



Postural cueing increases multifidus activation during stabilization exercise in participants with chronic and recurrent low back pain: An electromyographic study

Brooke Winder*, Paul A. Keri, David E. Weberg, George J. Beneck

California State University, Long Beach, Department of Physical Therapy, 1250 N. Bellflower Blvd., Long Beach, CA 90840-7201, United States

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ABSTRACT

Persons with low back pain (LBP) have demonstrated altered morphology and function of the deep multifidus (DM). This study examined the effects of postural cueing for increased lumbar lordosis on DM and longissimus thoracis (LT) activation during lumbar stabilization exercises (LSE) performed by persons with LBP. Nine adults with a history of chronic or recurrent LBP were recruited. Fine-wire EMG data was collected while participants performed 10 LSE's in neutral posture and with postural cueing. Percent maximum voluntary isometric contraction of L5 DM and T12 LT, and ratios of activation (DM/LT) were analyzed. There was a significant main effect for posture on DM activation ($p < 0.001$), indicating greater activation levels during exercises performed with postural cueing vs. neutral posture. LT activation did not increase significantly with postural cueing. Following a significant 1-way repeated measures ANOVA ($p = 0.034$) for the postural cueing condition, pairwise comparisons demonstrated significantly higher DM/LT activation ratios for prone leg lift, variable-angle Roman chair at 15°, bridging, and bilateral arm and leg lift. These results suggest postural cueing can be used across a range of LSE intensities to increase DM activation without a significant increase in LT activation in patients with chronic or recurrent LBP.

1. Introduction

Low back pain is one of the most common health complaints among the general public, and is a leading cause of activity limitation and work absence throughout the world (Hoy et al., 2010). Following lumbar spine injury, spinal stability can be compromised due to tissue damage to passive structures and alterations in proprioception and muscle activation (Brumagne et al., 2000; McGill et al., 2003; Panjabi, 2006). Exercises used to improve coordination, endurance and strength are recommended treatment interventions for patients with subacute and chronic low back pain who demonstrate movement impairments (Delitto et al., 2012).

Coordinated muscle function plays a large role in lumbar spine stabilization (McGill et al., 2003; Panjabi, 1992). The lumbar spine extensors are an integral part of this stabilizing system and can be classified into global muscles (those that transfer loads directly from thorax to the pelvis, such as the longissimus thoracis) or local muscles (those that attach directly to the vertebrae, provide segmental stability and control intervertebral motion, such as the multifidus) (Bergmark, 1989). The multifidus muscle is considered particularly important for

lumbar spine stability, due to its large physiologic cross sectional area (Ward et al., 2009), ability to control motion and increase intervertebral stiffness (Wilke et al., 1995), and its optimally designed fiber length (Ward et al., 2009). In subjects with acute, subacute, recurrent and chronic low back pain, lumbar multifidus function is altered and its cross sectional area reduced (Beneck and Kulig, 2012; Fortin and Macedo, 2013; Hides et al., 2008; MacDonald et al., 2009; Sánchez-Zuriaga et al., 2015; Smith and Kulig, 2016; Wallwork et al., 2009). Multifidus atrophy is most significant at the symptomatic region of pain or pathology (Beneck and Kulig, 2012; Hides et al., 2008; MacDonald et al., 2009; Wallwork et al., 2009), and most prominent near L5 (Fortin and Macedo, 2013). The deep fibers of the multifidus in particular demonstrate reduced magnitude of activation, altered timing and fatigability (Beneck et al., 2013; Macdonald et al., 2010; MacDonald et al., 2009; Smith and Kulig, 2016). An improved understanding of which stabilization exercises largely activate the deep multifidus in persons with low back pain could help clinicians optimally address this muscle deficit.

Stabilization programs aimed at addressing motor control deficits in patients with low back pain initially use low-load exercises (Kennedy

* Corresponding author.

E-mail address: Brooke.Winder@csulb.edu (B. Winder).

