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Protocol study for a randomized, controlled, double-blind, clinical trial involving virtual reality and anodal transcranial direct current stimulation for the improvement of upper limb motor function in children with Down syndrome

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Authors' contributions

All authors made substantial contributions.

Conceived and designed the experiments: JL LG RC HL ND IM GM AD MG CO. Acquisition of data: JL LG RC HL ND IM GM AD MG CO. Interpretation of data: JL LG RC HL ND IM GM AD MG CO. Contributed analysis tools: JL LG RC HL ND IM GM AD MG CO Wrote the paper: JL LG RC HL ND IM GM AD MG CO. Final approval of the version: JL LG RC HL ND IM GM AD MG CO

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The study received approval from the ethics committee of University Nove de Julho (Sao Paulo, Brazil) under protocol number 1.540.113 and is registered with the Brazilian Registry of Clinical Trials (RBR3PHPXB)

The authors declare that there are no additional unpublished data from the study.

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Abstract

Introduction: Down syndrome results in neuromotor impairment that affects selective motor control, compromising the acquisition of motor skills and functional independence,. The aim of the proposed study is to evaluate and compare the effects of anodal transcranial direct current stimulation and sham stimulation over the primary motor cortex during upper limb motor training involving virtual reality on motor control, muscle activity, cerebral activity, and functional independence.**Methods and Analysis:**A randomized, controlled, double-blind, clinical trial. The calculation of the sample size will be defined based on the results of a pilot study involving the same methods. The participants will be randomly allocated to two groups. Evaluations will be conducted prior, after and one month after the end of the intervention process. At each evaluation, three-dimensional analysis of upper limb movement will be performed with the SMART-D 140® system (BTS Milan, Italy) following the *SMARTup* protocol, will be measured using electromyography (FREEEMG®BTS), cerebral activity will be measured using an electroencephalogram system (BrainNet), and intellectual capacity will be assessed using the Wechsler Intelligence Scale for Children . Virtual reality training will be held three times per week for a total of ten 20-minute sessions. Transcranial stimulation will be administered simultaneously to the training. The results will be analyzed statistically, with a p-value ≤ 0.5 considered indicative of statistical significance. **Ethical aspects and publicity**:The present study received approval from the Institutional Review Board of *Universidade Nove de Julho* (Sao Paulo,Brazil) under process number 1.540.113 and is registered with the Brazilian Registry of Clinical Trials (N° RBR3PHPXB).The participating institutions have presented a declaration of participation. The volunteers will be permitted to drop out of the study at any time with no negative repercussions. The results will be published and will contribute evidence regarding the use of this type of intervention on children with Down syndrome.

Keywords: Down syndrome; transcranial direct current stimulation; upper limb.

Strengths and limitations of this study

1. The proposed project involves the combination of virtual reality activities for upper limb motor training and anodal transcranial direct current stimulation over the primary motor cortex with the aim of optimizing motor control and function of the upper limbs in children with Down syndrome (DS).
2. Adequate upper limb motor control enables individuals to perform daily, functional, and academic activities in an independent fashion.
3. The use of virtual reality(VR) activities to improve motor control is a promising therapeutic resource that has demonstrated satisfactory results in the scientific literature, including for individuals with DS.
4. Non-invasive brain stimulation techniques, specifically anodal transcranial direct current stimulation, are currently considered effective means by which to facilitate motor cortical excitability of brain regions underlying the stimulation electrode, leading to improvements in motor control and motor learning. Despite the lack of reports on the effects of transcranial stimulation in children with DS, studies involving pediatric patients have demonstrated that the technique is safe, with little or no adverse effects.
5. We believe that the administration of anodal transcranial direct current stimulation over the primary motor cortex, specifically the areas that correspond to upper limb motor control (C3 and C4 of the 10-20 electroencephalogram system), during upper limb motor training with the use of virtual reality activities will enhance the cortical excitability of motor regions and optimize cerebral activity, thereby potentiating the effects of upper limb motor therapy.

1. INTRODUCTION

Down syndrome (DS) is a highly prevalent genetic disease caused by the inheritance of an additional chromosome 21 and is one of the most frequent causes of mental impairment, affecting approximately 20% of the total number of individuals with mental disability.[1] The incidence in the United States is one out of every 700 births and it is estimated that at least 100 thousand individuals in Brazil are diagnosed with the syndrome.[2-4]

The nervous system of children with DS exhibits structural and functional abnormalities. Diffuse brain damage and peculiar electrical functioning during cognitive development result in poor analysis, synthesis, and speech skills. Moreover, such children demonstrate difficulties in selecting and directing a stimulus due to the fatigue of the connections. These abnormalities result in neurological disorders that vary in terms of manifestation and intensity.[5]

According to Flórez and Troncoso (1997),[6-7] the brain of an individual with DS is smaller in volume in comparison to an individual without this condition. Hypoplasia of the frontal and occipital lobes is a common finding. A unilateral or bilateral reduction in the temporal lobe is found in up to 50% of cases and reductions in the corpus callosum, anterior commissure, and hippocampus are found. According to Bomono & Rosetti (2010), the neuromotor abnormalities in DS include hypotonia, diminished primitive reflexes, delayed motor and cognitive development, and lower levels of learning.[8]

Studies have been conducted to understand why individuals with DS have slow, unharmonious movements.[10-22] The investigation of electromyographic activity and muscle torque demonstrates this deficiency, which can be corrected by the repetition of a given movement during motor training activities. Motor control strategies used in the execution of complex activities, such as a reaching task, have been investigated in this population.[13,17-22].

