Pathophysiology of adductor spasmodic dysphonia: a TMS study

A DISSERTATION
SUBMITTED TO THE FACULTY OF THE GRADUATE SCHOOL
OF THE UNIVERSITY OF MINNESOTA
BY

Sharyl Anne Samargia

IN PARTIAL FULFILLMENT OF THE REQUIREMENTS
FOR THE DEGREE OF
DOCTOR OF PHILOSOPHY

Advisor: Teresa Jacobson Kimberley, PhD PT

September, 2012

© Sharyl Anne Samargia, 2012

Acknowledgements

I would like to thank my doctoral advisor, Dr. Teresa Jacobson Kimberley for continually challenging me, guiding me and providing countless opportunities for personal and professional growth throughout this journey.

In addition, I would like to thank:

The members of my doctoral committee:

James Carey, Ph.D., PT
Mark DeRuiter, M.B.A., Ph.D., CCC-A/SLP
Teresa Kimberley, Ph.D., PT
Carl Kukulka, Ph.D., PT
George Goding, Jr., M.D.

Past and present members of the brain plasticity lab:

Michael Borich, Ph.D., PT
Kristen Pickett, Ph.D.
Joan Deng, M.D., Ph.D.
Bernadette Gillick, Ph.D., PT
Jessica Cassidy, Ph.D. candidate
Rebecca Schmidt, B.S. research assistant
Laura Golob, undergraduate research assistant

Family and friends

Dedication

This dissertation is dedicated to my beautiful daughter, Alexandra for selflessly and patiently supporting me over the past 5 years.

Alex, you have maturity, compassion and insight far beyond your years. You have not only encouraged me through this incredible journey but you have provided me much needed distraction to keep me balanced. No matter how difficult my day was, I could look forward to seeing your smiling face and feeling your unconditional love and support.

I can only hope, through my experience, you have learned to always challenge yourself and believe in yourself.

Your words echoed through my mind on many days, good and bad,

"Mommies can do anything"

You are the light of my life

Abstract

Adductor spasmodic dysphonia (AdSD) is a neurologically-based voice disorder affecting the firing rate and pattern of upper motor neurons responsible for the laryngeal musculature(). AdSD is characterized by intermittent hyperadduction of the true vocal folds during connected speech resulting in interruptions in phonation and a strained/struggled vocal quality. Differential diagnosis of AdSD can be difficult as the clinical symptoms present similar to those found in muscle tension dysphonia (MTD). *Purpose:* The purpose of this study was to identify and compare differences in intracortical inhibition and facilitation in the primary motor cortex between those with AdSD, MTD and healthy controls, if those differences were widespread; affecting both the cortical spinal and corticobulbar tract and, if differences in cortical excitability are related to the perceptual severity of the voice disorder. An additional purpose was to determine if measures of intracortical inhibition and facilitation are viable methods to assist in the differential diagnosis between AdSD and MTD. *Methods:* Transcranial magnetic stimulation (TMS) was used to measure intracortical inhibition and facilitation through the following measures: cortical silent period (CSP), short interval intracortical inhibition (SICI), intracortical facilitation (ICF) and the stimulus response curve (SR curve) in first dorsal interosseus and masseter in those with AdSD, MTD and healthy controls. The Consensus of Auditory Perceptual Evaluation of Voice (CAPE-V) was used as a tool to evaluate the nature and severity of voice characteristics of those with AdSD and MTD. *Results:* Masseter and FDI CSP were the most sensitive in capturing between group differences. Those with AdSD had significantly shorter CSP duration in masseter than those with MTD and healthy controls. Those with AdSD also had significantly shorter CSP duration in FDI than healthy controls, but not those with MTD indicating widespread dysfunction of the $GABA_B$ mechanism is a feature of AdSD, similar to other forms of focal dystonia. Use of TMS is feasible in assisting in the differential diagnosis of AdSD and MTD.

Table of Contents

List of Tables……. v

List of Figures…… vi

Chapter 1: Introduction……... 1-43

Chapter 2: Specific Aims……. 44

Chapter 3: Preliminary Data…….. 45-48

Chapter 4: Methods……. 49-59

Chapter 5: Results…… 60-70

Chapter 6: Discussion…….. 71-84

References……..... 85-94

Appendices…… 95-152

Appendix A: Consent forms…….. 95-101

Appendix B: Inclusion/Exclusion criteria…… 102-103

Appendix C: Recruitment information…… 104-112

Appendix D: Subject binder…….. 113-116

Appendix E: Perceptual voice measures…….. 117-120

Appendix F: Data processing information….. 121-130

Appendix G: SPSS printouts…… 131-152

List of Tables

Table	Title	Page
1	Cortical excitability results from single subject with OMD	47
2	Group demographics; age and gender	50
3	Individual subject demographics	50
4	MEP thresholds and definitions	55
5	Cortical excitability outcome measures and definitions	55
6	Mean SICI indexes for healthy and AdSD groups	63
7	Mean ICF indexes for healthy and AdSD groups	64
8	Post hoc power analysis	67
9	Correlation analysis of CAPE-V and CSP values	68
10	Mean CAPE-V scores for AdSD and MTD groups	68
11	Mean masseter latency values for all groups	69

List of Figures

Figure	Title	Page
1	Frequency of perceived triggers of AdSD	1
2	Schematic of descending motor pathways	5
3	EMG of laryngeal musculature: healthy compared to AdSD	7
4	Example of co-contraction in focal hand dystonia	10
5	Role of basal ganglia on motor cortex excitability	17
6	Illustrations of the emotional and volitional speech pathways	25
7	Task specificity in those with AdSD compared to MTD	26
8	Illustration of optimal coil placement for masseter	36
9	Comparison of coil orientation for stimulation of masseter	38
10	Photo of TMS coil placement for stimulation of masseter	45
11	EMG traces of masseter and FDI CSP in individual with OMD compared to healthy	46
12	Photo of electrode placement: contralateral masseter	54
13	Photo of TMS coil placement for stimulation of masseter	54
14	Comparison of mean CSP duration in masseter and FDI	60
15	EMG traces of masseter CSP in healthy, AdSD and MTD	61
16	EMG traces of FDI CSP in healthy, AdSD and MTD	61
17	EMG traces of masseter paired pulse in healthy and AdSD	63
18	EMG traces of FDI paired pulse in healthy and AdSD	64
19	Comparison of mean AUC values between all groups	65
20	Comparison of Mean slope values between all groups	66

CHAPTER 1

BACKGROUND:

Adductor spasmodic dysphonia (AdSD), also referred to as laryngeal dystonia (LD), is a neurologically-based movement disorder affecting the firing rate and pattern of upper motor neurons responsible for the laryngeal musculature()()()()()(). There are three types of laryngeal dystonia: 1) Adductor laryngeal dystonia (AdSD), characterized by intermittent hyperadduction of the false and true vocal folds during connected speech resulting in interruptions in phonation and a strained/struggled vocal quality, 2) Abductor laryngeal dystonia (AbSD), characterized by intermittent hyperabduction of the false and true vocal folds during connected speech resulting in excessively breathy voice with periods of aphonia, and 3) Mixed laryngeal dystonia which is a combination of the above. AdSD is the most common form of laryngeal dystonia, accounting for 80% of cases (Brin, Blitzer, & Stewart, 1998) and will be the focus of this research.

AdSD is typically idiopathic affecting more women (60-85%) than men (Adler, Edwards, & Bansberg, 1997). In general, older female patients tended to have more severe vocal symptomatology and a higher likelihood of associated movements (Lundy, Ling, Casiano, & Zue, 1998).

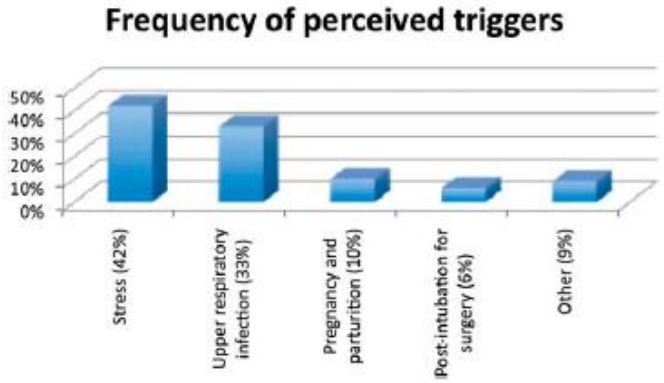


Figure 1. Frequency of perceived triggers of AdSD reported by patients (n=350)(Childs, 2011).

Average age of onset of 46 years ±1years has been reported and 83% of 150 patients reported intense voice use in their occupation (Tanner, 2011). As high as 20% of patients with SD have other forms of focal dystonia (Schweinfurth, Billante, & Courey, 2002) and reportedly < 8% have family members with a form of focal dystonia (C. L. Ludlow, 2011). Progression is gradual the first year then becoming chronic (Izdebski, dedo, & Boles, 1984). The onset for some has been perceived as sudden and linked to a traumatic event and others can identify a specific trigger of the onset of AdSD however, others described the onset as gradual with no identifiable trigger (Childs, 2011) (Fig.1). Differential diagnosis of AdSD can be difficult as the clinical symptoms present similar to those of muscle tension dysphonia (MTD) but has a different etiology and treatment. The current treatments for AdSD include: injections of botulinum toxin into the true vocal folds, dennervation of the recurrent laryngeal branch of the vagus nerve and, in some cases, laryngeal framework surgery (C. Ludlow, 2009). The efficacy of all three currently available treatment paradigms is limited. Additionally, the cost to benefit ratio of each modality is high due to the invasive nature of the treatment in comparison to the relatively low expectation of full recovery.

“Spastic dysphonia” was first described by Traube in 1871 and was considered to be psychogenic in nature due to the task specificity of the disorder (C. Sapienza & Hoffman-Ruddy, 2009). As such, it was treated with a variety of techniques including psychotherapy, acupuncture, hypnosis, behavioral speech/voice therapy, biofeedback, electroshock therapy, medications including tranquilizers, muscle relaxants, meditation and respiratory therapy; none of which proved to be effective in the management of this

disorder (H. Dedo & Izdebski, 1983). In the mid-1970s unilateral resection of the recurrent laryngeal nerve in conjunction with behavioral voice therapy was introduced and found to be more effective than past treatments (H. Dedo & Izdebski, 1983; H. H. Dedo, 1976). Aronson et al. (1968) suggested a neurogenic basis when he documented a high incidence of vocal tremor in those with AdSD indicating the disorder was not psychogenic but neurologically-based; however, it was not known if the impairment stemmed from the central or peripheral nervous system (Aronson, Brown, Litin, & Pearson, 1968). Evidence supporting a central causation was provided by Finitzo-Hieber, et al (1982) when they reported abnormal latencies from auditory brain stem reflex testing (Finitzo-Hieber, Freeman, Gerling, Dobson, & Schaefer, 1982). Laryngeal EMG indicated no evidence of "spasms" in the thyroarytenoid muscle and EMG patterns appeared similar to those with other forms of dystonia providing the first evidence that AdSD may be a form of focal dystonia (Blitzer et al., 1985). And later, improved EMG responses from thyroarytenoid muscle were reported in response to botulinum toxin injection in those with AdSD suggesting the injection led to a reduction in central motor neuron activity (Bielamowicz & Ludlow, 2000). Aminoff, Dedo, & Izdebski (1978) first described spasmodic dysphonia as a "widespread neurological disorder"(Aminoff, Dedo, & Izdebski, 1978).

NORMAL LARYNGEAL FUNCTION DURING PHONATION

The myoelastic-aerodynamic theory of voice production was first introduced by Muller in 1843 and states continued air pressure is developed and built up below the vocal folds to a degree great enough to displace the inertial property of the vocal folds tissue and

sustain the vibration of the vocal folds over time (van den Berg, 1958). The intrinsic muscles of the larynx, particularly the lateral cricoarytenoid and thyroarytenoid contract to pull the vocal folds into a nearly adducted position, referred to as vocal folds approximation. Vocal folds approximation occurs during the beginning phase of exhalation, causing an increase in pressure below the level of the vocal folds (subglottic). When this pressure reaches a particular point, it displaces the vocal folds laterally and, due to their elastic properties, the vocal folds return to their nearly adducted position again creating resistance to airflow. This cycle can be continuous as long as the subglottic pressure is sustained and vocal folds are healthy. The Bernoulli Effect is a feature of the myoelastic-aerodynamic theory of phonation. As the airway narrows (when the vocal folds are returning to their nearly adducted position), air velocity increases, as the glottal space narrows air velocity increases more causing greater negative pressure and a greater pulling force on the vocal folds toward midline to the fully adducting position. This repetitive cycle results in the vibratory sound source known as voice is measured in hertz (Hz). The fundamental frequency (F_o) refers to the number of cycles per second the vocal folds open and close. The gender and age of individual as well as the length and mass of the vocal folds are contributing factors in determining this frequency. Typically, the F_o for males is approximately 125-150 Hz and for females 230-250 Hz (C. Sapienza & Hoffman-Ruddy, 2009). Extrinsic laryngeal muscles primarily support the larynx and are responsible for elevating or depressing the larynx as a whole; however, it is the intrinsic laryngeal muscles that are responsible for initiating and modulating the characteristics of voice such as pitch and volume. The muscles responsible for adduction of the vocal folds, particularly the thyroarytenoids, lateral cricoarytenoids and transverse

and oblique interarytenoids are innervated by the recurrent laryngeal nerve (RLN) which is a branch of cranial nerve X (vagus nerve). The only muscle responsible for abduction of the vocal folds is the posterior cricoarytenoid (PCA) which is innervated by the posterior branch of the RLN (C. Sapienza & Hoffman-Ruddy, 2009). Muscle contraction required for vocal folds adduction reportedly starts 150-400 ms prior to actual voicing and is referred to as pre-phonatory tuning of the vocal folds (Izdebski & Shipp, 1978). Motor control of the larynx begins with upper motoneurons in the primary motor cortex and in the frontal precentral gyrus that project to the medulla via the internal capsule and descend through the corticobulbar pathway, specifically vagus nerve (Khedr & Aref, 2002). Most fibers decussate at the pons or medulla through the medial lemniscus which then project onto the motoneuron pools in the nucleus ambiguous which gives rise to the efferent fibers of the vagus nerve. Some fibers remain ipsilateral while they descend which results in bilateral innervation of some muscles of the larynx for review, (C. L. Ludlow, 2004) (Fig.2).

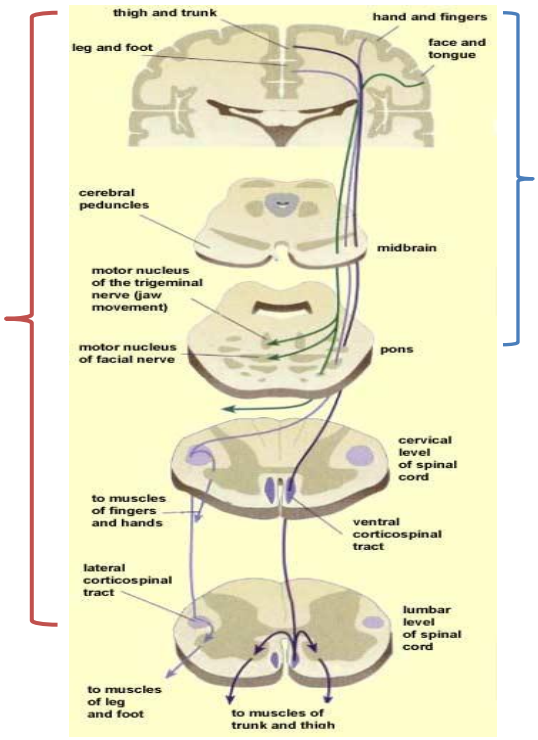


Figure 2. Schematic of the cortical spinal tract (blue bracket) and corticobulbar (red bracket) tract (Mosby, 2000).

Transcranial magnetic stimulation (TMS) is a tool that has been used to evaluate the integrity of the corticolaryngeal pathway in healthy subjects by recording motor evoked potentials (MEP) in laryngeal muscles in response to direct current magnetic stimulation to the motor cortex. Amplitude and latency are two parameters of an MEP that can be recorded and compared. Latency of the MEP is defined as the time from the point of stimulation presentation to the point of the evoked muscle contraction (spike) after stimulation. Khedr & Aref (2002) used TMS to establish latency norms in contralateral and ipsilateral responses between the superior laryngeal branch (SLN) of the vagus nerve, represented by EMG responses from the cricothyroid (CT) and the recurrent laryngeal branch (RLN) of the vagus nerve, represented by EMG responses from the thyroarytenoid (TA) (Khedr & Aref, 2002). Rodel, Olthoff, & Tergau, 2004 also used TMS in healthy subjects to determine response latency of the CT and TA muscles to determine if areas innervated by the superior laryngeal nerve (SLN) and recurrent laryngeal nerve (RLN) have separate cortical representation (Rodel, Olthoff, & Tergau, 2004). Both authors reported similar response latencies for CT and TA. Significant differences in MEP latency between the left and right thyroarytenoid were reported (Khedr & Aref, 2002). These results are not surprising as they reflect the difference in length between the right and left RLN route; the left RLN courses down and around the aortic arch before traveling back into the larynx.

Rodel et al., 2004 reported statistically significant differences in responses as a function of site of cortical stimulation suggesting areas innervated by SLN and RLN have separate cortical maps with some overlap (Rodel et al., 2004).

No evidence regarding MEP latency and amplitude at rest has been recorded in those with AdSD, however, increased TA activity has been reported during phonation using EMG (Hillel, 2001; Hirose, 1977; Schaefer, Watson, & Freemon, 1987; van Pelt & Ludlow, 1994). Bursts of TA and CT activation have been associated with phonatory breaks in those with AdSD (Fritzell, Feuer, Haglund, Knutsson, & Schiratzki, 1982; Hillel, 2001; Shipp, Izdebski, Reed, & Morrissey, 1985). During a sustained vowel phonation task ("eee"), increased amplitude and latency in TA prior to the onset of phonation as well as post phonation was reported in those with AdSD as well as increased amplitude during phonation providing EMG evidence of the overcontraction in both left and right TA in AdSD during sustained vowel production (Fig.3). During sentence reading, prolonged latencies and increased amplitude in TA were recorded throughout the sentence and phonatory breaks were associated with bursts of activation in TA and LCA (Hillel, 2001).

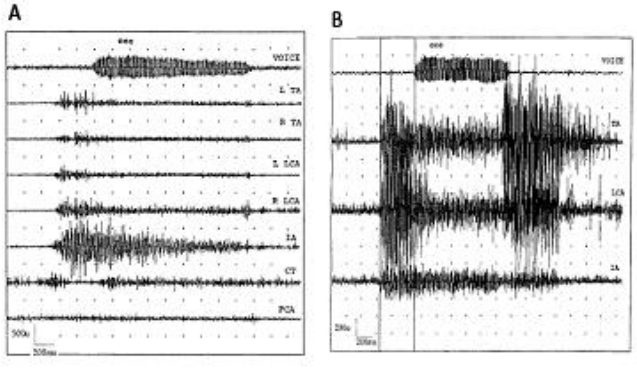


Figure 3. Left EMG trace represents normal intrinsic laryngeal muscle activation prior to, during and post vocalization of sustained /i/. Right EMG trace represents pre, during and post vocalization of sustained /i/ in a subject with AdSD (Hillel 2001).

s) of AdSD has been sparse, there is a small body of literature which indicates the presence of a measurable difference between individuals with AdSD and healthy individuals at the anatomical level. Upon histological examination of muscle and nerve fibers, Chhetri et al. (2003) reported the average diameter of the adductor branch of the recurrent laryngeal nerve was 8.3μm in those with AdSD compared to 7.1μm in healthy controls (p-value < 0.001). Additionally,

a high predominance of type 2 muscle fibers (fast twitch) in the lateral cricoarytenoid muscle (along with the TA, they adduct the vocal folds) was reported. No evidence of type 1 muscle fiber loss was apparent; however, the authors suggest "selective degeneration of motoneurons innervating type 1 muscle fibers could also lead to type 2 muscle fiber predominance" providing evidence for central nervous system involvement in AdSD (Chhetri, Blumin, Vinters, & Berke, 2003). In healthy participants, recruitment of intrinsic laryngeal muscles recorded using fine wire EMG revealed the thyroarytenoid muscle has a phasic firing pattern, spiking at the start of phonation then resting and the interarytenoid had sustained activation throughout phonation. In contrast, patients with AdSD show increased amplitude of activation throughout phonation (Hillel, 2001). Although the evidence has provided valuable information, additional neurophysiological evidence is needed to further elucidate the pathophysiology of this condition and eventually contribute to the advancement in relevant treatment paradigms. Of additional interest, perceptual similarities between AdSD and MTD are striking and result in frequent misdiagnosis. Neurophysiological features could also aid in differential diagnosis between these two conditions.

AdSD as a form of Dystonia

AdSD has been classified as a form of primary dystonia (Blitzer et al., 1985) and is the third most prevalent form of focal dystonia (estimated 1 in 100,000) after cervical dystonia and blepharospasm (Castelon Konkiewitz, Trender-Gerhard, & Kamm, 2002). As with other forms of dystonia, AdSD is task specific; only evident during speech but not other vocal tasks such as laughing or crying. Clinically, patients can exhibit preserved

nonspeech laryngeal tasks such as swallowing, throat clearing and valsalva maneuver as well as sustained vowel production, however during meaningful speech tasks, they exhibit a strained, harsh, tremulous voice, soft in volume and low in pitch and often demonstrate irregular stoppages of voice (Aminoff et al., 1978).

Many authors have reported patients with AdSD to have symptoms of focal dystonia affecting other areas of the body such as face, neck and hand (Aminoff et al., 1978; Lundy et al., 1998). Lundy et al., 1998 reported that of 68 patients studied with AdSD, 26.5% had a family history positive for neurologic disease, 29.4% had observed associated dystonic-like movements in other areas of the body; four involving head or neck, five involving abnormal eye blinking, one in limb, two with lower facial movement, eight with a combination of body movements and two with Meige's syndrome (Lundy et al., 1998). Rosenfield, 1988 reported of 100 subjects with AdSD, 71 had essential tremor, 25 had Meige syndrome and 27 had another form of focal dystonia (Rosenfield, 1988). AdSD has also been associated with writer's cramp (Blitzer et al., 1985; Schweinfurth et al., 2002). Due to the adult onset, task specificity and EMG characteristics, Blitzer, et al. (1985) identified spasmodic dysphonia, including adductor form, as a form of primary focal dystonia (Blitzer et al., 1985). As in other forms of focal dystonia, the pathophysiology of AdSD is unknown and there has been a paucity of research in this area.

PRIMARY DYSTONIA

Primary dystonia refers to a group of disorders characterized by excessive excitability of the efferent activity in motor neurons in specified areas (Courey et al., 2000).

It is characterized by prolonged muscle contractions including co-contractions and overflow to adjacent muscles which results in sustained twisting movements. This mechanism results in abnormal postures of a particular area(s) of the body (Berardelli et al., 1998). The result is a disturbance in function of the affected body part, specifically in control of fine motor, voluntary, task specific movements. Voluntary movements are described as bradykinetic and individuals often have difficulty transitioning from one posture to another while attempting to complete a complex movement. In 1976 the medical community recognized dystonia as a neurological condition. There are primarily two classifications of focal dystonia: 1) Primary dystonia, which is described as dystonic symptoms without evidence of structural abnormality or anatomical pathology within the CNS and can include: focal hand dystonia (FHD) or writer's cramp, blepharospasm and cervical dystonia or torticollis. 2) Secondary dystonia is described as dystonic symptoms secondary to another neurological diagnosis such as Parkinson's disease.

Figure 4. Example of co-contraction during a writing task in a patient with focal hand dystonia.

PATHOPHYSIOLOGY OF PRIMARY FOCAL DYSTONIA

The etiology of primary dystonia is not known; however, it has been described as a disorder of sensorimotor integration both of which offer insights toward the pathophysiology of this disease (Abbruzzese & Trompetto, 2002; Berardelli et al., 1998). In people with primary dystonia, electrophysiologic evidence has suggested a lack of selectivity of the muscle or muscle groups resulting in

overflow of activation into remote muscles (van der Kamp et al., 1989) hindering voluntary movement (Fig.4). In addition to overflow to remote muscles, excessive EMG activity of the effected muscle(s) during a specific task can occur. The prolonged contraction of the agonist muscle group can result in an overlap of muscle activation with the antagonist muscles which results in a co-contraction of the affected body segment. The observed manifestations of the affected musculature suggested to early investigators that impaired reciprocal inhibition (RI) may be a possible underlying mechanism. This theory has since evolved significantly with still no clear understanding of the pathophysiology of the disorder.

Neurophysiological features of focal dystonia

Many studies have investigated plausible mechanisms for the pathophysiology of primary dystonia. This body of literature has resulted in the current understanding that a variety of pathophysiological markers are associated with dystonia. Of the many etiologies that have been explored, three have emerged: 1) abnormal processing of sensory information 2) lack of homeostatic plasticity, long term potentiation (LTP) or depression (LTD) and 3) impaired inhibition (disinhibition).

Abnormal processing of sensory information

Differences in processing in the primary sensory cortex (S1) have been documented in patients with primary dystonia compared to healthy controls (Byl et al., 1997; Hallett, 1995). Decreased temporal and spatial discrimination and an altered somatotopic map in those with primary dystonia have been reported (Bara-Jimenez, Catalan, & Hallett,

1998). In addition, Lerner et al., 2004 correlated the amount of over activity in S1 to the severity of the dystonic symptoms and reported peripheral sensory stimulation to modulate clinical symptoms of focal dystonia and normalize pathologic regional blood flow patterns (Lerner et al., 2004). Clinically, it is known that so called 'sensory tricks' can modulate the dystonic spasm. This may include touching the side of the neck in cervical dystonia, wearing tight-fitting gloves in hand dystonia or placing a finger over the upper lip in embouchure dystonia. The application of a vibratory stimulus to the dystonic limb has been shown to induce dystonic movements and result in significantly less increase in blood flow in the sensorimotor cortex (Tempel & Perlmutter, 1990). Conversely, removing a sensory stimulus has been shown to improve dystonic symptoms, although not consistently. Similarly, the task-specificity of AdSD leads one to conclude the lack of sensory feedback from the laryngeal region may play a role in AdSD. In a neuroimaging study, Ali, et al. (2006) reported increased activity in the sensorimotor cortex in those with AdSD to normalize after injection of botulinum toxin injection suggesting the reduction of muscle force in the thyroarytenoid may reduce sensory feedback in the sensorimotor cortex (Ali et al., 2006).

Some dystonias are strongly associated with overuse. As new motor skills are acquired, the plasticity of the nervous system allows for change. In patients with focal dystonia, plasticity may be abnormally enhanced, such that excessive neural adaptation drives cortical and thalamic reorganization of sensory inputs and motor outputs. This reorganization could lead to undesirable neural changes and the resultant impaired motor control. Examples of aberrant cortical mapping of the hand have been reported in animal models with induced dystonia (Byl et al., 1997) and

humans with hand and generalized dystonia (Quartarone et al., 2003; Quartarone et al., 2005). In the somatosensory, motor cortex, and thalamus overlapping representations of adjacent digits, adjacent digit segments, dorsal and glabrous surfaces have been reported in a nonhuman primate model of focal hand dystonia (Byl et al., 1997). Sheehy & Marsden (1982) reported 25% of those with writer's cramp who learned to write with the opposite hand developed dystonia in the opposite hand (Sheehy & Marsden, 1982). Clinically, when one is receiving rehabilitation for hand dystonia and alternatives are sought to minimize use of the hand, voice recognition software is often suggested, but patients are instructed to use with caution to avoid laryngeal overuse that may lead to AdSD. Relatedly, singer's dystonia has been described as a type of laryngeal dystonia characterized by the same symptoms as those with AdSD only during the task of singing (Chitkara, Meyer, Keidar, & Blitzer, 2006). This has primarily been reported in professional level singers and provides evidence of overuse leading to maladaptive plasticity as a possible feature of AdSD.

Lack of homeostatic plasticity, LTP or LTD

Long term potentiation (LTP) is based on Hebbian Learning which is a frequency (temporal) dependent, local event that promotes efficiency of synaptic connections. If one neuron synapses onto another neuron repetitively, neurotransmission at that site will be more efficient. "Cells that fire together, wire together". Cells that do not synapse repeatedly upon one another will have a weaker connection and may eventually lose connection entirely. LTP refers to the molecular modifications in the post synaptic membrane that occur as a response to a repetitive high frequency stimulus. These changes

in the post synaptic membrane can lead to changes in gene expression and eventually, alter synaptic connections (Purves et al., 2008). Long term depression (LTD) refers to the molecular modifications in the post synaptic membrane in response to low frequency stimulation over a period of time to weaken the synaptic connectivity. During skilled motor practice, there is a tendency to form excessive connections between sensory inputs and motor outputs (heightened potentiation) and an inability to weaken existing associations (deficient depotentiation) which has been suggested to lead to maladaptive cortical reorganization (Quartarone, Siebner, & Rothwell, 2006). It can be speculated that there is an imbalance between inhibitory and excitatory mechanisms resulting in an inability to modulate inhibitory pathways in response to increased excitability in the cortical spinal tract. This may lead to maladaptive plasticity. Variation in the size, shape and location of the cortical map representing the affected region(s) of the body had been reported in subjects with dystonia, which is suggestive of maladaptive plasticity (M. L. Thompson et al., 1996).

Paired associative stimulation (PAS) involves applying electrical stimulation to the periphery and magnetic stimulation to the cortex and has been used as an experimental intervention to induce LTP. Quartarone, et al. (2008) used PAS to abductor pollicis brevis and first dorsal interosseus (neither having dystonic symptoms) in individuals with cervical and cranial dystonia, hemifacial spasm (HFS), a non-dystonic neurological condition, and healthy controls. They reported a larger increase in cortical spinal excitability and a loss of topographical specificity of the targeted regions in those with dystonia compared to those with HFS and healthy controls. Patients with dystonia appear to have excessive plasticity throughout not only the motor cortex, but the entire

sensorimotor system including areas that are not expressing dystonic symptoms (Quartarone, Rizzo, & Morgante, 2008). This suggestion of a more generalized topographical map is supported clinically in the prevalence of patients with more than one form of dystonia. Along with a more generalized topographical map, a generalized impairment in intracortical inhibition has also been reported in those with focal dystonia (Cakmur, Donmez, Uzunel, Aydin, & Kesken, 2004; Quartarone, Rizzo, et al., 2008). In summary, evidence suggests that in people with a focal dystonia of one body part may be at risk for developing another form of dystonia within the same or different descending motor pathway. However, a systematic investigation of the pathophysiology of the cortical spinal and corticobulbar tracts in focal dystonia has not been done.

Abnormal inhibition

Spinal level

Reduced reciprocal inhibition (RI) is a mechanism that occurs at the level of the spinal cord that inhibits the antagonist muscle upon contraction of the agonist and vice versa. Normal range of motion (ROM) and function of muscles innervated by the cortical spinal tract is dependent upon intact RI. If RI is impaired, co-contraction of the agonist and antagonist muscles occur in a manner similar to that observed in persons with focal dystonia. RI has also been identified as a possible cause of dystonia, due to the lack of muscle selectivity that results in overflow. Nakashima et al., (1989b) suggested dystonia led to a change in the descending control of spinal interneurons mediating one or both phases of reciprocal inhibition. While it appears as though RI at the spinal level may, in some cases, play a role in dystonic symptom presentation it does not appear as though it

provides a causative mechanism (Nakashima et al., 1989). Although reflex studies indicate a reduction in spinal cord and brainstem inhibition, this cannot account for the effect of dystonia on voluntary motor movement nor provide explanation of the task specificity of primary dystonia.

Cortical

TMS studies in primary dystonia have revealed normal central conduction time in the cortical spinal tract (P. D. Thompson et al., 1986). However, the amplitude of EMG responses increased to a greater degree in response to an increase in stimulus intensity and an increase in background contraction more than in healthy adults (Ikoma, Samii, Mercuri, Wassermann, & Hallett, 1996; Mavroudakis, Caroyer, Brunko, & Aamodt, 1995). The abnormal twisting movements in dystonia are due to co-contraction of the agonist and antagonist muscle groups and volitional movement appears to exacerbate the co-contraction (Murase, Duque, Mazzocchio, & Cohen, 2004) as EMG studies of the muscles at rest are normal. This suggests an impairment of intracortical inhibition which is driven by the inhibitory effect the basal ganglia has on the thalamus that then leads to overexcitation of the premotor and/or primary motor cortex. (Fig.5) The lack of inhibition or disinhibition observed in patients with dystonia has been linked to a disruption of the basal ganglia-cortical loop and provides the foundation of evidence that suggests dystonia is, at least in part, a disorder of the basal ganglia, a subcortical structure. The basal ganglia-cortical loop is a circuit beginning with projections from the cortex to the basal ganglia and returns to the cortex via the thalamus. The basal ganglia influences motor

output of the cortex via excitatory projections from the motor cortex to the striatum which inhibits the globus pallidus which inhibits the thalamus. The thalamus is also getting inhibitory projections from the reticular area of the substantia nigra. These inhibitory projections result in disinhibition, or excitation of the thalamus which in turn sends excitatory projections back into the motor cortex (Nolte, 2002). At rest, neurons in the medial globus pallidus of those with dystonia have been shown to have a lower firing rate and discharge in an irregular fashion (Vitek, Zhang, DeLong, Mewes, & Bakay, 1997). A reduction in inhibitory output from the medial globus pallidus would theoretically result in an increase in activity in the thalamus (both V_{oa} and V_{op}) which would have an inhibitory effect on the motor cortex. However, the V_{op} neurons appear to have reduced discharge rates (Berardelli et al., 1998). The basal ganglia loop does influence the motor cortex (M1, PMA and SMA), however, it cannot solely account for cortical differences of those with dystonia.