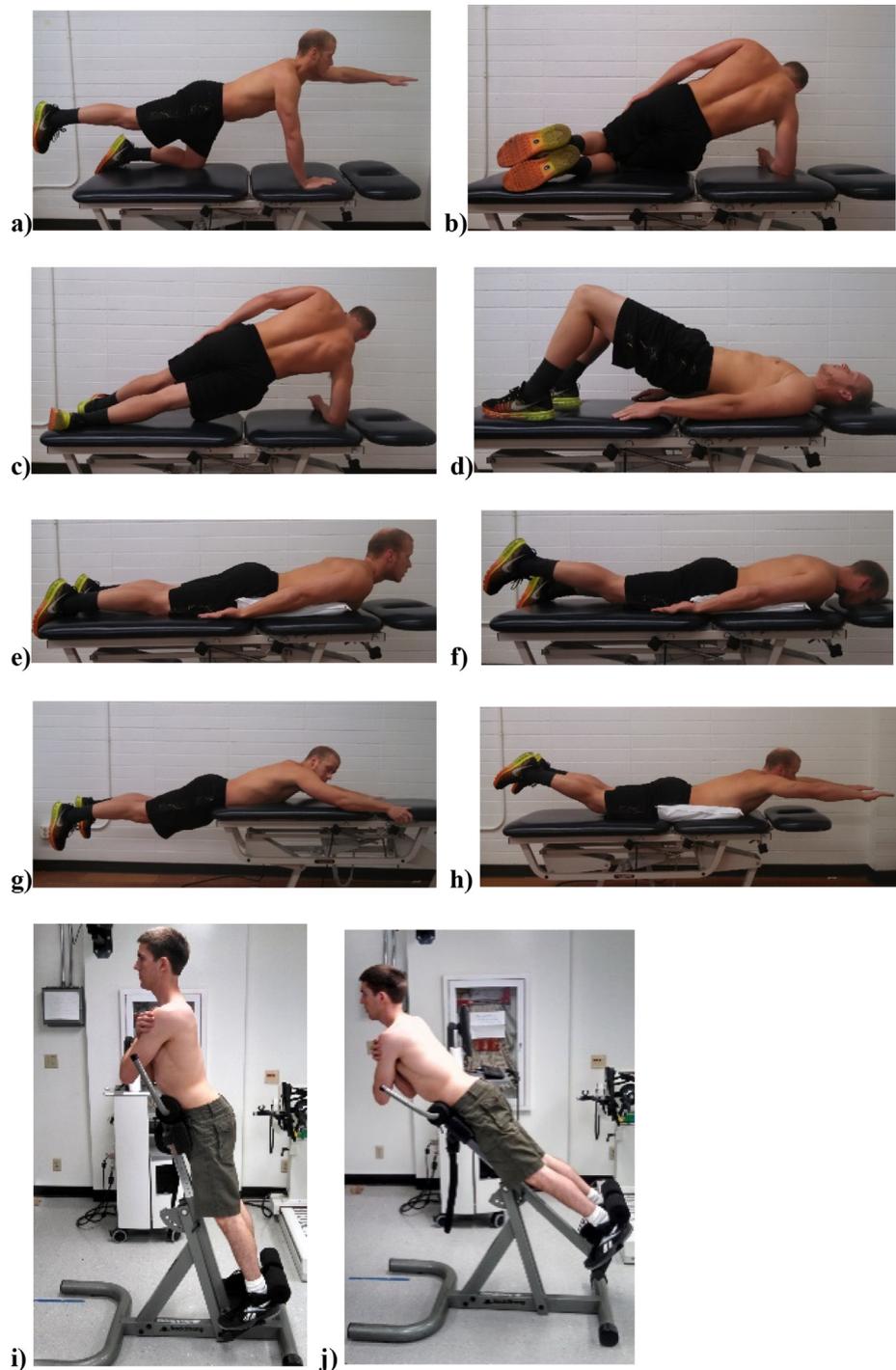


Fig. 1. (a) Quadruped leg lift with opposite arm raise, (b) Sidebridge on knees, (c) Sidebridge on feet, (d) Bridging, (e) Prone trunk lift, (f) Prone leg lift, (g) Bilateral leg lift, (h) Bilateral arm and leg lift, (i) Variable angle Roman chair at 15°, (j) Variable angle Roman chair at 45°.

