

Human Cerebral Cortex Plasticity in Response to Long-term Bedrest as an Analog to
Microgravity

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Principal Investigator

Donna Roberts, M.D.
Department of Radiology
Medical University of South Carolina
171 Ashley Avenue
PO Box 250322
Charleston, SC 29452
843-792-9343
email: robertdr@musc.edu

Co-Investigator

David Ramsey, MS
Research Services
South Carolina Research Authority
5300 International Blvd.
North Charleston, SC 29418
843-760-4538
email: ramsey@scra.org

TMS Bedrest Team

Kevin Johnson, Ph.D., Jejo Kola, Raffaella Ricci, Ph.D., Jeffrey Borckardt Ph.D., Charles Epstein, M.D.,
Mark S. George, M.D., Ph.D.

I. INTRODUCTION

Human performance in microgravity is crucial to the success of long-term space station and interplanetary missions. To assure maximum performance capability in the space environment, the acute and long-term effects of weightlessness on the central nervous system and specifically the sensory-motor system must be investigated. The purpose of this study was (1) to investigate motor cortex plasticity induced by a long-term bedrest model of microgravity, (2) to correlate cortical plasticity with alterations in performance using NASA standard measures of performance, and (3) to purposefully modulate cortical maps to ameliorate the known degradation in performance that occurs during periods of disuse. The end-product would be the development of a rehabilitative strategy for astronauts on long-term missions that would be suitable for use on-board the spacecraft to ensure maximal performance upon landing on a planetary surface.

The NASA Bioastronautics Roadmap has identified several potential risks resulting in impaired astronaut performance. Impaired sensory-motor capability adversely affecting performance of operational tasks during and following missions throughout re-adaptation is a potential risk (Risks #13 and #14). Furthermore, human performance failure may occur due to conditions such as neurobehavioral and cognitive problems (Risk #25). Human performance failure may also occur due to a mismatch between crew cognitive capabilities and task demands (Risk #26). This study specifically addresses the role of sensory-motor cortical plasticity in performance degradation in a NASA-relevant model and suggests a potential countermeasure (on-board cortical stimulation) for attenuating human performance failure which would be useful for long-term missions, such as to the Moon or Mars.

Research on the effects of microgravity on the nervous system using animal models ^[1-5] has shown changes in neurotransmitter concentrations and neural architecture plasticity. In addition, ground-based research including pilot data from our laboratory suggest that alterations in cortical excitability occur in a model for microgravity (lower extremity immobilization) ^[6]. Functional cortical reorganization can be seen as a form of motor learning following, for example, the practicing of a complex motor task or during periods of adjustment to altered sensory inputs. For an in-depth review of cortical plasticity see for example, ^[7,8]. It is likely that cortical motor map reorganization occurs as an adaptive response to the microgravity environment. Although this adaptation may be useful in a microgravity environment, it may not be optimal for return to Earth, operations on a planetary surface, or in the case of an emergency landing where optimal motor performance is required for safe crew egress. The identification and proper understanding of cortical plasticity in microgravity and its relationship to astronaut performance is a critical step in preparing for future long-term human space habitation. Even though normal movement clearly involves vestibular systems along with cerebellar control, motor cortical areas are still the final common pathway for control of movement and therefore play a pivotal role in adaptation to microgravity.

Multiple non-invasive methods of imaging brain function are now available for researching the neurobiology of brain plasticity. Most of these are not portable and require expensive scanners (functional MRI or positron emission tomography). However, one method, Transcranial Magnetic Stimulation (TMS), allows non-invasive, painless stimulation and evaluation of the cortex through the intact human skull using time-varying magnetic fields. TMS is relatively

inexpensive and is portable -- making actual onboard space flight usage a future possibility. TMS has been shown to be a useful method for motor cortex mapping^[9] and its safety has been established in both animal and human studies^[10]. TMS has many additional therapeutic applications, some of which are close to FDA approval for clinical use. These include improving mental health^[11, 12], improving analogical reasoning^[13], and speeding up reaction times^[14-16], and improving motor performance^[17-21]. Already TMS is being used to improve motor function following stroke^[18, 19, 22, 23].

Although requiring further scientific study such as undertaken here, TMS may have the potential to enhance astronaut performance and mental health onboard a long-duration flight vehicle, or immediately following space flight upon return to Earth or landing on a foreign planet. For example, a light-weight, portable TMS system has been developed in our lab (WIPO Patent Publication Number WO/2003/082405). Such a countermeasure would be useful to NASA, particularly on missions to the Moon and Mars where astronauts will be subjected to lengthy periods of altered gravity, followed by the need for rapid recovery. In recent flights, the time for an astronaut to recover fine and gross motor skills was flexible. In the future, loss of these skills and protracted recovery times on a distant planet implies loss of time to achieve program objectives, extra weight and volume for consumables to span the recovery period, increased risk due to poor reaction times, or component breakage due to lack of limb coordination. Therefore astronauts will simply have to have the same motor skills on the day of landing on a foreign surface, as the day they left Earth. This will require the development of a countermeasure, such as onboard TMS, to ensure maximal operational capability.

II. SPECIFIC AIMS AND HYPOTHESES

As an initial step in this direction, we used TMS to evaluate motor cortex plasticity in a long-term bedrest model. We hypothesized that alterations in afferent signaling, as a result of disuse, would induce cortical reorganization by altering cortical excitability. We additionally hypothesized that altered cortical excitability correlates with motor function and that cortical excitability can be modulated in useful ways to enhance performance. Our intention was also to favorably alter cortical plasticity in order to improve post-bedrest performance leading to the development of a countermeasure to microgravity-induced performance degradation. Our specific aims were:

Specific aim #1: To evaluate cortical plasticity in response to 60-90 days of bedrest. This experiment tested the following hypothesis:

1. Sixty to ninety days of strict bedrest will induce changes in the neurophysiological properties of lower extremity motor cortex as measured by TMS-derived recruitment curves. These changes will be maximal 24 hours following return to normal activity.

Status: Completed.

Specific aim #2: Using the results obtained during the first bedrest campaign, we would then test whether it is possible to alter bedrest induced cortical plasticity in a useful way to ameliorate the degradation in performance that occurs during periods of disuse.

Status: Not accomplished, due to early termination of study, however based on the data that we have acquired, we could complete this aim if given access to 16 additional subjects.