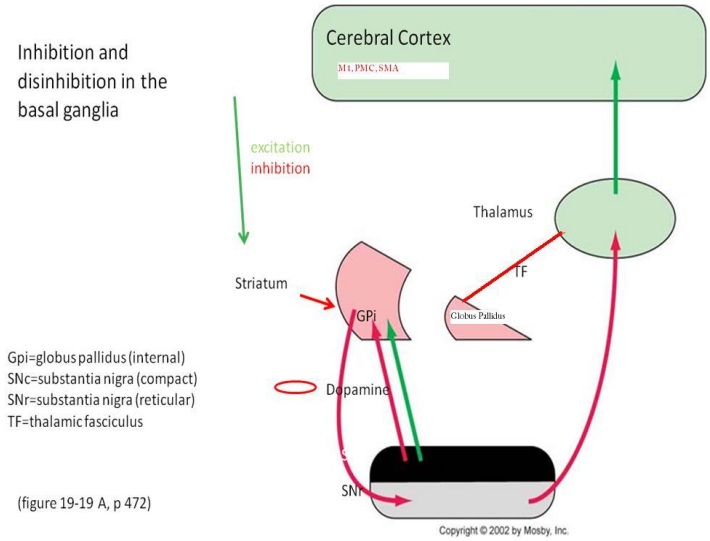


Figure 5. Role of basal ganglia on motor cortex excitability. (Mosby, 2002)

There is evidence to suggest patients with dystonia also have differences in activation excitability within the cortex itself, including the previously mentioned differences in homeostatic plasticity. As with any theory of disinhibition, one must consider the chemical mechanisms at work, particularly the role of GABA. It has been suggested,

patients with dystonia have decreased inhibition due to abnormal GABAergic mechanisms which provides a rationale for the overexcitation of the motor cortex in this population. Levy & Hallett (2002) found reduced inhibitory GABA levels in the sensorimtor cortex and lenitform nuclei in patients with focal hand dystonia (Levy & Hallett, 2002). Other studies have shown dystonic-like movements in primates after introduction of GABA antagonist bicuculline (Matsumura, Sawaguchi, & Kubota, 1992). Some evidence suggests an increase in dopaminergic function (due to a decrease in GABA activity) within the nigro-striatal pathway in dystonia. When Baclofen, a GABA-agonist drug was given to five human subjects with a combination of blepharospasm (dystonia of the eye muscles) and oromandibular dystonia, (dystonic postures affecting the function of the jaw referred to as Meige's syndrome), one subject showed marked improvement another had moderate improvement in both blepharospasm and oromandibular dystonia symptoms. Another subject exhibited moderate improvement of limb dystonia but no change in orofacial dystonic symptoms (De Andrade & Bertolucci, 1985).

TMS studies have revealed differences in cortical excitability in patients with dystonia compared with healthy peers providing further evidence to the idea that dystonia is a disorder of disinhibition (Ikoma et al., 1996; Mavroudakis et al., 1995). TMS using a paired pulse (PP) paradigm (explained further below) revealed subjects with FHD to have a significant decrease in cortical inhibitory mechanisms (Ridding, Sheean, Rothwell, Inzelberg, & Kujirai, 1995). These authors reported a significant decrease in intracortical inhibition in both hemispheres of those with FHD compared with the left hemisphere of

healthy controls when presented with a PP protocol that included a subthreshold conditioning pulse followed by a test pulse at the 1mV threshold and a duration of 1-6ms between the two pulses (inhibitory). This difference was not found in the facilitory paradigm of 7-15ms between the condition and test pulse (Ridding, Sheean, et al., 1995) indicating widespread decrease in intracortical inhibition in those with FHD. In addition, no significant differences in active motor threshold or resting motor threshold were found between those with FHD and healthy subjects (Ridding, Sheean, et al., 1995). Low frequency repetitive TMS (rTMS), thought to induce LTD, has been used as an intervention to promote inhibition of the targeted cortical areas. Murase & Rothwell (2005) studied the effects of low frequency rTMS over the primary motor cortex (M1), supplementary motor area (SMA) and the premotor cortex (PMC) in patients with dystonia compared to controls using cortical silent period (CSP) and computer-analyzed handwriting analysis as outcome measures. Results indicated rTMS to PMC, not M1 resulted in significant improvement of the handwriting rating and prolonged CSP duration. Stimulation over M1 and SMA and sham stimulation did not reveal any significant changes in cortical excitability or handwriting rating suggesting the overexcitabilty in PMC may be a primary mechanism in dystonia rather than overexcitability in M1. In addition, Borich, et al. (2009) reported decreased cortical excitability (as measured by prolonged CSP) and improvement in handwriting lasting at least 10 days in those with FHD after rTMS to PMC (Borich, Arora, & Kimberley, 2009). Given the state of evidence, it is likely that the pathophysiology of dystonia is due to a combination of factors highlighted above including impairments in: homeostatic

plasticity, intracortical inhibition, cortical-basal ganglia loop and sensorimotor processing.

Neuroanatomical features of focal dystonia

Focal dystonia of the arm and hand

Ceballos-Baumann et al., (1995) conducted a study using H_2 (15)O and positron emission tomography (PET) to identify cortical differences in patients with focal dystonia compared to controls. During a joystick task that required decision making, the authors reported differences in blood flow in patients with focal arm dystonia. Increased activation in those with focal arm dystonia was found in the following areas: contralateral PMC, rostral SMA, Brodmann area 8 (posterior frontal cortex), anterior cingulate, ipsilateral dorsolateral prefrontal cortex and bilateral lentiform nucleus. Underactivation, compared to controls, was seen in those with dystonia in the following areas: caudal SMA, bilateral sensorimotor cortex, posterior cingulate and mesial parietal cortex. In a follow up study using a continuous writing task that did not require decision making, these authors reported blood flow changes representative of overactivation of the ipsilateral premotor area, insula, parietal cortex and cerebellar vermis and underactivation of contralateral primary motor cortex, caudal SMC, anterior cingulated, mesial parietal and thalamus (Ceballos-Baumann, Sheean, Passingham, Marsden, & Brooks, 1997). In a functional MRI (fMRI) study, larger activation of the contralateral sensorimotor cortex and underactivation of the premotor cortex was reported in guitar players with focal hand dystonia compared to healthy guitar players during a scale task using a modified guitar

neck (Pujol et al., 2000). These results lend support to the idea of widespread impaired inhibition of cortical areas during specific tasks in those with dystonia.

Focal dystonia of head and neck

Oromandibular dystonia (OMD) is a form of focal dystonia affecting either the muscles responsible for closing or opening the jaw. Depending upon the subtype, speech and/or chewing and swallowing can be negatively affected. Blepharospasm affects the muscles around the eyes resulting in involuntary closing of the eyes and difficulty opening eyes voluntarily. In a comparison of subjects with blepharospasm and those with a combination of blepharospasm and oromandibular dystonia, referred to as Meige's syndrome, fMRI revealed decreased activation of the mouth representation area of the primary motor and ventral premotor cortex during a whistling task in those with Meige's syndrome when compared to healthy controls and those with isolated blepharospasm. However, activation in the somatosensory areas and the caudal supplementary motor area showed increased activation in both dystonia groups as compared to the control group. This finding suggests the enhanced somatosensory and SMA activation is independent of motor activation; thus indicating that enhanced activation of the somatosensory and caudal SMA regions is present in orofacial dystonia (Dresel, Haslinger, Castrop, Wohlschlaeger, & Ceballos-Baumann, 2006).

AdSD

Differences in fMRI activation patterns in those with AdSD compared with healthy individuals have been shown (Haslinger, Erhard, & Dresel, 2005; Hirano et al., 2001; K. Simonyan & Ludlow, 2010). Increased activation of left motor cortex, Broca's area,

cerebellum and auditory cortices and reduced activation of SMA were reported in one subject with AdSD (Hirano et al., 2001). However, reduced activation of primary sensorimotor cortices, premotor cortex (PMC) and sensory association areas were reported in those with AdSD (Haslinger et al., 2005). While one might expect increased activation in the primary sensorimotor cortex, a reduction in activation may actually be observed in those with impaired inhibition due to maladaptive plasticity. That is to say, overuse has caused the cortical map to expand, resulting in reduced intensity of activation due to larger spread (Haslinger et al., 2005). The stimulus tasks in the Haslinger, et al. (2005) study included production of prolonged vowels which is a primarily asymptomatic task in those with AdSD (C. M. Sapienza, Walton, & Murry, 2000). To further investigate the cortical activation differences as a function of vocal tasks, an fMRI study was conducted while subjects engaged in symptomatic vocal tasks (2 repetitions of vowel "i"), asymptomatic vocalization tasks (whimpering) and asymptomatic laryngeal tasks (coughing). Results indicated a significant increase in the extent of activation, as measured by ROI analysis, in the primary motor and somatosensory cortices during symptomatic and asymptomatic tasks. In addition, an increase in the extent of activation in the basal ganglia, thalamus and cerebellum during the symptomatic task and decreased during asymptomatic tasks. The intensity of activation, measured by percent signal change in each ROI, was significantly increased in only the primary somatosensory cortex during symptomatic tasks but not asymptomatic task productions (K. Simonyan & Ludlow, 2010). Ali and colleagues (2006) used positron emission tomography (PET) to investigate neural activation during a narrative speech task in patients with AdSD compared to healthy controls. Results indicated increased activation of the ventral

sensorimotor, auditory and anterior cingulate cortices, insula and cerebellum and decreased activation in the SMA, posterior supramarginal and posterior middle temporal gyri and the periaqueductal gray in those with AdSD (Ali et al., 2006).

The current evidence suggests overactivation of sensorimotor cortex appears to be present in those with AdSD however, overactivation of SMA does not appear to be a feature of AdSD as has been reported in FHD (Murase, Shimadu, Urushihara, & Kaji, 2006) and orofacial dystonia (Dresel et al., 2006). However this underactivation was only found in a single subject study. Considering SMA activation has been reported during voluntary phonation in healthy subjects (Hirano et al., 1996), electrical stimulation to the SMA region has resulted in speech arrest (Penfield & Welch, 1951) and considering SMA is responsible for motor planning, it is feasible to conclude that impairment of this region would result in impairment of voluntary phonation.

In addition to various regions of the cortex being implicated in AdSD, there have also been reports of bilateral cortical abnormalities using quantitative topographic electrophysiological mapping (QTE) to analyze cortical electrophysiology and single-photon emission computed tomography (SPECT) to evaluate cerebral perfusion in subjects with ADSD. Multifocal abnormalities in cerebral perfusion in the left hemisphere were reported in 46% of subjects. Of that 46%, 35% of this group showed abnormalities in the right hemisphere as well. Of the 17 (65%) subjects who demonstrated electrophysiological abnormalities of the cortex, 15 of them had multifocal bilateral abnormalities (Devous, Pool, Finitzo, & Freeman, 1990). Simonyan, Tovar-Moll, Ostuni, & Hallett, (2008) reported a bilateral increase in water diffusivity in the white matter along the corticobulbar tract and the cortical spinal tract in subjects with

AdSD which correlated with postmortem findings including a decrease in axonal density and myelin content in various regions. The changes in diffusivity and clinical symptoms of ADSD were highly correlated (K. Simonyan, Tovar-Moll, Ostuni, & Hallett, 2008). These findings lend strong evidence to suggest a bilateral, multifocal cortical basis for AdSD.

Abnormalities in brainstem anatomy and physiology in AdSD

Evidence in focal cranial dystonias (blepharospasm, oromandibular dystonia and cervical dystonia) suggests a key pathophysiological feature is overexcitability of interneurons at the brainstem level (Berardelli, Rothwell, Day, & Marsden, 1985; Cruccu, Pauletti, Agostino, Berardelli, & Manfredi, 1991). A postmortem study in two patients with AdSD revealed mild neuronal degeneration and depigmentation of the pars compact of the substantia nigra and the locus ceruleous that were not present in the healthy specimens (K. L. Simonyan, C.; Vortmeyer, A., 2009). Of additional importance, the post mortem investigation identified clusters of microglia/macrophage in the reticular formation surrounding the lower brainstem nuclei (K. L. Simonyan, C.; Vortmeyer, A., 2009). These differences may result in focal brainstem inflammation altering sensorimotor processing at the brainstem level and may impact the ascending projections that control voice and speech (C. L. Ludlow, 2011). However, these differences do not account for the classic feature of AdSD; task specificity. Results from animal and human research indicate the presence of two separate vocalization tracts. The first, the mammalian vocalization system is responsible for biologic, primitive vocalizations that are not learned, such as crying etc. The second system, volitional speech, is highly adaptable, dynamic and learned. Not only do the functions of these tracts differ, but the anatomic

pathways differ as well. The mammalian vocalization pathway originates in the anterior cingulate and travels to the periaqueductal gray and the reticular system within the medulla which provides input to the laryngeal motor neurons within the nucleus ambiguous (Jurgens, 2002).

Conversely, the volitional speech system has a direct corticobulbar path from the laryngeal motor cortex to the nucleus ambiguous in the reticular system of the medulla then to the laryngeal musculature via vagus nerve. In addition to the nucleus ambiguous, the laryngeal motor cortex also sends projections to the supplementary motor area (SMA), frontal opercular speech system (FOP), primary motor cortex (M1), posterior superior temporal gyrus (pSTG) and the supramarginal gyrus (SMG) (Kuypers, 1958; Rodel et al., 2004; Kristina Simonyan, Ostuni, Ludlow, & Horwitz, 2009) (C. L. Ludlow, 2011) (Fig 6). If the pathophysiology of AdSD was solely in the brainstem, both primitive sounds and volitional speech would be impacted, as the reticular system is a common point for both the mammalian vocalization pathway as well as the volitional

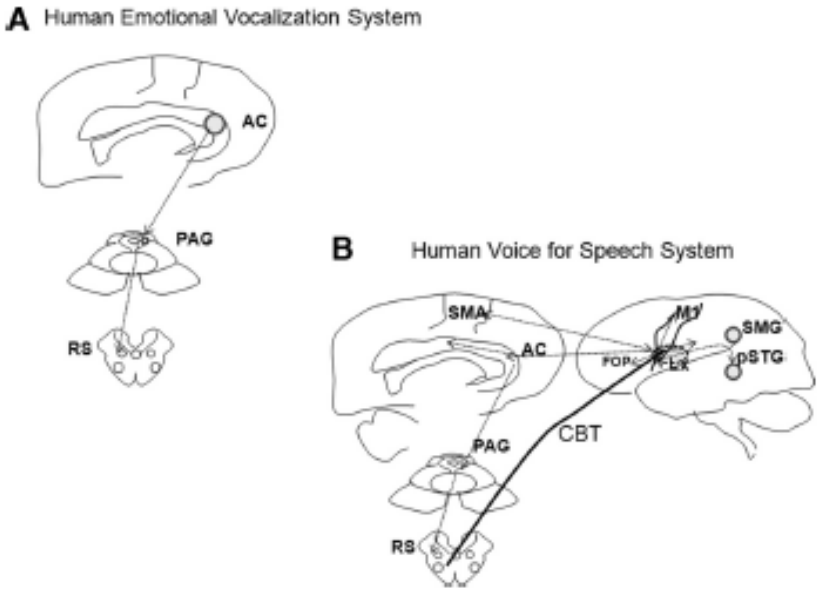


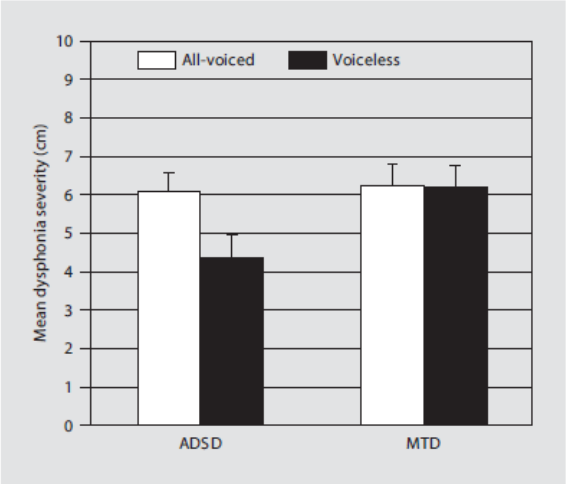
Figure 6. Illustrations of the emotional vocalization system and volitional voice/speech system in humans. AC=anterior cingulate, PAG=periaqueductal gray, RS= reticular system, SMA=supplementary motor area, FOP= frontal opercular speech system, M1= primary motor cortex, pSTG= posterior superior temporal gyrus, SMG=supramartinal gyrus and CBT= corticobulbar tract (C. L. Ludlow, 2011).

speech pathway. Since those with AdSD only have symptoms during connected speech, it follows that cortical mechanisms influence volitional speech in these individuals, but what are they?

In addition to the neurophysiological and neuroanatomical evidence, there is histological and clinical evidence that may help further elucidate the pathophysiology of AdSD.

Comparisons between AdSD and MTD

At this time, there are no assessment protocols that definitively diagnose AdSD; it is a diagnosis of exclusion and is often misdiagnosed as MTD due to similar voice perceptual qualities observed in both disorders (Roy, Gouse, Mauszycki, Merrill, & Smith, 2005; C. Sapienza & Hoffman-Ruddy, 2009). MTD is a functional voice disorder characterized by hyperfunction of the laryngeal musculature, both extrinsic and intrinsic but is not neurogenic in nature (Altman, Atkinson, & Lazarus, 2005; C. Sapienza & Hoffman-Ruddy, 2009). Accurate diagnosis is necessary for appropriate treatment recommendations. A series of studies were conducted to identify other diagnostic markers to differentiate ADSD from MTD (Roy et al., 2005; Roy, Smith, Allen, & Merrill, 2007; Roy, Whitchurch, Merrill, Houtz, & Smith, 2008). These authors identified the importance of including task-specific components to the clinical assessment of voice



Figuure 7. Increase in voice severity rating in AdSD during speech task with all voiced phonemes compared to a speech task loaded with voiceless phonemes. Note those with MTD did not demonstrate a difference in voice severity rating based on speech task (Roy et al. 2007).

to aid in more accurate differential diagnosis between AdSD and MTD. In a blinded controlled study, these authors identified a significant difference between severity of voice during connected speech and sustained vowel /a/ in patients with ADSD, with connected speech being significantly more impacted than sustained vowel production. No significant difference was reported between these two tasks in patients with MTD (Roy et al., 2005). In a follow up study, Roy and colleagues (Roy, Smith, et al., 2007) investigated the difference in the perception of dysphonia severity as a function of voiced vs voiceless phonetic loaded sentences. They reported a significant difference in the perception of dysphonia severity of those with AdSD between voiceless and voiced phoneme loaded sentences ($p < 0.001$). Sentences loaded with voiced phonemes resulted in a more severe dysphonia rating in those with AdSD. There was no difference in dysphonia severity between the two conditions in those with MTD (Fig.7).

The differences in production of sustained vowels and connected speech, voiced and voiceless loaded sentences and other aberrant acoustic events as a function of speech task supports the author's conclusion that there is task specificity in AdSD that is not present in MTD (Roy et al., 2005; Roy, Mauszycki, Merrill, Gouse, & Smith, 2007; C. M. Sapienza et al., 2000). In yet another study, these authors investigated the usefulness of phonatory break analysis in differential diagnosis of AdSD and MTD. Presence, frequency and duration of phonatory breaks, defined as complete interruption of phonation within words, were assessed in voice samples of sentences containing all voiced phonemes, of those with MTD and AdSD. A significantly higher number of phonatory breaks were documented in those with AdSD (Roy et al., 2008). Although these results are helpful in creating more useful diagnostic methods, the task specific

tasks relied on perceptual analysis which is subjective and may be vary based on clinician experience. The phonatory break analysis is a strong diagnostic marker but is time consuming and may not be practical in a clinical setting due to access to equipment, experience and time. Currently, many professionals base their diagnostic decision on response to treatment. Because AdSD is considered a neurologically-based voice disorder and MTD a functional voice disorder, treatment of the two is vastly different. Differential diagnosis is often confirmed after either a positive or negative response to treatment is documented. For example, those with MTD respond favorably to traditional behavioral voice therapy, those with AdSD do not (Roy et al., 2005; C. M. Sapienza et al., 2000). Those with AdSD typically respond favorably to injections of botulinum toxin into the vocal folds, those with MTD do not. This method of diagnosis is not ideal as it is time consuming and can result in a patient receiving an invasive procedure needlessly. There are a variety of vocal folds imaging techniques, none of which can provide definitive diagnosis of AdSD. Direct laryngoscopy (flexible or rigid endoscope) with a halogen light source provides a clear view of the integrity of the vocal folds and is primarily used to rule out vocal fold pathology. Videostroboscopy incorporates direct laryngoscopy with a strobe light source, utilizing Talbot's Law in order to observe the pattern of vocal fold vibration. Sapienza et al.(2000) stated that the use of videostroboscopy can be used to identify key features of AdSD including glottic and supraglottic compression and intermediate laryngeal spasm (C. M. Sapienza et al., 2000). However, some voice experts contend that videostroboscopy does not have the capability to capture overcontraction of the thyroarytenoid muscles because it utilizes a visual illusion as its basis and therefore is not recording in real time. Often, those with MTD

present with anterior to posterior as well as lateral compression of the supraglottic region at rest and during phonation to indicate excessive laryngeal tension. However, these features are not consistent and therefore videostroboscopy cannot be used as a definitive diagnostic tool for AdSD and MTD.

At this time, there is no gold standard diagnostic method to differentiate AdSD and MTD, although modifications to the perceptual analysis of voice and the use of vocal fold imaging, particularly videostroboscopy have been suggested, neither provide a definitive diagnosis.

Transcranial magnetic stimulation (TMS) offers a noninvasive method of evaluating cortical excitability and has been used to measure cortical excitability in other forms of dystonia such as focal hand dystonia (FHD). TMS may provide invaluable information in determining a differential diagnosis between AdSD and MTD.

TRANSCRANIAL MAGNETIC STIMULATION

As eluded to above, transcranial magnetic stimulation (TMS) is a tool used to assess: 1) integrity of the cortical spinal and corticobulbar tract 2) cortical excitability and 3) cortical mapping and 4) conduction time in health and pathological individuals (Pascual-Leone, Rothwell, Davey, Wasserman, & Puri, 2002).

Mechanism

TMS is a neural stimulation tool based on the principals of electromagnetic conduction which was discovered in 1831 by Michael Faraday (Wassermann et al., 2008). The TMS coil conducts an electrical current into a magnetic field which can pass painlessly through the skull creating an electrical field within the neuronal tissue and causing current to flow

into an excitable neuron. There are parameters that can be altered to modify the effect of the TMS including the intensity, interstimulus spacing and stimulus duration; if the parameters are set in such a way, magnetic stimulation can induce depoloarization of the neuronal membrane leading to an action potential, referred to as a motor evoked potential.

What is being stimulated?

The magnetic field induced by TMS is thought to excite pyramidal neurons transsynaptically, inducing I-waves (indirect waves) in contrast to transcranial electrical stimulation, which directly excites pyramidal tract axons, inducing D-waves (Terao, 2002). Therefore, the use of TMS is optimal for assessing cortical excitability as I-waves are subject to changes in cortical excitability more than D-waves (Kujirai et al., 1993). When the intensity of TMS stimulation is set at or above the resting motor threshold (RMT), it induces depolarization of the alpha motor neuron, resulting in a muscle response, or motor evoked potential (MEP). If the stimulus intensity is below that of the RMT, it can inhibit depolarization which, in turn abolishes an action potential from taking place.

Coil type and orientation

There are two types of coils commonly used in TMS research; a round coil and figure of eight. The figure of eight coil consists of two interconnected coils; one with a clockwise directed current and the other counter clockwise. Due to the interaction of the directed currents, the juncture between the two coils is the point of greatest stimulator output and therefore offers a more focal point of stimulation (sweet spot) than a round coil. The direction of the current, dependent on the orientation of the handle of the coil, stimulates

different axons based on speed of conduction (Pascual-Leone et al., 2002). Since motor neurons for the hand are oriented perpendicular to the precentral gyrus and interneurons interacting with them are oriented parallel to the surface of the cortex, a horizontal current, particularly posterior-anterior current flow is optimal for stimulation. Therefore, optimal handle position for stimulating the hand motor cortex is a posterior-lateral orientation as it projects an anterior to posterior current flow perpendicular to the central sulcus. In contrast, an inferior handle position leads to current flow that is parallel to the central sulcus, optimal for stimulation of the masseter motor cortex (Guggisberg, Dubach, Hess, Wuthrich, & Mathis, 2001).

The depth of stimulation attained by TMS is dependent upon the dimensions and type of coil, range from 4mm (Pascual-Leone et al., 2002) to 2cm (Epstein, Schwartzberg, Davey, & Sudderth, 1990) which is optimal for stimulating cortical neurons. The TMS coil can produce a maximum magnetic field of 1.5-2.0T, depending upon the design (Epstein et al., 1990). As mentioned above, the figure of eight coil allows for a more focal presentation of the stimulus which is beneficial in controlling for spread of signal to areas of the cortex, or periphery that are not of interest. Only if the stimulator is set at 150% of the subject's threshold can the side loops of a figure of eight coil influence the cortex beneath (Wassermann et al., 2008). Therefore, use of a figure of eight coil at intensities less than 150% of subject's threshold will result in more focal presentation of stimulation and avoid effects of spread.

Safety

Historically, single pulse TMS used for measuring cortical excitability has resulted in few reports of adverse effects. In animal studies, no adverse effects were reported after rats

(100 pulses at 2.8 Tesla) and rabbits (5000 pulses at 2.4 Tesla) were presented with single pulse TMS. However, a permanent increase in auditory thresholds in animals after TMS stimulation has been reported (Counter, Borg, Lofquist, & Brismar, 1990). In addition, a temporary increase in auditory thresholds in humans following TMS has been reported (Pascual-Leone, Valls-Sole, Wassermann, & Hallett, 1994). A few authors have reported seizures induced by single pulse TMS but only in patients with large cerebral infarcts, specifically cortical lesions, for review see (Pascual-Leone et al., 2002). These findings have resulted in strict guidelines in the use safe use of TMS (Chen et al., 2008). Contraindications for single pulse TMS include: pacemaker, metal implants such as aneurysm clips or coils, and those with pre-existing epilepsy due to the potential of TMS to provoke a seizure which continues to be debated (Chen et al., 2008). In his review of the 1996 workshop on the risks and safety issues of TMS and rTMS, Wassermann concluded there are no substantial risks in the use of TMS in health and disease (for review see Wassermann, 1998. However, Kratz (2011) reported a TMS induced seizure in an individual who was not predisposed to seizures during single pulse motor threshold testing (Kratz, 2011) re-emphasizing the importance of screening, full disclosure of risks to potential subjects and a plan of care in the event of a seizure.

Cortical Excitability Measures

Single pulse

Single Pulse (SP) TMS measures the integrity and excitability of the cortical spinal and corticobulbar tract. A single stimulus pulse at a predetermined starting intensity is presented to the target area of the primary motor cortex and the EMG response of the target muscle is recorded. From this both the amplitude and the latency of the MEP can

be evaluated to determine a person's susceptibility to TMS (Pascual-Leone et al., 2002). Although, the latency of MEPs can be consistent over time in healthy subjects, the amplitude has high variability in healthy participants with an even higher variability in patients with CNS disorders (Catano, Houa, Caroyer, Ducarne, & Noel, 1996). Single pulse MEP values are used as the unconditioned response when calculating paired pulse responses. The single pulse MEP latency can be used to calculate total conduction time which helps evaluate the integrity of the descending motor pathway.

Stimulus response curve (SR Curve)

SR curve is a basic measure of excitability that measures how the MEP changes as a function of increases in the intensity of the stimulus. Although there is high variability both between subjects and within subjects, a pattern in MEP responses has been reported in the healthy population; as the stimulus intensity is increased, the MEP amplitude of the target muscle increases until saturation is reached. In peripherally stimulated muscles, responses will eventually saturate before the action potential is elicited, as such the SR curve is considered a cortical measure. Factors influencing the size if the MEP may include the number of motor neurons discharging more than once in response to the stimulus and voluntary contraction of the target muscle which leads to reduced threshold, shorter latency and increased amplitude of the MEP (Wassermann et al., 2008).

Paired Pulse: Short interval cortical inhibition (SICI)

SICI is a technique that uses a conditioning / test paradigm that is sensitive to measuring intracortical inhibitory mechanisms by stimulating intracortical synapses, particularly $GABA_A$ connections (Manganotti et al., 2002). The effect, reflected by EMG response is

a function of two parameters: conditioning and test stimulus intensity as well as the duration between the conditioning and test pulse, referred to as the interstimulus interval (ISI). A subthreshold conditioning pulse was found to reduce the amplitude of synaptically evoked cortical spinal volleys (I-waves) (Di Lazzaro et al., 1999) and is thought to excite $GABA_A$ergic intracortical inhibitory interneurons that have latencies between 1-5 ms (Ziemann et al., 1996). Therefore, the effect of the conditioning stimulus is driven by the interval between the conditioning pulse and the test pulse. Pulses separated by a 3 ms interval have been shown to have an inhibitory effect due to excitation of the above mentioned $GABA_A$ergic inhibitory interneurons. The resulting effect is referred to as short interval cortical inhibition (SICI) (Kujirai et al., 1993). SICI responses appear to be present in muscles at rest and in slight contraction (Ridding, Sheean, et al., 1995).

Paired Pulse: Intracortical facilitation (ICF)

ICF is similar to the technique described above, use of a subthreshold conditioning stimulus followed by a suprathreshold test stimulus. However in ICF, an ISI of 10-15ms is used. Responses to ICF appear to be influenced by current direction more than SICI and are optimal when an anterior current flow is presented (Ziemann, Rothwell, & Ridding, 1996). Currently, ICF is thought to reflect the excitability of the glutamatergic receptor circuitry of excitatory interneurons (Ziemann, Chen, Cohen, & Hallett, 1998). The resulting effect is referred to as intracortical facilitation (ICF). It has been reported low level contraction reduces or abolishes the ICF responses in healthy subjects (Ridding, Sheean, et al., 1995).

Cortical silent period

Cortical silent period (CSP) is a measure of the effect of the stimulus on the target muscle during contraction and is useful in measuring cortical inhibition in health and disease. CSP provides information regarding intracortical inhibition. After TMS induced MEP, a silent period in both limb muscles (E. M. Wassermann et al., 2008) and facial muscles (Jaberzadeh, Sakuma, Zoghi, Miles, & Nordstrom, 2008) have been reported. The duration of the CSP is influenced by the intensity of the stimulus but not the level of contraction in the target muscle. In the cortical spinal tract, the CSP can be explained using a segmental level rationale. The initial portion of the CSP is thought to be due to suppression of H-reflexes, due to Renshaw inhibition which is a spinal mechanism (Cantello, Gianelli, Civardi, & Mutani, 1992; Chen, Lozano, & Ashby, 1999). The later portion of CSP is thought to be due to suppression of voluntary motor drive by $GABA_B$ receptor-mediated inhibition in the cortex (Tergau et al., 1999). The mechanisms for CSP in the trigeminal motor system are still under investigation and evidence of these mechanisms will be discussed in the next section.

TMS studies in healthy masseter

The masseter is responsible for closing the jaw by elevating the mandible and plays a key role in voluntary biting, mastication, swallowing and speech production. Descending projections course along the corticobulbar tract from the facial region of M1 to the trigeminal motor nuclei housed within the pons and finally, through the mandibular branch of the trigeminal nerve to innervate the masseter. TMS has been used to study the neural circuitry of the masseter in healthy adults. However, differences between

trigeminal motor and spinal motor systems need to be taken into consideration when investigating the intracortical inhibition of those systems. The trigeminal and spinal motor systems differ in a number of ways. Firstly, facial muscles do not act on joints and do not have proprioceptors. Secondly, motoneurons of jaw-closing muscles such as the masseter do not act on reciprocal inhibition (Nakamura, 1980) and lastly, animal research has shown the trigeminal system to lack axon collaterals and recurrent inhibition from Renshaw cells(Kidokoro, 1986). In addition, MEPs from facial and masticatory muscles cannot be elicited at rest, the muscle must be in active contraction in order for an MEP to be elicited (Cruccu, Berardelli, Inghilleri, & Manfredi, 1990; Jaberzadeh, Pearce, Miles, Turker, & Nordstrom, 2007; Ortu et al., 2008). Because of these differences, it cannot be assumed the corticobulbar system will have the same inhibitory responses and mechanisms as the cortical spinal system.