and Levesque, 2015; Kim et al., 2015; Richardson et al., 1999) to train coordinated activity of the local muscles while reducing over-activation of global muscles (Barr et al., 2007; Costa et al., 2009; Kennedy and Levesque, 2015; Richardson and Jull, 1995). Low loading is often recommended due to concern for deficits in the load-bearing capabilities of patients with low back injury (McGill and Karpowicz, 2009; Richardson et al., 1999) as well as the possibility of inducing pain, muscle inhibition responses, or promoting imbalanced muscle recruitment (Hibbs et al., 2008; Richardson et al., 1999; Russo et al., 2018). However, a progression to increased loads may be needed to prepare patients for higher-level functional activities. Though training at both

low and high loads to muscle fatigue seem to equivalently induce muscle hypertrophy (Schoenfeld et al., 2017), higher-load training may present additional advantages: developing optimal neural adaptations, achieving greater strength gains and requiring less exercise volume to produce effects (Jenkins et al., 2017; Schoenfeld et al., 2017). When progressing to higher load exercises, concern remains for monitoring proper muscle activation patterns (Richardson et al., 1999). Though presentations vary, some patients with low back pain may demonstrate a tendency toward global muscle over-activation (Alexiev, 1994; Arena et al., 1989; Arendt-Nielsen et al., 1995; Kaigle et al., 1998; van der Hulst et al., 2010). Understanding the ratio of local/global extensor

muscle activation produced by patients with low back pain during lumbar stabilization progressions can further assist clinicians in choosing optimal exercises for patients with both multifidus atrophy and a tendency toward global-muscle dominance.

Beneck et al used fine-wire electromyography to assess the activation of the lumbar multifidus and longissimus thoracis during 6 lumbar stabilization exercises performed in neutral posture and with postural cueing to increase lumbar lordosis. The results indicated that postural cueing to increase lumbar lordosis increased multifidus activation during the exercises without increasing longissimus thoracis activation (Beneck et al., 2016). It is not clear if persons with a history of low back pain will demonstrate such activation patterns using postural cueing. Ratios between the multifidus and the erector spinae have been assessed for certain small groups of lumbar stabilization exercises, but mostly in persons without a history of low back pain or using a small number of stabilization exercises (Kim et al., 2013; Yoon et al., 2015).

Therefore, the purpose of this study was to examine the effects of postural cueing to increase lumbar lordosis during the performance of trunk stabilization exercises in persons with chronic and recurrent low back pain. The first objective was to observe whether postural cueing could increase activation of the lumbar multifidus without a significant increase in longissimus thoracis activation. Contingent on the first objective, the second objective was to examine the ratio of lumbar multifidus activation to longissimus thoracis activation across the 10 exercises performed with and without postural cueing.

2. Materials and methods

2.1. Participants

Nine adults with a history of low back pain (6 males, 3 females, mean \pm SD age, 31.4 \pm 11.6 years) were recruited from a large public university campus community. To determine eligibility for the study, participants were screened with a medical health history questionnaire. Those included in the study reported a history of chronic or recurrent low back pain between L3 and the upper buttocks. Chronic low back pain was defined as back pain present on at least half of the days in a 6-month period in single or multiple episodes (Korff, 2011). Recurrent low back pain was defined as back pain present on less than half of the days in a 12-month period, occurring in multiple episodes over the year (Korff, 1994). All subjects reported an episode of back pain occurring within the most recent 6 months prior to the study. Participants were excluded if they had a history of neuromuscular pathology, prior lower back surgery, spinal malignancy, spinal infection, demonstrated clinical signs of radiculopathy upon objective testing (e.g. positive straight leg raise test, weakness, diminished reflexes, loss of sensation), or were currently pregnant. Written consent was obtained from each participant. The California State University, Long Beach Institutional Review Board reviewed and approved the study protocol.