Specific aim #3: To collaborate with NASA investigators to correlate our measures of cortical excitability with their measures of musculoskeletal lower limb functionality. The following hypothesis was tested.

1. Measures of cortical excitability will correlate with measures of lower limb functional recovery.

Status: Completed but need more subjects for statistical significance.

As in our original grant proposal, we had intended to carry out specific aim #1 in the first bedrest campaign. According to our original power analysis, we hypothesized that a sample size of 18 would be required for a power of 0.90 and an alpha of 0.05 for a two-tailed t-test, effect size 1.85 to detect changes in cortical plasticity in the long-term bedrest model. We were asked to participate in campaigns 3a and 3b. In campaign 3a, we were provided with 4 subjects that completed the protocol. In campaign 3b, we were provided with an additional 5 subjects, however, due to a nature disaster (Hurricane Rita in the wake of Hurricane Katrina in the fall of 2005) this campaign was abruptly terminated early without an opportunity to acquire the necessary post-bedrest data. Therefore, we were provided with a total of four subjects that carried out our experimental protocol towards specific aims #1 and #3 and the results we are reporting here are based on the data from these four subjects.

To carry out specific aim #2, leading to countermeasure development, we would require an additional 16 subjects, if the opportunity were to become available.

III. MATERIALS AND METHODS

Data Collection

In order to evaluate cortical plasticity induced by long-term bedrest, four experimental subjects (2 men, all right-handed) underwent TMS to assess motor cortex excitability prior to, during, and following 90 days of bedrest. Subjects also underwent functional mobility testing (FMT) as a NASA standard measure of motor performance. Finally, subjects underwent functional brain imaging to further assess motor cortex activation changes induced by long-term bedrest. This research was performed as part of a multi-investigator study and therefore all subjects underwent additional experimental measures reported elsewhere. The bedrest study was carried out within the University of Texas Medical Branch GCRC and the subjects were monitored continuously throughout to ensure compliance. Diet was strictly controlled. Written signed informed consent was obtained from all subjects and the study was approved by the Medical University of South Carolina, the University of Texas Medical Branch, and the Johnson Space Center Institution Review Boards.

In order to establish baseline measurements of cortical excitability and motor performance, TMS and FMT were performed on the subjects prior to bedrest. TMS was performed on all subjects for 4 days immediately prior to bedrest (BR-4, BR-3, BR-2, BR-1). FMT was performed on BR-

10 and BR-6. All subjects were then placed at bedrest in a 6 degrees head-down tilt position. On day 60 of bedrest, with the subjects maintained in the head-down tilt position, TMS was again performed. The following day the subjects were allowed out of bed to perform the FMT and then placed back at bedrest. Following 90 days of bedrest, TMS was again performed. Following this TMS session, the subjects were allowed to return to normal activity, but remained at the GCRC for an additional 2 weeks. During these two weeks, TMS sessions were repeated on days BR+1, BR+2, BR+3, BR+5, BR+8, BR+ 10 and BR+13. FMT was performed on BR+0, BR+1, BR+3, and BR+12. See Figure 1.

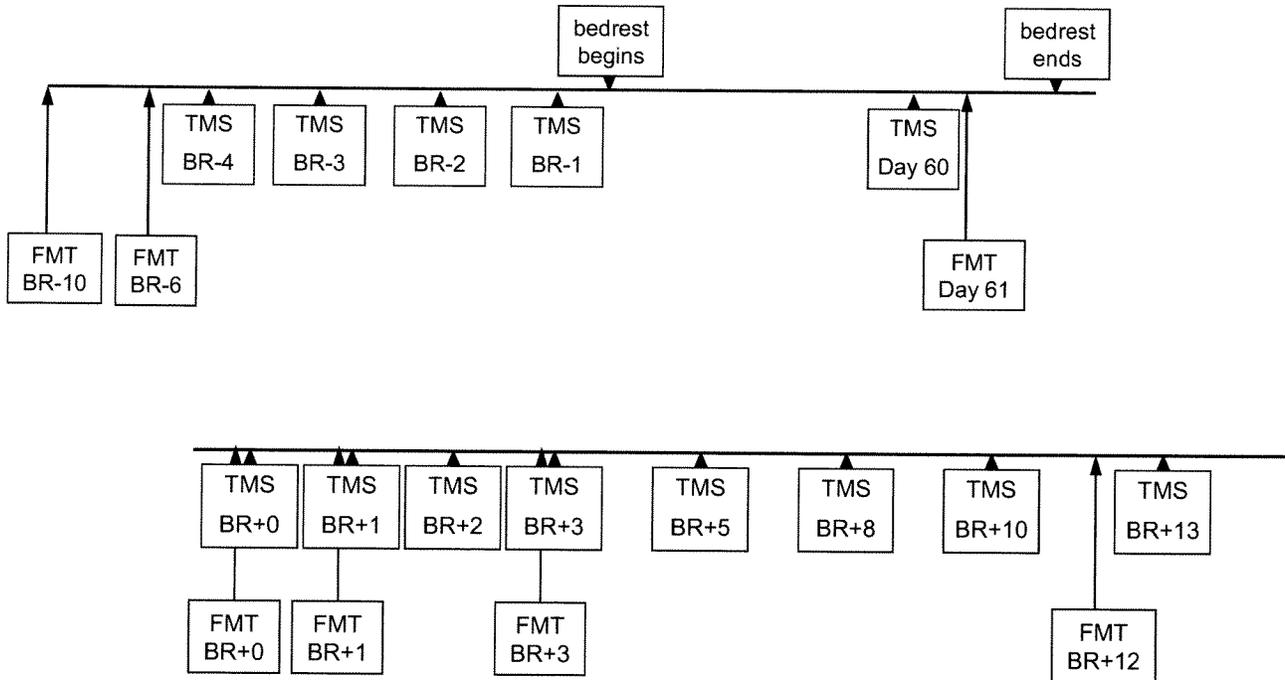


Figure 1. Timeline of study showing the data collection points for transcranial magnetic stimulation (TMS) and functional mobility testing (FMT).