Although some reports conclude masseter to be innervated in a symmetrical fashion; equal projections from ipsilateral and contralateral primary motor cortices (Guggisberg et al., 2001; Kuypers, 1958), others report cortical projections to masseter to be primarily contralateral (Butler, Miles, Thompson, & Nordstrom, 2001; Cruccu, Berardelli, Inghilleri, & Manfredi, 1989; McMillan, Graven-Nielsen, Romaniello, & Svensson, 2001; M. A. Nordstrom et al., 1999; Pearce, Miles, Thompson, & Nordstrom, 2003). The latency

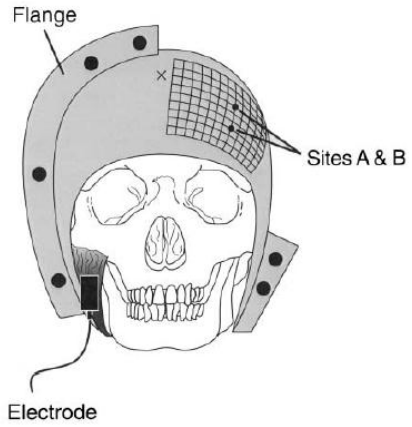


Figure 8: Image of stimulation site for masseter (Blitzer, Lovelace, Brin, Fahn, & Fink, 1985; McMillan, Watson, & D., 1998)

values suggest corticobular neurons project monosynaptically onto trigeminal motoneurons. Evidence from a single motor unit study conducted by Pearce, et al (2003) confirmed masseter motoneurons receive asymmetrical monosynaptic input from the primary motor cortex and the majority of masseter motoneurons received short-latency excitatory input from the contralateral hemisphere (Pearce et al., 2003). Because masseter is a muscle rarely at complete rest, differences in TMS technique must be employed in order to obtain optimal, reliable responses. Unlike the hand, preactivation of the masseter must occur in order to record an MEP (Butler et al., 2001; Cruccu et al., 1989; Macaluso, Pavesi, Bonanini, Mancia, & Gennari, 1990; McMillan et al., 1998). The standard preactivation level established for optimal MEP recording ranges from 10% of maximal contraction (McMillan et al., 1998) to 20% of maximal contraction (Desiato, Bernardi, Hagi, Boffa, & Caramia, 2002).

A reliable cortical map of the human masseter muscle was found using TMS during a teeth clenching task (Blitzer et al., 1985; McMillan et al., 1998) (Fig.8). This site was confirmed by cortical topography study conducted by Watson, et al. (2000). Previous work in animals and humans has suggested cortical projections to the jaw and small muscles in the hand may have task-specific organization (Hoffman & Luschei, 1980). The use of TMS confirmed masseter cortical maps were task dependent (Watson, Walshaw, & McMillan, 2000). The amplitude of masseter MEP is significantly higher during a bilateral biting task compared to a unilateral biting task (Butler et al., 2001). Cruccu, et al (1989) reported a short-latency response of ~3ms in the ipsilateral masseter representing stimulation of the trigeminal root ganglion. Considering the location of the

trigeminal root ganglion, coil orientation is of utmost importance when targeting the masseter motor cortex to avoid peripheral stimulation.

Guggisberg, Dubach, Hess, Wuthrich, & Mathis, (2001) studied the influence of coil orientation on responses from masseter using TMS to determine optimal coil position. These authors hypothesized the pyramidal cells of the masticatory muscles are less abundant compared to hand muscles, and as such they may be less responsive to trans-synaptic stimulation which is achieved in the hand with a 45 degree postero-lateral coil orientation. Positioning the coil at a 120 degree position resulted in lower thresholds and reliable MEPs of greater amplitude in the contralateral masseter muscle (Fig.9). The authors suggest this orientation excites the pyramidal fibers directly rather than trans-synaptically which is optimal for stimulating masseter (Guggisberg et al., 2001). Another author reported rotating the coil 120 degrees away from the midline with the handle pointing forwards and laterally resulted a posteriormedial current flow and reliable responses in masseter (Ortu et al., 2008). In addition to coil orientation, Guggisberg et al. (2001) also described the optimal scalp position for eliciting reliable MEPs from masseter as an area between 4-10 cm lateral to the vertex and 0-4 cm frontal to the bi-auricular

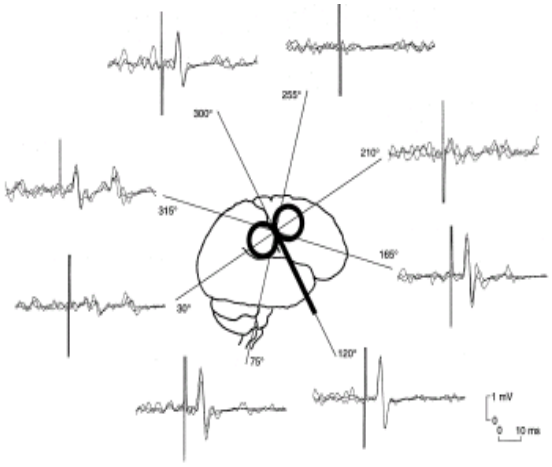


Figure 9. MEP from the masseter at different TMS coil orientation. The greatest and most consistent MEPs were found at 120 and 300 degrees (Guggisberg, 2001).

line. These findings are in agreement with past studies reporting optimal scalp landmarks for eliciting masseter responses (McMillan et al., 1998; Ortu et al., 2008; Watson et al., 2000). In addition, Ortu, et al. (2008) reported a coil position between 6-8 cm laterally from the vertex resulted in an ipsilateral masseter response due to stimulation of the trigeminal nerve (Ortu et al., 2008).

CSP

TMS is a viable method for investigating intracortical inhibition of the facial muscles in health and disease, particularly masseter (Cruccu, Inghilleri, Berardelli, Romaniello, & Manfredi, 1997). Although the CSP in the cortical spinal system reflects the suppression of motor cortical output through mechanisms discussed previously, the CSP in the corticobulbar tract, particularly the trigeminal system is not completely understood. Like the silent period in the spinal system, the silent period in the trigeminal system is a central mechanism, not peripheral as no silent period was reported in masseter after stimulation to the periphery (Cruccu et al., 1997). Unlike the spinal motor system, the trigeminal system has neither Renshaw cells nor axon collaterals to account for the silent period. Sowman, et al (2008) reported eliciting a silent period after a jaw jerk reflex and suggested the masseter silent period to be primarily due to suppression of the motoneurons in the trigeminal nucleus in the pons rather than purely a cortical phenomenon. In addition, they reported the descending volleys that act on the brainstem interneurons either 1) inhibit masseter motor neuron pools or 2) increase presynaptic inhibition of Ia afferent terminals (Sowman, Flavel, McShane, Miles, & Nordstrom, 2008). However, there are no Ia inhibitory interneurons at the brainstem level of the

trigeminal system, they are under cortical control (Pearce et al., 2003). Jaberzadeh, et al (2008) found an absence of an MEP and presence of a silent period when the individual was presented with the same stimulus intensity and suggested the silent period is primarily intracortical due to no corticobulbar projection to the brainstem to elicit the MEP (Jaberzadeh, Sakuma, Zoghi, Miles, & Nordstrom, 2008). All authors conclude the CSP in the trigeminal motor system appears to be segmental, similar to the spinal motor system, with brainstem and cortex having roles in the phenomenon (Cruccu et al., 1997; Jaberzadeh et al., 2008; Ortu et al., 2008; Pearce et al., 2003; Sowman et al., 2008). CSP duration in masseter is not dependent upon level of muscle contraction, but rather is dependent upon stimulus intensity and coil orientation, (Jaberzadeh et al., 2008; Ortu et al., 2008) which may explain the variability in CSP values across past studies (Table 1) (Cruccu et al., 1997; Jaberzadeh et al., 2008; Ortu et al., 2008; Sowman et al., 2008).

SICI

The use of paired pulse TMS has been used to investigate the short-interval intracortical inhibition (SICI) in the trigeminal motor system, specifically the digastric muscle (Jaberzadeh et al., 2007) and masseter (Ortu et al., 2008) in healthy adults. Results indicate SICI is present in the trigeminal motor system.

In the only known study conducted measuring SICI in masseter of healthy individuals, Ortu, et al. (2008) was able to record consistent inhibitory responses and reported SICI in masseter appeared to be influenced by intensity of the conditioning and test stimuli as well as the level of voluntary contraction but not coil orientation (Ortu et al., 2008). Reliable SICI responses were recorded during 10% of maximum voluntary contraction

(MVC) but the response was reduced or abolished when the contraction levels were at 25% of MVC (Ortu et al., 2008). Jaberzadeh, et al (2007) also found the effect of a conditioning stimulus was reduced during active contraction of the digastric muscle, which is within the trigeminal motor system as well (Jaberzadeh et al., 2007). In addition, Ortu, et al (2008) reported a conditioning stimulus intensity of 70% of AMT (90% of AMT resulted in no inhibitory response) and test pulse intensity of .5mV threshold resulted in reliable SICI responses.

ICF

At this time little is known about the SICF response in masseter. In the only known study of SICF in masseter, a strong *inhibitory* response to a conditioning stimulus of 70% of AMT and a test stimulus of .5mV threshold at ISIs of 10 and 15ms was reported, which was unexpected as ISIs of 10-15 ms are typically facilitory in other areas of M1 (Ortu et al., 2008). This response was thought to be due to the auditory reflex which was stimulated by the subthreshold discharge of the coil causing suppression of the MEP (Ortu et al., 2008). In addition, voluntary contraction not only reduces the threshold of the SICF but can completely abolish the response in hand (Ridding, Sheean, et al., 1995) and could account for the suppression of MEP in masseter. The effects of auditory reflex and voluntary contraction make obtaining a SICF response in masseter challenging.

Cortical excitability in focal hand dystonia

TMS to motor cortex with FDI as the target muscle resulted in shortened CSP in those with focal hand dystonia, indicating dysfunction of inhibitory interneurons within the motor cortex in those with FHD (Borich et al., 2009; Murase & Rothwell, 2005; Rona,

Berardelli, Vacca, Inghilleri, & Manfredi, 1998). After low frequency rTMS to PMC, significant prolongation of CSP was reported which indicates the lack of inhibition in the primary motor cortex in dystonia may be due to overexciatation in the premotor cortex (Murase & Rothwell, 2005). In addition, individuals with FHD have demonstrated less inhibition to SICI than healthy individuals while the target muscle is at rest (Ridding, Sheean, et al., 1995; Cathy M. Stinear & Winston D. Byblow, 2004). Another study revealed no significant differences in SICI responses between healthy controls and individuals with FHD in a slightly contracted muscle suggesting a muscle in slight contraction may mask between group differences (Rona et al., 1998). In addition, SICI responses during a selective task were increased in healthy controls but not in those with FHD suggesting dysfunctional task dependent SICI mechanisms in those with FHD (Butefisch, Boroojerdi, Chen, Battaglia, & Hallett, 2005)

Cortical Excitability in cranial and cervical dystonia

Limited investigations of cortical excitability in cranial and cervical dystonia have reported shortened CSP duration in those with spastic torticollis (Amadio et al., 2000), blepharospasm and oromandibular dystonia (Curra, Romaniello, Berardelli, Cruccu, & Manfredi, 2000). In addition, a suppression of SICI responses was reported in those with blepharospasm (Sommer, 2002).

Quartarone et al., (2008) presented paired associative stimulation to abductor pollicis brevis and first dorsal interosseus (neither having dystonic symptoms) to subjects with cervical and cranial dystonia, hemifacial spasm (HFS) and healthy controls. They reported a larger increase in cortical spinal excitability and a loss of topographical

specificity of the targeted regions in those with dystonia compared to those with HFS and healthy controls (Quartarone, Morgante, et al., 2008).

To provide evidence to support focal dystonia as a more generalized disorder of inhibition, TMS was used to measure cortical excitability in orbicularis oculi, orbicularis oris, sternocleidomastoid (SCM) and abductor pollicis brevis (APB) in two forms of focal dystonia (blepharospasm and cervical dystonia) compared to controls. Single pulse TMS revealed no differences in MEP latency or amplitude in the target muscles across groups. When compared to controls, the mean duration of CSP was significantly shorter in both orbicularis oris and orbicularis oculi, but not APB, in both the blepharospasm and cervical dystonia groups. Only people with cervical dystonia exhibited a significantly shorter CSP in SCM (Cakmur et al., 2004). This raises the question, is the impaired cortical inhibition in dystonia limited to the cortical pathway of the affected areas of the body?

The purpose of this study is to investigate if individuals with dystonia, particularly AdSD have generalized cortical excitability across both cortical spinal and corticobulbar tracts and if the differences in cortical excitability can be used in the differential diagnosis of AdSD versus MTD.

CHAPTER 2

Specific Aims

Transcranial magnetic stimulation (TMS) will be used to quantify and compare the cortical excitability in subjects with AdSD, MTD and healthy controls through a comprehensive evaluation of cortical excitability measures from the first dorsal interosseus and masseter muscle contralateral to stimulated hemisphere, including: (a) stimulus response curve (SR curve) (b) cortical silent period (CSP), (c) short interval cortical inhibition (SICI) and d) intracortical facilitation (ICF).

Aim 1: Determine and compare the cortical excitability of cortical spinal (CS) tract and corticobulbar (CB) tract in subjects with AdSD, MTD and healthy controls.

> **$H_{1.A)}$** *The cortical excitability in subjects with AdSD will be significantly different than healthy controls and those with MTD in two descending motor pathways (CS tract and CB tract). Those with AdSD will show: decreased slope of SR curve, shortened CSP duration, less inhibition (SICI) and greater facilitation (ICF) indicating higher excitability. There will be no significant difference in cortical excitability in CS and CB tracts between healthy controls and subjects with MTD.*

Aim 2. Determine the relationship between cortical excitability measures and perceptual voice characteristics.

> **$H._{2.A.)}$** *A strong negative correlation will be found between the total CAPE-V score and the cortical silent period suggesting the greater the disinhibition, the more severe the voice disorder.*

Aim 3. Determine if measures of intracortical inhibition and facilitation can be used to assist in differential diagnosis of AdSD from MTD.

> **$H._{3.A.)}$** *TMS can be used to assist in the differential diagnosis of AdSD.*

CHAPTER 3

PRELIMINARY DATA

Past TMS studies have shown differences in cortical excitability in subjects with FHD and healthy controls (Borich et al., 2009; N. Murase & Rothwell, 2005). Preliminary data was on a single subject with oromandibular dystonia (OMD) to test the feasibility of the applying TMS to a patient population with dystonia involving muscles innervated by corticobulbar neurons.

Methods

A single subject design was used to compare cortical excitability between age matched healthy controls (n=5) and one subject with OMD. All subjects were seated comfortably in a reclining chair and fit with a swim cap in order to mark cranial landmarks. Surface electrodes were placed on the contralateral and ipsilateral masseter and on the belly of the first dorsal interosseus (FDI) in a belly tendon montage. Using a figure of eight TMS coil positioned with the handle 45° posterolateral (FDI) and 100 ° anterolateral (masseter) to the midsagittal line of the head (Fig. 10). The vertex and approximate hand region were marked on the swim cap and single pulses at 55% of maximum stimulator output were delivered until a motor evoked potential (MEP) was elicited. The coil was systematically moved in an anterior and lateral orientation until a maximum MEP was elicited. This location was labeled as the "hot spot" and used to document the resting

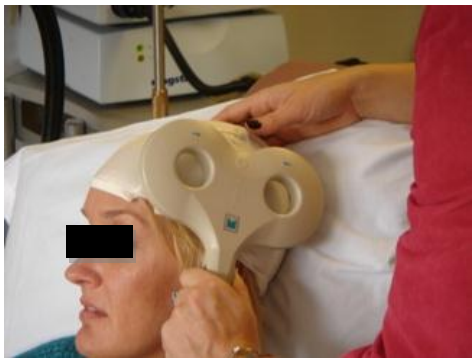


Figure 10. Optimal TMS coil placement to stimulate face/mouth region of primary motor cortex.

motor threshold (RMT), defined as the lowest intensity that induced an MEP of ~100 mV in approximately 50% of 10-20 consecutive trials (Rossini et al., 1999). Cortical excitability measures were taken contralaterally and included: single pulse (SP), cortical silent period (CSP), paired pulse including short interval cortical inhibition (SICI) and intracortical facilitation (ICF) were obtained for the masseter and the FDI. Ipsilaterally, only single pulse measures were taken.

Results

Full wave rectified EMG traces for FDI CSP and masseter CSP were analyzed by

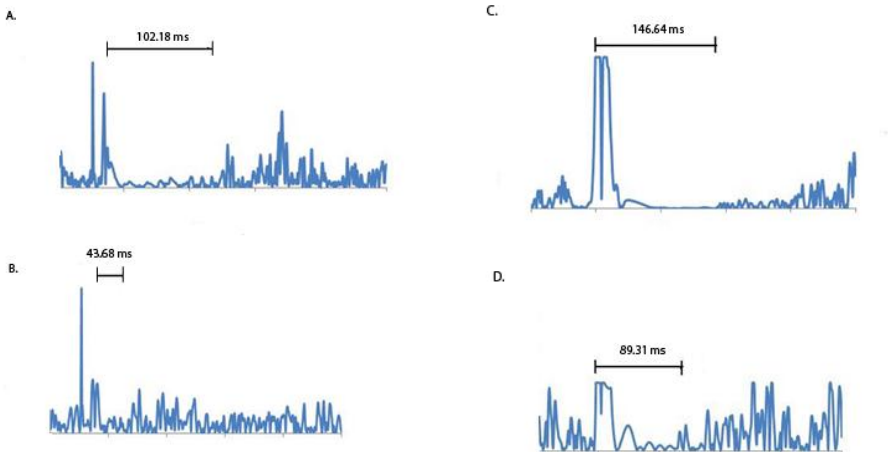


Figure 11. Rectified EMG trace of cortical silent period (CSP) in (left) masseter in: A) healthy (102.18 ms) and B) subject with oromandibular dystonia (43.68 ms) and (right) first dorsal interosseus in C) healthy (146.64 ms) and D) subject with oromandibular dystonia (89.31 ms).

identifying the onset of the CSP, defined as the MEP facilitated by the TMS pulse and the offset, defined as the point at which the EMG signal returned to 50% of prestimulus activation (FDI) and average prestimulus activation (masseter) (Fig. 11). An outlier in the healthy group was removed (n=4). The subject with OMD demonstrated shorter masseter CSP duration, mean 60.04 ms $\pm$19.04 [95% CI 64.15-108.62] and FDI, mean 100.82 ms ± 22.90 [95% CI 102.67-189.58] (Table 2).

Table 1. Cortical excitability results for subject with OMD

Outcome Measure	Mean	SD	Healthy 95% CI
CSP in FDI	100.82ms	22.90	102.67 - 189.58
CSP in Masseter	60.04ms	19.04	64.15 - 108.62
SP at 100% Contralateral	50.45mV	31.30	0 - 233.81
SP at 120% Contralateral	77.82mV	49.04	0 - 547.68
SP at 100% Ipsilateral	102.24mV	74.85	0 - 405.15
SP at 120% Ipsilateral	245.43mV	247.82	87.52-373.63
MEP Latency in Masseter	10.08ms	3.47	7.32-13.89

Motor evoked potential MEPs were recorded for the masseter muscle both contralateral and ipsilateral to the stimulated hemisphere at 100% and 120% of RMT to determine optimal response. Single pulse data was analyzed by calculating the peak to peak value for each trial and averaging those values (Table 1). The mean SP at both intensities is not different from the healthy group. The mean MEP latency for masseter in healthy controls was 10.61ms $\pm$ 2.64 [95% CI 7.32-13.89] for the subject with OMD, 10.08 ms $\pm$ 3.47, supporting a central pathophysiology in OMD.

Discussion

These preliminary data show likely success in using the proposed TMS techniques to investigate the pathophysiology of other forms of focal dystonia, specifically, AdSD. Although a difference in SP between healthy subjects and subject with OMD was not observed, it should be noted, the SP measure is highly variable in healthy subjects as well as those with neurologic conditions (Catano et al., 1996) which is also reflected in these data. In order to control for this variability, a measure that is more reliable and sensitive to intracortical inhibition, such as a 1mV threshold or active motor threshold (AMT) instead of RMT, further described in methods section, could be used (E. M. Wassermann et al., 2008). Chen and Curra (2004) (R. Chen & Curra, 2004) found motor neurons

recruited at higher intensity (higher than RMT) were more susceptible to measures of intracortical inhibition.

In addition, these preliminary data are in agreement with past research that has shown a decrease in CSP duration in subjects with dystonia. In healthy subjects, a CSP for the first dorsal interosseus (FDI) has been identified as approximately 141 ms (Borich et al., 2009). A shortened or absent CSP suggests impaired cortical inhibition, specifically an abnormality in inhibitory interneurons and has been reported in subjects with FHD (Borich et al., 2009; N. Murase & Rothwell, 2005; A. Quartarone, Rizzo, & Morgante, 2008). These data also demonstrate that impaired cortical inhibition can be widespread in a subject with dystonia which is supported by the findings of Cakmur, et al (2004) and Quarterone, et al. (2008) (Cakmur et al., 2004; A. Quartarone et al., 2008) who reported an increase in cortical excitability across the sensorimotor system exhibited in muscles that did and did not have dystonic symptoms. This suggests a more generalized disturbance of intracortical inhibition is imperative in understanding the pathophysiology of dystonia.

CHAPTER 4

CURRENT STUDY

The purpose of the proposed study is to determine if subjects with AdSD exhibit increased excitability across cortical pathways and to determine if cortical excitability measures can be used to assist in differentiating between AdSD and MTD. Specific aims for this study are: 1) Determine the cortical excitability of the primary motor cortex as it projects to cortical spinal (CS) tract and corticobulbar (CB) tract in subjects with AdSD, MTD and healthy controls 2) Compare differences in cortical excitability in the primary motor cortex in subjects with AdSD, MTD and healthy controls 3) Determine if cortical excitability measures are feasible to assist in the differential diagnosis of AdSD and MTD 4) Determine the relationship between cortical excitability measures on the severity of perceptual voice characteristics and 5) Determine the relationship between clinician-based perceptual measures and client-based measures in the AdSD and MTD populations.

Experimental Design and Methods

An *a priori* power analysis was conducted based on data from previous work comparing cortical excitability as measured by CSP in healthy subjects and those with FHD (Borich et al., 2009). True difference between the means = 28, Sigma1 = 22, Sigma 2 = 20.47 with 80% power and an alpha level of .05 revealed a sample size of 10 subjects per group would be needed to identify a large effect (d=.8). Due to difficulty recruiting subjects diagnosed with MTD, the number of subjects in that group was 5.

Subjects

Of eighty potential subjects initially contacted by mail or phone, fifty-five right handed subjects, ages 18-75 were screened for eligibility. Of those, twenty-one were excluded for 1) not meeting inclusion criteria (n=5), 2) declining participation (n=10) and 3) other reasons (n=6) including transportation, geography and scheduling conflicts. Thirty-two subjects were enrolled in the study: AdSD group (n=12), MTD group (n=5) and healthy control group (n=15). Data from two subjects in the AdSD group were not used due to one subject having been diagnosed with other forms of dystonia after participating and data from the other was not used due to difficulty finding an age matched peer. Of 15 subjects in the healthy control group, data from ten subjects was used in the final analysis. Five were excluded from the

Table 2. Group age and gender demographic data.

Group	**N**	**Gender**	**Age**			
			Mean (years)	**SD**	**CI lower**	**CI upper**
Healthy	10	Male(5) Female(5)	53.5	6.9	53	54
AdSD	10	Male(5) Female(5)	53.9	6.7	53	55
MTD	5	Male(2) Female(3)	51.6	16.1	48	55

Table 3. Individual subject demographics

Subject ID	**Gender**	**Age (yrs)**	**Diagnosis**	**Symptom duration (months)**		**Healthy controls ID**	**Gender**	**Age (yrs)**
02	M	53	AdSD	120		15	M	54
03	M	20	MTD	6		N/A		
04	M	50	AdSD	408		14	M	52
07	F	55	AdSD	96		05	F	53
08	M	54	AdSD	96		10	M	56
09	M	66	AdSD	228		11	M	64
10	F	58	MTD	12		N/A		
11	F	51	MTD	12		N/A		
12	F	62	AdSD	216		09	F	63
13	F	45	AdSD	36		13	F	43
14	F	60	AdSD	36		12	F	58
15	M	56	MTD	120		N/A		
16	F	50	AdSD	48		01	F	51
17	M	44	AdSD	18		07	M	43
18	F	63	MTD	24		N/A		

analysis due to 1) uninterpretable data (n=2) and 2) no age match peers in the AdSD group (n=3). Data for all subjects in the MTD group was used. Thus, final analysis was conducted on twenty-five subjects (Table 3).

All subjects were screened for voice symptoms and TMS safety and gave written, informed consent according to the Declaration of Helsinki prior to participation (Appendix A). The study was approved by the University of Minnesota Clinical Translational Science Institute (CTSI) and Institutional Review Board (IRB) (approval #0608M91226).

Research group 1: AdSD. Subjects in this group were diagnosed with AdSD based on a comprehensive voice evaluation by an experienced speech-language pathologist (SLP). As an additional assurance of the diagnosis of AdSD all subjects had documented 1) symptom task specificity 2) little to no benefit from behavioral voice therapy as documented by progress reports and verbal report from the subject; and 3) a positive outcome after botulinum toxin injection as reflected by their responses to screening questions. Any treatment with botulinum toxin injection must have been received $\geq$ 3months prior to study participation. Inclusion criteria of task specificity, increased difficulty with voiced versus voiceless phonemes and little to no benefit from traditional voice therapy were selected based on past reports of differential diagnosis between AdSD and MTD (C. L. Ludlow et al., 2008; Schlotthauer, 2010).

Research group 2: MTD. Subjects in this group were diagnosed with MTD based on a comprehensive voice evaluation by an experienced SLP. As an additional assurance of

the diagnosis of MTD, all subjects had documented 1) voice symptoms that are not task specific 2) experienced benefit from traditional voice therapy and/or 3) have had little to no benefit from low dose botulinum toxin injection as reflected by their responses to screening questions. Benefit from behavioral voice therapy was determined by progress reports written by the primary SLP and verbal report from the subject. The subjects in this group had a perceivable voice disorder at the time of the study. Individuals with a history of MTD but who have since been fully rehabilitated through traditional voice therapy were excluded.

All subjects in the AdSD and MTD groups had a comprehensive voice evaluation at the Lion's Voice Clinic including a perceptual voice assessment to document the nature, frequency and severity of various perceptual voice characteristics and a videostroboscopy interpreted by an otolaryngologist to rule out vocal fold pathology and an SLP to describe the movement patterns of the vocal folds. Results of the voice evaluation were documented in an evaluation report along with a diagnosis of MTD or AdSD (Appendix B).

Control group. Subjects in this group had no history of speech, voice or swallowing disorders, laryngeal surgery, gastroesophageal reflux disease, dystonia or any neurological condition that could affect laryngeal function.

Subjects in all groups were screened for TMS safety including no history of seizures, neurologic conditions such as stroke or degenerative disease, metal implanted devices

(excluding dental fillings) and pregnancy. Subjects in the AdSD and MTD groups were recruited through the Lion's Voice Clinic at the University of Minnesota, other voice clinics in the Minneapolis / St. Paul area and posting on the National Spasmodic Dysphonia Association (NSDA) website. Healthy control subjects were recruited through other posted advertisements (Appendix C). Data collection was carried out at University of Minnesota Clinical Translational Science Institute (CTSI).

Procedures

Voice Sample

A voice sample of all research subjects was video recorded prior to the presentation of TMS. The severity of voice characteristics was assessed by an expert listener, blinded to the subject's diagnosis, at a later date using the Consensus of Auditory Perceptual Evaluation of Voice (CAPE-V). The CAPE-V is a reliable, valid measure of the severity of an individual's voice characteristics (Zraick et al., 2011) . The Voice Handicap Index (VHI) was also presented to all research subjects for them to complete as a measure of the magnitude of the voice disorder on the subject's quality of life. The VHI is a reliable 30 item subjective rating tool completed by those with voice disorders. There are three areas including: physical subscale, functional subscale and social subscale (Jacobson et al., 1997). Copies of all perceptual voice measures can be found in Appendix E.

TMS

TMS was used to test the cortical excitability of the primary motor cortex (M1). Electromyographic (EMG) traces from target muscles were acquired using a bandpass filter width of 20-2000 Hz and sensitivity of 100 μV (FDI) and 500 μV (masseter).

(Cadwell Laboratory, Washington). Each subject was seated comfortably in a reclining chair and silver chloride disc electrodes (1 cm in diameter) were affixed to the contralateral and ipsilateral masseter and FDI using a belly/tendon montage. The reference electrode for the masseter was placed on the angle of the mandible as described by Guggisberg, et al. 2001 (Guggisberg et al., 2001) (Fig. 12). Recordings were first taken from the masseter with the ground electrode placed on the chin. The ground electrode was then moved to the outer surface of the hand for FDI recordings. The subject was then fit with a swim cap to allow cranial landmarks to be first approximated and later confirmed using TMS. The midpoint of the sagittal plane was identified by locating the midpoint between nasal bone and occiput. The midpoint of the coronal plane was identified by locating the midpoint between the left and right tragus. The location at which these two points intersected is referred to as the vertex. To identify the location of the hand region of M1, markings were then made 5 cm anterior and 5cm lateral from the vertex and connected with straight lines to create a triangle. The hypotenuse of this triangle is the estimated hand region of M1. A constellation of 4 spots in a north, south, east, west

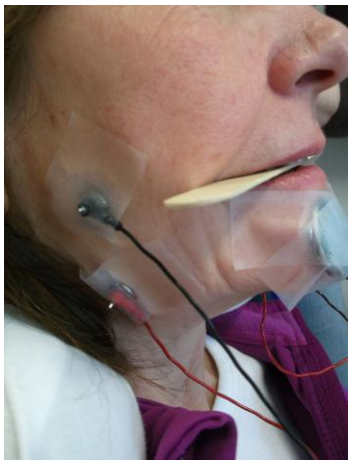


Figure 12. Image of electrode placement for right masseter.

Figure 13. Image of coil orientation to elicit masseter response.

pattern around the presupposed "hotspot" were then marked to guide the exact location of the hand region. Markings were made at 4cm and 5cm laterally from the vertex on the bi-auricular line to identify the face region of M1 (Guggisberg et al., 2001). Approximately 4cm lateral to the vertex resulted in reliable, optimal masseter responses. To find the optimal position for activating FDI and masseter, a 70-mm figure of eight TMS coil (Magstim Co. LTD, Whitland, UK) was placed over the subject's head and directed orthogonally to the scalp with the handle directed postero-laterally 45° for FDI (Borich et al., 2009) and antero-laterally 100° for masseter (Guggisberg et al., 2001) (Figure 13). Single pulses were delivered at approximately 0.1 Hz starting at an intensity of 55% of the maximum stimulator output. The

Table 4. Motor evoked potential (MEP) thresholds and definitions

Threshold	Definition
Resting motor threshold (RMT)	Lowest intensity of stimulation needed to elicit reproducible MEPs of ~50μV in the target muscle in approximately 50% of trials (Rossini, 1994).
Active motor threshold (AMT)	Lowest intensity of stimulation needed to elicit reproducible MEPs of ~100μV in the target muscle in approximately 50% of consecutive trials while the subject is maintaining a slight contraction, 8-12% of maximal contraction (Wasserman, 2008).
.3mV threshold	Lowest intensity of stimulation needed to elicit reproducible MEPs of .3mV (300μV) in the target muscle in 3 out of 5 trials.
1mV threshold	Lowest intensity of stimulation needed to elicit reproducible MEPs of 1mV (1000μV) in the target muscle in 3 out of 5 consecutive trials

Table 5. Cortical Excitability Outcome Measures

Term		Measure
SR Curve	Stimulus response curve	The MEP that is elicited in response to a single pulse at various stimulation intensities
CSP	Cortical silent period	Period of electromyographic suppression caused by TMS in an actively contracting muscle.
SICI	Short interval intracortical inhibition; paired pulse	Suppression of a test MEP by a subthreshold conditioning stimulus 1-5 ms earlier.
ICF	Intracortical facilitation; paired pulse	Facilitation of a test MEP by a subthreshold condition stimulus 10-15 ms earlier.

intensity level was adjusted until an MEP was elicited, at which time the coil was systematically moved 1cm anterior, posterior, medial and lateral to the presupposed "hotspot" until a maximal, consistent MEP was observed. The individual "hotspots" for FDI and masseter were then marked on the swim cap and used to record thresholds including; resting motor threshold (RMT) and 1mV threshold for FDI and active motor threshold (AMT) and .5mV threshold for masseter (Table 4). Single pulse recordings at the 1mV threshold (FDI) and .3mV threshold (masseter) were taken to determine latency in masseter and to be used as a baseline for paired pulse measures. In addition, four measures of cortical excitability were collected, including: stimulus response curve (SR curve), paired pulse (SICI and SICF) and cortical silent period (CSP) (Table 5).

Cortical excitability testing

Stimulus Response Curve

Eight pulses of each stimulus intensity 110%, 120%, 130% and 140% of RMT (FDI) and AMT (masseter) were presented in a predetermined random fashion with an interpulse interval of between 5-10 ms.

Paired Pulse

Twenty paired pulse stimuli (10 SICI and 10 SICF) interspersed with single pulse stimuli were presented in a predetermined random fashion. In FDI, a conditioning pulse at 80% of RMT was followed by a test pulse at the 1mV threshold, in masseter the conditioning pulse was 80% of AMT and test pulse was .3 mV threshold. SICI was determined with 3ms interstimulus interval and SICF with a 10ms interstimulus interval (Kujirai et al., 1993; Wassermann et al., 2008).

Cortical Silent Period

In another test of intracortical inhibition, the CSP was collected by asking subjects to perform a slight contraction (25% of maximum contraction) while a suprathreshold pulse was delivered to the predetermined "hotspot". In FDI, the subjects performed a finger abduction task against a strain gauge at 25% of maximal voluntary contraction. Force was displayed on an oscilloscope to indicate target contraction level. In masseter, subjects used a tongue depressor in a transverse plane (bilateral bite) to perform a contraction that was approximately 25% of MVC. The test pulse applied was the 1mV threshold (FDI) and .3mV threshold (masseter) 3-5 seconds after the contraction was initiated and subjects were then asked to relax 2-3 seconds after stimulation. Ten CSP measurements were taken with 30 second rest interval between each trial for each muscle.