2.2. Training

On days prior to the data collection, participants received 1–5 instructional sessions in execution of the 10 exercises to be tested with and without postural cueing (sidebridge on knees, sidebridge on feet, prone leg lift, variable-angle Roman chair at 15°, bridging, prone trunk lift, quadruped leg lift with opposite arm raise, variable-angle Roman chair at 45°, bilateral leg lift, and bilateral arm and leg lift). (Fig. 1). The exercises were initially taught and practiced in a neutral lumbar spine posture before adding postural cueing of the lumbar spine to the exercise. During the training sessions, the investigators monitored and measured with a goniometer predefined joint angles for each exercise. Given the number of exercises and the higher level of difficulty of some, care was taken not to aggravate the participants' low back condition while exercising. While lying prone over a pillow, participants received postural cueing to increase the lower lumbar lordosis and increase

multifidus activation. (Beneck et al., 2016; Claus et al., 2009) An investigator gave the cue, "slightly push your tailbone up and over your head." When needed, additional cues were used to elicit the desired muscle recruitment pattern. Mimicking clinical practice, the investigator used palpation and observation to monitor the small anterior pelvic tilt intended to increase lower lumbar lordosis and lumbar multifidus activation. Participants were then instructed to perform the activation procedure during each of the 10 exercises. Easier exercises were performed first followed by those the subject found more challenging. During the exercises, additional tactile and verbal cues were given to prevent hip flexor activity, excessive lordosis and excessive erector spinae activity. Each training session lasted approximately 15 min and subjects were asked to practice daily at home. Once the investigator deemed the subject could correctly perform the exercises consistently with and without postural cueing, and without aggravation of low back pain, participants were scheduled for EMG (electromyographic) testing.

2.3. Instrumentation

Muscle activation during the exercises was recorded with intramuscular electrodes and a 10-channel MA300 EMG system.^(a) MA-416 discrete preamplifiers^(a) transmitted the electrical signals. The preamplifiers have a common mode rejection ratio greater than 100 dB at 65 Hz, gain at 1 kHz \times 20% \pm 1%, input impedance greater than 100M Ω , and a signal bandwidth of 15–3500 Hz. The raw EMG signal sampling rate was 2000 Hz.

Intramuscular electrode insertions were performed by a physical therapist certified in kinesiological EMG in the state of California. The electrodes were inserted bilaterally into the deep fibers of the multifidus at L5 and the longissimus thoracis at T12 using ultrasound guidance (Toshiba Nemio 30, Model: SSA-550A-20^(b)) as previously described (Moseley et al., 2002). The investigator confirmed electrode placement using stimulation of the muscles as previously described (Beneck et al., 2013). A reference electrode was taped over the area of the C7 spinous process.

2.4. Testing procedures

Participants then performed a maximum voluntary isometric contraction (MVIC) test by holding the original Sorenson test position (Demoulin et al., 2006), i.e. anterior–superior iliac spines placed at the edge of the table, lower extremities strapped to the table, arm folded across the chest, and the trunk held in a horizontal position. An investigator gradually applied a downward manual force to the upper back using the highest force that the subject could resist without causing spinal flexion. This MVIC test was practiced during the exercise training sessions to maximize performance on the day of testing. The MVIC test was used to normalize the EMG data for the muscles analyzed.

Following resting and MVIC EMG testing, participants performed each of the 10 exercises with a neutral posture and with increased lumbar lordosis. The order of the 10 exercises and 2 postural conditions was randomized. Body alignment was monitored by investigators during all exercises to maintain the homogeneity of the postures. Subjects were also monitored verbally throughout the testing process to ensure that their low back pain was not provoked. During the EMG recordings, each exercise was held statically for a minimum of 5 s. A rest period of two minutes was given between each exercise. No subjects reported aggravation of pain during the testing.