Transcranial Magnetic Stimulation (TMS): All TMS sessions included resting motor threshold (rMT) determination followed by the acquisition of recruitment curves (RC) for the right upper and lower extremities. The rMT is defined as the minimal magnetic pulse causing a contraction of the target muscle as detected by electromyography (EMG) recordings of the motor evoked potential (MEP). The rMT value was then used to set TMS intensities for obtaining recruitment curves. Recruitment curves are created by measuring the MEP amplitudes in a target muscle over a range of TMS intensities. As the TMS output is increased, MEPs of increasing amplitude are obtained until a plateau level is reached^[24]. RC can be characterized by a maximum slope value and a plateau threshold. Changes in the slope of the RC reflect changes in cortical excitability and have been used to detect changes in motor cortex output maps induced for example by ischemia or amputation^[25]. Additional TMS measures of cortical excitability

(cortical silent period and paired pulse) were also acquired; however analysis of this data is not yet complete.

All TMS sessions were performed using MRI guidance. For TMS, the subjects were positioned supine with their lower extremity fully relaxed as verified by EMG recordings. Surface electrodes were placed along the medial gastrocnemius muscle belly. The peak-to-peak amplitudes of the MEPs were recorded using equipment and software from Cambridge Electronic Design, the signal was converted from analog to digital using the Micro 1401 MK II and conditioned using the CED1902 signal conditioner. For cortical stimulation, a TMS device with a figure-of-eight coil was used (SuperRapid, Whitland, South West Wales, The Magstim Company Limited). For the first TMS session, the optimal location for stimulus induction (the location that gave the maximum MEP amplitude) for each muscle was identified and the coil was secured in place by a holder. During subsequent sessions, the TMS coil was positioned using a neuronavigational guidance system (Brainsight™, Rogue Research Inc., Montreal Quebec) at the same location identified during the first session. At this location, the rMT was determined as the intensity needed to evoke an MEP in the relaxed muscle of more than 50 μ V in 5 out of 10 consecutive trials. RC's were then obtained. 10 stimuli were delivered at each of 8 intensities starting 5 points below the rMT and increasing by 5 increments (% maximum machine output: -5, 0, +5, +10, +15, +20, +25, +30, +35, +40) or until 100% of stimulator output was reached. The stimulus intensities were randomized throughout the RC acquisition. This procedure was performed during rest.

Functional Mobility Testing (FMT): The FMT is part of the NASA Standard Measures and was conducted by NASA personnel. Briefly, the FMT was an obstacle course set up on a base of 10 cm thick medium density foam (Sunmate Foam, Dynamic Systems, Inc., Leicester, NC). The foam base introduced a proprioceptive challenge into the otherwise normal walking task. The 6.0m X 4.0m course consisted of five vertical foam pylons arranged in a slalom fashion, a 46cm foam hurdle, a “portal”, and a “gate”. The portal obstacle was comprised of two successive 31cm foam hurdles with a horizontal foam bar suspended between them. The horizontal foam bar was adjusted to the height of each subject’s shoulders. The portal required subjects to step over hurdles while bending at the waist to duck under the horizontal foam bar. The gate obstacle was comprised of two vertical foam pylons adjusted to each subject’s shoulder width, requiring the subject to ‘squeeze’ through the pylons without touching them. Subjects walked in bare feet or socks at a preferred pace through the course, beginning and ending each trial at a start/finish line marked on the foam floor. They were instructed to complete the course as quickly as possible without running and without touching any of the obstacles. This task was performed three times in the clockwise direction and three times in the counterclockwise direction in a randomized order for a total of six trials during any given session. The dependent measure for each trial was time to complete the course (TCC), measured in seconds.

Functional Magnetic Resonance Imaging (fMRI): Also, changes in motor brain activity were explored using functional brain imaging of these four subjects before and following bedrest^[26]. Functional images were acquired on a 1.5T scanner (GE Medical Systems, Milwaukee WI) using a standard alternating block design (30s rest, 30s task, 6min total). A random finger tapping task was used to localize cortical control of hand movement and a random foot movement task was used to localize cortical control of foot movement. Analysis FSL 3.3.3 (University of Oxford) was used for time course extraction.

Data Analysis

For data analysis we used Hierarchical Linear Modeling (HLM) to assess the effects of time (pre-bedrest versus post-bedrest) and functional outcome (FMT scores) on individual recruitment curves. HLM has been shown to appropriately handle nested models with serially dependent data points^[27, 28] and it allows for modeling of variables at the individual subject-level (e.g., each subject's individual recruitment curves), at the broader organizational level to which each individual is assigned, and with respect to random effects nested within individuals (pre-bedrest versus post-bedrest).

MEP values that comprised the recruitment curves were log-transformed to correct for non-linearity and non-normality of the curves for statistical analyses. The linearity of the transformed recruitment curves and the normality of the residuals were verified by fitting an ordinary least squares model and examining normal probability plots in SPSS (Figure 2 - 4). For HLM analysis, "proc mixed" was used in SAS^[29], and the estimation method of the model was Restricted Maximum Likelihood (REML). The covariance structure was "Unstructured." Intercepts at the individual subject level were entered into the model as random effects at level-1.

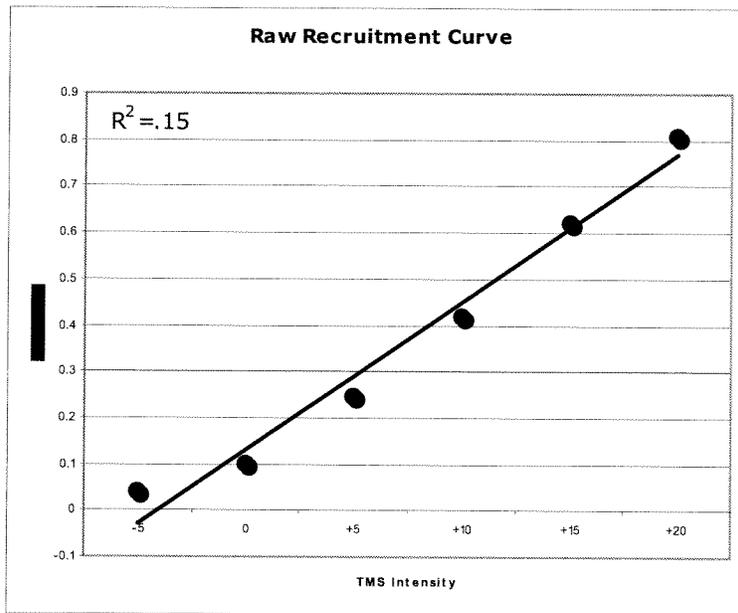


Figure 2. Linearity of the transformed recruitment curves.

