BRD; y1, y2, y3: lateral axis of BB, BRA and BRD; z1, z2, z3: transverse axis of BB, BRA and BRD) of the three muscle pairs (MP1: BB & BRA, MP2: BRA & BRD and MP3: BRD & BB) during the (a) flexion, (b) pronation and (c) supination exercises.

of muscle fibres due to changes in the pennation angle, specifically during the pronation and supination exercises, while maintaining various elbow joint angles. Additionally, the identification and effect of variations in the filter between muscles, which comprises non-muscular structures, on crosstalk could be better understood using ultrasound imaging. Similarly, a study on the effect of variation in anthropometric variables such as skin thickness and inter-muscular distance at various levels of muscle effort on crosstalk levels in MMG signals might provide additional insights.

## Conclusion

MMG signals recorded from the BB, BRA and BRD muscles reflected the biomechanical functions of these muscles during sustained isometric forearm flexion, pronation and supination exercises. MMG signals in the direction lateral and transverse to the muscle fibre axis produced the highest and lowest MMG RMS values. The lowest crosstalk values (common signal) in the recorded MMG signals were observed to be in the direction transverse to the muscle fibre axis and they were statistically significant among all nine possible pairs of fibre axes with mostly medium effect sizes. Furthermore, the post hoc analysis revealed that 58% of the comparisons between the transverse axes and remaining eight axes pairs were statistically significant. The range of the mean crosstalk values observed for the elbow flexors (synergistic muscle pairs) was relatively lower than that observed for agonist/antagonist muscle pairs in the literature. Since MMG still remains at an early stage of development, our findings provide additional supporting evidence on the use of MMG as an alternative and potential tool for muscle function assessment and the development of related applications.

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*Author's contribution*

*I.T and K.S conceived, designed and performed the experiments and analysed the data. All authors contributed reagents/materials/analysis tools and wrote the paper. I.T takes responsibility of integrity of data.*

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