2.5. EMG analysis

The digital EMG signal for each muscle was imported into MATLAB R2013A software^(c) and filtered with a 10-Hz Butterworth high pass filter. A root-mean-square (RMS) calculation using a 75-millisecond

moving window was used to smooth maximum voluntary isometric contraction data to determine the highest 1-s average. The RMS of 5 s of each trial was calculated. Normalized values of the EMG signal were determined by dividing the RMS of the exercise trial by the RMS of the highest 1 s of an MVIC.

2.6. Statistical analysis

All statistical analyses were performed with SPSS Version 24.0^(d). Muscle activation (percent MVIC) of multifidus and longissimus thoracis was compared between sides using paired *t* tests for each of the 10 exercises in the neutral-posture trials. For exercises that demonstrated no side to side difference in the paired *t* test, the data from right and left sides were averaged for each trial. For exercises that demonstrated side to side differences, data from the side with the greatest muscle activity (highest percent MVIC) was used for analysis. To analyze the effect of postural cueing on muscle activation across exercises, a 2-way repeated measures analysis of variance (ANOVA) (posture by exercise) was used for L5 deep multifidus and T12 longissimus.

To examine relative muscle activity, the ratio of muscle activation of the L5 multifidus to the T12 longissimus thoracis (percent MVIC multifidus/percent MVIC longissimus) was calculated for the exercise trials with and without postural cueing. To compare the muscle ratios between exercises, a 1-way repeated-measures analysis of variance (ratio of multifidus activation to longissimus activation by exercise) was performed across exercises. In the case of a significant ANOVA, paired *t*-tests (2-tailed) were used to identify differences between exercises. Significance was set at an alpha level of 0.05 for all analyses.

3. Results

Demographic information for the subject sample included in the study is given in Table 1. Subjects required an average of 4 training sessions to safely learn the 10 exercises and demonstrate a correct increase in lower lumbar lordosis with each exercise. Subjects' disability scores were low at the time of inclusion (Table 1).

Paired *t*-tests demonstrated significant differences ($p < 0.05$) in muscle activation between sides only for the following exercises: side-bridge on feet, side-bridge on knees, and quadruped leg lift with opposite arm raise. Therefore, the results of the primary and secondary analyses consist of values from the side with the highest muscle activation for these three exercises combined with side-to-side averaged values from the remaining exercises.

For the 2-way ANOVA, data sphericity was not assumed, therefore the Greenhouse-Geisser correction was used. No significant exercise by posture interaction was found for either the longissimus thoracis ($p = 0.159$, $F = 2.004$) or lumbar multifidus ($p = 0.472$, $F = 0.904$). Results showed a significant main effect for posture on the deep multifidus activation at L5 ($p < 0.001$, $F = 35.904$), indicating significantly greater multifidus activation levels during exercises performed with postural cueing vs. neutral posture. The average increase in deep multifidus activation was $5.9\% \pm 3.0\%$ MVIC when postural cueing was used. The statistical power calculated from the mean

Table 1

Subject Demographics: (N = 9). Values below represent either counts or means \pm SD.

Gender	Female: 3, Male: 6
Pain location	Unilateral: 4, Bilateral: 5
Type of low back pain	Recurrent: 3, Chronic: 6
Age	31.4 \pm 11.6
Height (in)	67.6 \pm 4.4
Weight (lb.)	152.3 \pm 46.1
Duration of low back pain (years)	7.0 \pm 4.0
Training sessions	4.0 \pm 0.7
Oswestry Disability Index	13.6% \pm 0.1

difference of the marginal means was 0.999. There was no significant increase in longissimus T12 activation with postural cueing ($p = 0.191$, $F = 2.045$), with an average increase of $1.6\% \pm 3.3\%$ MVIC. Table 2 shows the %MVIC for each muscle during each exercise under the two conditions.