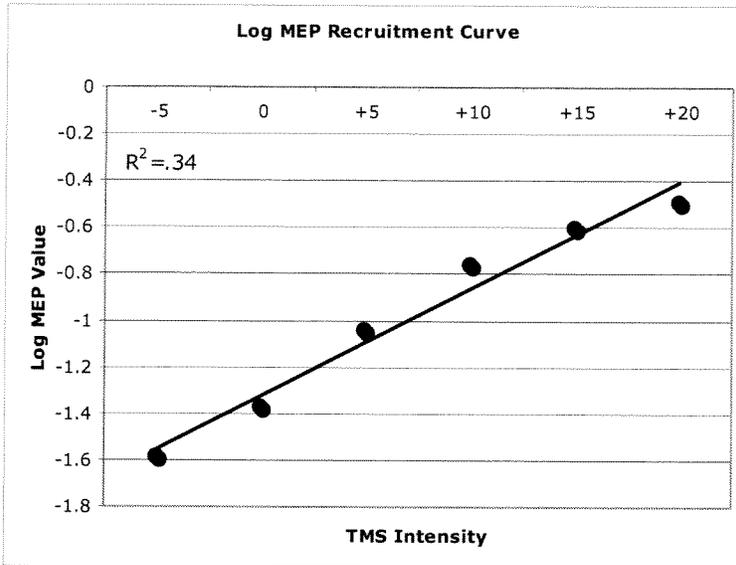


Figure 3. Linearity of the transformed recruitment curves.

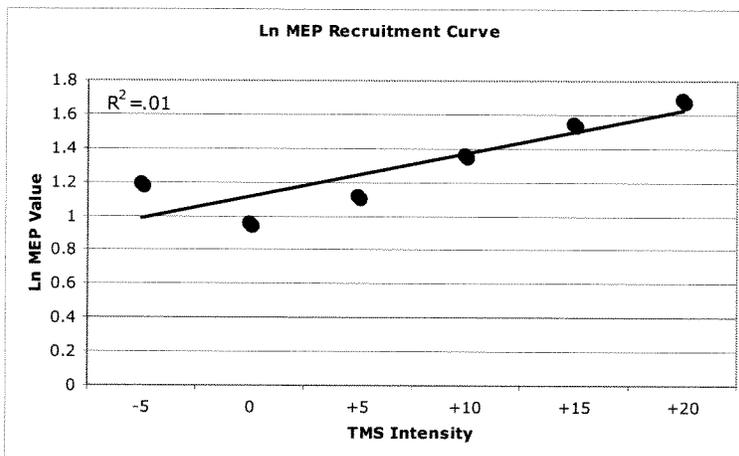


Figure 4. Linearity of the transformed recruitment curves.

HLM is a powerful method for evaluating changes in recruitment curves over time. Its power to detect significant changes is affected by a number of factors but the primary factors of interest are: 1) the number of participants, 2) the number of data points collected per participant, and 3) the effect-size. The power ($1-\beta$) for this study was 0.84.

If additional subjects became available, we would have adequate power under the following recruitment conditions:

04 participants	3 measurements per session	12 sessions	$1-\beta = 0.84$
06 participants	3 measurements per session	12 sessions	$1-\beta = 0.95$
08 participants	3 measurements per session	12 sessions	$1-\beta = 0.99$

IV. RESULTS

All of the subjects tolerated the study well without immediate or delayed adverse side effects from TMS. One subject became diaphoretic during his first TMS session, however vital signs remained stable and the subject quickly recovered to baseline. The subject attributed this reaction to anxiety and did not become diaphoretic on subsequent sessions. Two subjects were withdrawn from the bedrest study due to reasons unrelated to this experiment. Following 90 days of bedrest, one of the subjects demonstrated delayed functional recovery and at two weeks post-bedrest had not returned to the subject's pre-bedrest baseline as evidenced by performance on the functional mobility task.

Significant effects were observed on recruitment curve intercepts relative to baseline ($F(8, 5146) = 15.82, p < .0001$) and on recruitment curve slopes ($F(8, 5147) = 10.73, p < .0001$). Significant intercept increases were observed 2, 3, 5 and 8 days after bedrest ($t(5146) = 2.89, p = .004$; $t(5146) = 8.74, p < .0001$; $t(5146) = 6.09, p < .0001$; $t(5146) = 6.90, p < .0001$, respectively) suggesting an increase in cortical excitability following bedrest. Significant effects were also observed with respect to recruitment curve slopes on the same days ($t(5146) = 2.47, p = .013$; $t(5146) = 7.10, p < .0001$; $t(5146) = 4.28, p < .0001$; $t(5146) = 5.84, p < .0001$, respectively). These effects also support the notion that there is increased cortical excitability through at least 2-weeks following bedrest. See Figure 5 for linear model fits of the recruitment curves across measurement periods. Figure 6 shows the means (and standard errors) of the log MEP values across all study measurement periods, highlighting the significant effects described above.

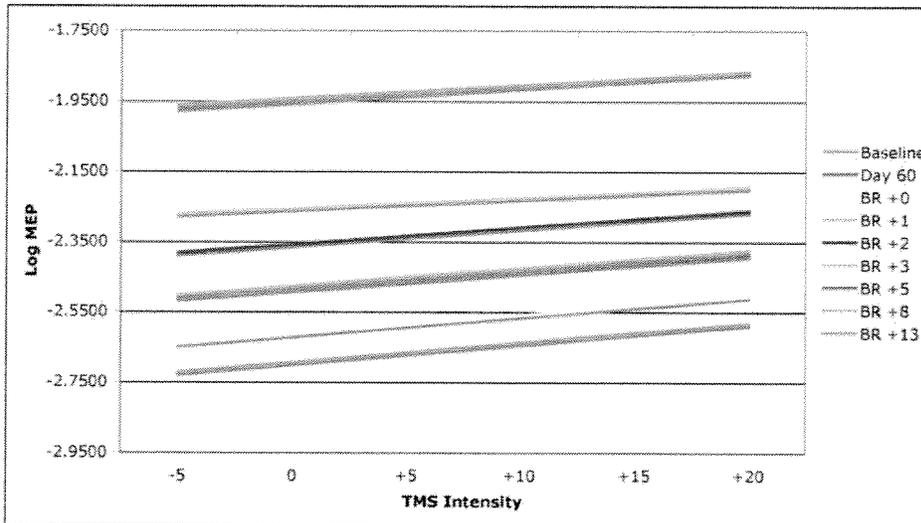


Figure 5. Linear model fits of the recruitment curves across measurement periods.