Data Processing

MEP values of each measure were determined by calculating the peak to peak amplitude by identifying the data point with the highest (peak) amplitude and lowest (trough) amplitude. The peak to peak amplitude was calculated as the difference between the peak and trough for each trial. For detailed description of data processing procedures, see Appendix F.

Cortical excitability measures

Stimulus Response Curve

MEP responses for the SR curve protocol were analyzed by calculating the peak to peak amplitude for each trial and finding the average for each stimulus intensity, 110%, 120%, 130% and 140% of the RMT (FDI) and AMT (masseter) for each subject. The SR curve

was then plotted for each subject within a group and a mean SR curve was created for each group. Regression slopes and areas under the curve were then calculated for each muscle in each subject.

Single Pulse and Paired Pulse

MEP amplitude was measured by calculating the peak-to-peak values for each trial and then averaged for each subject. The averaged SICI and SICF MEP values were used as the numerator to establish a ratio between the paired pulse MEP to the single pulse MEP *(PPmean / SPmean)*. This ratio provides a mechanism to examine the amplitude of the conditioned MEP relative to the unconditioned MEP and can be expressed as a percent difference (Kujirai et al., 1993). Past research has reported the SICI MEP to be smaller than the single pulse MEP, represented by a value <1 and the SICF MEP to be larger than the single pulse MEP, represented by a value >1.

MEP Latency was measured after single pulse presentation only by calculating the difference between the TMS artifact and the MEP, the value was translated into a duration(ms) based on the sweep of the recording.

CSP

Techniques for manual analysis of CSP duration and reliability has been described (Kimberley et al., 2009) and were used in this analysis. CSP onset was defined as the TMS induced MEP, the offset was identified as the point at which the muscle contraction returned to 50% of the prestimulus average, for FDI (Kimberley et al., 2009) and the prestimulus average, for masseter.

Perceptual Voice Measures

CAPE-V

A licensed, certified speech language pathologist with experience in voice disorders, referred to as "expert listener", listened to the audio recordings of subjects in the MTD and AdSD groups and scored each using the CAPE-V. The listener was blinded to the subject group and had experience with scoring the CAPE-V.

Statistical analysis:

All variables were assessed for normality. Log transformation was conducted for all variables violating the assumption of normality. Following transformation satisfactory normality was achieved and back transformation was conducted for interpretation. A correlation analysis was conducted to identify covariates and an ANCOVA was conducted as appropriate.

A 3 group one way ANOVA was conducted for each response variable with an alpha value of 0.05 used to determine group differences. Post hoc analyses were run as appropriate with bonferroni to correct for multiple comparison (.05/2=.025). Effect sizes, 0.10 (small effect), 0.30 (medium effect) and 0.50 (large effect) were reported as suggested by Coehn (1992). A post hoc power analysis was conducted for all nonsignificant findings.

Chapter 5

Results

The primary purpose of this study was to determine and compare the cortical excitability in people with AdSD, MTD and healthy controls. The first hypothesis stated the cortical excitability in subjects with AdSD would be significantly different than healthy controls in both descending motor pathways. Specifically, participants with AdSD would show: decreased slope of SR curve, increased area under the curve, shortened CSP duration, less inhibition (SICI) and greater facilitation (SICF) indicating higher excitability compared to the other two groups and that there would be no difference between MTD and healthy control participants. This hypothesis was partially supported; suggesting AdSD may have different cortical neurophysiology compared to healthy controls. All data are reported as mean ± SE [95% CI lower bound, 95% CI upper bound]. All statistical analyses were conducted using SPSS, version 20 and statistical output tables can be found in Appendix E.

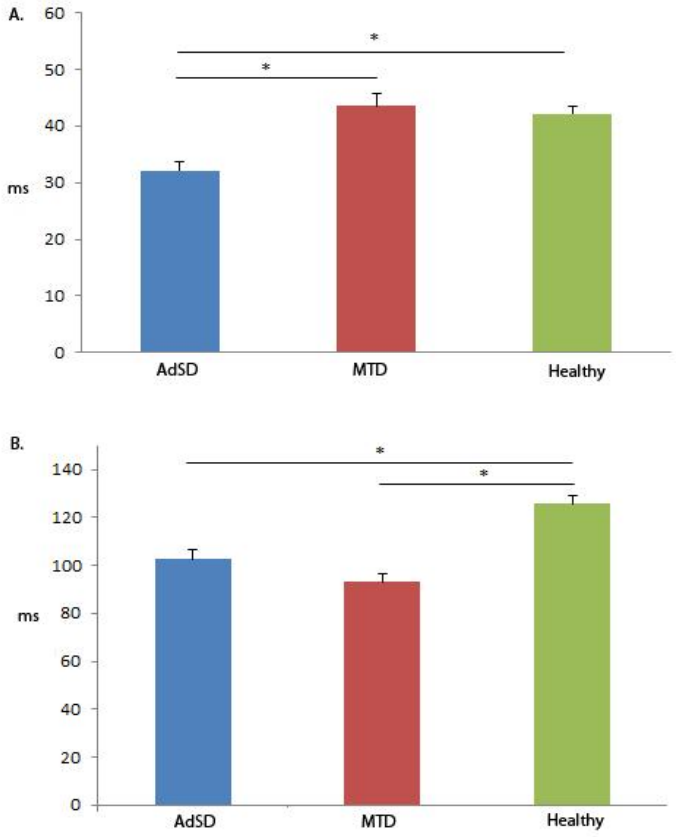


Figure 14. Mean CSP in masseter (A.) and FDI (B) in AdSD (n=10), MTD(n=5) and healthy (n=10) groups. * = p<0.001

Cortical Silent Period

A correlation analysis revealed

age to be a covariate of CSP in FDI, r(244) = -0.135, p = 0.035 and masseter, r(227) = 0.186, p = 0.005. Masseter CSP was compared across groups using an ANCOVA with age as a covariate: F(2,223) = 21.461, p < 0.001 which revealed significant differences in

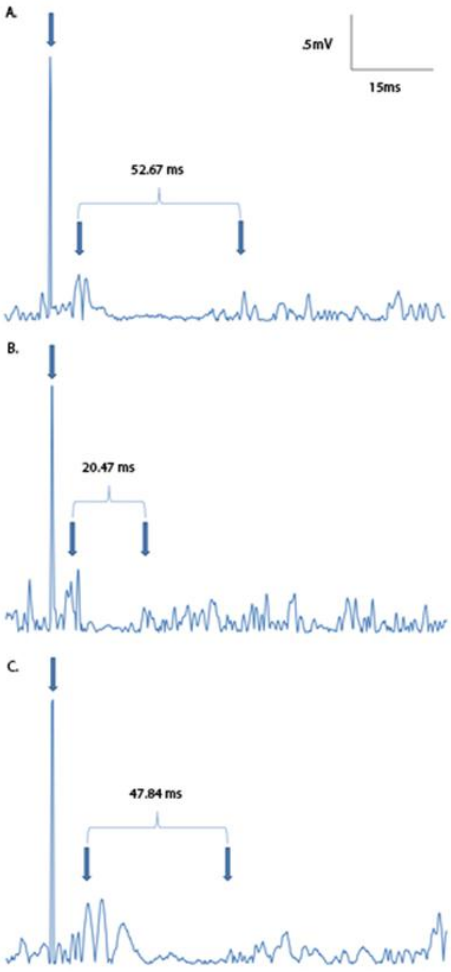


Figure 15. Rectified EMG traces of cortical silent period in masseter in (A) Healthy, (B) AdSD and (C) MTD.

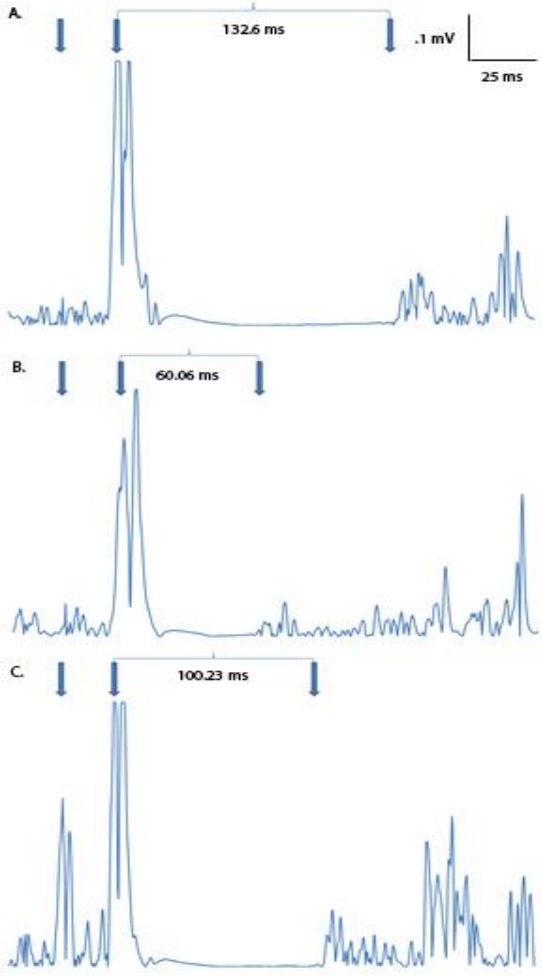


Figure16 . Rectified EMG traces of cortical silent period in FDI in (A) Healthy, (B) AdSD and (C) MTD.

masseter CSP across groups. A post hoc pairwise comparison revealed no significant difference in masseter CSP between the healthy and MTD groups (p = 0.192). However, the AdSD group demonstrated significantly shorter masseter CSP duration than the MTD group (p < 0.001) and the healthy group (p < 0.001), with a medium to large effect size, r

= 0.44 supporting the above hypothesis. Specifically, the healthy control group had a mean CSP of 42.12 ms ± 1.33 [40, 45]; AdSD, 32.06 ms ± 1.6 [29,35] and MTD, 43.57 ms ± 2.18 [39, 48] (Fig. 14). Rectified EMG of individual masseter CSP traces can be reviewed in Figure 15. Results from an ANCOVA, with age as a covariate, also revealed statistically significant differences in FDI CSP duration across groups, $F(2,240) = 24.142$, $p < 0.001$. A post hoc pairwise comparison revealed statistically significant differences between the AdSD and healthy groups ($p < 0.001$) supporting the hypothesis, however differences in FDI CSP between MTD and healthy groups were significant ($p < 0.001$) and no significant differences were found between the AdSD and MTD groups ($p = 0.551$) with a medium to large effect size, $r = 0.43$. These results do not uphold the above hypothesis. Mean FDI CSP duration in the healthy group was 125.72 ms ± 3.43 [119, 133], AdSD group, 102.62 ms ± 3.9 [95, 110] and the MTD group, 92.99 ms ± 3.36 [86, 100] (Fig.14). Rectified EMG of individual FDI CSP traces can be reviewed in Figure 16. The AdSD group demonstrated significantly shorter CSP duration in both masseter and FDI compared to both the healthy group and in masseter compared to the MTD group supporting the hypothesis that decreased intracortical inhibition compared to healthy subjects is a feature of AdSD. However, no significant differences were found between the healthy and MTD groups in FDI CSP, which didsupport the hypothesis.

SICI

In SICI, another measure of intracortical inhibition, an ANOVA revealed no significant differences in mean SICI between groups in both masseter, $F(2,20) = .348$, $p = 0.710$ with a small effect size, $r = 0.18$ and FDI $F(2,22) = 3.03$, $p = 0.069$ having a medium to large effect size, $r = 0.46$, showing a trend toward significance. The mean masseter SICI

indexes revealed the healthy group, 1.05 ± 0.06 [0.9, 1.2] and MTD group, 1.06 ± .08 [0.08, 1.3] to have, on average, excitatory (ratio > 1) responses whereas the AdSD group demonstrated inhibitory (ratio < 1) responses, 0.99 ± .05 [0.9, 1.1) (Table 7). Mean FDI SICI indexes for each group were inhibitory; healthy group 0.60 ± 1.27 [0.2, 1.7], AdSD group 0.51 ±1.27 [0.2, 1.4] and MTD group 0.24 ± 1.44 [0.06, 0.9] (Table 6). Results from the statistical analysis do not support the hypothesis. Examples of individual EMG traces for the SICI measure can be reviewed for masseter (Fig. 17) and FDI (Fig. 18).

Table 6. Number of subjects who demonstrated inhibitory or facilitory responses to short interval cortical inhibition (SICI) and the mean index.

	Masseter		FDI	
	Mean Inhibitory index Ratio <1	Mean Facilitory index Ratio >1	Mean Inhibitory index Ratio <1	Mean Facilitory index Ratio >1
Healthy group	0.92 (n=6)	1.25 (n=4)	0.50 (n=7)	1.31 (n=3)
AdSD group	0.86 (n=3)	1.23 (n=7)	0.50 (n=8)	1.26 (n=2)

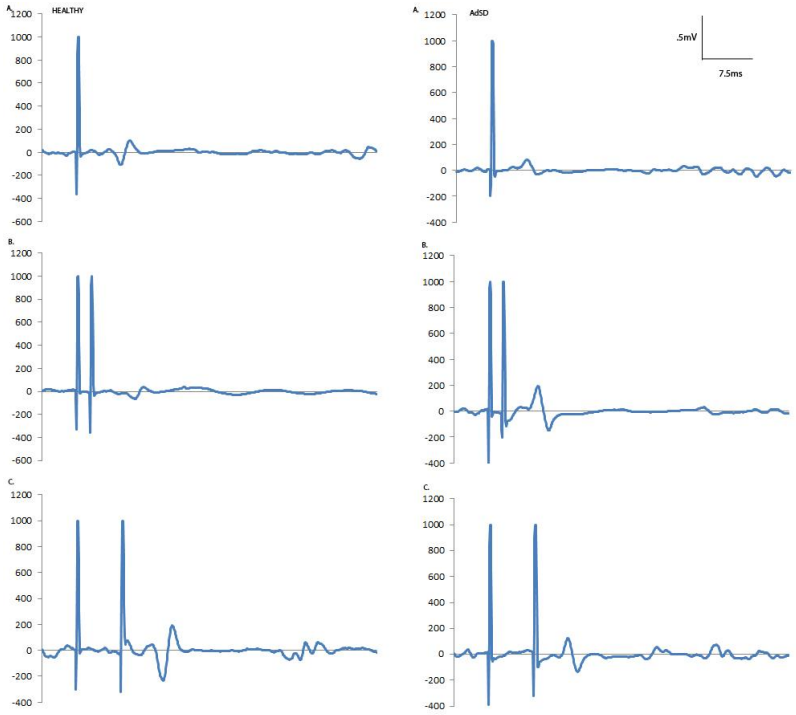


Figure 17. Masseter A) single pulse (SP), B) short interval cortical inhibition (SICI) and C) intracortical facilitation (ICF) in healthy (left) and AdSD (right).

ICF

In ICF, a measure of intracortical facilitation, an ANOVA revealed no significant differences between groups in masseter, F(2,22) = 0.635, p = 0.540, with a small to medium effect size, r = 0.23 and FDI, F(2,22) = 0.502, p = 0.612 with a small to medium effect size, r = 1.21. Mean masseter ICF indexes were facilitory (>1) in the healthy group, 1.0 ± 0.06 [0.88, 1.1] and the AdSD group, 1.06 ± 0.13 [0.8, 1.35] and inhibitory (<1) in the MTD group, 0.86 ± 0.17 [0.4, 1.32]. In FDI, all groups demonstrated a facilitory response: healthy group, 1.63 ± 0.13 [1.3, 1.9], AdSD group 1.52 ± 0.35 [0.7, 2.3] and MTD group, 1.2 ± 0.3 [0.4, 2.0] (Table 7). Examples of individual EMG traces for the ICF measure can be reviewed for masseter (Fig. 17) and FDI (Fig. 18).

Table 7. Number of subjects having inhibitory or facilitory responses to intracortical facilitation (ICF) and the mean differences in the SICF index of the AdSD and healthy groups.

	Masseter		FDI	
	Mean Inhibitory index Ratio <1	Mean Facilitory index Ratio >1	Mean Inhibitory index Ratio <1	Mean Facilitory index Ratio >1
Healthy group	0.81 (n=4)	1.13 (n=6)	0.90 (n=1)	1.71 (n=9)
AdSD group	0.79 (n=6)	1.46 (n=4)	0.80 (n=4)	2.0 (n=6)

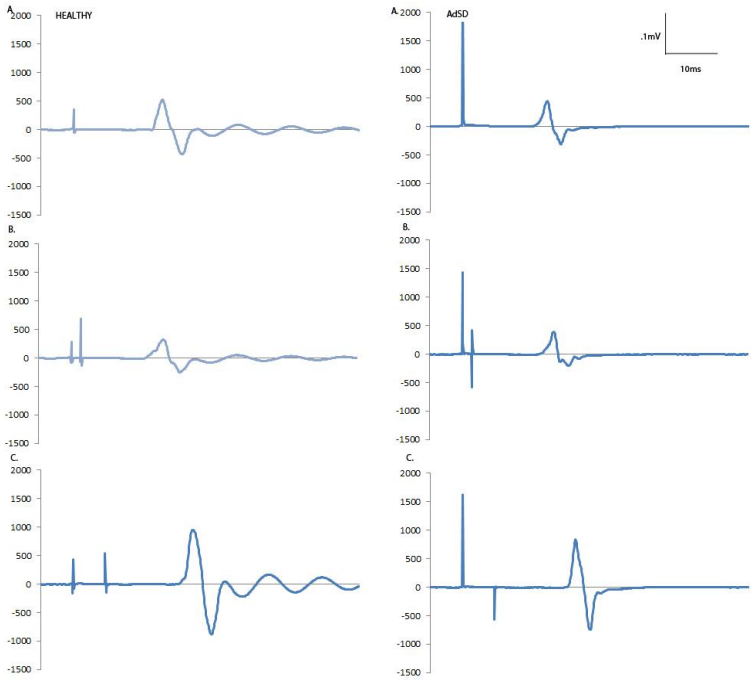


Figure 18 . FDI A) single pulse (SP), B) short interval cortical inhibition (SICI) and C) intracortical facilitation (ICF) in healthy (left) and AdSD (right).

SR Curve

The slope and area under the curve (AUC) provided additional information regarding cortical excitability. AUC values between groups were analyzed using an ANOVA which revealed no statistically significant differences between groups in contralateral masseter, F(2,22) = 0.058, *p* = 0.358, r = 0.18, ipsilateral masseter, F(2,22) = 0.378, *p* = 0.690, r = 0.18 and FDI, F(2,21) = 1.48, *p* = 0.250, r =0.35. Mean area under the curve values in contralateral masseter for healthy, 6627.41µV ± 345.92 [-1434, 14689]; AdSD, 7055.68µV ± 2635.79 [1093, 13018] and MTD, 3280.83µV ±556.22 [1736, 4825]. Ipsilateral masseter in healthy, 6496.03µV ± 1911.87 [2087, 10904]; AdSD, 11794.89µV ± 3926.06 [2913, 20676]; MTD, 7697.53µV ±2240.96 [1475, 13919]. And FDI in healthy, 21541.78µV ± 4725.20 [10645, 32438]; AdSD, 52508.72µV ± 16740.17 [14639, 90377]; MTD, 37750.80µV ± 8784.25 [13361, 62139] (Fig. 19). A correlation analylsis for slope identified gender to be a significant covariate for contralateral masseter, r(21)=-0.582, *p* = 0.014.

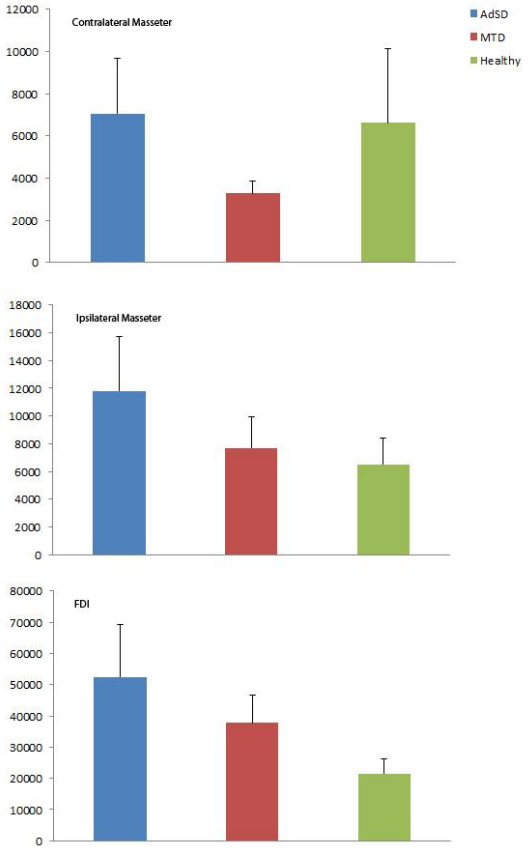


Figure 19. Mean area under the curve values (µV) for all groups.

An ANCOVA with gender as a covariate revealed no significant differences in slope between groups in contralateral masseter, F(2,17) = 0.739, *p* =0.492, large effect size, r = 0.58. An ANOVA revealed no significant differences in the slope of the SR curve between groups in ipsilateral masseter, F(2,22) = 1.62, *p* = 0.220, medium effect size, r = 0.36 and FDI, F(2,21) = 0.947, *p* =0.404 with a small to medium effect size, r = 0.29. The mean slope values for contralateral masseter in healthy, 0.91 ± 1.68 [0.20,4.03]; AdSD, 0.98 ± 1.33 [0.22, 5.39] and MTD, 2.04 ± 2.04 [0.21, 18]. For ipsilateral masseter in healthy, 3.04 ± 0.68 [1.43,4.65]; AdSD, 4.08 ± 0.85 [2.08, 6.08]; MTD, 6.56 ± 0.88 [3.7, 9.3]. And, for FDI in healthy, 37.2 ± 9.5 [14.68, 59.72]; AdSD, 66.97 ± 20.56 [18.35, 115.58] and MTD, 46.64 ± 4.22 [33.21, 60.07] (Fig. 20). The CSP in masseter was the most sensitive measure to determine differences between groups. The other measures

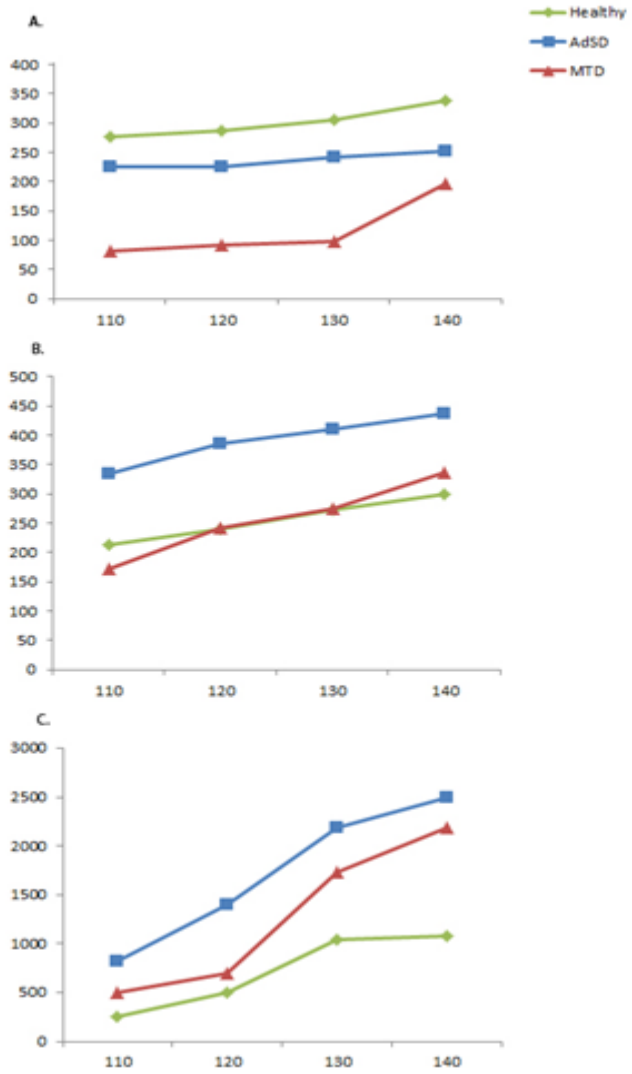


Figure 20 . Mean stimulus response curve slopes across groups in (A) contralateral masseter, (B) ipsilateral masseter and(C) FDI.

including SICI, ICF, slope and AUC did not support the hypothesis. A post hoc power analysis revealed sample sizes needed to obtain significance (Table 8). It is notable that an n of 40 in each group would have revealed differences between AdSD and healthy in FDI slope and FDI AUC. In addition, an n of 50 in each group would have revealed differences between AdSD and MTD in masseter slope, masseter AUC and FDI SICI. CSP in masseter and FDI in AdSD were significantly shorter than those in the healthy group supporting the hypothesis of widespread disinhibition in AdSD. To further investigate the CSP between the CS and CB tracts, a correlation analysis was conducted. A statistically significant linear relationship of moderate strength was found in CSP between FDI and masseter in the AdSD group, $r(85) = 0.506$, $p < 0.001$. This relationship was not found in the healthy group, $r(89) = 0.161$, $p = 0.126$ or the MTD group, $r(41) = 0.056$, $p = 0.723$. Another purpose of this study was to determine the relationship between neurophysiological and perceptual measures in those with AdSD. It was hypothesized a significant negative correlation would be found between the total CAPE-V score and the cortical silent period. Although a correlation analysis revealed a negative relationship between CSP and CAPE-V scores, it was weak in strength and did not reach significance

Table 8. Post hoc power analysis. Sample sizes required to find significance.

Measure	**Muscle**	**AdSD and Healthy**	**AdSD and MTD**	**Healthy and MTD**
CSP	FDI	n/a	155	n/a
Slope	Masseter (C)	70	46	153
	Masseter (I)	70	13	6
	FDI	38	67	72
Area under the curve	Masseter (C)	471	41	39
	Masseter (I)	93	85	39,960
	FDI	26	116	19
SICI	Masseter	115	89	16090
	FDI	257	15	12
ICF	Masseter	447	61	71
	FDI	921	123	27

(Table 9). These results do not support the above hypothesis and suggest there is no significant relationship between the neurophysiological response and the perceptual characteristics of the voice or CSP was not a sensitive measure to detect a true relationship. Significant correlations were found in those with AdSD between FDI RMT and CAPE-V total scores, r (11) = -.552, p = 0.05 and masseter AMT and CAPE-V total scores, r (11) = -.583, p = 0.037. These results indicate as the severity of the voice disorder increases, the motor threshold for masseter and FDI decreases.

Table 9. Correlation analysis between CAPE-V scores and cortical excitability measures.

	CAPE-V Total Score		
	Pearson's r	Significance	N
CSP Masseter	-.160	.681	9
CSP FDI	-.304	.427	9

Results from a two group ANOVA revealed no significant differences between the AdSD and MTD groups in the following variables of the CAPE-V: Breathy F(1,11) = 0.323, p = 0.581; Rough, F(1,11) = 0.020, p = 0.890; Strain, F(1,11) = 2.403, p = 0.149; Pitch, F(1,11) = 0.006, p = 0.939; Loudness, F(1,11) = 1.146, p = 0.307 and Total CAPE-V score, F(1,11) = 0.902, p = 0.363. Mean scores are reported in Table 10. Results indicate no differences in perceptual voice characteristics between groups supporting the need for additional differential diagnosis techniques. Based on the results of the cortical excitability testing, masseter CSP is a feasible method to assist in the differential diagnosis of AdSD.

Table 10. Mean scores of the Consensus Auditory – Perceptual Evaluation of Voice (CAPE-V) by group.

	AdSD				**MTD**			
	Mean	SE	CI lower	CI upper	Mean	SE	CI lower	CI upper
Breathy	21	6.65	6	36	27.75	9.62	-3	58
Rough	36.44	7.3	20	53	34	12.6	-6	75
Strain	39.8	7.15	23	56	22	5.5	4	40
Pitch	12.7	3.8	4	22	12.25	5.3	-5	29
Loudness	11	3.9	2	20	20.5	10.3	-12	53
Total	40.8	5.5	28	54	29.5	13.4	-13	72

Although, motor thresholds and MEP latency in masseter were not direct aims of this study, analysis was conducted to determine if differences existed between groups. Gender was identified as a covariate of masseter latency through correlation analysis of contralateral masseter, r(431) = -0.159, $p = 0.001$ and ipsilateral masseter, r(218) = -0.423, $p < 0.001$. An ANCOVA, with gender as a covariate, revealed differences between groups in latency duration of contralateral masseter, F(2,427) = 24.951, $p < 0.001$ and ipsilateral masseter F(2, 214) = 0.193, $p = 0.825$. A post hoc analysis of contralateral masseter revealed the AdSD group demonstrated a shorter latency compared to the healthy group that was statistically significant ($p < 0.001$). In addition, the MTD group had a significantly shorter latency than those in the healthy group ($p < 0.001$). There was no significant difference in contralateral masseter latency between the AdSD and MTD groups ($p = 0.147$). Mean latency values for masseter are reported in Table 11. Motor thresholds between groups using ANOVA revealed: masseter AMT, F(2,40) = 2.762, $p = 0.075$ and FDI RMT, F(2,39) = 1.34, $p = 0.272$. A post hoc power analysis revealed sample sizes of 18-72 per group were needed to detect differences in AMT and between 30-256 individuals per group to detect differences in RMT.

Table 11. Motor evoked potential latency in contralateral and ipsilateral masseter across groups.

	Contralateral Masseter				Ipsilateral Masseter			
	Mean	SE	CI lower	CI upper	Mean	SE	CI lower	CI upper
Healthy	7.83ms	1.02	7.1	8.6	4.36	1.02	4.3	4.8
AdSD	7.04ms	1.01	6.7	7.4	4.26	1.02	3.7	4.9
MTD	6.76ms	1.02	6.3	7.2	4.13	1.04	3.4	4.9

Each subject was asked a series of adverse effect questions after the TMS procedure (Appendix D). Two subjects, both in the AdSD group reported slight headache during the TMS procedure. The band of the swimcap was cut and relief was expressed by both

individuals. However, one continued to report headache after the TMS had concluded. A follow up phone call the next day revealed the headache subsided approximately 2 hours after the experiment.

Chapter 6

Discussion

The primary findings of this study revealed individuals with AdSD demonstrated an impaired intracortical inhibitory mechanism, specifically $GABA_B$ as measured by CSP, when compared to healthy controls and MTD. Differences were found in both corticobulbar and cortical spinal tract suggesting those with AdSD have dysfunction of intracortical inhibition that is not localized to the affected musculature or one descending motor system. Tests of other neurophysiological mechanisms were not sensitive enough to find differences between groups, suggesting dysfunction of $GABA_B$ mechanisms may be a primary pathophysiological feature in AdSD and $GABA_A$ and glutamateric mechanisms either play a supplementary role or are not impaired in the cortical pathways of the unaffected hand and masseter in AdSD. Additionally, the severity of voice characteristics did not correlate with the CSP suggesting the severity of the clinical features of AdSD may not be reflected by neurophysiological measures and therefore cannot be used to predict the severity of voice quality in individuals nor can the perceptual voice quality predict the neurophysiological state.

Is lack of intracortical inhibition a feature of AdSD?

This is the first known study to investigate intracortical inhibition in AdSD and determine if lack of intracortical inhibition may be a pathophysiological feature in AdSD similar to that reported in other forma of focal dystonia (Borich et al., 2009; Butefisch et al., 2005; Cakmur et al., 2004; Ikoma et al., 1996; Mavroudakis et al., 1995; Murase & Rothwell, 2005; Ridding, Sheean, et al., 1995; Rona et al., 1998).

Cortical Silent Period

The AdSD group demonstrated significantly shorter CSP duration in both masseter and FDI compared to the healthy group and in masseter when compared to MTD suggesting dysfunction of the $GABA_B$ mechanism within the primary motor cortex may be a pathophysiological marker in AdSD, similar to that observed in FHD (Borich et al., 2009; Quartarone et al., 2003; Siebner et al., 1999) and cranial dystonias (Curra et al., 2000). A retrospective power analysis revealed a sample size of 155 was needed in order to detect differences between AdSD and MTD in FDI CSP, suggesting CSP FDI is not a sensitive measure for differential diagnosis.

The duration of masseter CSPs were shorter than FDI CSPs in healthy individuals which is in agreement with past TMS studies of the trigeminal motor system (Pearce et al., 2003). This difference may be due to monosynaptic input to masseter which leads to less neurotransmitter release at the neuromuscular junction compared to poly synaptic inputs to muscles of the hand. As a result, $GABA_B$ inhibitory circuits in M1 have a weaker effect on corticotrigeminal neurons that innervate masseter, reflected in a shorter CSP duration compared to those that innervate the cortical spinal system (Cruccu et al., 1989; Di Lazzaro et al., 1998; Ortu et al., 2008; Pearce et al., 2003). Masseter CSP duration in healthy individuals in this study was similar to that found by Ortu, et al. (2008). CSP values were consistently elicited in FDI in all subjects, however, CSP in masseter were less consistent and a few traces were uninterruptable in the AdSD group due to a lack of EMG quiescence or due to the lack of inhibition which eliminated the presence of the CSP.