For the ratio analysis, data sphericity was not assumed, therefore the Greenhouse-Geisser correction was used. For the neutral posture condition, the 1-way ANOVA was not significant (p -value = 0.164). Mean \pm SD values for the ratio of multifidus activation/longissimus activation per exercise in neutral posture are listed in Table 3. The 1-way ANOVA within-subject p -value was significant ($p = 0.034$) for the postural cueing condition, and pairwise comparisons indicated exercises demonstrating significantly higher multifidus/longissimus activation ratios. The prone leg lift (Mean \pm SD ratio = 5.0 \pm 3.5) demonstrated significantly higher relative activation ratios vs. several exercises: prone trunk lift ($p = 0.007$), variable angle Roman chair at 15° ($p = 0.031$), variable angle Roman chair at 45° ($p = 0.008$), bilateral leg lift ($p = 0.007$), and the bilateral arm and leg lift ($p = 0.018$). The variable angle Roman chair at 15° (Mean \pm SD ratio = 2.7 \pm 2.0) showed significantly higher ratios compared to the prone trunk lift ($p = 0.034$), variable angle Roman chair at 45° ($p = 0.016$) and bilateral leg lift ($p = 0.028$). Bridging (Mean \pm SD ratio = 2.7 \pm 2.2) demonstrated significantly higher ratios vs. prone trunk lift ($p = 0.038$) and bilateral leg lift ($p = 0.049$). The bilateral arm and leg lift exercise (Mean \pm SD ratio = 1.7 \pm 0.7) showed significantly higher ratios compared to prone trunk lift ($p = 0.035$). Mean \pm SD values for the ratio of muscle activation per exercise in the postural cueing condition are listed in Table 3.

4. Discussion

The purpose of this study was to examine the effects of postural cueing to increase lumbar lordosis during the performance of trunk stabilization exercises in persons with chronic and recurrent low back pain. The first objective was to observe whether postural cueing could increase lumbar multifidus activation in these subjects without a significant increase in global muscle activation.

Participants demonstrated significantly greater deep multifidus activation (5.9% MVIC) across the 10 exercises when performed with postural cueing, without a significant increase in longissimus thoracis activation. This finding is similar to the 6.0% MVIC increase in deep multifidus activation demonstrated by healthy subjects during trunk stabilization exercises with postural cueing in a previous study (Beneck et al., 2016). This current study explored a wider range of lumbar stabilization exercises (16.0–65.4% MVIC) compared to a range found in the previous study (12.4–44.6% MVIC) by including four additional exercises. Despite increased loading with the exercises, there was no exercise by posture interaction found, indicating that higher-load exercises did not interfere with the ability to increase deep multifidus activation with postural cueing. In our study, subjects were carefully monitored during training and performance to ensure that they could perform each exercise without pain. It seems that with proper training and monitoring by a clinician, subjects successfully achieved the desired pattern of muscle activation with postural cueing, possibly through increasing neural drive to the muscle. If the subjects had experienced pain, it is possible that different activation patterns could have been observed due to inhibition or muscle guarding (Ghamkhar and Kahlae, 2015; Hodges and Tucker, 2011; van Dieën et al., 2003).

Deep multifidus activation ranged from 16.0–65.4% MVIC in neutral posture for the 10 exercises studied, and increased to a range of 21.1–69.6% MVIC with postural cueing. For clinicians looking to optimize multifidus activation levels to maximize potential for hypertrophy and neuromuscular adaptation, it is useful to observe that out of the 10 exercises analyzed, the following three exercises demonstrated the highest levels of multifidus activation overall: bilateral arm and leg lift, bilateral leg lift, and variable angle Roman chair at 45°, especially

Table 2Values represent mean \pm SD percent maximum voluntary isometric contraction (%MVIC) for L5 deep multifidus and T12 longissimus thoracis.