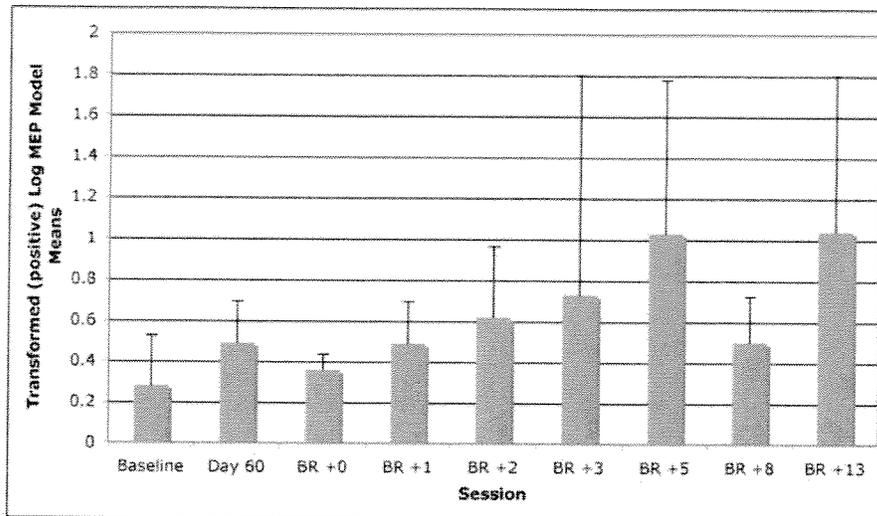


Figure 6. Means (and standard errors) of the log MEP values across all study measurement periods.

Next, log MEP values were correlated with functional mobility performance data from all participants across all measurement periods. The Pearson correlation coefficient was not statistically significant ($r=.30$, $p=.16$) with our limited n-size, but as can be seen in Figure 7, there appears to be an emergent trend supporting a positive relationship between cortical excitability and maze performance on functional mobility testing.

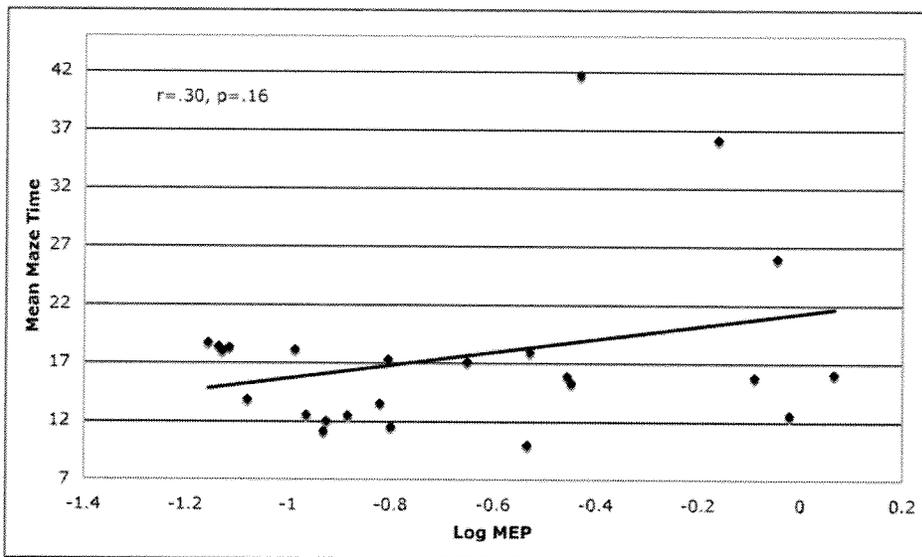


Figure 7. Log MEP values correlated with maze performance data.

Analysis of the fMRI data demonstrated that the hand fMRI signal remained stable or decreased slightly with bedrest^[26]. The leg function was of primary interest, as the legs are virtually unused during the bedrest period. For three out of the four subjects, the leg fMRI signal increased after 90 days of bedrest. See Figure 8 and Figure 9. This finding is again consistent with increased cortical excitability in the immediate post-bedrest period.

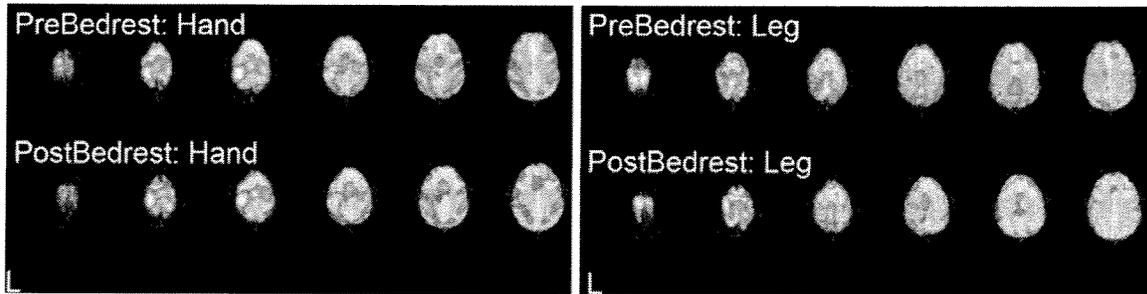


Figure 8. Example of fMRI activation patterns (Subject 1)