SICI

Although mean masseter indexes showed inhibitory responses (ratio < 1) in the AdSD group and facilitory responses (ratio > 1) in the healthy and MTD groups, observation of individual subject indexes revealed 70% of individuals in the AdSD group demonstrated facilitory responses to SICI compared to 40% in the healthy group. Facilitory responses to this measure indicates reduced intracortical inhibition, specifically in the $GABA_A$ mechanism which has been found in past reports that SICI was "less inhibited" in those with FHD (Quartarone et al., 2005; Ridding, Sheean, et al., 1995; Rona et al., 1998; Sommer, 2002; Cathy M. Stinear & Winston D. Byblow, 2004) and blepharospasm (Sommer, 2002) suggesting the $GABA_A$ mechanism in this population to be impaired. The majority of individual subjects in the healthy group had inhibitory responses to SICI, not reflected in the mean value, which is in agreement with past research of masseter (Ortu et al., 2008) and digastric (Jaberzadeh et al., 2007) which both reported inhibitory responses to SICI in a healthy population. The amount of contraction of a muscle does impact the SICI measure. It is feasible that the healthy individuals who demonstrated facilitory responses had an increased contraction of the masseter muscle during the measure, ~~as~~ however, this was not quantitatively monitored. No statistically significant differences were found between groups however, a retrospective power analysis revealed that with an n of 115 (masseter) and 257 (FDI) ~~,~~ a statistical difference could have been found.

Mean FDI indexes revealed inhibitory responses in all groups. Results in AdSD were not in agreement with past research which again reported reduced SICI in FDI in those with

FHD (Quartarone et al., 2005; Ridding, Sheean, et al., 1995; Rona et al., 1998; Sommer, 2002; Cathy M. Stinear & Winston D. Byblow, 2004) and blepharospasm (Sommer, 2002). This does not indicate that $GABA_A$ mechanisms are intact in those with AdSD, but rather this measure was not sensitive enough to detect differences in this size sample. A retrospective power analysis revealed a sample size of 257 was needed to detect differences between AdSD and healthy controls. Although SICI may not be sensitive enough to detect between group differences and no significance was found, observation of the number of individuals with facilitory vs inhibitory responses indicates if there may be a dysfunction of $GABA_A$ in those with AdSD which affects the corticobulbar tract to a greater degree than the cortical spinal tract.

ICF

The expected response to this measure is facilitation. Mean masseter indexes revealed facilitory responses (ratio > 1) in both the healthy and AdSD groups and inhibitory responses (ratio < 1) in the MTD group, however differences did not reach significance. Observation of individual indexes revealed 60% of the healthy group had facilitory responses compared to only 40% of those in the AdSD group. It is difficult to speculate why the majority of the individuals in the AdSD group had an inhibitory response to masseter ICF in masseter as no known reports of ICF in masseter have been published in health or disease. A possible explanation for inhibition in 40% of the healthy population leads one back to the monosynaptic nature of innervation in masseter. Similar to CSP, mean masseter ICF indexes in healthy were smaller, mean of 1.13 compared to FDI, mean of 1.63. Intracortical inhibitory circuits have been found to be weaker in the

corticotrigeminal system due to the monosynaptic input, it would be feasible to consider glutamatergic mechanisms may also be weaker, resulting in less glutamate at the neuromuscular junction resulting in inhibitory responses to ICF in masseter. Visual inspection of the mean indexes of those who had facilitory responses to ICF revealed larger mean facilitory response in AdSD (1.46) compared to healthy (1.13) although differences did not reach significance, this increased facilitation pattern is similar to that found in FHD in other forms of focal dystonia.

In FDI the responses were more consistant in the healthy group, with 90% displaying a facilitory responses, fewer individuals, 60% in the AdSD group demonstrated facilitory responses. Visual analysis revealed the mean facilitory index was larger in AdSD (2.0) compared to healthy (1.71) suggesting exaggerated facilitory responses in some individuals with AdSD that did not reach or trend toward significance. These results are similar to those found in individuals with FDI and blepharospasm where slightly exaggerated facilitatory responses, that trended toward significance, were found in FDI (Sommer, 2002). A retrospective power analysis revealed a sample size of 447 (masseter) and 921 (FDI) would have detected differences suggesting high variability in the measure. Enhanced ICF in unaffected musculature suggests dysfunction of surround inhibition (Sommer, 2002) which has been associated with focal dystonia (Berardelli et al., 1998). However, this study was unable to idendify this to be a feature in AdSD due to the variability in the ICF measure. Future work is needed to better understand the glutamatergic system in both the cortical spinal and corticobulbar tract in focal dystonia.

Stimulus Response Curve

Although differences in mean slope values did not reach significance, visual analysis revealed steeper slope in those with AdSD compared to healthy controls across all muscles tested and in FDI compared to individuals in the MTD group. In addition, visual analysis of mean SR curves revealed individuals with AdSD demonstrated consistently larger, but not significant, AUC values across all muscles compared to healthy and in both ipsilateral and contralateral masseter compared to MTD. Although these differences did not reach statistical significance, the pattern is striking and is similar to that reported in FHD (Ikoma et al., 1996; Mavroudakis et al., 1995) providing support that increased cortical excitability may be a feature of AdSD. A retrospective power analysis revealed a sample size for slope of 70 (masseter) and 38 (FDI) and AUC of 471 (masseter) and 26 (FDI) were needed to detect group differences between healthy and AdSD suggesting this neurophysiological measure appears to be more sensitive when testing the cortical spinal tract than the corticobulbar tract when comparing AdSD to healthy. To detect group differences in slope between AdSD and MTD, a sample size of 46 (masseter) and 67 (FDI) were needed and in AUC, 41 (masseter), and 116 (FDI). The slope and AUC of the SR curve are more sensitive measures for detecting cortical excitability differences than ICF in this population.

Is cortical disinhibition widespread in AdSD?

Results from the current study indicate impaired cortical inhibition is a pathophysiological feature of AdSD. Specifically, dysfunction of the $GABA_B$ mechanism in both corticobulbar and cortical spinal tracts, reflected in shortened CSP duration in asymptomatic muscles, masseter and FDI, supporting evidence of widespread

disinhibition in focal dystonia (Cakmur et al., 2004; Quartarone, Morgante, et al., 2008). These findings are in agreement with past reports of widespread disinhibition in unaffected muscles in FHD (Quartarone, Rizzo, et al., 2008; Sommer, 2002), blepharospasm (Cakmur et al., 2004; Sommer, 2002) and cervical dystonia (Cakmur et al., 2004).

The reason for this wide spread dysfunction isn't clearly understood, other authors have suggested the existence of a generalized disturbance of the inhibitory system in focal dystonia (Curra et al., 2000; Rona et al., 1998) and bilateral dysfunction of the inhibitory mechanism in those with unilateral focal dystonia has been reported with the use of paired pulse TMS (Ridding, Sheean, et al., 1995; Rona et al., 1998). In addition, shortened CSP duration has been found bilaterally with those with cervical dystonia (Curra, Berardelli, Rona, Fabri, & Manfredi, 1998). In addition, impairment of the inhibitory interneurons is not just a bilateral motor cortex phenomenon. Shortened CSP duration in regions not associated with affected musculature have been found in those with cranial dystonia (Cakmur et al., 2004; Curra et al., 2000), blepharospasm (Cakmur et al., 2004) and now in AdSD. This evidence supports widespread reduced intracortical inhibition to be a feature of focal dystonia, including AdSD providing rationale for the presence of more than one form of dystonia in some individuals.

Are neurophysiological measures correlated with perceptual voice severity?

Contrary to our hypothesis, this study revealed no correlation between CAPE-V total score and CSP in either masseter or FDI suggesting the severity of the voice disorder does not appear to be reflected in the degree of reduced intracortical inhibition in the

areas assessed in those with AdSD. Importantly however, this study did not target affected musculature. One might hypothesize symptom severity would more likely be correlated with CSP of the thyroarytenoid musculature. However Ridding, et al (1995) reported no correlation between symptom severity and neurophysiological measures in hand in those with simple writer's cramp (less severe symptoms) and dystonic writer's cramp (more severe symptoms). Both groups had reduced inhibition (at rest) with no significant differences between groups (Ridding, Sheean, et al., 1995). This evidence suggests that neurophysiological measures cannot predict symptom severity therefore it is imperative to include perceptual or behavioral measures as well as neurophysiologic measures when investigating focal dystonias. There is a lack of evidence surrounding the nature of symptom severity in dystonia which leads one to wonder what drives symptom severity? Future work is needed to determine if any neurophysiologic measure does correlate with symptom severity to help further elucidate the pathophysiology of this disorder.

Can CAPE-V differentially diagnose AdSD and MTD?

Results from this study confirm past research indicating perceptual voice assessment measures are not sensitive enough to differentially diagnose AdSD from MTD which leads to frequent misdiagnosis and ineffective treatment. There is a need for alternative diagnostic methods to differentially diagnose the two disorders to ensure accurate diagnosis and treatment. This study showed the feasibility in using TMS to assist in the diagnostic process and identified masseter CSP to be the most sensitive in detecting differences between groups.

Are cortical excitability characteristics in AdSD similar to those found in other movement disorders?

Decreased intracortical inhibition, as measured by shortened CSP duration and decreased inhibition to SICI, have been reported in movement disorders including Parkinson's disease (PD), Tourette's syndrome (TS), primary dystonia (Wassermann et al., 2008), focal hand dystonia (Butefisch et al., 2005; Ridding, Taylor, & Rothwell, 1995; C. M. Stinear & W. D. Byblow, 2004), cervical dystonia, blepharospasm (Sommer, 2002) and amyotrophic lateral sclerosis (ALS) (Vucic, Cheah, & Kiernan, 2011; Zanette et al., 2002). And now this decrease in intracortical inhibition, particularly $GABA_B$ has also been identified as a feature of AdSD. With more power, it is likely decreased inhibition through SICI may have been observed in those with AdSD similar to findings in other movement disorders, particularly other focal dystonias. Dysfunction of cortical inhibitory interneurons appears to be a shared feature of multiple movement disorders.

Exaggerated facilitory responses have also been reported in some movement disorders. For example, a significantly steeper slope of the stimulus response curve, interpreted as an increase in cortical excitability, has been reported in ALS (Zanette et al., 2002) and focal hand dystonia (Ikoma et al., 1996). In resting muscles in individuals with PD, a steeper slope and larger area under the curve was observed consistently, but did not reach significance (Valls-Sole et al., 1994) similar to that found in those with AdSD in the current study. The glutamatergic mechanism in these populations is not well understood as ICF in movement disorders has not been investigated to the same extent as SICI. However Zanette, et al. (2002) reported no differences in ICF between individuals with ALS and healthy controls; similar to results found in this study suggesting dysfunction of

the glutamatergic system may not be a primary pathophysiological feature of movement disorders or ICF is not a sensitive measure to detect glutamatergic dysfunction. There is more evidence to suggest movement disorders share a common feature; dysfunction of $GABA_A$ and $GABA_B$ mechanisms.

No differences in MEP threshold between those with AdSD and healthy were found, similar to that reported in PD, primary dystonia (Wassermann et al., 2008) and ALS (Vucic et al., 2011). Results from this study revealed a significant negative correlation between severity of symptoms and MEP thresholds; as severity of symptoms increased, MEP thresholds decreased. An area for future study could investigate changes in MEP threshold over time as symptom severity changed. If individual MEP thresholds did indeed decrease over time as symptom severity increased, it would indicate dysfunctional intracortical inhibitory system; the pathways are more excitable, making it easier to elicit an MEP.

How does cortical excitability in AdSD compare to stroke?

Much research has been done to investigate intracortical inhibition in individuals after stroke. Although, not classified as a movement disorder, comparisons of cortical excitability between movement disorders and stroke can assist in identifying similarities and differences in pathophysiological features. Similar to that found in dystonia, individuals with stroke have demonstrated decreased intracortical inhibition in the affected hemisphere as shown by decreased SICI (Wassermann et al., 2008). In addition, disinhibition in the affected hemisphere appears to be stronger in those with cortical lesions compared to subcortical lesions and it appears to be correlated with spasticity

(Liepert, 2006). In a longitudinal study, Manganotti, et al (2008) investigated intracortical inhibition in individuals status post cortical or subcortical stroke and reported all individuals had significantly reduced SICI in the acute phase (5-7 days post stroke). At 30 days post stroke, those that recovered hand function also demonstrated SICI had returned to "normal" and in those with persistent motor impairment demonstrated reduced SICI which is indicative of the relationship between motor symptoms and neurophysiological measures (Manganotti, Acler, Zanette, Smania, & Fiaschi, 2008). However, Hummel, et al (2009) reported reduction in SICI at rest and during movement preparation in individuals ≥ 1 year status post subcortical stroke, who had "good recovery of hand function" (Hummel et al., 2009) which indicates the status of intracortical inhibition cannot necessarily be predicted by motor recovery. Reduced inhibition has also been reported in the unaffected hemisphere in those with cortical stroke but not subcortical stroke suggesting location of the stroke influences cortical excitability outcomes in the unaffected hemisphere (Oh, Kim, & Paik, 2010). However, Butefisch, et al. (2008) reported reduced SICI in contralesional M1 in those with cortical and subcortical stroke (Butefisch, Wessling, Netz, Seitz, & Homberg, 2008). Reduced SICI in both hemispheres has also been reported focal dystonia (Berardelli et al., 1998; Ridding, Sheean, et al., 1995) suggesting dysfunction of intracortical inhibitory circuitry may be similar between dystonia and stroke. Reduced SICI in contralesional hemisphere may be an adaptive process of loss of interhemispheric inhibition (IHI) from lesioned to nonlesioned hemisphere (Butefisch et al., 2008). Loss of IHI has been reported in focal hand dystonia (Neslon, 2010) and stroke (Butefisch et al., 2008).

CSP is also altered as a result of stroke, specifically lesions outside the motor cortex result in an increased duration of CSP and lesions within the motor cortex result in decreased duration of CSP (Wassermann et al., 2008), similar to that found in focal dystonia and as shown in AdSD in this study. These findings suggest dysfunction of inhibitory interneurons within the primary motor cortex to be similar in those with cortical stroke and those with focal dystonia. However, since cortical lesions are not a characteristic of AdSD, shortening of the CSP in focal dystonia is more likely due to altered basal ganglia input which affects the activity of the intracortical inhibitory interneurons compared to damage to those neurons in stroke.

LIMITATIONS

As in all research, this study had limitations that should be considered. Of utmost importance is the lack of subjects in the MTD group to achieve a balanced study design. However, the post hoc power analysis revealed many variables would still have been underpowered even if the sample sizes were balanced. The a priori power analysis was conducted using mean and standard deviation values from past CSP findings and therefore resulted in other cortical excitability variables to be underpowered to detect significance. This suggests that CSP is the more sensitive measure, whereas other measures are more variable with less drastic differences between groups. In CSP testing, a load cell is generally used to keep the amount of effort applied consistent. While we did use a load cell for the FDI measure, no load cell was used to measure masseter contraction levels. This could have affected masseter SICI and MEP latency results since level of contraction does influence these variables. A bite force load cell was piloted in

the early stages of study design, but it required unilateral biting which has been associated with reduced excitability of corticobulbar neurons in contralateral M1. This device was abandoned due to technical difficulties and the unilateral biting position. In future work, a bilateral biting task should be used for examining the cortical excitability of the masseter (Michael A. Nordstrom, 2007). It may also be of benefit to assess cortical excitability using TMS during a symptom triggering task.

MTD and AdSD are known to be difficult to differentially diagnose, hence the purpose of this project. However, this difficulty is another limitation of the study, as another consideration is the difficult differential diagnosis of MTD and AdSD as it pertains to group assignment. It is possible the screening form created for this study may not have been sensitive enough for consistently accurate group assignment. This is unlikely however, as the criteria used in the screening questions included the primary perceptual features used by experienced speech language pathologists and otolaryngologists that distinguish the two disorders

Conclusion

This was the first known study investigating intracortical inhibitory and facilitory mechanisms in AdSD using TMS. Differences in intracortical inhibition in M1 were observed in those with AdSD compared with healthy controls, and in some instances, those with MTD. Masseter and FDI CSP were the most sensitive measures to capture these differences. SICI, ICF, slope and AUC were not sensitive enough to capture differences or simply are not dysfunctional in asymptomatic musculature in AdSD. However, differences in these measures may have been found in symptomatic

musculature, the thyroarytenoid, by stimulating laryngeal motor cortex, which should be an area of future investigation. Disinhibition was observed across central pathways, reflected in decreased CSP duration, affecting both corticobulbar and cortical spinal tracts suggesting disinhibition in AdSD is not focal, but widespread and is present in pathways leading to asymptomatic musculature. This provides rationale for individuals who have multiple forms of focal dystonia. Evidence from the current study indicates AdSD to be a disorder of generalized disinhibition, specifically of the $GABA_B$ mechanism similar to other forms of focal dystonia.

REFERENCES

. (Journal Article).

Abbruzzese, G., & Trompetto, C. (2002). Clinical and Research Methods for Evaluating Cortical Excitability. *J Clin Neurophysiol, 19(4)*, 307-321.

Adler, C. H., Edwards, B. W., & Bansberg, S. F. (1997). Female predominance in spasmodic dysphonia. *J Neurol Neurosurg Psychiatry, 63*(Journal Article), 688.

Ali, S. O., Thomassen, M., Schulz, G. M., Hosey, L. A., Varga, M., Ludlow, C. L., & Braun, A. R. (2006). Alterations in CNS activity induced by botulinum toxin treatment in spasmodic dysphonia: an H215O PET study. *Journal of Speech Language & Hearing Research, 49*(5), 1127-1146.

Altman, K. W., Atkinson, C., & Lazarus, C. (2005). Current and emerging concepts in muscle tension dysphonia: A 30 month review. *Journal of Voice, 19(2)*(Journal Article), 261-267.

Amadio, S., Panizza, M., Pisano, F., Maderna, L., Miscio, C., Nilsson, J., . . . Galardi, G. (2000). Transcranial magnetic stimulation and silent period in spasmodic torticollis. *American Journal of Physical Medicine & Rehabilitation, 79*(4), 361-368.

Aminoff, M. J., Dedo, H. H., & Izdebski, K. (1978). Clinical aspects of spasmodic dysphonia. *Journal of Neurology, Neurosurgery & Psychiatry, 41*(4), 361-365.

Aronson, A. E., Brown, J. R., Litin, E. M., & Pearson, J. S. (1968). Spastic dysphonia. I. Voice, neurologic, and psychiatric aspects. *Journal of Speech & Hearing Disorders, 33*(3), 203-218.

Bara-Jimenez, W., Catalan, M. J., & Hallett, M. (1998). Abnormal somatosensory homunculus in dystonia of the hand. *Ann Neurol, 44*, 828-831.

Berardelli, A., Rothwell, J. C., Day, B. L., & Marsden, C. D. (1985). Pathophysiology of blepharospasm and oromandibular dystonia. *Brain, 108*(Pt 3), 593-608.

Berardelli, A., Rothwell, J. C., Hallett, M., Thompson, P. D., Manfredi, M., & Marsden, C. D. (1998). The pathophysiology of primary dystonia. *Brain, 121*(Pt 7), 1195-1212.

Bielamowicz, S., & Ludlow, C. L. (2000). Effects of botulinum toxin on pathophysiology in spasmodic dysphonia. *Annals of Otology, Rhinology & Laryngology, 109*(2), 194-203.

Blitzer, A., Lovelace, R. E., Brin, M. F., Fahn, S., & Fink, M. E. (1985). Electromyographic findings in focal laryngeal dystonia (spastic dysphonia). *Annals of Otology, Rhinology & Laryngology, 94*(6 Pt 1), 591-594.

Borich, M., Arora, S., & Kimberley, T. J. (2009). Lasting effects of repeated rTMS application in focal hand dystonia. *Restorative Neurology & Neuroscience, 27*(1), 55-65.

Brin, M. F., Blitzer, A., & Stewart, C. (1998). Laryngeal dystonia: observations of 901 patients treated with botulinum toxin. *Adv Neurol., 78*, 237-252.

Butefisch, C. M., Boroojerdi, B., Chen, R., Battaglia, F., & Hallett, M. (2005). Task-dependent intracortical inhibition is impaired in focal hand dystonia. *Movement Disorders, 20*(5), 545-551.

Butefisch, C. M., Wessling, M., Netz, J., Seitz, R. J., & Homberg, V. (2008). Relationship between interhemispheric inhibition and motor cortex excitability in subacute stroke patients. *Neurorehabilitation & Neural Repair, 22*(1), 4-21.

Butler, S. L., Miles, T. S., Thompson, P. D., & Nordstrom, M. A. (2001). Task-dependent control of human masseter muscles from ipsilateral and contralateral motor cortex. *Experimental Brain Research, 137*(1), 65-70.

Byl, N. N., Merzenich, M. M., Cheung, S., Bedenbaugh, P., Nagarajan, S. S., & Jenkins, W. M. (1997). A primate model for studying focal dystonia and repetitive strain injury: effecs on the primary somatosensory cortex. *Physical Therapy, 77(3)*(Journal Article), 269-284.

Cakmur, R., Donmez, B., Uzunel, F., Aydin, H., & Kesken, S. (2004). Evidence of widespread impairment of motor cortical inhibition in focal dystonia: a transcranial magnetic stimulation study in patients with blepharospasm and cervical dystonia. *Advances in Neurology, 94*, 37-44.

Cantello, R., Gianelli, M., Civardi, C., & Mutani, R. (1992). Magnetic brain stimulation: the silent period after the motor evoked potential. *Neurology, 42*(10), 1951-1959.

Castelon Konkiewitz, E., Trender-Gerhard, I., & Kamm, C. (2002). Service-based survey of dystonia in Munich. *Neuroepidemiology, 21*, 202-206.

Ceballos-Baumann, A. O., Sheean, G., Passingham, R. E., Marsden, C. D., & Brooks, D. J. (1997). Botulinum toxin does not reverse the cortical dysfunction associated with writer's cramp. A PET study. *Brain, 120*(Pt 4), 571-582.

Chen, R., Cros, D., Curra, A., Di Lazzaro, V., Lefaucheur, J. P., Magistris, M. R., . . . Ziemann, U. (2008). The clinical diagnostic utility of transcranial magnetic stimulation: report of an IFCN committee. *Clinical Neurophysiology, 119*(3), 504-532.

Chen, R., Lozano, A. M., & Ashby, P. (1999). Mechanism of the silent period following transcranial magnetic stimulation. Evidence from epidural recordings. *Experimental Brain Research, 128*(4), 539-542.

Chhetri, D., Blumin, J., Vinters, H., & Berke, G. (2003). Histology of nerves and muscles in adductor spasmodic dysphonia. *Ann Otol Rhinol Laryngol, 112*(Journal Article), 334-341.

Childs, L. R., S.; Murry, T.; Blitzer, A.; Sulica, L. (2011). Patient perceptions of factors leading to spasmodic dysphonia: a combined clinical experience of 350 patients. *Laryngoscope, 121*(October), 2195-2198.

Chitkara, A., Meyer, T., Keidar, A., & Blitzer, A. (2006). Singer's dystonia: first report of a variant of spasmodic dysphonia. *Annals of Otology, Rhinology & Laryngology, 115*(2), 89-92.

Counter, S. A., Borg, E., Lofquist, L., & Brismar, T. (1990). Hearing loss from the acoustic artifact of the coil used in extracranial magnetic stimulation. *Neurology, 40*(Journal Article), 1159-1162.

Courey, M. S., Garrett, C. G., Billante, C. R., Stone, R. E., Portell, M. D., Smith, T. L., & Netterville, J. L. (2000). Outcomes assessment following treatment of spasmodic dysphonia with botulinum toxin. *Annals of Otology, Rhinology & Laryngology, 109*(9), 819-822.

Cruccu, G., Berardelli, A., Inghilleri, M., & Manfredi, M. (1989). Functional organization of the trigeminal motor system in man. A neurophysiological study. *Brain, 112*(Pt 5), 1333-1350.

Cruccu, G., Berardelli, A., Inghilleri, M., & Manfredi, M. (1990). Corticobulbar projections to upper and lower facial motoneurons. A study by magnetic transcranial stimulation in man. *Neuroscience letters, 117*(1-2), 68-73.

Cruccu, G., Inghilleri, M., Berardelli, A., Romaniello, A., & Manfredi, M. (1997). Cortical mechanisms mediating the inhibitory period after magnetic stimulation of the facial motor area. *Muscle & nerve, 20*(4), 418-424.

Cruccu, G., Pauletti, G., Agostino, R., Berardelli, A., & Manfredi, M. (1991). Masseter inhibitory reflex in movement disorders. Huntington's chorea, Parkinson's disease, dystonia, and unilateral masticatory spasm. *Electroencephalography & Clinical Neurophysiology, 81*(1), 24-30.

Curra, A., Berardelli, A., Rona, S., Fabri, S., & Manfredi, M. (1998). Excitability of motor cortex in patients with dystonia. *Advances in Neurology, 78*, 33-40.

Curra, A., Romaniello, A., Berardelli, A., Cruccu, G., & Manfredi, M. (2000). Shortened cortical silent period in facial muscles of patients with cranial dystonia. *Neurology, 54*(1), 130-135.

De Andrade, L. A., & Bertolucci, P. H. (1985). [Treatment of Meige disease with GABA receptor agonist]. [Portuguese] Abstract. *Arquivos de Neuro-Psiquiatria, 43(3)*, 260-266.

Dedo, H., & Izdebski, K. (1983). Intermediate results of 306 recurrent laryngeal nerve sections for spastic dysphonia. *Laryngoscope, 93*, 9-16.

Dedo, H. H. (1976). Recurrent laryngeal nerve section for spastic dysphonia. *Ann Otol Rhinol Laryngol, 85*, 451-459.

Desiato, M. T., Bernardi, G., Hagi, H. A., Boffa, L., & Caramia, M. D. (2002). Transcranial magnetic stimulation of motor pathways directed to muscles supplied by cranial nerves in amyotrophic lateral sclerosis. *Clinical Neurophysiology, 113*(1), 132-140.

Devous, M. D., Pool, K. D., Sr., Finitzo, T., & Freeman, F. J. (1990). Evidence for cortical dysfunction in spasmodic dysphonia: regional cerebral blood flow and quantitative electrophysiology. *Brain and Language, 39*, 331-344.

Di Lazzaro, V., Restuccia, D., Oliviero, A., Profice, P., Ferrara, L., Insola, A., . . . Rothwell, J. C. (1998). Effects of voluntary contraction on descending volleys evoked by transcranial stimulation in conscious humans. *Journal of Physiology, 508*(Pt 2), 625-633.

Dresel, C., Haslinger, B., Castrop, F., Wohlschlaeger, A. M., & Ceballos-Baumann, A. O. (2006). Silent event-related fMRI reveals deficient motor and enhanced somatosensory activation in orofacial dystonia. *Brain, 129*(Pt 1), 36-46.

Epstein, C. M., Schwartzberg, D. G., Davey, K. R., & Sudderth, D. B. (1990). Localizing the site of magnetic brain stimulation in humans. *Neurology, 40*(4), 666-670.

Finitzo-Hieber, T., Freeman, F. J., Gerling, I. J., Dobson, L., & Schaefer, S. D. (1982). Auditory brainstem response abnormalities in adductor spasmodic dysphonia. *Am J Otolaryngol, 3*, 26-30.

Fritzell, B., Feuer, E., Haglund, S., Knutsson, E., & Schiratzki, H. (1982). Experiences with recurrent laryngeal nerve section of spastic dystonia. *Folia Phoniatr, 34*, 160-167.

Guggisberg, A. G., Dubach, P., Hess, C., Wuthrich, C., & Mathis, J. (2001). Motor evoked potentials from masseter muscle induced by transcranial magnetic stimulation of the pyramidal tract: teh importance of coil orientation. *Clinical Neurophysiology, 112*, 2312-2319.

Hallett, M. (1995). Is dystonia a sensory disorder?[Editorial]. *Ann Neurol, 38*(Journal Article), 139-140.

Haslinger, B., Erhard, P., & Dresel, C. (2005). "Silent event-related" fMRI reveals reduced sensorimotor activation in laryngeal dystonia. *Neurology, 65*(Journal Article), 1562-1569.

Hillel, A. D. (2001). The study of laryngeal muscle activity in normal human subjects and in patients with laryngeal dystonia using multiple fine-wire electromyography. *The Laryngoscope, 111*(Journal Article), 1-47.

Hirano, S., Kojima, H., Naito, Y., Honjo, I., Kamoto, Y., Okazawa, H., . . . Konishi, J. (1996). Cortical speech processing mechanisms while vocalizing visually presented languages. *Neuroreport, 8*(1), 363-367.

Hirano, S., Kojima, H., Naito, Y., Tateya, I., Shoji, K., Kaneko, K., . . . Konishi, J. (2001). Cortical dysfunction of the supplementary motor area in a spasmodic dysphonia patient. *American Journal of Otolaryngology, 22*(3), 219-222.

Hirose, H. (1977). Electromyography of the larynx and other speech organs *Dynamic Aspects of Speech Production* (pp. 49-67). Tokyo: University of Tokyo Press.

Hoffman, D. S., & Luschei, E. S. (1980). Responses of monkey precentral cortical cells during a controlled jaw bite task. [Research Support, U.S. Gov't, P.H.S.]. *Journal of neurophysiology, 44*(2), 333-348.

Hummel, F. C., Steven, B., Hoppe, J., Heise, K., Thomalla, G., Cohen, L. G., & Gerloff, C. (2009). Deficient intracortical inhibition (SICI) during movement preparation after chronic stroke. [Research Support, N.I.H., Intramural

Research Support, Non-U.S. Gov't]. *Neurology, 72*(20), 1766-1772.

Ikoma, K., Samii, A., Mercuri, B., Wassermann, E. M., & Hallett, M. (1996). Abnormal cortical motor excitability in dystonia. *Neurology, 46*(Journal Article), 1371-1376.

Izdebski, K., dedo, H. H., & Boles, L. (1984). Spastic dysphonia: a patient profile of 200 cases. *Am J Otolaryngology, 5*(Journal Article), 7-14.

Izdebski, K., & Shipp, T. (1978). Minimal reaction times for phonatory initiation. *J Speech Hear Res, 21*(Journal Article), 638-651.

Jaberzadeh, S., Pearce, S. L., Miles, T. S., Turker, K. S., & Nordstrom, M. A. (2007). Intracortical inhibition in the human trigeminal motor system. *Clinical Neurophysiology, 118*(8), 1785-1793.

Jaberzadeh, S., Sakuma, S., Zoghi, M., Miles, T. S., & Nordstrom, M. A. (2008). Focal transcranial magnetic stimulation of motor cortex evokes bilateral and symmetrical silent periods in human masseter muscles. *Clinical Neurophysiology, 119*(3), 693-703.

Jurgens, U. (2002). Neural pathways underlying vocal control. [Review]. *Neuroscience & Biobehavioral Reviews, 26*(2), 235-258.

Khedr, E. M., & Aref, E. E. (2002). Electrophysiological study of vocal-fold mobility disorders using a magnetic stimulator. *European Journal of Neurology, 9*(3), 259-267.

Kidokoro, Y. K., K.; Shuto, S.; Sumino, R. (1986). Reflex organization of cat masticatory muscles. *Journal of neurophysiology, 31*, 695-708.

Kimberley, T. J., Borich, M. R., Prochaska, K. D., Mundfrom, S. L., Perkins, A. E., & Poepping, J. M. (2009). Establishing the definition and inter-rater reliability of cortical silent period calculation in subjects with focal hand dystonia and healthy controls. *Neuroscience letters, 464*(2), 84-87.

Kratz, O. (2011). Reply to the letter to the editor "response to Kratz et Al, seizure in a nonpredisposed individual induced by single-pulse transcranial magnetic stimulation". [Comment]. *Journal of ECT, 27*(2), 177.

Kujirai, T., Caramia, M. D., Rothwell, J. C., Day, B. L., Thompson, P. D., Ferbert, A., . . . Marsden, C. D. (1993). Corticocortical inhibition in human motor cortex. *J of Physiology, 471*(Journal Article), 501-519.

Kuypers, H. G. (1958). Corticobular connexions to the pons and lower brain-stem in man: an anatomical study. *Brain, 81*(3), 364-388.

Lerner, A., Shill, H., Hanakawa, T., Bushara, K., Goldfine, A., & Hallett, M. (2004). Regional cerebral blood flow correlates of the severity of writer's cramp symptoms. *Neuroimage, 21(3)*(Journal Article), 904-913.

Levy, L. M., & Hallett, M. (2002). Impaired brain GABA in focal dystonia. *Annals of Neurology, 51*(1), 93-101.

Liepert, J. (2006). Motor cortex excitability in stroke before and after constraint-induced movement therapy. *Cognitive & Behavioral Neurology, 19*(1), 41-47.

Ludlow, C. (2009). Treatment for spasmodic dysphonia: limitations of current approaches. *Curr Opin Otolaryngol Head Neck Surg, 17*, 160-165.

Ludlow, C. L. (2004). Recent advances in laryngeal sensorimotor control for voice, speech and swallowing. *Current Opinion in Otolaryngology & Head & Neck Surgery, 12*(3), 160-165.

Ludlow, C. L. (2011). Spasmodic dysphonia: a laryngeal control disorder specific to speech. *Journal of Neuroscience, 31*(3), 793-797.

Ludlow, C. L., Adler, C. H., Berke, G. S., Bielamowicz, S. A., Blitzer, A., Bressman, S. B., . . . Woodson, G. E. (2008). Research priorities in spasmodic dysphonia. *Otolaryngology - Head & Neck Surgery, 139*(4), 495-505.