	L5 Neutral (%MVIC \pm SD)	L5 Postural cueing (%MVIC \pm SD)	T12 Neutral (%MVIC \pm SD)	T12 Postural cueing (%MVIC \pm SD)
Sidebridge on knees	16.0 \pm 10.0	21.1 \pm 8.9*	25.8 \pm 25.0	25.0 \pm 17.0
Sidebridge on feet	17.8 \pm 18.4	20.2 \pm 17.0*	22.4 \pm 21.6	32.8 \pm 28.3
Prone leg lift	28.8 \pm 19.0	36.4 \pm 18.7*	13.3 \pm 13.9	11.2 \pm 10.0
Variable angle Roman chair at 15°	35.2 \pm 12.3	50.0 \pm 23.7*	29.5 \pm 25.1	30.2 \pm 21.2
Bridging	38.4 \pm 11.9	41.1 \pm 14.3*	27.8 \pm 24.6	25.5 \pm 24.3
Prone trunk lift	44.4 \pm 13.0	51.2 \pm 19.5*	43.1 \pm 11.2	46.6 \pm 11.1
Quadruped leg lift opposite arm raise	47.3 \pm 25.3	53.5 \pm 24.8*	36.8 \pm 29.3	32.3 \pm 26.5
Variable angle Roman chair at 45°	53.2 \pm 22.0	59.6 \pm 27.1*	55.5 \pm 48.4	61.9 \pm 47.4
Bilateral leg lift	59.4 \pm 21.3	67.5 \pm 25.3*	53.3 \pm 23.8	58.2 \pm 27.6
Bilateral arm and leg lift	65.4 \pm 16.4	69.6 \pm 19.6*	46.4 \pm 17.4	46.3 \pm 18.4

* Significantly higher activation than the neutral posture when averaged across all exercises.

Table 3Ratio of L5 deep multifidus activation to T12 longissimus thoracis activation per exercise in each posture. Values represent means \pm SD.

	Neutral	Postural Cueing
Sidebridge on knees	1.4 \pm 2.0	1.8 \pm 2.2
Sidebridge on feet	1.1 \pm 3.2	1.6 \pm 2.3
Prone leg lift	3.9 \pm 3.2	5.0 \pm 3.5****
Variable angle Roman chair at 15°	2.1 \pm 1.6	2.7 \pm 2.0***
Bridging	2.1 \pm 1.4	2.7 \pm 2.2**
Prone trunk lift	1.1 \pm 0.4	1.2 \pm 0.4
Quad leg lift opposite arm raise	1.9 \pm 2.2	2.2 \pm 2.4
Variable angle Roman chair at 45°	1.4 \pm 1.0	1.4 \pm 1.1
Bilateral leg lift	1.3 \pm 0.6	1.3 \pm 0.7
Bilateral arm and leg lift	1.6 \pm 0.9	1.7 \pm 0.7*

**** Statistically significant difference compared to variable angle Roman chair at 15°, variable angle Roman chair at 45°, bilateral leg lift, and bilateral arm and leg lift.

*** Statistically significant difference compared to prone trunk lift, variable angle Roman chair at 45°, and bilateral leg lift

** Statistically significant difference compared to prone trunk lift and bilateral leg lift

* Statistically significant difference compared to prone trunk lift

when performed with postural cueing (Table 2).

The second objective of this study was to examine the ratio of lumbar multifidus to longissimus thoracis activation in these subjects during the selected exercises with and without postural cueing. The results revealed that specific exercises demonstrated significantly higher relative muscle activation ratios (Table 3). This information could be helpful for clinicians to determine which stabilization exercise most optimally fits certain goals. For example, the prone leg lift exercise with postural cueing demonstrated the highest relative muscle activation ratio (5.0 \pm 3.5), indicating that it could be an optimal exercise in early stages of rehabilitation for low back pain: it can specifically target deep multifidus impairment while maintaining a lower relative longissimus thoracis activation, thus decreasing the risk of compression-induced pain. The prone leg lift, however, demonstrates the third lowest %MVIC for the deep multifidus. Therefore, with a goal of progressing the deep multifidus to higher levels of activation while minimizing risk for compression-induced pain, variable angle Roman chair at 15° and bridging performed with postural cueing could be ideal, demonstrating significantly high relative muscle activation ratios (Table 3) as well as increased %MVIC for deep multifidus compared to the prone leg lift.