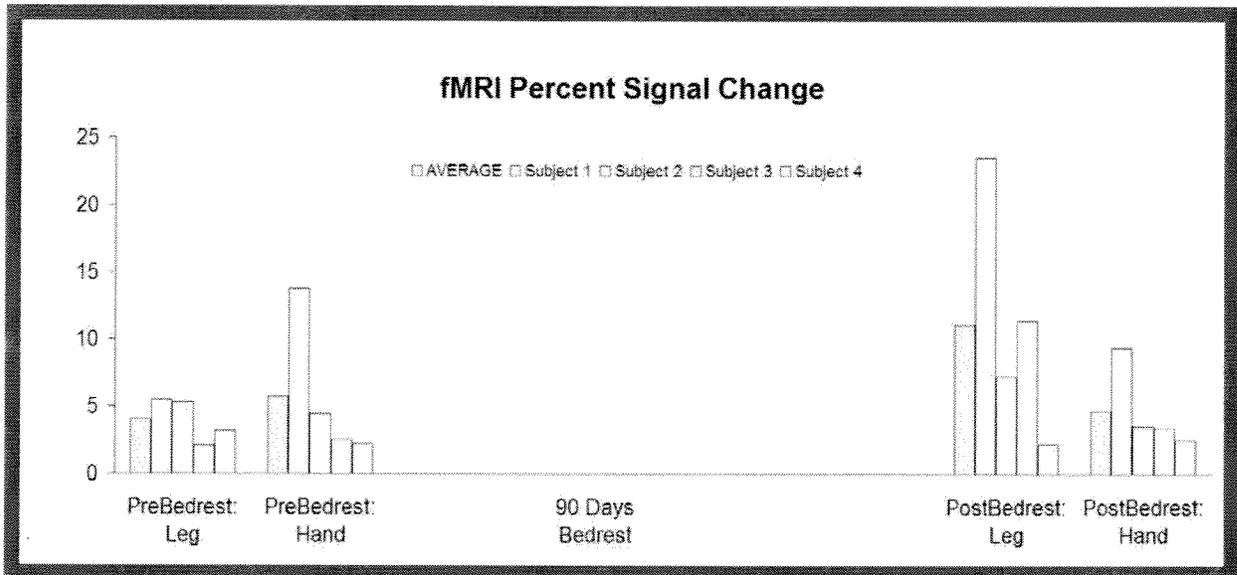


Figure 9. fMRI percent signal change for single voxel with highest z-value.

V. DISCUSSION

The purpose of this study was: (1) to evaluate lower extremity motor cortex plasticity in response to long-term bedrest, (2) to correlate alterations in cortical excitability with measures of lower limb functionality, and then (3) to purposefully modulate cortical maps (through the application of repetitive TMS) to ameliorate the known degradation in performance that occurs during periods of disuse.

In comparison with pre-bedrest measurements, we observed the development of increased lower extremity primary motor cortex excitability over the course of 90 days of bedrest as evidenced by statistically significant increases in motor cortex recruitment curve intercepts and slopes on days 2, 3, 5 and 8 days post bedrest following return to normal activity. These effects support the notion that there is increased cortical excitability for at least 2-weeks following bedrest. fMRI activation also supported this finding with increased activation of the leg motor cortex, but not the hand motor cortex, in the post bedrest period. Our results are in line with previous

studies which have demonstrated increased cortical excitability after limb disuse^[6, 30-32] but unlike prior studies by Liepert et al. and Zanette et al. which involved patients undergoing casting for bony fractures, our study involved healthy subjects without concomitant pathology.

The finding of increased cortical excitability during the recovery period following long-term bedrest with associated reduced motor output might reflect an abrupt increase in motor output drive during return to normal activity. Additionally, increased excitability of motor cortex has also been observed in other conditions such as experimentally-induced deafferentation^[33-36] or prolonged changes in limb position^[8, 37, 38] and therefore our findings may also reflect reduced sensory inputs from the lower extremity over the 90 days bedrest due to the lack of weight-bearing. The subjects in this study, however, were allowed to freely move the lower extremities throughout the study, and were only required to maintain the head-down tilt position. Finally, increased excitability of primary motor cortex may reflect learning (or in our case “relearning”) of lower extremity control during ambulation as a result of long-term disuse.

There is evidence for the critical role of primary motor cortex in early motor consolidation^[39, 40] and increased excitability has been shown to occur during acquisition of new motor skills^[14, 39, 41-45]. For example, Lotze and colleagues^[42] showed that a 30 minute period of hand motor training significantly increased motor cortex excitability as measured by TMS (i.e. increased recruitment curves and intracortical facilitation), fMRI and motor performance with respect to passive training. Perez and co-workers^[45] reported a similar finding for the leg motor area. Thirty-two minutes of motor skill training, but not passive or non-skill training, was found to increase motor cortex excitability as indexed by higher recruitment curves and decreased intracortical inhibition. Taken together, our data along with others, suggest that increased cortical excitability is associated with, or is a marker for, reorganization of the motor cortex. The corticospinal changes we observed during the recovery period after 90 days of bedrest may represent a marker of motor relearning with higher excitability serving as an index of ‘motor readjustment’ to the tasks of normal ambulation.

While numerous studies have demonstrated cortical plasticity in response to various environmental alterations, such as disuse as in our study, the functional implications of altered cortical maps are not yet completely understood. Many previous studies did not include a measure of motor performance or did not report correlations between the magnitude of excitability change and behavioral response. In our current study, we correlated TMS measures of cortical excitability with maze performance data from functional mobility testing for all participants across all measurement periods. While the Pearson correlation coefficient was not statistically significant due to our limited n-size, there appeared to be an emergent trend supporting a positive relationship between cortical excitability and maze performance.

Although further work is necessary to clarify the functional relevance of changes in cortical excitability in response to limb non-use, investigators have recently shown that altering corticospinal excitability can improve measures of functionality in chronic stroke patients. Changes in excitability of corticomotor circuits can be induced by peripheral nerve stimulation (PNS)^[46, 47] and by rTMS^[48]. PNS can improve motor function when delivered in isolation^[49] or together with TMS^[50]. More recently, rTMS^[18, 19, 22, 51] and electrical stimulation^[52-54] have been shown to constitute potentially safe and painless treatments for the reduction of motor impairments. In our laboratory, we have recently tested the safety and feasibility of daily rTMS as a ‘treatment’ to inhibit cortical reorganization due to lower extremity casting.