Lundy, D., Ling, F., Casiano, R., & Zue, J. (1998). The effect of patient factors on response outcomes to botox treatment of spasmodic dysphonia. *J of Voice, 12(4)*(Journal Article), 460-466.

Macaluso, G. M., Pavesi, G., Bonanini, M., Mancia, D., & Gennari, P. U. (1990). Motor-evoked potentials in masseter muscle by electrical and magnetic stimulation in the intact alert man. *Archs oral biol, 35*(8), 623-628.

Manganotti, P., Acler, M., Zanette, G. P., Smania, N., & Fiaschi, A. (2008). Motor cortical disinhibition during early and late recovery after stroke. *Neurorehabilitation & Neural Repair, 22*(4), 396-403.

Matsumura, M., Sawaguchi, T., & Kubota, K. (1992). GABAergic inhibition of neuronal activity in the primate motor and premotor cortex during voluntary movement. *J Neurophysiol, 68*(Journal Article), 692-702.

Mavroudakis, N., Caroyer, J. M., Brunko, E., & Aamodt, A. (1995). Abnormal motor evoked responses to transcranial magnetic stimulation in focal dystonia. *Neurology, 45*(Journal Article), 1671-1677.

McMillan, A. S., Graven-Nielsen, T., Romaniello, A., & Svensson, P. (2001). Motor potentials evoked by transcranial magnetic stimulation during isometric and dynamic masseter muscle contraction in humans. *Archives of Oral Biology, 46*(4), 381-386.

McMillan, A. S., Watson, C., & D., W. (1998). Improved reproducibility of magnetic stimulation-evoked motor potentials in the human masseter by a new method for locating stimulation sites on the scalp. *Archives of Oral Biology, 43*, 665-668.

Murase, N., Duque, J., Mazzocchio, R., & Cohen, L. G. (2004). Influence of interhemispheric interactions on motor function in chronic stroke. *Annals of Neurology, 55*(3), 400-409.

Murase, N., & Rothwell, J. C. (2005). Subthreshold low-frequency repetitive transcranial magnetic stimulation over premotor cortex modulates writer's cramp. *Brain, 128*(Journal Article), 104-115.

Murase, N., Shimadu, H., Urushihara, R., & Kaji, R. (2006). Abnormal sensorimotor integration in hand dystonia. *Supplements to Clinical Neurophysiology, 59*(Journal Article), 283-287.

Nakamura, Y. (1980). Brainstem neuronal mechanisms controlling the trigeminal motoneuron activity. In J. E. Desmedt. (Ed.), *Spinal and supraspinal mechansims of voluntary motor control and locomotion. Progress in Clinical Neurophysiology* (Vol. 8, pp. 181-202): Basel and London.

Nakashima, K., Thompson, P. D., Rothwell, J. C., Day, B. L., Stell, R., & Marsden, C. D. (1989). An exteroceptive reflex in the sternocleidomastoid muscle producted by electrical stimulation of the supraorbital nerve in normal subjects and patietns with spasmodic torticollis. *Neurology, 39*(Journal Article), 1354-1358.

Neslon, A. J. H., T.; Gunraj, C.; Ni, Z.; Chen, R. (2010). Impaired interhemispheric inhibition in writer's cramp. *Neurology, 75*, 441-447.

Nolte, J. (2002). *The Human Brain: An Introduction to it's Functional Anatomy* (Vol. Fifth). St. Louis, MO: Mosby.

Nordstrom, M. A. (2007). Insights into the bilateral cortical control of human masticatory muscles revealed by transcranial magnetic stimulation. *Archives of Oral Biology, 52*(4), 338-342.

Nordstrom, M. A., Miles, T. S., Gooden, B. R., Butler, S. L., Ridding, M. C., & Thompson, P. D. (1999). Motor cortical control of human masticatory muscles. *Progress in brain research, 123*(Journal Article), 203-214.

Oh, B.-M., Kim, D.-Y., & Paik, N.-J. (2010). Disinhibition in the unaffected hemisphere is related with the cortical involvement of the affected hemisphere. [Research Support, Non-U.S. Gov't]. *International Journal of Neuroscience, 120*(7), 512-515.

Ortu, E., Deriu, F., Suppa, A., Giaconi, E., Tolu, E., & Rothwell, J. C. (2008). Intracortical modulation of cortical-bulbar responses for the masseter muscle. *Journal of Physiology, 586*(14), 3385-3404.

Pascual-Leone, A., Rothwell, J., Davey, N., Wasserman, E. M., & Puri, B. (2002). *Handbook of Transcranial Magnetic Stimulation*: Hodder Arnold.

Pascual-Leone, A., Valls-Sole, J., Wassermann, E. M., & Hallett, M. (1994). Responses to rapid-rate transcranial magnetic stimulation of the human motor cortex. *Brain, 117*(Pt 4), 847-858.

Pearce, S. L., Miles, T. S., Thompson, P. D., & Nordstrom, M. A. (2003). Responses of single motor units in human masseter to transcranial magnetic stimulation of either hemisphere. *Journal of Physiology, 549*(Pt 2), 583-596.

Penfield, W., & Welch, K. (1951). The supplementary motor area of the cerebral cortex: A clinical and experimental study. *Arch Neurol Psychiatry, 66*(Journal Article), 289-294.

Pujol, J., Roset-Llobet, J., Rosines-Cubells, D., Deus, J., Narberhaus, B., Valls-Sole, J., . . . Pascual-Leone, A. (2000). Brain cortical activation during guitar-induced hand dystonia studied by functional MRI. *NeuroImage, 12*(3), 257-267.

Purves, D., Brannon, E., Cabeza, R., Huettel, S., LaBar, K., Platt, M., & Woldorff, M. (2008). *Principles of Cognifive Neurosciences*. Sunderland, MA: Sinauer Associates.

Quartarone, A., Bagnato, S., Rizzo, V., siebner, H., Dattola, V., Scalfari, A., . . . Girlanda, P. (2003). Abnormal associative plasticity of the human motor cortex in writer's cramp. *Brain, 126*(Journal Article), 2586-2596.

Quartarone, A., Morgante, F., Sant'angelo, A., Rizzo, V., Bagnato, S., Terranova, C., . . . Girlanda, P. (2008). Abnormal plasticity of sensorimotor circuits extends beyond the affected body part in focal dystonia. *Journal of Neurology, Neurosurgery & Psychiatry, 79*(9), 985-990.

Quartarone, A., Rizzo, V., Bagnato, S., Morgante, F., Sant'Angelo, A., Romano, M., . . . Siebner, H. R. (2005). Homeostatic-like plasticity of the primary motor hand area is impaired in focal hand dystonia. *Brain, 128*(Pt 8), 1943-1950.

Quartarone, A., Rizzo, V., & Morgante, F. (2008). Clinical features of dystonia: a pathophysiological revisitation. *Current opinion in neurology, 21*(4), 484-490.

Quartarone, A., Siebner, H. R., & Rothwell, J. C. (2006). Task-specific hand dystonia: can too much plasticity be bad for you? *Trends in neurosciences, 29*(4), 192-199.

Ridding, M. C., Sheean, G., Rothwell, J. C., Inzelberg, R., & Kujirai, T. (1995). Changes in the balance between motor cortical excitation and inhibition in focal, task specific dystonia. *Journal of Neurology, Neurosurgery & Psychiatry, 59*(5), 493-498.

Ridding, M. C., Taylor, J. L., & Rothwell, J. C. (1995). The effect of voluntary contraction on cortico-cortical inhibition in human motor cortex. [Comparative Study

Research Support, Non-U.S. Gov't]. *Journal of Physiology, 487*(Pt 2), 541-548.

Rodel, R., Olthoff, A., & Tergau, F. (2004). Human cortical motor representation of the larynx as assessed by transcranial magnetic stimulation (TMS). *The Laryngoscope, 114*(Journal Article), 918-922.

Rona, S., Berardelli, A., Vacca, L., Inghilleri, M., & Manfredi, M. (1998). Alterations of motor cortical inhibition in patients with dystonia. *Mov Disord, 13*(Journal Article), 118-124.

Rosenfield, D. B. (1988). Spasmodic dysphonia. *Advances in Neurology, 49*(Journal Article), 317-327.

Roy, N., Gouse, M., Mauszycki, S. C., Merrill, R. M., & Smith, M. E. (2005). Task specificity in adductor spasmodic dysphonia versus muscle tension dysphonia. *Laryngoscope, 115*(2), 311-316.

Roy, N., Mauszycki, S. C., Merrill, R. M., Gouse, M., & Smith, M. E. (2007). Toward improved differential diagnosis of adductor spasmodic dysphonia and muscle tension dysphonia. *Folia Phoniatrica et Logopedica, 59*(2), 83-90.

Roy, N., Smith, M. E., Allen, B., & Merrill, R. M. (2007). Adductor spasmodic dysphonia versus muscle tension dysphonia: examining the diagnostic value of recurrent laryngeal nerve lidocaine block. *Annals of Otology, Rhinology & Laryngology, 116*(3), 161-168.

Roy, N., Whitchurch, M., Merrill, R. M., Houtz, D., & Smith, M. E. (2008). Differential diagnosis of adductor spasmodic dysphonia and muscle tension dysphonia using phonatory break analysis. *Laryngoscope, 118*(12), 2245-2253.

Sapienza, C., & Hoffman-Ruddy, B. (2009). *Voice Disorders*. San Diego, CA: Plural Publishing.

Sapienza, C. M., Walton, S., & Murry, T. (2000). Adductor spasmodic dysphonia and muscular tension dysphonia: acoustic analysis of sustained phonation and reading. *Journal of Voice, 14*(4), 502-520.

Schaefer, S. D., Watson, B. C., & Freemon, F. J. (1987). Vocal tract electromyographic abnormalities in spasmodic dysphonia. *Trans Am Laryngol Assoc, 108*(Journal Article), 187-196.

Schlotthauer, G. T., M.; Jackson-Menaldi, M. (2010). A pattern recognition approach to spasmodic dysphonia and muscle tension dysphonia automatic classification. *Journal of Voice, 24(3)*, 346-353.

Schweinfurth, J. M., Billante, M., & Courey, M. S. (2002). Risk factors and demographics in patients with spasmodic dysphonia. *Laryngoscope, 112*(Journal Article), 220-223.

Sheehy, M. P., & Marsden, C. D. (1982). Writer's cramp: a focal dystonia. *Brain, 105 (Pt3)*(Journal Article), 461-480.

Shipp, T., Izdebski, K., Reed, C., & Morrissey, P. (1985). Intrinsic laryngeal muscle activity in a spastic dysphonia patient. *J Speech Hear Disord, 50*(Journal Article), 54-59.

Siebner, H. R., Tormos, J. M., Ceballos-Baumann, A. O., Auer, C., Catala, M. D., Conrad, B., & Pascual-Leone, A. (1999). Low-frequency repetitive transcranial magnetic stimulation of the motor cortex in writer's cramp. *Neurology, 52*(3), 529-537.

Simonyan, K., & Ludlow, C. L. (2010). Abnormal activation of the primary somatosensory cortex in spasmodic dysphonia: an fMRI study. *Cerebral Cortex, 20*(Journal Article), 2749-2759.

Simonyan, K., Ostuni, J., Ludlow, C. L., & Horwitz, B. (2009). Functional but not structural networks of the human laryngeal motor cortex show left hemispheric lateralization during syllable but not breathing production. [Research Support, N.I.H., Intramural]. *Journal of Neuroscience, 29*(47), 14912-14923.

Simonyan, K., Tovar-Moll, F., Ostuni, J., & Hallett, M. (2008). Focal white matter changes in spasmodic dysphonia: a combined diffusion tensor imaging and neuropathological study. *Brain, 131*(Journal Article), 447-459.

Simonyan, K. L., C.; Vortmeyer, A. (2009). Brainstem pathology in spasmodic dysphonia. *Laryngoscope, 120*(January), 121-124.

Sommer, M. R., D.; Tergau, R.; Beuche, W.; Altenmuller, E.; Paulus, W. (2002). Intracortical exitability in the hand motor representation in hand dystonia and blepharospasm. *Movement Disorders, 17*(5), 1017-1025.

Sowman, P. F., Flavel, S. C., McShane, C. L., Miles, T. S., & Nordstrom, M. A. (2008). Transcranial magnetic stimulation reduces masseter motoneuron pool excitability throughout the cortical silent period. *Clinical Neurophysiology, 119*(5), 1119-1129.

Stinear, C. M., & Byblow, W. D. (2004). Impaired modulation of intracortical inhibition in focal hand dystonia. [Clinical Trial

Controlled Clinical Trial

Research Support, Non-U.S. Gov't]. *Cerebral Cortex, 14*(5), 555-561.

Stinear, C. M., & Byblow, W. D. (2004). Modulation of corticospinal excitability and intracortical inhibition during motor imagery is task-dependent. *Experimental Brain Research, 157*(3), 351-358.

Tanner, K. R., N.; Merrill, R.; Sauder, C.; Houtz, D.; Smith, M. (2011). Spasmodic dysphonia: onset, course, socioemotional effects and treatment response. *Annals of Otology, Rhinology & Laryngology, 120(7)*, 465-473.

Tempel, L. W., & Perlmutter, J. S. (1990). Abnormal vibration-induced cerebral blood flow responses in idiopathic dystonia. *Brain, 113*(Journal Article), 691-707.

Terao, Y. U., Y. (2002). Basic mechanisms of TMS. *Journal of Clinical Neurophysiology, 19(4)*, 322-343.

Tergau, F., Wanschura, V., Canelo, M., Wischer, S., Wassermann, E. M., Ziemann, U., & Paulus, W. (1999). Complete suppression of voluntary motor drive during the silent period after transcranial magnetic stimulation. *Experimental Brain Research, 124*(4), 447-454.

Thompson, M. L., Thickbroom, G. W., Sacco, P., Wilson, S. A., Stell, R., & Mastaglia, F. L. (1996). Changes in the organization of the corticomotor projection to the hand in writer's cramp [Abstract]. *Mov Disord, 11 Suppl 1*(Journal Article), 219.

Thompson, P. D., Dick, J. P., Day, B. L., Rothwell, J. C., Berandelli, A., & Kachi, T. (1986). Electrophysiology of the corticomotoneurone pathways in patients wtih movement disorders. *Mov Disord, 1*(Journal Article), 113-117.

Valls-Sole, J., Pascual-Leone, A., Brasil-Neto, J. P., Cammarota, A., McShane, L., & Hallett, M. (1994). Abnormal facilitation of the response to transcranial magnetic stimulation in patients with Parkinson's disease. *Neurology, 44*(4), 735-741.

van den Berg, J. (1958). Myoelastic-aerodynamic theory of voice production. *J of Sp Hearing Research, 1(3)*(Journal Article), 227-244.

van der Kamp, W., Berandelli, A., Rothwell, J. C., Thompson, P. D., Day, B. L., & Marsden, C. D. (1989). Rapid elbow movements in patients with torsion dystonia. *J Neurol Neurosurg Psychiatry, 52*(Journal Article), 1043-1049.

van Pelt, F., & Ludlow, C. (1994). An evaluation of laryngeal muscle activation in patients with voice tremor. *Ann Otol Rhinol Laryngol, 103*(Journal Article), 192-200.

Vitek, J. L., Zhang, J., DeLong, M. R., Mewes, K., & Bakay, R. A. E. (1997). Neuronal activity in the pallidum in patients with medically intractable dystonia. In S. Fahn, M. DeLong & C. D. Marsden (Eds.), *Advances in Neurology*. Philadelphia: Lippincott-Raven.

Vucic, S., Cheah, B. C., & Kiernan, M. C. (2011). Maladaptation of cortical circuits underlies fatigue and weakness in ALS. [Research Support, Non-U.S. Gov't]. *Amyotrophic Lateral Sclerosis, 12*(6), 414-420.

Wassermann, E. M., Epstein, C. M., Ziemann, U., Walsh, V., Paus, T., & Lisanby, S. (2008). *The Oxford Handbook of Transcranial Magnetic Stimulation*. Oxford: Oxford University Press.

Watson, C., Walshaw, D., & McMillan, A. S. (2000). Effect of motor tasks on the cortical topography of the human masseter muscle. [Research Support, Non-U.S. Gov't]. *Archives of Oral Biology, 45*(9), 767-773.

Zanette, G., Tamburin, S., Manganotti, P., Refatti, N., Forgione, A., & Rizzuto, N. (2002). Different mechanisms contribute to motor cortex hyperexcitability in amyotrophic lateral sclerosis. *Clinical Neurophysiology, 113*(11), 1688-1697.

Ziemann, U., Chen, R., Cohen, L. G., & Hallett, M. (1998). Dextromethorphan decreases the excitability of the human motor cortex. [Research Support, Non-U.S. Gov't]. *Neurology, 51*(5), 1320-1324.

Ziemann, U., Rothwell, J. C., & Ridding, M. C. (1996). Interaction between intracortical inhibition and facilitation in human motor cortex. *Journal of Physiology, 496*(Pt 3), 873-881.

Zraick, R., Kempster, G., Conner, N., Thibeault, S., Klaben, B., Bursac, Z., . . . Glaze, L. (2011). Establishing Validity of the Consensus Auditory-Perceptual Evaluation of Voice (CAPE-V). *American Journal of Speech-Language Pathology, 20*, 14-22.

Appendix A.

Consent Forms

Consent to Participate in Research Study:

Pathophysiolgoy of AdSD: A TMS Study– Healthy Control

You are invited to participate in a study examining the effect of brain stimulation using a device called a transcranial magnetic stimulator. This study is being done by Teresa J. Kimberley, PhD, PT in the Program in Physical Therapy, Sharyl Samargia, MA, CCC-SLP in the program in Rehabilitation Science and George Goding, MD in the Lion's Labs at the University of Minnesota at the University of Minnesota.
Please read this form and ask any questions before agreeing to be in this study.

Introduction

This study is exploring a new experimental procedure in dystonia called transcranial magnetic brain stimulation (TMS). The purpose of the study is to use TMS to determine the excitability of the brain between healthy subjects, those diagnosed with muscle tension dysphonia (MTD) and those diagnosed with adductor spasmodic dysphonia (AdSD) to gain a better understanding of the disorder. TMS works through a mechanism in which changes in magnetic fields in the brain induce electrical currents, which allow researchers to measure the excitability of portions of the brain. People without the diagnosis of this disorder are needed to serve as healthy controls in comparison to people with the disorder.

Procedures

If you agree to participate, you will be seen for one visit at which time you will be asked questions about your health history including your seizure history and medication use. People with a history of seizure or bipolar disorder will not be allowed to participate. You may also be asked to answer questions designed to test your memory and perform repeated motor movements of varying types. A sample of your voice may be obtained through video recording. A physician may review your medical history to determine that you qualify for the study.

Following this initial screening, you will receive a single session of diagnostic TMS. For this, you will be seated in a chair. First, we will determine your motor threshold, which is the lowest intensity of stimulation that produces a measurable response in your muscle. Small surface electrodes will be attached to the skin and connected to an EMG machine that shows the electrical response in your muscle as it is stimulated. Next, a swim cap will be applied to your head so that we can make measurements and mark spots for the brain stimulation. A figure-8 coil will be positioned over your scalp over a site that

corresponds to the target area of your brain. A very brief pulse of electrical current will pass through the coil once every 10 seconds and this will create a magnetic stimulus that will pass through your skull and activate the brain. For this you may feel a small tapping sensation on the scalp. After each pulse of stimulation we will check for a response from your muscle and lower the stimulation intensity for subsequent pulses until no response is seen. We will then move to another nearby site and do the same procedure. We will repeat this about eight times. At the end of this we will mark the best location for producing a response in your muscle with the lowest intensity of stimulation. We will do this for all muscles of interest. We will then perform various tests through stimulus pulses at a particular calculated intensity that will allow us to measure the level of excitability in that part of the brain.

The Food and Drug Administration approval exists for the stimulation that we will give you. Total time for participation will be approximately 2 hours.

Benefits of Study Participation

There is no benefit to you to participate in this study.

Risks of Study Participation

There have been reports of a seizure and induced mania from repetitive TMS but none of these side effects have been reported with the type of single pulse TMS that will be used in this study. However, as an additional precaution, we will not include any subjects with a history of seizure or bipolar disorder. The effect of TMS on the unborn fetus is not known and participating women should not be pregnant.

The possibility exists for a temporary headache due to the TMS or the tight swim cap surrounding the head. There is also a risk for dental pain. If either of these pains occur, we will manage them by administering acetaminophen. The effects of TMS on thinking, memory and mood in subjects are not known. We may discontinue the session without your consent if we recognize any abnormal signals in muscle recordings or any abnormal behavioral responses.

In the event that this research activity results in an injury, treatment will be available, including first aid, emergency treatment and follow-up care as needed. Care for such injuries will be billed in the ordinary manner, to you or your insurance company. If you think that you have suffered a research-related injury, please let us know right away.

Compensation

You will receive no payment for participating in this study.

Confidentiality

The records of this study will be kept private. In any publications or presentations, we will not include any information that will make it possible to identify you as a subject. Your record for the study may, however, be reviewed by departments at the University with appropriate regulatory oversight. To these extents, confidentiality is not absolute. The video recording will be stored on a secure, password protected computer drive for 2 years using your assigned subject code, not your name to ensure confidentiality. It is possible you could be identified on the video recording as it will be a recording of your face and an audio recording of your voice, however only co-investigators will have access to the video recordings. The video recording will not be used for any purposes other than classifying characteristics of your voice disorder. Once that information is obtained, the video recording will be destroyed. A separate optional consent form will be provided requesting your permission to use the video recording for educational purposes. Your decision to sign or not sign that consent form will not affect your participation in the study.

Protected Health Information (PHI)
Your PHI created or received for the purposes of this study is protected under the federal regulation known as HIPAA. Refer to the attached HIPAA authorization for details concerning the use of this information.

Voluntary Nature of the Study
Participation in this study is voluntary. Your decision whether or not to participate in this study will not affect your current or future relations with the University of Minnesota. If you decide to participate, you are free to withdraw at any time without affecting those relationships.

Contact People
You may ask questions now. If you have any questions later, you are encouraged to contact Teresa Kimberley at (612/626-4096). If you have any questions or concerns regarding the study and would like to talk to someone other than the researcher(s), you are encouraged to contact the Research Subject Advocate Program (RSA) of the Clinical Translational Science Institute at (612) 624-2621.

You will be given a copy of this form to keep for your records. You are making a decision whether or not to participate. Your signature indicates that you have read the information provided above and have decided to participate.

Signature/Patient:_____ Date:_____
Time of day: _____

Signature/Investigator:_____ Date:_____

Time of day: _____

Consent to Participate in Research Study:

Pathophysiology of AdSD: A TMS Study– AdSD Subjects

You are invited to participate in a study examining the diagnostic effect of brain stimulation using a device called a transcranial magnetic stimulator. This study is being done by Teresa J. Kimberley, PhD, PT in the Program in Physical Therapy, Sharyl Samargia, MA, CCC-SLP in the program in Rehabilitation Science and George Goding, MD in the Lion's Labs at the University of Minnesota.

Please read this form and ask any questions before agreeing to be in this study.

Introduction

You are being invited to participate in this study because you have been diagnosed with a condition called adductor spasmodic dysphonia (AdSD) or muscle tension dysphonia (MTD).This study is exploring a new experimental procedure in dystonia called transcranial magnetic brain stimulation (TMS). The purpose of the study is to use TMS to determine the excitability of the brain between healthy subjects, those diagnosed with muscle tension dysphonia (MTD) and those diagnosed with adductor spasmodic dysphonia (AdSD) to gain a better understanding of the disorder. TMS works through a mechanism in which changes in magnetic fields in the brain induce electrical currents, which allow researchers to measure the excitability of portions of the brain.

Procedures

If you agree to participate, you will be seen for one visit at which time you will be asked questions about your health history including your seizure history and medication use. People with a history of seizure or bipolar disorder will not be allowed to participate. You may also be asked to answer questions designed to test your memory and perform repeated motor movements of varying types. A sample of your voice may be obtained through video recording. A physician may review your medical history to determine that you qualify for the study.

Following this initial screening, you will receive a single session of diagnostic TMS. For this, you will be seated in a chair. First, we will determine your motor threshold, which is the lowest intensity of stimulation that produces a measurable response in your muscle. Small surface electrodes will be attached to the skin and connected to an EMG machine that shows the electrical response in your muscle as it is stimulated. Next, a swim cap will be applied to your head so that we can make measurements and mark spots for the brain stimulation. A figure-8 coil will be positioned over your scalp over a site that corresponds to the target area of your brain. A very brief pulse of electrical current will pass through the coil once every 10 seconds and this will create a magnetic stimulus that

will pass through your skull and activate the brain. For this you may feel a small tapping sensation on the scalp. After each pulse of stimulation we will check for a response from your muscle and lower the stimulation intensity for subsequent pulses until no response is seen. We will then move to another nearby site and do the same procedure. We will repeat this about eight times. At the end of this we will mark the best location for producing a response in your muscle with the lowest intensity of stimulation. We will do this for all muscles of interest. We will then perform various tests through stimulus pulses at a particular calculated intensity that will allow us to measure the level of excitability in that part of the brain.

The Food and Drug Administration approval exists for the stimulation that we will give you. Total time for participation will be approximately 2 hours.

Benefits of Study Participation

There is no benefit to you to participate in this study.

Risks of Study Participation

There have been reports of a seizure and induced mania from repetitive TMS but none of these side effects have been reported with the type of single pulse TMS that will be used in this study. However, as an additional precaution, we will not include any subjects with a history of seizure or bipolar disorder. The effect of TMS on the unborn fetus is not known and participating women should not be pregnant.

The possibility exists for a temporary headache due to the TMS or the tight swim cap surrounding the head. There is also a risk for dental pain. If either of these pains occur, we will manage them by administering acetaminophen. The effects of TMS on thinking, memory and mood in subjects are not known. We may discontinue the session without your consent if we recognize any abnormal signals in muscle recordings or any abnormal behavioral responses.

Research Related Injury

In the event that this research activity results in an injury, treatment will be available, including first aid, emergency treatment and follow-up care as needed. Care for such injuries will be billed in the ordinary manner, to you or your insurance company. If you think that you have suffered a research-related injury, please let us know right away.

Compensation

You will receive no payment for participating in this study.

Confidentiality

The records of this study will be kept private. In any publications or presentations, we will not include any information that will make it possible to identify you as a subject. Your record for the study may, however, be reviewed by departments at the University with appropriate regulatory oversight. To these extents, confidentiality is not absolute. The video recording will be stored on a secure, password protected computer drive for 2 years using your assigned subject code, not your name to ensure confidentiality. It is possible you could be identified on the video recording as it will be a recording of your face and an audio recording of your voice, however only co-investigators will have access to the video recordings. The video recording will not be used for any purposes other than classifying characteristics of your voice disorder. Once that information is obtained, the video recording will be destroyed. A separate optional consent form will be provided requesting your permission to use the video recording for educational purposes. Your decision to sign or not sign that consent form will not affect your participation in the study.

Protected Health Information (PHI)
Your PHI created or received for the purposes of this study is protected under the federal regulation known as HIPAA. Refer to the attached HIPAA authorization for details concerning the use of this information.

Voluntary Nature of the Study
Participation in this study is voluntary. Your decision whether or not to participate in this study will not affect your current or future relations with the Universityof Minnesota . If you decide to participate, you are free to withdraw at any time without affecting those relationships.

Contact People
You may ask questions now. If you have any questions later, you are encouraged to contact Teresa Kimberley at (612/626-4096). If you have any questions or concerns regarding the study and would like to talk to someone other than the researcher(s), you are encouraged to contact the Research Subject Advocate Program (RSA) of the Clinical Translational Science Institute at (612) 624-2621.

You will be given a copy of this form to keep for your records. You are making a decision whether or not to participate. Your signature indicates that you have read the information provided above and have decided to participate.

Signature/Patient:_____ Date:_____
Time of day: _____

Signature/Investigator:_____ Date:_____

Time of day: _____

Appendix B.

Inclusion and Exclusion Criteria

Subject Inclusion Criteria/Recruitment

Groups	Inclusion Criteria
Research group 1 (AdSD)	Adults between 18-75 years. Right handed Demonstrate little benefit from traditional voice therapy as measured by post treatment perceptual voice evaluation and/or patient report. Demonstrate positive benefit from botulinum toxin as measured by post treatment perceptual voice evaluation and/ or patient report. Voice evaluation by an experienced SLP
Research group 2 (MTD)	Adults between 18-75 years Right handed Demonstrate benefit from traditional voice therapy as measured by post treatment perceptual voice evaluation and/or patient report Demonstrate no positive benefit from botulinum toxin OR required high dose (>)botulinum toxin to achieve an effect as measured by post treatment perceptual voice evaluation and/or patient report. Voice evaluation by experienced SLP Voice disorder must still be perceivable
Control group (Healthy)	Adults between 18-75 years Right handed

Exclusion Criteria for subjects in all groups: Subjects will be excluded if they have a history of: vocal fold pathology, essential tremor, neurological condition (other than dystonia), focal hand dystonia or orthopedic hand or wrist dysfunction, past neurological insults, laryngeal surgery, chronic gastroesophageal reflux disease. If they have received botulinum toxin injections in the past, the last injection must be $\geq$ 4months prior to participation in the study. Subjects will also be excluded if they have a history of seizures, have a pacemaker or any other implanted metal medical device (excluding dental fillings) or if they are currently pregnant.

Appendix C.

Recruitment Materials

5/24/2011

University of Minnesota
Department of Rehabilitation Science
MMC 388, 420 Church St. SE
Minneapolis, MN, 55455

RE: Research Study in Laryngeal Dystonia/Spasmodic Dysphonia

The Department of Rehabilitation Science at the University of Minnesota is currently conducting a research study titled: *Pathophysiology of Adductor Spasmodic Dysphonia: a TMS study.* The purpose of the study is to better understand the differences between adductor laryngeal dystonia, (also referred to as spasmodic dysphonia) and muscle tension dysphonia.

If you have been diagnosed with adductor laryngeal dystonia (or spasmodic dysphonia) or muscle tension dysphonia, you could be eligible to participate.

The study requires a 2 ½ - 3 hour appointment at Clinical and Translational Science Institute at the University of Minnesota in Minneapolis. Unfortunately, no compensation is able to be provided for your participation.

Participation in this study is voluntary. Your decision whether or not to participate in this study will not affect your current or future relations with the University, the Fairview-University Medical Center or the Lion's Voice Clinic. If you decide to participate, you are free to withdraw at any time without affecting those relationships.

If you are interested in learning more about this study, please contact:

Sharyl Samargia by phone: (715)-441-2444 or by email: cleme046@umn.edu

Thank you

Sharyl Samargia, MA CCC-SLP
PhD Candidate
University of Minnesota
Department of Rehabilitation Science

RESEARCH STUDY IN LARYNGEAL DYSTONIA (SPASMODIC DYSPHONIA) AND MUSCLE TENSION DYSPHONIA

The Department of Rehabilitation Science at the University of Minnesota is currently conducting a research study to compare brain excitability between healthy adults, those diagnosed with adductor laryngeal dystonia (spasmodic dysphonia) and those diagnosed with muscle tension dysphonia.

Purpose of the study

The purpose of the study is to improve the current understanding of adductor laryngeal dystonia and help to differentiate it from other voice disorders such as muscle tension dysphonia. Participation will take approximately 2 hours at the University of Minnesota campus. This study has been approved by the University of Minnesota Institutional Review Board

If you are interested in learning more about this study, please contact:
Sharyl Samargia, MA CCC-SLP Doctoral Candidate:
by phone at (715) 441-2444 or email at cleme046@umn.edu

Sharyl Samargia
(715) 441-2444
cleme046@umn.edu

Sharyl Samargia
(715) 441-2444
cleme046@umn.edu

Sharyl Samargia
(715) 441-2444
cleme046@umn.edu

Sharyl Samargia
(715) 441-2444
cleme046@umn.edu

Sharyl Samargia
(715) 441-2444
cleme046@umn.edu

Sharyl Samargia
(715) 441-2444
cleme046@umn.edu

Sharyl Samargia
(715) 441-2444
cleme046@umn.edu

HEALTHY ADULT VOLUNTEERS NEEDED FOR RESEARCH STUDY

The Department of Rehabilitation Science at the University of Minnesota is currently conducting a research study to compare brain excitability between healthy adults, those diagnosed with adductor laryngeal dystonia (spasmodic dysphonia) and those diagnosed with muscle tension dysphonia.

Purpose of the study

The purpose of the study is to improve the current understanding of adductor laryngeal dystonia and help to differentiate it from other voice disorders such as muscle tension dysphonia. Participation will take approximately 2 hours at the University of Minnesota campus. This study has been approved by the University of Minnesota Institutional Review Board.

We are currently recruiting healthy males and females between the ages if 51 and 68 years. If you are interested in learning more about this study, please contact: **Sharyl Samargia, MA CCC-SLP Doctoral Candidate by phone at (715) 441-2444 or email at cleme046@umn.edu**

Sharyl Samargia
(715) 441-2444
cleme046@umn.edu
Sharyl Samargia
(715) 441-2444
cleme046@umn.edu
Sharyl Samargia
(715) 441-2444
cleme046@umn.edu
Sharyl Samargia
(715) 441-2444
cleme046@umn.edu
Sharyl Samargia
(715) 441-2444
cleme046@umn.edu
Sharyl Samargia
(715) 441-2444
cleme046@umn.edu

Email Script

Hi, my name is Sharyl Samargia, I am a doctoral candidate at the University of Minnesota studying spasmodic dysphonia (or laryngeal dystonia) and muscle tension dysphonia.