This study is the first to our knowledge to examine lumbar multifidus and longissimus thoracis muscle activity using intramuscular EMG during stabilization exercises in subjects with low back pain. Previous studies have examined muscle activity during lumbar stabilization exercises in healthy subjects and persons with low back pain, typically with surface electromyography (Arokoski et al., 2004; Colado et al., 2011; Ekstrom et al., 2008; Feldwieser et al., 2012) Yoon et al. assessed

trunk muscle activation during three quadruped stabilization exercises in 10 individuals with chronic low back pain, including a measure of local vs. global muscle activation (Yoon et al., 2015). In their study, the authors only assessed three quadruped activities: quadruped single arm raise, quadruped single leg lift, and quadruped leg lift with opposite arm raise. They found that the local/global muscle ratios of the back muscles (multifidus vs. iliocostalis lumborum) were higher in the exercises involving a raise of the leg compared to just raising one arm (Yoon et al., 2015). Because surface electromyography was used for multifidus, the possibility of cross-talk cannot be ruled out (Stokes et al., 2003) and the ability of surface EMG to record deep muscle activity is limited.

4.1. Study limitations

The first limitation of this study is that the lumbar lordosis of the two postures was not specifically quantified. A second limitation is that other global muscles were not monitored during the performance of the exercises, including the iliocostalis lumborum, external obliques, and rectus abdominis. Activation of these muscle would likely result in additional spinal loading. Last, the results can only be generalized to persons low levels of disability as the average Oswestry Disability score was 13.6% (Fairbank and Pynsent, 2000).

5. Conclusions

Postural cueing can be used during 10 common lumbar stabilization exercises of varied intensity to increase deep multifidus activation without a significant increase in longissimus thoracis activation in patients with chronic or recurrent low back pain. Knowledge of the relative activation of these two muscles across exercises can assist clinicians in using them optimally during a progression of rehabilitation exercises.

Suppliers

- (a) Motion Lab Systems, Inc.
- (b) Toshiba America Medical Systems, Inc.
- (c) The MathWork, Inc.
- (d) IMB Corporation.

Conflicts of Interest

None declared.

Ethical statements

The work described in this research article was reviewed and approved by the California State University, Long Beach Institutional Review Board. Subjects provided written consent prior to participation in the study.

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Brooke R. Winder is an Assistant Professor in the Department of Dance at California State University, Long Beach. She received a BFA in Dance from Chapman University (2004), a Doctorate in Physical Therapy from the University of Southern California (2010), and completed a post-professional residency in Orthopedic Physical Therapy at the University of Southern California (2011). Her research focuses on the low back and pelvic pain as it relates to muscle performance and exercise interventions, both preventative and rehabilitative.



Paul A. Keri is a practicing physical therapist working in both orthopedics and home health. He received his B.Sc in Kinesiology Exercises Science (2011), and Doctorate in Physical Therapy (2015) both at California State University, Long Beach.



David Weberg is a practicing physical therapist at Kaiser Permanente South Bay. He received the B.Sc. in Health, Physical Education, and Leadership at Southern Oregon University (2011), and Doctorate in Physical Therapy at California State University, Long Beach (2015). He completed a post-professional residency in orthopedic physical therapy in 2017, and continued on to a spine fellowship in physical therapy in 2018.



George J. Beneck is an Associate Professor in the Department of Physical Therapy at California State University Long Beach. He received the B.Sc. in Physical Therapy at Temple University (1984), M.Sc. in Biomechanics at Michigan State University (1991), and Ph.D. in Biokinesiology at the University of Southern California (2010). His research examines how spinal motion and paraspinal muscle performance are altered in association with spinal disorders, and how various manual and exercise interventions alter spinal motion and muscle performance.