NASA has previously studied optimizing performance of astronauts in space through exercise strategies. Training regimes have been shown by TMS to induce motor cortex plasticity as evidenced by increased MEP values ^[45, 55, 56], increased recruitment curve slopes ^[55], enlarged cortical maps ^[57], and altered short-interval intracortical inhibition ^[45]. Interestingly, Jensen et al. found that while 4 weeks of visuo-motor skill training lead to increased recruitment curve slopes, 4 weeks of strength training lead to decreased recruitment curve slopes implying that increased cortical excitability is induced by highly skilled training and not strength training alone. Similar results were found by Perez et al. and Beck et al. in investigating lower extremity training ^[45, 58]. This is also in line with prior animal studies in which cortical representational changes occurred in squirrel monkeys following motor skill acquisition but was not seen following repetitive movements alone ^[59]. Therefore TMS derived measures are markers for training regimes that are more efficient at driving cortical plasticity and might prove useful in assessing NASA prescribed exercise routines.

While muscular fitness is crucial for astronaut performance, there is also evidence that optimization of motor output may in part be dependent on central mechanisms. While muscle fatigue is a major limitation in the performance of endurance sports, several studies have demonstrated that motor cortex fatigue, or central fatigue, also plays a critical role in performance limitation ^[60, 61]. In a group of marathon runners, response to peripheral stimulation of the peroneal nerve was not reduced following a marathon run, while maximal voluntary contraction and the amplitude of MEP to TMS were both reduced ^[62]. Similar results of MEP depression were found in health subjects following an exercise protocol on a rowing ergometer, but not in elite rowers completing the same protocol likely reflecting less central fatigue within corticospinal pathways of back muscles of elite rowers ^[63]. TMS derived measures are also useful markers for detecting both peripheral and supraspinal limitations to performance and TMS may even be useful in overcoming these limitations. As described above several protocols using TMS with or without peripheral applied stimulation are under investigation as adjunctive rehabilitative treatment options ^[64]. A recent review article even hinted at the possible future use of this technology by Olympic athletes ^[65]. Given the portability of TMS and its ease of usage (which is less physically exhausting than substantial daily exercise regimes), TMS may prove to be a useful adjunct to current countermeasures enhancing astronaut performance on long-duration missions.

Limitations of the current study include a very small sample size and the lack of complete immobilization of the lower extremities. In fact, one subject reported daily abdominal exercises although he remained in a head down tilt position throughout the study. While three of the subjects reported clumsiness in the first days following return to ambulation, this subject reported no such difficulties, which was also documented on functional mobility testing. Interestingly, this subject also demonstrated the largest increase in cortical excitability as assessed by TMS (steepest recruitment curve slope out of the four subjects). In our study, the contribution to plasticity following immobilization of other levels along the corticospinal system was not assessed. However, none of previous immobilization studies, which also investigated other levels of the nervous system, found any significant relationship between changes of TMS measures related to immobilization and measures of spinal excitability and/or neuromuscular transmission ^[30-32]. Liepert and colleagues (1995) did not find any correlation between neuroplastic changes as measured by TMS and spinal excitability as measured by F waves. Results from upper limb immobilization studies performed on patients ^[31, 32] and from studies on

motor learning^[42, 45] indicate that increased MEP recruitment curves following limb disuse or a phase of motor learning are mainly associated with imbalance between intracortical inhibition and facilitation resulting in motor cortical hyperexcitation (as shown by the paired pulse technique). These data would strongly suggest that the observed effects are likely to mainly rely on cortical mechanisms, even though contribution from other structures along the corticospinal pathway can not be excluded.

VI. CONCLUSIONS

In four subjects, we have identified increased motor cortex excitability following 90 days of head down tilt bedrest. Additionally, we have demonstrated a correlation between increased cortical excitability and performance on a functional mobility task during the two week recovery period. Results of this research include a better understanding of the basic neurophysiology underlying cortical plasticity occurring during lower extremity disuse and suggest a possible intervention for remediating astronaut performance prior to landing on a planetary surface. On Earth, these data may lead to applications in modulating cortical maps furthering the rehabilitation of patients suffering from brain injury or prolonged confinement to bed. ***Furthermore, if given the opportunity to complete the final specific aim of our original proposal (to alter bedrest induced cortical plasticity in a useful way), our results might lead to a specific deliverable - a rehabilitative strategy suitable for on-board spacecraft usage assuring maximal astronaut performance on long-term missions such as to the Moon and Mars.***

VII. SUPPLEMENTAL STUDY – BRAIN VOLUMETRICS FOLLOWING BEDREST

While acquiring the fMRI data reported above, during the same scanning session a standard anatomical MRI scan of the brain was also acquired on each bedrest subject. These scans were performed on the four subjects from the first bedrest campaign (campaign 3a) before bedrest, at day 60, and following bedrest at day 90. Also these anatomical brain scans were acquired on four subjects who participated in the second bedrest campaign (campaign 3b) before bedrest and at day 60 of bedrest. We do not have post bedrest day 90 data because campaign 3b was terminated early at day 60. The anatomical brain scans on these subjects were acquired just before they were evacuated from the facility in the wake of Hurricane Rita in the fall of 2005. Therefore, using the day 60 data of the subjects of campaign 3a, we have complete data sets of anatomical brain scans on 8 subjects before and following 60 days of bedrest. We used this data to perform a supplemental analysis of the effects of long-term bedrest on brain volumetrics^[66].

Objective: The US Public Health Service (1972) has reported that approximately 0.2% of the US population is confined to bed due to chronic illness. Extensive research has evaluated cardiovascular, musculoskeletal, metabolic, and endocrine changes associated with long term bedrest. Here we report structural changes that occurred in the brain of normal volunteers during long-term bedrest.

Material and Methods: 8 normal subjects underwent 60 days of strict bedrest as part of an ongoing NASA study. The subjects were allowed to elevate their head on one arm for a total of 30 minutes per day at meal times. Otherwise strict head down bedrest was maintained. The subjects were positioned in 6° headdown tilt but were free to move about the bed. The subjects were maintained on a strict diet throughout the study. Caloric intake was designed to assure that the subjects' weights remained stable. Additionally fluid intake and output were maintained. T1

weighted 3D SPGR MRI were acquired approximately 3 days before undergoing bedrest. All 8 subjects were then rescanned on day 60 of bedrest. The subjects remained in the 6° head down tilt position until being placed supine on the MRI table. We applied deformation based morphometry^[67] to the MR image pairs to map changes in location and size of tissue over time. We then examined the common patterns of local volume change over time^[68] across the participants using a voxel-wise averaging across subjects.