The purpose of the study is to better understand the differences in the brain's excitability level in those with spasmodic dysphonia compared to those with muscle tension dysphonia using transcranial magnetic stimulation (TMS). TMS is a safe, non-invasive method of using magnetic current to measure the excitability level in a very specific portion of the brain. No adverse side effects, other than slight headache have been reported by our past patients. Exclusion criteria are a history of seizure, bipolar disorder, lesions on the vocal cords, and pregnancy.

This study will help more definitively diagnose SD from muscle tension dysphonia and will lay a foundation for future studies for alternative treatment options.

If you choose to participate and meet the qualifications, you will be scheduled for a single appointment lasting approximately 2 hours at the University of Minnesota and there is no compensation for your time.

If you are interested, the next step would be to find out if you qualify for the study by asking you some screening questions, some of which include personal history questions regarding past medical diagnoses, seizure history and medication use. You are not required to answer any questions you are not comfortable with, however this may exclude you from the study.

If you think you would be interested in participating, I can arrange a time to call you and go over the screening questions with you, or if email is more convenient, I can email the screening questions to you.

Thank you for your interest and I look forward to talking more with you.

Phone Script

Hi, my name is Sharyl Samargia, I am a doctoral candidate at the University of Minnesota studying spasmodic dysphonia (or laryngeal dystonia) and muscle tension dysphonia.

The purpose of the study is to better understand the differences in the brain's excitability level in those with spasmodic dysphonia compared to those with muscle tension dysphonia using transcranial magnetic stimulation (TMS). TMS is a safe, non-invasive method of using magnetic current to measure the excitability level in a very specific portion of the brain. There have been no adverse side effects, other than slight headache reported by our past patients. Exclusion criteria are a history of seizure, bipolar disorder, lesions on the vocal cords, and pregnancy.

This study will help more definitively diagnose SD from muscle tension dysphonia and will lay a foundation for future studies for alternative treatment options.

If you choose to participate and meet the qualifications, you will be scheduled for a single appointment lasting approximately 2 hours at the University of Minnesota and, there is no compensation for your time.

Do you have questions about the study?

If you are interested, the next step would be to find out if you qualify for the study by asking you some screening questions, some of which include personal history questions regarding past medical diagnoses, regarding seizure history and medication use. You are not required to answer any questions you are not comfortable with, however this may exclude you from the study.

If you think you would be interested in participating, I can ask you screening questions now or contact you at a more convenient time to determine if you would qualify for the study. If email is more convenient, I can email the screening questions to you.

Based on individual's answer:

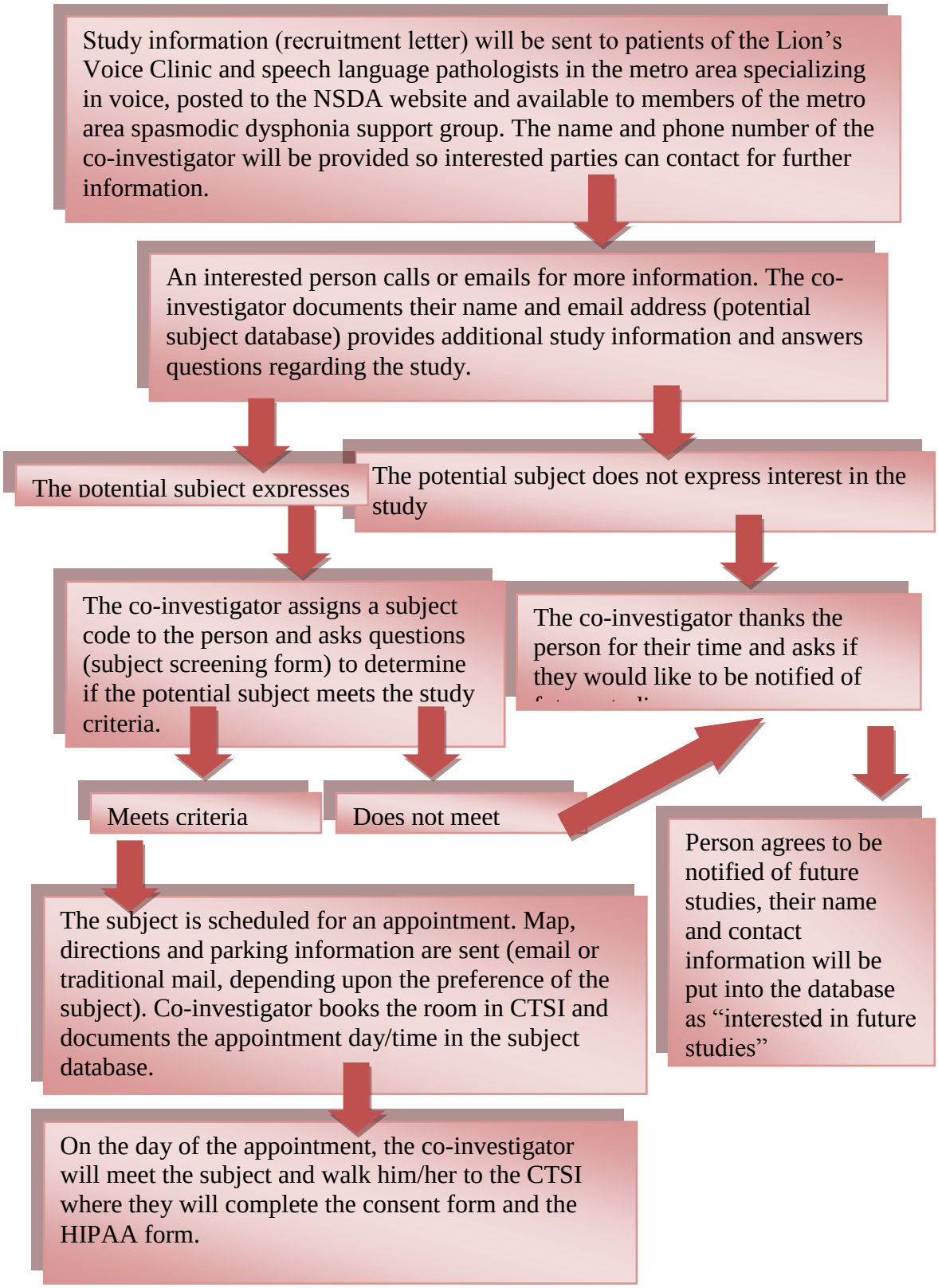
If yes: ask screening questions from the form and follow subject recruitment flow sheet based on their qualification

If yes, but different time: arrange another time to call

If no: thank them for their time and find out if they would be interested in participating in future studies.

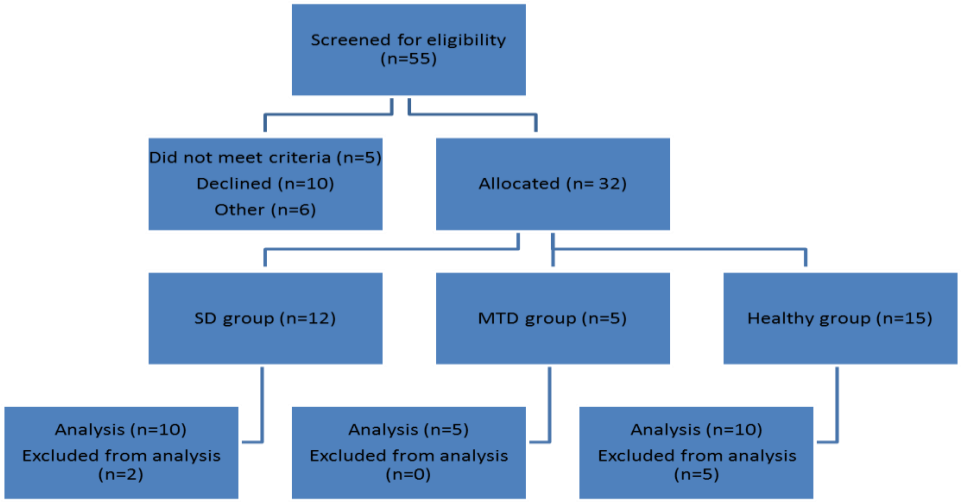
Thank you for your time, have a nice day.

Recruitment Flow Sheet



Subject Recruitment, Allocation and Screening Flow Sheet

80 people initially contacted



Pathophysiology of AdSD: a Transcranial Magnetic Stimulation study
Subject Screening Form

Initials: _____ Subject Code:_____

TMS Screening Questions	Yes	No	Comments
Are you currently taking any medications?			
If you are female, might you be pregnant?			
Do you have other forms of dystonia?			
Have you ever had a seizure?			
Does in anyone in your family have epilepsy			
Have you ever had a stroke?			
Do you suffer from frequent or severe headaches?			
Do you have any implanted devices such as a pacemaker?			
Do you have any metal inside your head (excluding dental fillings)?			
Do you have any other medical conditions?			
Do you need further explanation of TMS and its risks?			

Voice Screening Questions	Yes	No	Comments
Have you been diagnosed with a voice disorder?			What type? When?
Have you been diagnosed with a voice tremor?			
Have you ever had laryngeal surgery?			
Do you have other forms of dystonia?			
Do you have chronic GERD?			Is it effectively managed?
*Do your voice symptoms change depending upon the on the task (speaking vs laughing vs whispering)?			
Are you able to sing with little to no symptoms?			
Are you able to speak in a high pitch with little to no symptoms?			
*Are you able to say voiceless sounds easier than voiced ?			
*Have you had minimal benefit from traditional voice therapy?			
Are you currently being treated with botulinum toxin?			When: Muscles: Dose:
*Have you experienced benefit from *low dose* botulinum toxin as compared to traditional voice therapy?			
Have you experienced little to no benefit from low dose botulinum toxin?			Is high dose better?
Would you like to be contacted for any future studies?			

Appendix D.

Subject Binder Materials

Adverse Effects Questionnaire

Subject Id: _____

Date: _____

Mark yes or no next to the following symptoms:

	Immediate		Delayed (24-48 hours)	
Symptom	**YES**	**NO**	**YES**	**NO**
Seizure				
Headache				
Neck pain				
Dental pain				
Hearing				
Nausea				
Abnormal voice characteristics				
Dizziness				
Abnormal sleep				
Difficult concentration				
Anxiety				
Impaired memory				
Mood disturbance				
Impaired balance				
Impaired use of opposite hand				
Other				

Comments:

Subjective Dystonia Scale

Sujbect Id: _____

Date: _____

Compared to a typical day, today my dystonia is:

WORSE			NEUTRAL		BETTER	
Much	Moderately	Slightly	No Change	Slightly	Moderately	Much
-3	-2	-1	0	+1	+2	+3

Comments:

TMS Protocol Flowsheet

Subject ID _____ PI: Teresa J. Kimberley / Sharyl Samargia
Group: Healthy / AdLD / MTD Pathophysiology of AdSD: A TMS Study
Hemisphere of stimulation: L R

DATE	
Consent	
HIPAA	
Pregnancy test Yes/ No	
Screening form	
Edinbourgh handedness	
Adverse effect sheet	
CAPE-V and VHI	
Dystonia symptoms/subjective scale	
THRESHOLDS:Notch ON; Sweep@10(FDI) and 7.5 (masseter); FILTER Lo 20 High 2000	
FDI_RMT (GAIN @ 50 look for 1 div MEP):_____ 1mV _____	
80% of RMT: FDI_____	
110% of RMT: FDI_____	
120% of RMT: FDI_____	
130% of RMT: FDI_____	
140% of RMT: FDI_____	
Masseter_AMT (Gain @ 200; look for 1-2 div MEP)_____ .3mV _____	
80% of AMT: Masseter _____	
110% of AMT: Masseter _____	
120% of AMT: Masseter _____	
130% of Masseter_____	
140% of Masseter _____	
CORTICAL EXCITABILITY MEASURES: Filter Lo 20 HIGH 2000	
FDI_SP_S/R Curve@ varying intensties of RMT: GAIN@ 100 NOTCH ON	
Masseter_SP_S/R Curve@ varying intensities of AMT: GAIN @500 NOTCH ON	
FDI_SP 20@ 1mV threshold: GAIN@100 NOTCH ON	
Masseter_SP 20@ .3mV threshold: GAIN @500 NOTCH ON	
FDI_PP 10 with 3ms ISI: COND 80% of RMT: TEST 1mV thr: GAIN@100: NOTCH ON	
FDI_PP 10 with 10ms ISI: COND 80% RMT: TEST 1mV thr: GAIN@100: NOTCH ON	
Masseter_PP 10 with 3ms ISI: COND 80% AMT: TEST .3mV thr: GAIN@500: NOTCH ON	
Masseter_PP 10 with 10ms ISI: COND 80% AMT: TEST .3mV thr: GAIN@500: NOTCH ON	
FDI_CSP 10 @ 1mV threshold: GAIN@100 NOTCH OFF SWEEP@25	
Masseter_CSP 10 @ .3mV threshold: GAIN@500 NOTCH OFF SWEEP@15	

Appendix E.

Perceptual Voice Measures

Consensus Auditory-Perceptual Evaluation of Voice (CAPE-V)

Voice Sample #:_____

The following parameters of voice quality will be rated upon completion of the following tasks:
1. Sustained vowels, /a/ and /i/ for 3-5 seconds duration each.
2. Sentence production:
   a. The blue spot is on the key again.
   b. How hard did he hit him?
   c. We were away a year ago.
   d. We eat eggs every Easter.
   e. My mama makes lemon muffins.
   f. Peter will keep at the peak.
3. Spontaneous speech in response to: "Tell me about your voice problem." or "Tell me how your voice is functioning."

Legend: C = Consistent I = Intermittent
MI = Mildly Deviant
MO =Moderately Deviant
SE = Severely Deviant

			SCORE
Overall Severity	_____ MI MO SE	C I	/100
Roughness	_____ MI MO SE	C I	/100
Breathiness	_____ MI MO SE	C I	/100
Strain	_____ MI MO SE	C I	/100
Pitch	(Indicate the nature of the abnormality): _____ _____ MI MO SE	C I	/100
Loudness	(Indicate the nature of the abnormality): _____ _____ MI MO SE	C I	/100
_____	_____ MI MO SE	C I	/100
_____	_____ MI MO SE	C I	/100

COMMENTS ABOUT RESONANCE: NORMAL OTHER (Provide description):_____

_____

ADDITIONAL FEATURES (for example, diplophonia, fry, falsetto, asthenia, aphonia, pitch instability, tremor, wet/gurgly, or other relevant terms):

Clinician:_____

Name_____ Date_____Follow-up #_____

Voice Handicap Index (VHI)
(Jacobson, Johnson, Grywalski, *et al.*)

Instructions: These are statements that many people have used to describe their voices and the effects of their voices on their lives. Check the response that indicates how frequently you have the same experience.

(Never = 0 points; Almost Never = 1 point; Sometimes = 2 points; Almost Always = 3 points; Always = 4 points)

	Never	Almost Never	Sometimes	Almost Always	Always
F1. My voice makes it difficult for people to hear me.					
P2. I run out of air when I talk					
F3. People have difficulty understanding me in a noisy room					
P4. The sound of my voice varies throughout the day.					
F5. My family has difficulty hearing me when I call them throughout the house.					
F6. I use the phone less often than I would like.					
E7. I'm tense when talking with others because of my voice.					
F8. I tend to avoid groups of people because of my voice.					
E9. People seem irritated with my voice.					
P10. People ask, "What's wrong with your voice?"					
F11. I speak with friends, neighbors, or relatives less often because of my voice.					
F12. People ask me to repeat myself when speaking face-to-face.					
P13. My voice sounds creaky and dry.					

	Never	Almost Never	Sometimes	Almost Always	Always
P 14. I feel as though I have to strain to produce voice					
E15. I find other people don't understand my voice problem.					
F16. My voice difficulties restrict my personal and social life.					
P17. The clarity of my voice is unpredictable.					
P18. I try to change my voice to sound different.					
F19. I feel left out of conversations because of my voice.					
P20. I use a great deal of effort to speak.					
P21. My voice is worse in the evening.					
F22. My voice problem causes me to lose income.					
E23. My voice problem upsets me.					
E24. I am less out-going because of my voice problem.					
E25. My voice makes me feel handicapped.					
P26. My voice "gives out" on me in the middle of speaking.					
E27. I feel annoyed when people ask me to repeat.					
E28. I feel embarrassed when people ask me to repeat.					
E29. My voice makes me feel incompetent.					
E30. I'm ashamed of my voice problem.					

Please circle the word that matches your voice today.

Normal Mild Moderate Severe

P_____F_____E_____ Total_____

Appendix F.

Data Processing Tutorial and Forms

SD Study Data Analysis Tutorial

For an example of each of these, refer to the folder labeled: LDH_02_LK_FINAL. PLEASE DO NOT alter any files within this folder as the data within it has been fully processed. It is meant to be a guide to labeling and what your sheets should look like.

Access: I have invited all of you into Dropbox folders labeled DATA_IN PROGRESS and DATA_TO BE REVIEWED. Access the files that need to be processed in the 'in progress' folder. When a file is completed, move it from "in progress" folder to the designated subject folder within the 'to be reviewed' folder (there should never be two copies of same file in both folders). I will then review the data and check for any errors and compare to my calculations to be assured all data is accurate.

Tracking: Whenever any data is processed, be sure to check off what you did and your initials on the Data Processing Tracking sheet in the upper cabinet. It is VERY important we keep this up to date since so many people will have access to this data.

Tagging data: Any values that appear to be "off" from others (example peak to peak amplitudes of 120s-130s and you find one that is in the 30s), highlight in yellow so I can look at it. For CSP traces, any trace that you think seems to be an excellent example, highlight in green (makes it easier to go back and find good ones for figures, etc later). Those you question, highlight in yellow for me to review.

Delimiting Data: This transfers text files into excel files. I will be doing this on all data files. Open Excel; select open from file tab, select all files (lower right corner), select the text file you are working with from the subject data folder. Select delimit and next. Select tab, comma, space and in other type in a colon. Select finish. Now you should see data in columns in excel format. Often the columns will be off by one, if that is the case, cut and paste the columns so they match the headings.

S/R Curve: Open the MACRO_SR_Curve_FDI.xlsm file and the MACRO_SR_Curve_Masseter.xlsm from the MACRO folder in DATA IN PROGRESS. You will need to keep these open in order to run the macro in a new file. If you get a bar at the top saying "this macro has been disabled". Depending upon which version of excel you are working in: if 2010, choose enable, if older version click on options and enable. If you have an older version of excel, the macro may not run properly. This step enables the macro to be applied to other files.

Calculate in first sheet

1) Amplitude for each column: designate A668 as Peak, A669 as Trough and A670 PPAmpl

FDI	Masseter
Peak =max(b2223:b326)	=max(b120:b226)
Trough = min(b223:b326)	=min(b120:b226)
PPAmp = (b668-b669)	PPAmp (b668-b669)

2) Create a new sheet labeled RAW

3) Copy columns and rows from first sheet including **all raw data, peak, trough and pp ampl** (NOT the averages/SD values), and **paste special: values** (do not use paste special formulas) into the sheet labeled RAW.

4) Create 2 additional sheets within masseter file labeled: contralateral and ipsilateral and 4 additional sheets within FDI labeled: 110, 120, 130 and 140.

5) Be sure you are in the RAW sheet in cell A1. Run macro.

Masseter Contralateral: **ctrl +a**

Masseter Ipsilateral: **ctrl + b**

FDI 110: **ctrl + c**

FDI 120, 130 and 140: **ctrl + d**

6) Calculate mean and SD across 4 intensities (110%, 120%, 130% and 140%) for both FDI and Masseter (contralateral and ipsilateral). **

**please note: not all subjects received all intensities even though there will be values in all columns (if 140% is too high, we will fire it into the air and move to next) see their AdLD TMS protocol sheet within their subject binder for notes. Make note on the bottom of excel sheet so we do not include these values in processing.

7) SAVE as a macro enabled document ie: LD_14_JM_SRCurve_FDI.xslm into the TO BE REVIEWED folder.

SP_PP Open the MACRO_SP_PP__FDI.xlsm file and the MACRO_SP_PP_Masseter.xlsm from the MACRO folder in DATA IN PROGRESS. You will need to keep these open in order to run the macro in a new file. If you get a bar at the top saying "this macro has been disabled". Depending upon which version of excel you are working in: if 2010, choose enable, if older version click on options and enable. If you have an older version of excel, the macro may not run properly. This step enables the macro to be applied to other files.

Within your subject's data FDI file, create 4 sheets labeled RAW, PP_3 PP_10 and SP and in the Masseter file, create 5 sheets labeled RAW, PP_3, PP_10, SP_Contra and SP_Ipsi (you must label exactly as you see here or the macro won't run properly). Copy all columns and rows of raw data and paste them into the RAW sheet by choosing **paste special: values.** At this point, the macro can be run. This function will derandomize the data allowing for faster calculation.

Masseter paired pulse: **ctrl + e** will derandomize the paired pulse data

Masseter single pulse: **ctrl + f** will derandomize the single pulse data

FDI paired pulse and single pulse: **ctrl +g**

Now that the data has been derandomized into individual sheets, calculate the following within the individual sheets:

SP Calculate

1) Amplitude for each column: A643 as Peak, A644 Trough and A645 PPAmpl

FDI	Masseter(contra)	Masseter (ipsi)
Peak = max(b200:b300)	=max(b115:b200)	=max(b90:b190)
Trough=min(b200:n300)	=min(b115:b200)	=min(b90:b190)
PPAmpl= (b643-b644)	=(b643-b644)	=(b643-b644)

2) Amplitude Mean and SD across columns

3) Latency for each column (use tracking sheet if it's helpful)

Highlight entire column, click insert, select line graph. You will see a trace representation of the selected data, hover over the peak of the TMS stimulus artifact and record the data point. Hover over the peak of the MEP and record the data point. Do this for each column then enter the data into the excel sheet. Then subtract TMS onset – MEP onset and multiply by duration value (see page 2). For masseter, sweep is 7.5, so duration value is .12. For FDI, sweep is 10 so duration value is .16.

You don't need to save any of the graphs you make for SPs at this point. But, when you make them, document if it is an excellent example of an MEP (highlight that PPampl in green) or if it is a questionable MEP, looks funny or looks like no MEP at all (highlight in yellow).

4) Latency Mean and SD across columns

PP_3 Calculate

1) Amplitude for each column: A643 as Peak, A644 Trough and A645 PPAmpl

FDI	Masseter(contra)
Peak = max(b220:b320)	=max(b140:b240)
Trough=min(b220:b320)	=min(b140:b240)
PPAmpl= (b643-b644)	=(b643-b644)

2) Amplitude Mean and SD across columns
3) Normalize PP to SP mean amplitude: $\frac{PPmean-SPmean}{SPmean}$ to get "raw"value; multiply by 100 to get a percentage. Create cell for raw value and percentage.

PP_10 Calculate

1) Amplitude for each column: A643 as Peak, A644 Trough and A645 PPAmpl

FDI	Masseter(contra)
Peak = max(b260:b360)	=max(b195:b295)
Trough=min(b260:b360)	=min(b195:b295)
PPAmpl= (b643-b644)	=(b643-b644)

2) Amplitude Mean and SD across columns
3) Normalize PP to SP mean amplitude: $\frac{PPmean-SPmean}{SPmean}$ to get "raw"value; multiply by 100 to get a percentage. Create cell for raw value and percentage.
4) **SAVE as a macro enabled document ie: LD_14_JM_SP_PP_FDI.xslm**

CSP Open the MACRO_CSP.xlsm file from the MACRO folder in DATA IN PROGRESS. You will need to keep these open in order to run the macro in a new file. If you get a bar at the top saying "this macro has been disabled". Depending upon which version of excel you are working in: if 2010, choose enable, if

older version click on options and enable. If you have an older version of excel, the macro may not run properly. This step enables the macro to be applied to other files.

Calculate

1) Duration of CSP.

Create 1 sheet within the excel file labeled: RECTIFIED. Then, rectify the data and transfer it into RECTIFIED sheet. Be sure you are in the rectified Then run Macro

Masseter and FDI: **Ctrl + h**
This will make graphs of each CSP trace. If there is a bug, you might have to make a sheet for each trace (CSP_1, CSP_2, etc) then run macro.

For FDI, calculate the average prestimulus activity (mean of the first 63 data points) then calculate 50% of that value (that will be needed to determine the offset of the silent period)
Using the data tracking worksheet and looking at each trace, document the data point where TMS artifact is (usually around 66, 67), the ONSET: defined as the data point at the first superimposed EMG spike following the TMS stimulus (usually around 126, 127). Then document the OFFSET: defined as the data point where the trace returns to 50% of the prestimulus activation. DURATION: =(offset-onset)*.39 (for FDI because sweep is at 25)

For Masseter, calculate the average prestimulus activity (mean of the first 63 data points).
Using the data tracking worksheet and looking at each trace, document the data point where TMS artifact is (usually around 66, 67), the ONSET: defined as the data point at the first superimposed EMG spike following the TMS stimulus (usually around 97, 98). Then document the OFFSET: defined as the data point where the trace returns to the average prestimulus activation. DURATION: =(offset-onset)*.23 (for masseter, because sweep is at 15)

2) CSP duration Mean and SD

3) **For both FDI and Masseter:** Within the rectified sheet, create a column (M) labeled Average across rows, then calculate the average value for each row and carry down to duration calculation. This will give us one average CSP for this subject. Make a CSP trace of this data by highlighting column M, click insert, line graph. Move graph to a new sheet and label it CSP_Average_subject ID.

4) SAVE as a macro enabled document ie: LD_14_JM_CSP_FDI.xslm

Duration calculations depending upon sweep setting:

Calculation example: If sweep is 25: $\frac{250ms}{640dp} = \frac{0.39ms}{1dp}$

Sweep Value	Multiply by ___ to get duration
7.5	.12
10	.16
15	.23
25	.39

SD Data Processing Tracking Worksheet

Subject ID_____

CSP_Masseter										
	CSP_1	CSP_2	CSP_3	CSP_4	CSP_5	CSP_6	CSP_7	CSP_8	CSP_9	CSP_10
Average prestimulus activation										
TMS artifact										
MEP onset										
MEP offset										
CSP_FDI										
	CSP_1	CSP_2	CSP_3	CSP_4	CSP_5	CSP_6	CSP_7	CSP_8	CSP_9	CSP_10
50% prestim activation										
TMS artifact										
MEP onset										
MEP offset										

SP_Latency_Masseter_Contralateral																				
Trial	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
TMS onset																				
MEP onset																				

SP_Latency_Masseter_Ipsilateral																				
Trial	1	2	3	4	5	6	7	8	9	10										
TMS onset																				
MEP onset																				

SP_Latency_FDI																				
Trial	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
TMS onset																				
MEP onset																				

Page_____ of_____

Subject Data Processing Tracking

Subject ID	Delimit	SR Curve PPAmpl Mean/SD	SP PPAmpl MeanSD	PP_03 PPAmpl Mean/SD Ratio	PP_10 PP Ampl Mean/SD Ratio	CSP Identify landmarks on graphs	CSP Duration Mean/SD
FDI							
Masseter							
FDI							
Masseter							
FDI							
Masseter							
FDI							
Masseter							
FDI							
Masseter							
FDI							
Masseter							
FDI							
Masseter							
FDI							
Masseter							

Appendix G.

Statistical Analysis Output (SPSS)

Correlations

		gender	age	trial	CSP_M	CSP_F	PP_3_M	PP_10_M	PP_10_F	Slope_M_I	Slope_F	Log transformed MEP for contralateral masseter
gender	Pearson Correlation	1	.240**	.000	-.012	.078	.031	.342	-.128	.121	.288	-.301**
	Sig. (2-tailed)		.000	1.000	.855	.222	.889	.095	.541	.566	.172	.000
	N	500	500	500	227	244	23	25	25	25	24	482
age	Pearson Correlation	.240**	1	.000	.186**	-.135*	.066	.182	.431*	-.173	-.082	-.036
	Sig. (2-tailed)	.000		1.000	.005	.035	.764	.383	.031	.408	.704	.433
	N	500	500	500	227	244	23	25	25	25	24	482
trial	Pearson Correlation	.000	.000	1	.054	-.015	.c	.c	.c	.c	.c	-.035
	Sig. (2-tailed)	1.000	1.000		.420	.811	.000	.000	.000	.000	.000	.444
	N	500	500	500	227	244	23	25	25	25	24	482
CSP_M	Pearson Correlation	-.012	.186**	.054	1	.316**	.599**	.218	.209	.034	-.150	-.063
	Sig. (2-tailed)	.855	.005	.420		.000	.003	.296	.317	.874	.484	.350
	N	227	227	227	227	221	23	25	25	25	24	222
CSP_F	Pearson Correlation	.078	-.135*	-.015	.316**	1	-.161	-.480*	.085	-.142	.213	.133*
	Sig. (2-tailed)	.222	.035	.811	.000		.464	.015	.685	.499	.318	.041
	N	244	244	244	221	244	23	25	25	25	24	238
PP_3_M	Pearson Correlation	.031	.066	.c	.599**	-.161	1	.625**	.017	-.135	-.268	-.237
	Sig. (2-tailed)	.889	.764	.000	.003	.464		.001	.940	.538	.228	.287
	N	23	23	23	23	23	23	23	23	23	22	22
PP_10_M	Pearson Correlation	.342	.182	.c	.218	-.480*	.625**	1	-.015	.050	-.271	-.109
	Sig. (2-tailed)	.095	.383	.000	.296	.015	.001		.944	.813	.201	.612
	N	25	25	25	25	25	23	25	25	25	24	24
PP_10_F	Pearson Correlation	-.128	.431*	.c	.209	.085	.017	-.015	1	-.371	-.488*	.508*
	Sig. (2-tailed)	.541	.031	.000	.317	.685	.940	.944		.068	.016	.011
	N	25	25	25	25	25	23	25	25	25	24	24
Slope_M_I	Pearson Correlation	.121	-.173	.c	.034	-.142	-.135	.050	-.371	1	-.011	-.291
	Sig. (2-tailed)	.566	.408	.000	.874	.499	.538	.813	.068		.960	.168
	N	25	25	25	25	25	23	25	25	25	24	24
Slope_F	Pearson Correlation	.288	-.082	.c	-.150	.213	-.268	-.271	-.488*	-.011	1	-.221
	Sig. (2-tailed)	.172	.704	.000	.484	.318	.228	.201	.016	.960		.311
	N	24	24	24	24	24	22	24	24	24	24	23
Log transformed MEP for contralateral masseter	Pearson Correlation	-.301**	-.036	-.035	-.063	.133*	-.237	-.109	.508*	-.291	-.221	1
	Sig. (2-tailed)	.000	.433	.444	.350	.041	.287	.612	.011	.168	.311	
	N	482	482	482	222	238	22	24	24	24	23	482
Log transformed MEP for ipsilateral masseter	Pearson Correlation	-.042	-.019	-.012	.264**	.019	.258	.367	.000	.461*	-.178	.055
	Sig. (2-tailed)	.522	.773	.848	.000	.776	.247	.078	.999	.023	.416	.404
	N	239	239	239	221	233	22	24	24	24	23	234
Log_MEP_F	Pearson Correlation	.313**	.151**	.000	-.026	.070	-.094	.001	-.130	-.304	.578**	-.072
	Sig. (2-tailed)	.000	.002	.994	.699	.286	.671	.996	.537	.139	.003	.136
	N	437	437	437	220	234	23	25	25	25	24	426
Log transformed latency contralateral masseter	Pearson Correlation	-.159**	-.050	.069	-.066	.101	-.077	.043	.102	-.019	.005	.281**
	Sig. (2-tailed)	.001	.298	.155	.353	.145	.769	.862	.678	.938	.983	.000
	N	431	431	431	200	209	17	19	19	19	18	431
Log transformed latency ipsilateral masseter	Pearson Correlation	-.423**	-.052	.032	-.167*	-.177**	-.159	-.059	-.054	-.230	-.262	.132
	Sig. (2-tailed)	.000	.445	.636	.018	.010	.529	.805	.821	.330	.279	.055
	N	218	218	218	202	212	18	20	20	20	19	213
Log_PP_3_F	Pearson Correlation	-.119	.488*	.c	-.011	-.027	-.132	.181	.517**	-.145	-.299	.325
	Sig. (2-tailed)	.571	.013	.000	.958	.900	.548	.388	.008	.490	.156	.121
	N	25	25	25	25	25	23	25	25	25	24	24
Log_Slope_M_C	Pearson Correlation	-.528*	-.355	.c	.025	-.103	.056	-.151	-.105	.302	-.188	.143
	Sig. (2-tailed)	.014	.114	.000	.913	.657	.820	.515	.651	.184	.428	.546
	N	21	21	21	21	21	19	21	21	21	20	20
LogARC_M_C	Pearson Correlation	-.392	-.046	.c	-.034	-.164	.147	.160	-.017	.267	-.292	.376
	Sig. (2-tailed)	.053	.827	.000	.870	.434	.504	.444	.936	.197	.166	.071
	N	25	25	25	25	25	23	25	25	25	24	24
LogARC_M_I	Pearson Correlation	-.134	-.146	.c	-.036	-.237	.073	.217	-.233	.701**	-.152	-.102
	Sig. (2-tailed)	.522	.487	.000	.863	.253	.740	.297	.263	.000	.477	.637
	N	25	25	25	25	25	23	25	25	25	24	24
LogARC_FDI	Pearson Correlation	.034	-.078	.c	-.211	.251	-.144	-.441*	-.282	-.028	.743**	-.301
	Sig. (2-tailed)	.873	.718	.000	.321	.236	.521	.031	.182	.896	.000	.163
	N	24	24	24	24	24	22	24	24	24	24	23
COMPUTE Log_AMT=LG10 (AMT_Mass)	Pearson Correlation	.035	.012	-.390**	.347*	.150	.014	-.094	.053	.267	.105	-.339*
	Sig. (2-tailed)	.825	.942	.010	.048	.406	.952	.670	.811	.219	.643	.028
	N	43	43	43	33	33	21	23	23	23	22	42
COMPUTE Log_RMT=LG10 (RMT_FDI)	Pearson Correlation	.028	.109	-.268	.026	.214	-.108	-.308	.447*	-.106	-.344	.020
	Sig. (2-tailed)	.858	.492	.086	.889	.239	.650	.163	.037	.638	.126	.903
	N	42	42	42	32	32	20	22	22	22	21	41