Results: All subjects underwent 60 days of bedrest without any adverse events. Figure 10 shows an example map of displacements (scaled up for illustration) estimated in a single subject illustrating shift of the brain tissue upwards and posteriorly. Frontal lobe tissue appeared to be expanded while sulci along the vertex become more crowded. Figure 11 shows an illustration of a map of local volume changes in a single subject.

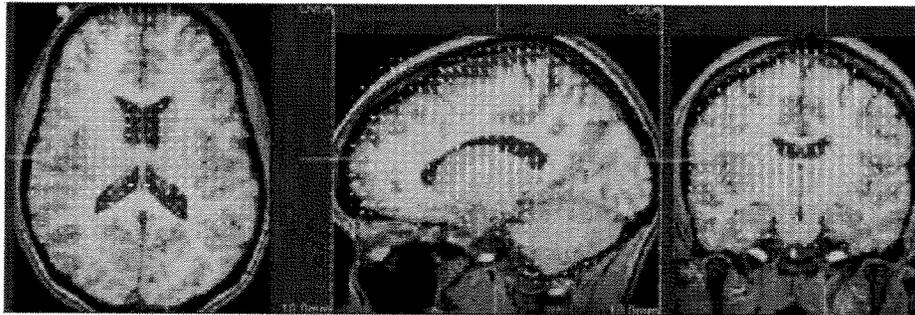


Figure 10. A map of estimated local displacements between scans for a single subject (shown with enlarged scale for visualization) displayed on the first time point scan.

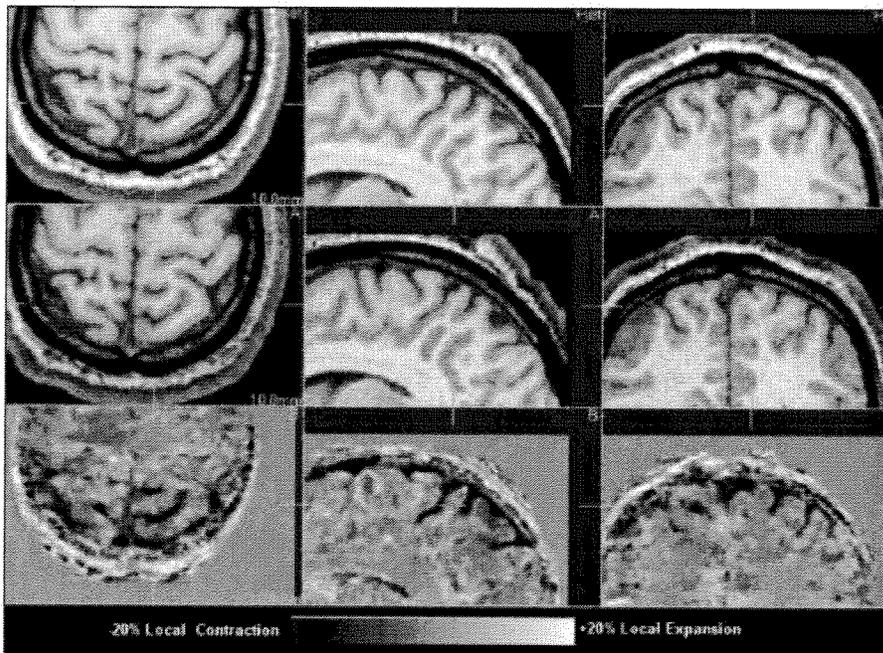


Figure 11. Enlarged views of 1st and 2nd scans of a single subject (Axial, Sagittal, Coronal) together with the estimated local volume change between the two shown as a grey scale of % change. Note focal contraction of sulcal CSF regions.

Although variability existed between subjects, consistent structural changes were shown to occur in a number of regions. Figure 12 shows the T statistic map of the difference between the observed atrophy pattern and zero change for all the 8 subjects, after mapping to a common template MRI and smoothing as in [68]. There is a consistent contraction (blue/cyan) of the CSF spaces along the vertex with expansion (red/yellow) of brain tissue and surrounding sulcal CSF in the anterior frontal lobes.

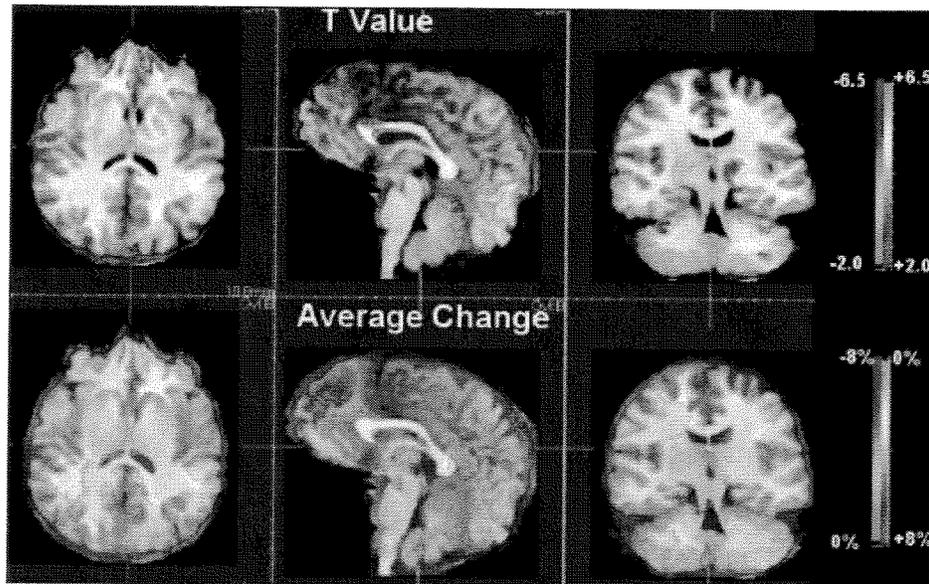


Figure 12. Statistics of the 8 subjects volume change maps transformed to a common template MRI Axial, Sagittal and Coronal views displayed on the Group Average MRI.

Conclusion: Morphological studies have demonstrated regional changes in brain volume associated with diseases such as Alzheimer's disease, mental illness, epilepsy, and with normal aging. Transient shifts can be seen in processes such as intracranial hypotension and downward shifts of the brain are seen in Chiari malformation. Here we report structural changes induced by long-term bedrest. Follow-up studies would include a larger subject population along with a control group.

VIII. REFERENCES

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