		Log transformed MEP for ipsilateral masseter	Log_MEP_F	Log transformed latency contralateral masseter	Log transformed latency ipsilateral masseter	Log_PP_3_F	Log_Slope_M_C	LogARC_M_C	LogARC_M_I	LogARC_FDI	COMPUTE Log_AMT=LG10 (AMT_Mass)	COMPUTE Log_RMT=LG10(RMT_FDI)
gender	Pearson Correlation	-.042	.313**	-.159**	-.423**	-.119	-.528*	-.392	-.134	.034	.035	.028
	Sig. (2-tailed)	.522	.000	.001	.000	.571	.014	.053	.522	.873	.825	.858
	N	239	437	431	218	25	21	25	25	24	43	42
age	Pearson Correlation	-.019	.151**	-.050	-.052	.488*	-.355	-.046	-.146	-.078	.012	.109
	Sig. (2-tailed)	.773	.002	.298	.445	.013	.114	.827	.487	.718	.942	.492
	N	239	437	431	218	25	21	25	25	24	43	42
trial	Pearson Correlation	-.012	.000	.069	.032	.c	.c	.c	.c	.c	-.390**	-.268
	Sig. (2-tailed)	.848	.994	.155	.636	.000	.000	.000	.000	.000	.010	.086
	N	239	437	431	218	25	21	25	25	24	43	42
CSP_M	Pearson Correlation	.264**	-.026	-.066	-.167*	-.011	.025	-.034	-.036	-.211	.347*	.026
	Sig. (2-tailed)	.000	.699	.353	.018	.958	.913	.870	.863	.321	.048	.889
	N	221	220	200	202	25	21	25	25	24	33	32
CSP_F	Pearson Correlation	.019	.070	.101	-.177**	-.027	-.103	-.164	-.237	.251	.150	.214
	Sig. (2-tailed)	.776	.286	.145	.010	.900	.657	.434	.253	.236	.406	.239
	N	233	234	209	212	25	21	25	25	24	33	32
PP_3_M	Pearson Correlation	.258	-.094	-.077	-.159	-.132	.056	.147	.073	-.144	.014	-.108
	Sig. (2-tailed)	.247	.671	.769	.529	.548	.820	.504	.740	.521	.952	.650
	N	22	23	17	18	23	19	23	23	22	21	20
PP_10_M	Pearson Correlation	.367	.001	.043	-.059	.181	-.151	.160	.217	-.441*	-.094	-.308
	Sig. (2-tailed)	.078	.996	.862	.805	.388	.515	.444	.297	.031	.670	.163
	N	24	25	19	20	25	21	25	25	24	23	22
PP_10_F	Pearson Correlation	.000	-.130	.102	-.054	.517**	-.105	-.017	-.233	-.282	.053	.447*
	Sig. (2-tailed)	.999	.537	.678	.821	.008	.651	.936	.263	.182	.811	.037
	N	24	25	19	20	25	21	25	25	24	23	22
Slope_M_I	Pearson Correlation	.461*	-.304	-.019	-.230	-.145	.302	.267	.701**	-.028	.267	-.106
	Sig. (2-tailed)	.023	.139	.938	.330	.490	.184	.197	.000	.896	.219	.638
	N	24	25	19	20	25	21	25	25	24	23	22
Slope_F	Pearson Correlation	-.178	.578**	.005	-.262	-.299	-.188	-.292	-.152	.743**	.105	-.344
	Sig. (2-tailed)	.416	.003	.983	.279	.156	.428	.166	.477	.000	.643	.126
	N	23	24	18	19	24	20	24	24	24	22	21
Log transformed MEP for contralateral masseter	Pearson Correlation	.055	-.072	.281**	.132	.325	.143	.376	-.102	-.301	-.339*	.020
	Sig. (2-tailed)	.404	.136	.000	.055	.121	.546	.071	.637	.163	.028	.903
	N	234	426	431	213	24	20	24	24	23	42	41
Log transformed MEP for ipsilateral masseter	Pearson Correlation	1	-.137*	-.036	-.091	.339	.072	.493*	.669**	-.147	.134	-.149
	Sig. (2-tailed)		.038	.597	.179	.105	.757	.014	.000	.503	.464	.417
	N	239	229	215	218	24	21	24	24	23	32	32
Log_MEP_F	Pearson Correlation	-.137*	1	.101*	-.103	.083	-.283	-.246	-.336	.454*	-.542**	-.556**
	Sig. (2-tailed)	.038		.048	.137	.692	.214	.236	.101	.026	.000	.000
	N	229	437	382	210	25	21	25	25	24	43	42
Log transformed latency contralateral masseter	Pearson Correlation	-.036	.101*	1	.424**	.354	.141	.222	.066	-.062	-.386*	-.430**
	Sig. (2-tailed)	.597	.048		.000	.138	.601	.362	.790	.808	.018	.008
	N	215	382	431	197	19	16	19	19	18	37	37
Log transformed latency ipsilateral masseter	Pearson Correlation	-.091	-.103	.424**	1	-.085	-.065	.077	-.120	-.205	-.491**	-.239
	Sig. (2-tailed)	.179	.137	.000		.722	.798	.748	.613	.400	.008	.220
	N	218	210	197	218	20	18	20	20	19	28	28
Log_PP_3_F	Pearson Correlation	.339	.083	.354	-.085	1	-.170	.239	.101	-.213	-.243	-.190
	Sig. (2-tailed)	.105	.692	.138	.722		.461	.250	.632	.318	.265	.396
	N	24	25	19	20	25	21	25	25	24	23	22
Log_Slope_M_C	Pearson Correlation	.072	-.283	.141	-.065	-.170	1	.398	.158	-.095	-.247	.039
	Sig. (2-tailed)	.757	.214	.601	.798	.461		.074	.494	.689	.309	.875
	N	21	21	16	18	21	21	21	21	20	19	19
LogARC_M_C	Pearson Correlation	.493*	-.246	.222	.077	.239	.398	1	.713**	-.237	-.369	-.297
	Sig. (2-tailed)	.014	.236	.362	.748	.250	.074		.000	.265	.084	.180
	N	24	25	19	20	25	21	25	25	24	23	22
LogARC_M_I	Pearson Correlation	.669**	-.336	.066	-.120	.101	.158	.713**	1	-.146	.062	-.328
	Sig. (2-tailed)	.000	.101	.790	.613	.632	.494	.000		.496	.778	.137
	N	24	25	19	20	25	21	25	25	24	23	22
LogARC_FDI	Pearson Correlation	-.147	.454*	-.062	-.205	-.213	-.095	-.237	-.146	1	.221	-.132
	Sig. (2-tailed)	.503	.026	.808	.400	.318	.689	.265	.496		.322	.568
	N	23	24	18	19	24	20	24	24	24	22	21
COMPUTE Log_AMT=LG10 (AMT_Mass)	Pearson Correlation	.134	-.542**	-.386*	-.491**	-.243	-.247	-.369	.062	.221	1	.570**
	Sig. (2-tailed)	.464	.000	.018	.008	.265	.309	.084	.778	.322		.000
	N	32	43	37	28	23	19	23	23	22	43	42
COMPUTE Log_RMT=LG10 (RMT_FDI)	Pearson Correlation	-.149	-.556**	-.430**	-.239	-.190	.039	-.297	-.328	-.132	.570**	1
	Sig. (2-tailed)	.417	.000	.008	.220	.396	.875	.180	.137	.568	.000	
	N	32	42	37	28	22	19	22	22	21	42	42

**. Correlation is significant at the 0.01 level (2-tailed).

*. Correlation is significant at the 0.05 level (2-tailed).

c. Cannot be computed because at least one of the variables is constant.

```
UNIANOVA CSP_M BY group WITH age
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /EMMEANS=TABLES(OVERALL) WITH(age=MEAN)
  /EMMEANS=TABLES(group) WITH(age=MEAN) COMPARE ADJ(BONFERRONI)
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
group	1	Healthy	93
	2	SD	87
	3	MTD	47

Descriptive Statistics

Dependent Variable: CSP_M

group	Mean	Std. Deviation	N
Healthy	42.4142	12.82521	93
SD	32.0609	14.94228	87
MTD	46.2838	16.51238	47
Total	39.2474	15.54742	227

Tests of Between-Subjects Effects

Dependent Variable: CSP_M

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Corrected Model	10397.342[a]	3	3465.781	17.473	.000	52.419
Intercept	3391.431	1	3391.431	17.098	.000	17.098
age	2644.492	1	2644.492	13.333	.000	13.333
group	8513.408	2	4256.704	21.461	.000	42.921
Error	44231.882	223	198.349			
Total	404290.598	227				
Corrected Total	54629.224	226				

a. R Squared = .190 (Adjusted R Squared = .179)

b. Computed using alpha = .05

2. group

Pairwise Comparisons

Dependent Variable: CSP_M

(I) group	(J) group	Mean Difference (I-J)	Std. Error	Sig.[b]	95% Confidence Interval for Difference[b]	
					Lower Bound	Upper Bound
Healthy	SD	10.544*	2.101	.000	5.475	15.613
	MTD	-4.713	2.531	.192	-10.818	1.393
SD	Healthy	-10.544*	2.101	.000	-15.613	-5.475
	MTD	-15.257*	2.565	.000	-21.444	-9.069
MTD	Healthy	4.713	2.531	.192	-1.393	10.818
	SD	15.257*	2.565	.000	9.069	21.444

Based on estimated marginal means

*. The mean difference is significant at the .05 level.

b. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: CSP_M

	Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Contrast	8513.408	2	4256.704	21.461	.000	42.921
Error	44231.882	223	198.349			

The F tests the effect of group. This test is based on the linearly independent pairwise comparisons among the estimated marginal means.

a. Computed using alpha = .05

```
UNIANOVA CSP_F BY group WITH age
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /EMMEANS=TABLES(OVERALL) WITH(age=MEAN)
  /EMMEANS=TABLES(group) WITH(age=MEAN) COMPARE ADJ(BONFERRONI)
  /PRINT=OPOWER DESCRIPTIVE PARAMETER
  /CRITERIA=ALPHA(.05)
  /DESIGN=age group.
```

Univariate Analysis of Variance

Notes

Output Created		26-JUN-2012 18:43:44
Comments		
Input	Data	C:\Users\w1089035\Documents\My Dropbox\U of MN Courses\Laryngeal Dystonia\Data Analysis\MASTER_Filtered for subjects_20120616.sav
	Active Dataset	DataSet1
	Filter	<none>
	Weight	<none>
	Split File	<none>
	N of Rows in Working Data File	1000
Missing Value Handling	Definition of Missing	User-defined missing values are treated as missing.
	Cases Used	Statistics are based on all cases with valid data for all variables in the model.
Syntax		UNIANOVA CSP_F BY group WITH age /METHOD=SSTYPE(3) /INTERCEPT=INCLUDE /EMMEANS=TABLES(OVERALL) WITH(age=MEAN) /EMMEANS=TABLES(group) WITH(age=MEAN) COMPARE ADJ(BONFERRONI) /PRINT=OPOWER DESCRIPTIVE PARAMETER /CRITERIA=ALPHA(.05) /DESIGN=age group.
Resources	Processor Time	00:00:00.06
	Elapsed Time	00:00:00.07

[DataSet1] C:\Users\w1089035\Documents\My Dropbox\U of MN Courses\Laryngeal Dystonia\Data Analysis\MASTER_Filtered for subjects_20120616.sav

Between-Subjects Factors

		Value Label	N
group	1	Healthy	98
	2	SD	100
	3	MTD	46

Descriptive Statistics

Dependent Variable: CSP_F

group	Mean	Std. Deviation	N
Healthy	125.2191	32.86517	98
SD	98.5732	36.34822	100
MTD	92.3537	22.11982	46
Total	108.1027	35.53178	244

Tests of Between-Subjects Effects

Dependent Variable: CSP_F

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Corrected Model	56018.083[a]	3	18672.694	17.871	.000	53.612
Intercept	137659.016	1	137659.016	131.746	.000	131.746
age	6816.403	1	6816.403	6.524	.011	6.524
group	50451.740	2	25225.870	24.142	.000	48.285
Error	250771.169	240	1044.880			
Total	3158220.786	244				
Corrected Total	306789.252	243				

2. group

Pairwise Comparisons

Dependent Variable: CSP_F

(I) group	(J) group	Mean Difference (I-J)	Std. Error	Sig.[b]	95% Confidence Interval for Difference[b]	
					Lower Bound	Upper Bound
Healthy	SD	26.372*	4.596	.000	15.291	37.452
	MTD	34.087*	5.797	.000	20.111	48.063
SD	Healthy	-26.372*	4.596	.000	-37.452	-15.291
	MTD	7.716	5.789	.551	-6.240	21.671
MTD	Healthy	-34.087*	5.797	.000	-48.063	-20.111
	SD	-7.716	5.789	.551	-21.671	6.240

Based on estimated marginal means

*. The mean difference is significant at the .05 level.

b. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: CSP_F

	Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Contrast	50451.740	2	25225.870	24.142	.000	48.285
Error	250771.169	240	1044.880			

```
UNIANOVA Log_Slope_M_C BY group WITH gender
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /EMMEANS=TABLES(OVERALL) WITH(gender=MEAN)
  /EMMEANS=TABLES(group) WITH(gender=MEAN) COMPARE ADJ(BONFERRONI)
  /PRINT=OPOWER DESCRIPTIVE PARAMETER
  /CRITERIA=ALPHA(.05)
  /DESIGN=gender group.
```

Univariate Analysis of Variance

```
 [DataSet1] C:\Users\w1089035\Documents\My Dropbox\U of MN
Courses\Laryngeal Dystonia\Data Analysis\MASTER_Filtered for
subjects_20120616.sav
```

Between-Subjects Factors

		Value Label	N
group	1	Healthy	9
	2	SD	8
	3	MTD	4

Descriptive Statistics

Dependent Variable: Log_Slope_M_C

group	Mean	Std. Deviation	N
Healthy	.0036	.61445	9
SD	-.0086	.35599	8
MTD	.3063	.60999	4
Total	.0566	.51636	21

Tests of Between-Subjects Effects

Dependent Variable: Log_Slope_M_C

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Corrected Model	1.794[a]	3	.598	2.873	.067	8.618
Intercept	1.361	1	1.361	6.537	.020	6.537
gender	1.485	1	1.485	7.134	.016	7.134
group	.308	2	.154	.739	.492	1.478
Error	3.539	17	.208			
Total	5.400	21				
Corrected Total	5.332	20				

2. group

Pairwise Comparisons

Dependent Variable: Log_Slope_M_C

(I) group	(J) group	Mean Difference (I-J)	Std. Error	Sig.[a]	95% Confidence Interval for Difference[a]	
					Lower Bound	Upper Bound
Healthy	SD	-.085	.225	1.000	-.682	.511
	MTD	-.333	.274	.726	-1.061	.396
SD	Healthy	.085	.225	1.000	-.511	.682
	MTD	-.247	.281	1.000	-.992	.497
MTD	Healthy	.333	.274	.726	-.396	1.061
	SD	.247	.281	1.000	-.497	.992

Based on estimated marginal means

a. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: Log_Slope_M_C

	Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Contrast	.308	2	.154	.739	.492	1.478
Error	3.539	17	.208			

```
UNIANOVA Log_Lat_M_C BY group WITH gender
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /EMMEANS=TABLES(OVERALL) WITH(gender=MEAN)
  /EMMEANS=TABLES(group) WITH(gender=MEAN) COMPARE ADJ(BONFERRONI)
  /PRINT=OPOWER DESCRIPTIVE PARAMETER
  /CRITERIA=ALPHA(.05)
  /DESIGN=gender group.
```

Univariate Analysis of Variance

```
[DataSet1] C:\Users\w1089035\Documents\My Dropbox\U of MN
Courses\Laryngeal Dystonia\Data Analysis\MASTER_Filtered for
subjects_20120616.sav
```

Between-Subjects Factors

	Value Label	N

group	1	Healthy	179
	2	SD	165
	3	MTD	87

Descriptive Statistics

Dependent Variable: Log transformed latency contralateral masseter

group	Mean	Std. Deviation	N
Healthy	.8990	.10545	179
SD	.8527	.05253	165
MTD	.8310	.05742	87
Total	.8676	.08415	431

Tests of Between-Subjects Effects

Dependent Variable: Log transformed latency contralateral masseter

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Corrected Model	.388[a]	3	.129	20.778	.000	62.333
Intercept	140.392	1	140.392	22561.522	.000	22561.522
gender	.058	1	.058	9.332	.002	9.332
group	.311	2	.155	24.951	.000	49.903
Error	2.657	427	.006			
Total	327.443	431				
Corrected Total	3.045	430				

2. group

Pairwise Comparisons

Dependent Variable: Log transformed latency contralateral masseter

(I) group	(J) group	Mean Difference (I-J)	Std. Error	Sig.[b]	95% Confidence Interval for Difference[b]	
					Lower Bound	Upper Bound
Healthy	SD	.045*	.009	.000	.025	.066
	MTD	.066*	.010	.000	.041	.091
SD	Healthy	-.045*	.009	.000	-.066	-.025
	MTD	.021	.010	.147	-.004	.046
MTD	Healthy	-.066*	.010	.000	-.091	-.041
	SD	-.021	.010	.147	-.046	.004

Based on estimated marginal means

*. The mean difference is significant at the .05 level.

b. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: Log transformed latency contralateral masseter

	Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Contrast	.311	2	.155	24.951	.000	49.903
Error	2.657	427	.006			

```
UNIANOVA Log_Lat_M_I BY group WITH gender
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /EMMEANS=TABLES(OVERALL) WITH(gender=MEAN)
  /EMMEANS=TABLES(group) WITH(gender=MEAN) COMPARE ADJ(BONFERRONI)
  /PRINT=OPOWER DESCRIPTIVE PARAMETER
  /CRITERIA=ALPHA(.05)
  /DESIGN=gender group.
```

Univariate Analysis of Variance

```
[DataSet1] C:\Users\w1089035\Documents\My Dropbox\U of MN
Courses\Laryngeal Dystonia\Data Analysis\MASTER_Filtered for
subjects_20120616.sav
```

Between-Subjects Factors

		Value Label	N
group	1	Healthy	94
	2	SD	80
	3	MTD	44

Descriptive Statistics

Dependent Variable: Log transformed latency ipsilateral masseter

group	Mean	Std. Deviation	N
Healthy	.6346	.10815	94
SD	.6264	.11493	80
MTD	.6099	.12118	44
Total	.6266	.11321	218

Tests of Between-Subjects Effects

Dependent Variable: Log transformed latency ipsilateral masseter

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Corrected Model	.502[a]	3	.167	15.720	.000	47.161
Intercept	39.590	1	39.590	3717.783	.000	3717.783
gender	.484	1	.484	45.437	.000	45.437
group	.004	2	.002	.193	.825	.386
Error	2.279	214	.011			
Total	88.370	218				
Corrected Total	2.781	217				

2. group

Pairwise Comparisons

Dependent Variable: Log transformed latency ipsilateral masseter

(I) group	(J) group	Mean Difference (I-J)	Std. Error	Sig.[a]	95% Confidence Interval for Difference[a]	
					Lower Bound	Upper Bound
Healthy	SD	.002	.016	1.000	-.036	.040
	MTD	.012	.019	1.000	-.034	.057
SD	Healthy	-.002	.016	1.000	-.040	.036
	MTD	.010	.019	1.000	-.037	.056
MTD	Healthy	-.012	.019	1.000	-.057	.034
	SD	-.010	.019	1.000	-.056	.037

Based on estimated marginal means

a. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: Log transformed latency ipsilateral masseter

	Sum of Squares	df	Mean Square	F	Sig.	Noncent. Parameter
Contrast	.004	2	.002	.193	.825	.386
Error	2.279	214	.011			

```
ONEWAY PP_3_M Log_PP_3_F PP_10_M PP_10_F LogARC_M_C LogARC_M_I
LogARC_FDI Log_AMT Log_RMT BY group
  /STATISTICS DESCRIPTIVES
  /MISSING ANALYSIS
  /POSTHOC=BONFERRONI ALPHA(0.05).
```

Oneway

[DataSet1] C:\Users\w1089035\Documents\My Dropbox\U of MN Courses\Laryngeal Dystonia\Data Analysis\MASTER_Filtered for subjects_20120616.sav

ANOVA

		Sum of Squares	df	Mean Square	F
PP_3_M	Between Groups	.022	2	.011	.348
	Within Groups	.633	20	.032	
	Total	.655	22		
Log_PP_3_F	Between Groups	.593	2	.297	3.029
	Within Groups	2.153	22	.098	
	Total	2.746	24		
PP_10_M	Between Groups	.136	2	.068	.635
	Within Groups	2.362	22	.107	
	Total	2.499	24		
PP_10_F	Between Groups	.667	2	.333	.502
	Within Groups	14.605	22	.664	
	Total	15.272	24		
LogARC_M_C	Between Groups	.116	2	.058	.358
	Within Groups	3.569	22	.162	
	Total	3.685	24		
LogARC_M_I	Between Groups	.107	2	.054	.378
	Within Groups	3.117	22	.142	
	Total	3.224	24		
LogARC_FDI	Between Groups	.829	2	.414	1.483
	Within Groups	5.870	21	.280	
	Total	6.698	23		
COMPUTE Log_AMT=LG10(AMT_Mass)	Between Groups	.034	2	.017	2.762
	Within Groups	.245	40	.006	
	Total	.279	42		
COMPUTE Log_RMT=LG10(RMT_FDI)	Between Groups	.011	2	.006	1.346
	Within Groups	.163	39	.004	
	Total	.174	41		

ANOVA

		Sig.
PP_3_M	Between Groups	.710
	Within Groups	
	Total	
Log_PP_3_F	Between Groups	.069
	Within Groups	
	Total	
PP_10_M	Between Groups	.540
	Within Groups	
	Total	
PP_10_F	Between Groups	.612
	Within Groups	
	Total	
LogARC_M_C	Between Groups	.703
	Within Groups	
	Total	
LogARC_M_I	Between Groups	.690
	Within Groups	
	Total	
LogARC_FDI	Between Groups	.250
	Within Groups	
	Total	
COMPUTE Log_AMT=LG10(AMT_Mass)	Between Groups	.075
	Within Groups	
	Total	
COMPUTE Log_RMT=LG10(RMT_FDI)	Between Groups	.272
	Within Groups	
	Total	

Post Hoc Tests

Multiple Comparisons

Bonferroni

Dependent Variable	(I) group	(J) group	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval

						Lower Bound
PP_3_M	Healthy	SD	.06370	.08438	1.000	-.1567
		MTD	-.00367	.09743	1.000	-.2582
	SD	Healthy	-.06370	.08438	1.000	-.2841
		MTD	-.06737	.10141	1.000	-.3323
	MTD	Healthy	.00367	.09743	1.000	-.2509
		SD	.06737	.10141	1.000	-.1976
Log_PP_3_F	Healthy	SD	.04044	.13992	1.000	-.3221
		MTD	.40259	.17136	.085	-.0414
	SD	Healthy	-.04044	.13992	1.000	-.4030
		MTD	.36215	.17136	.138	-.0819
	MTD	Healthy	-.40259	.17136	.085	-.8466
		SD	-.36215	.17136	.138	-.8062
PP_10_M	Healthy	SD	-.06030	.14655	1.000	-.4400
		MTD	.14169	.17948	1.000	-.3234
	SD	Healthy	.06030	.14655	1.000	-.3194
		MTD	.20198	.17948	.818	-.2631
	MTD	Healthy	-.14169	.17948	1.000	-.6068
		SD	-.20198	.17948	.818	-.6671
PP_10_F	Healthy	SD	.11071	.36438	1.000	-.8335
		MTD	.44438	.44628	.991	-.7120
	SD	Healthy	-.11071	.36438	1.000	-1.0549
		MTD	.33367	.44628	1.000	-.8227
	MTD	Healthy	-.44438	.44628	.991	-1.6008
		SD	-.33367	.44628	1.000	-1.4901
LogARC_M_C	Healthy	SD	.01309	.18013	1.000	-.4537
		MTD	.17634	.22061	1.000	-.3953
	SD	Healthy	-.01309	.18013	1.000	-.4798
		MTD	.16324	.22061	1.000	-.4084
	MTD	Healthy	-.17634	.22061	1.000	-.7480
		SD	-.16324	.22061	1.000	-.7349

Multiple Comparisons

Bonferroni

Dependent Variable	(I) group	(J) group	95% Confidence Interval
			Upper Bound
PP_3_M	Healthy	SD	.2841
		MTD	.2509
	SD	Healthy	.1567
		MTD	.1976
	MTD	Healthy	.2582
		SD	.3323
Log_PP_3_F	Healthy	SD	.4030
		MTD	.8466
	SD	Healthy	.3221
		MTD	.8062
	MTD	Healthy	.0414
		SD	.0819
PP_10_M	Healthy	SD	.3194
		MTD	.6068
	SD	Healthy	.4400
		MTD	.6671
	MTD	Healthy	.3234
		SD	.2631
PP_10_F	Healthy	SD	1.0549
		MTD	1.6008
	SD	Healthy	.8335
		MTD	1.4901
	MTD	Healthy	.7120
		SD	.8227
LogARC_M_C	Healthy	SD	.4798
		MTD	.7480
	SD	Healthy	.4537
		MTD	.7349
	MTD	Healthy	.3953
		SD	.4084

Multiple Comparisons

Bonferroni

Dependent Variable	(I) group	(J) group	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
						Lower Bound
LogARC_M_I	Healthy	SD	-.13575	.16833	1.000	-.5719
		MTD	-.00667	.20616	1.000	-.5409
	SD	Healthy	.13575	.16833	1.000	-.3004
		MTD	.12908	.20616	1.000	-.4051
	MTD	Healthy	.00667	.20616	1.000	-.5275
		SD	-.12908	.20616	1.000	-.6633
LogARC_FDI	Healthy	SD	-.36863	.24291	.432	-1.0005
		MTD	-.41025	.29488	.536	-1.1773
	SD	Healthy	.36863	.24291	.432	-.2633
		MTD	-.04162	.28957	1.000	-.7949
	MTD	Healthy	.41025	.29488	.536	-.3568
		SD	.04162	.28957	1.000	-.7117
COMPUTE Log_AMT=LG10(AMT_Mass)	Healthy	SD	-.03605	.02802	.617	-.1061
		MTD	-.08373	.03813	.102	-.1790
	SD	Healthy	.03605	.02802	.617	-.0340
		MTD	-.04768	.04225	.797	-.1533
	MTD	Healthy	.08373	.03813	.102	-.0116
		SD	.04768	.04225	.797	-.0579
COMPUTE Log_RMT=LG10(RMT_FDI)	Healthy	SD	-.01264	.02393	1.000	-.0725
		MTD	-.05126	.03147	.334	-.1300
	SD	Healthy	.01264	.02393	1.000	-.0472
		MTD	-.03862	.03540	.846	-.1272
	MTD	Healthy	.05126	.03147	.334	-.0275
		SD	.03862	.03540	.846	-.0499

Multiple Comparisons

Bonferroni

Dependent Variable	(I) group	(J) group	95% Confidence Interval
			Upper Bound
LogARC_M_I	Healthy	SD	.3004
		MTD	.5275

	SD	Healthy	.5719
		MTD	.6633
	MTD	Healthy	.5409
		SD	.4051
	Healthy	SD	.2633
		MTD	.3568
LogARC_FDI	SD	Healthy	1.0005
		MTD	.7117
	MTD	Healthy	1.1773
		SD	.7949
	Healthy	SD	.0340
		MTD	.0116
COMPUTE Log_AMT=LG10(AMT_Mass)	SD	Healthy	.1061
		MTD	.0579
	MTD	Healthy	.1790
		SD	.1533
	Healthy	SD	.0472
		MTD	.0275
COMPUTE Log_RMT=LG10(RMT_FDI)	SD	Healthy	.0725
		MTD	.0499
	MTD	Healthy	.1300
		SD	.1272

```
SORT CASES  BY group.
SPLIT FILE LAYERED BY group.
DESCRIPTIVES VARIABLES=MEP_M_I MEP_F CSP_M CSP_F PP_3_M PP_10_M PP_10_F
CAPE_V VHI_total Log_MEP_M_C Log_MEP_M_I Log_Lat_M_C Log_Lat_M_I
Log_PP_3_F Log_Slope_M_C LogARC_M_C LogARC_M_I LogARC_FDI
  /STATISTICS=MEAN STDDEV MIN MAX
  /SORT=NAME (A).
```

Descriptives

```
[DataSet1] C:\Users\w1089035\Documents\My Dropbox\U of MN
Courses\Laryngeal Dystonia\Data Analysis\MASTER_Filtered for
subjects_20120616.sav
```

Descriptive Statistics[a]

group		N	Minimum	Maximum	Mean	Std. Deviation
Healthy	CAPE_V	0				
	CSP_F	98	63.18	194.11	125.2191	32.86517

	CSP_M	93	23.46	80.73	42.4142	12.82521
	Log transformed latency contralateral masseter	179	.71	1.22	.8990	.10545
	Log transformed latency ipsilateral masseter	94	.35	.86	.6346	.10815
	Log transformed MEP for contralateral masseter	192	1.38	2.68	2.1375	.23132
	Log transformed MEP for ipsilateral masseter	99	1.89	3.06	2.3401	.30386
	Log_PP_3_F	10	-.87	.15	-.2205	.32659
	Log_Slope_M_C	9	-.90	.87	.0036	.61445
	LogARC_FDI	9	2.58	4.68	4.1264	.63512
	LogARC_M_C	10	3.15	4.54	3.6681	.48046
	LogARC_M_I	10	3.36	4.31	3.7603	.33537
	MEP_F	197	36.16	4851.91	1085.2848	956.38922
	MEP_M_I	99	76.97	1141.34	285.6122	241.57981
	PP_10_F	10	.90	2.32	1.6323	.42625
	PP_10_M	10	.74	1.32	1.0026	.19201
	PP_3_M	10	.77	1.39	1.0542	.20092
	VHI_total	0				
	Valid N (listwise)	0				
SD	CAPE_V	9	15.00	67.00	40.8889	16.64665
	CSP_F	100	42.12	183.30	98.5732	36.34822
	CSP_M	87	10.12	88.32	32.0609	14.94228
	Log transformed latency contralateral masseter	165	.61	1.04	.8527	.05253
	Log transformed latency ipsilateral masseter	80	.44	.90	.6264	.11493
	Log transformed MEP for contralateral masseter	190	1.34	2.76	2.0097	.27354
	Log transformed MEP for ipsilateral masseter	90	1.78	3.01	2.3360	.30211
	Log_PP_3_F	10	-.63	.10	-.2610	.26903

Descriptive Statistics[a]

group		N	Minimum	Maximum	Mean	Std. Deviation
SD	Log_Slope_M_C	8	-.49	.49	-.0086	.35599
	LogARC_FDI	10	3.48	5.25	4.4950	.52487
	LogARC_M_C	10	3.25	4.43	3.6550	.39279
	LogARC_M_I	10	3.31	4.57	3.8961	.39745
	MEP_F	177	42.56	4366.90	870.8566	768.27095
	MEP_M_I	90	60.06	1017.62	286.0907	260.46450
	PP_10_F	10	.63	4.10	1.5216	1.11272
	PP_10_M	10	.42	1.85	1.0629	.40556
	PP_3_M	8	.71	1.17	.9905	.14188
	VHI_total	10	47.00	109.00	68.8000	16.71194
	Valid N (listwise)	4				
MTD	CAPE_V	4	5.00	65.00	29.5000	26.85144
	CSP_F	46	49.53	147.03	92.3537	22.11982
	CSP_M	47	16.79	83.49	46.2838	16.51238
	Log transformed latency contralateral masseter	87	.72	1.02	.8310	.05742
	Log transformed latency ipsilateral masseter	44	.49	.84	.6099	.12118
	Log transformed MEP for contralateral masseter	100	1.35	2.58	1.9938	.25727
	Log transformed MEP for ipsilateral masseter	50	1.78	2.69	2.2703	.28297
	Log_PP_3_F	5	-1.07	-.20	-.6231	.36811
	Log_Slope_M_C	4	-.30	1.15	.3063	.60999
	LogARC_FDI	5	4.31	4.85	4.5367	.20194
	LogARC_M_C	5	3.34	3.69	3.4917	.16047
	LogARC_M_I	5	3.11	4.13	3.7670	.41317
	MEP_F	63	31.59	5624.63	1087.7219	1252.74592
	MEP_M_I	50	59.59	484.78	225.2735	131.21066
	PP_10_F	5	.17	2.00	1.1879	.67580
	PP_10_M	5	.49	1.40	.8609	.37087

PP_3_M	5	.92	1.35	1.0579	.17932
VHI_total	5	57.00	87.00	72.2000	12.55787
Valid N (listwise)	1				

a. No statistics are computed for one or more split files because there are no valid cases.