The optimal results achieved with virtual reality are believed to be related to training in an interactive environment that provides a broad range of activities and scenarios with multiple sensory channels, allowing the creation of exercises at an intensity that is promising for the needs of individuals with DS.[23-26] Virtual reality(VR) can be used as an auxiliary tool involving a playful, motivational objective that can facilitate the development of perception and motor skills, with the training of planning skills and motor control as well as the stimulation of the plasticity of the central nervous system.

Non-invasive brain stimulation methods have been employed in physical rehabilitation protocols due to the promising results achieved with regard to motor learning. Transcranial

direct current stimulation (TDCS) is a relatively low-cost, noninvasive brain stimulation technique that is easy to administer and offers minimal adverse effects. This method is known to produce lasting changes in motor cortical excitability [27]. Cortical modulation depends on the polarity of the current: anodal stimulation increases cortical excitability, favoring the depolarization of the neuronal membrane, whereas cathodal stimulation has an inhibitory effect due to the hyperpolarization of the neuronal membrane.[28,33]

TDCS has advantages over other transcranial stimulation techniques, such as providing a longer lasting modulatory effect on cortical function as well as its ease of use and lower cost. The results of clinical trials have demonstrated its considerable potential in the treatment of neurological disorders and the investigation of processes of cortical excitability modulation. Moreover, this type of intervention offers a better condition for sham stimulation, which confers greater specificity to the findings.[34] In the rehabilitation process, the aim of neuromodulating techniques is to enhance local synaptic efficiency and alter the maladaptive plasticity pattern that emerges after a cortical injury.[35-39].

Although DS is one of the most prevalent diseases in the pediatric population, no studies were found on the effects of TDCS in children with this syndrome. Thus, the lack of investigations on anodal TDCS over the primary motor cortex during motor training for children with DS constitutes a gap in the scientific literature.[40] Considering the high prevalence of DS, the motor limitations stemming from this disease, which exert a negative impact on functionality and independence, and the fact that TDCS is not contraindicated in most cases of this syndrome, the investigation of the effects of this noninvasive brain stimulation technique on children with DS is relevant.[38-39]

The proposed study could be used as the basis for the development of further projects conducted to broaden knowledge on this technique, enabling a novel intervention option for the optimization of motor control in individuals with DS.

2. OBJECTIVES

2.1 Primary objective

The aim of the proposed study is to evaluate and compare the effect of TDCS and sham stimulation over the primary motor cortex during upper limb motor training involving virtual reality on motor control (spatiotemporal variables and kinematics of a reaching task), activity of the elbow flexors and extensors, cerebral activity and functional independence in children with DS.

2.2 Secondary objectives

- Determine possible correlations between upper limb motor control (movement velocity and total duration of movement) and muscle activity (elbow flexors and extensors), cerebral activity (activity of the parietal lobe, specifically regions C3 and C4) and functional independence with regard to self-care.

- Identify possible prediction factors for the response of upper limb motor control (movement velocity and total duration of movement) in children with Down syndrome. The factors will be investigated: muscle activity of elbow flexors and extensors, cerebral activity (areas C3 and C4 of the 10-20 electroencephalogram system) and transcranial direct current stimulation (active and sham).

3. METHODS AND ANALYSIS

The following will be the inclusion criteria: 1) a diagnosis of DS; 2) adequate comprehension and cooperation during the procedures; 3) age six to 12 years; (4) compromised upper limb motor coordination; and 5) statement of informed consent signed by a legal guardian. The exclusion criteria will be 1) having undergone surgical procedures in the 12 months prior to the onset of the training sessions, 2) orthopedic deformity of the lower limbs or spinal column with an indication for surgery, 3) epilepsy, 4) metal implant in skull or hearing aids, 5) associated neurological disorder, and 6) use of a pacemaker.

3.1 Study Design

A Phase I-II study will be conducted: analytical, paired, randomized, controlled, double-blind, clinical trial

Figure 1: Flowchart of study following CONSORT statement

3.2 Sample size

The sample size will be calculated based on the results of a pilot study with the same methods as those of the main study. The pilot study will involve ten children randomly allocated to the experimental and control groups (five children in each group). The sample size will be calculated based on the mean of both groups considering total duration of movement as the primary outcome, with a unidirectional alpha of 0.05 and an 80% power. The sample will be increased by 20% to compensate for possible dropouts.

3.3 Randomization

Patients with DS who meet the eligibility criteria and agree to participate in the study will be submitted to an initial evaluation and will then be randomly allocated to two groups using a randomization method available on the site www.randomizacion.com. This process will be performed by a member of the research team who is not involved in the recruitment or development of the study. Experimental group: anodal TDCS over the primary motor cortex bilaterally combined with upper limb motor training involving the use of VR; Control group: sham TDCS over the primary motor cortex bilaterally combined with upper limb motor training involving the use of VR.

3.4 Evaluations

The participants will be submitted to three evaluations: Pre-intervention, post-evaluation (after ten training sessions), and follow up (one month after last training session).

3.4.1 Three-dimensional movement analysis:

Three-dimensional analysis of upper limb movement: the kinematics of upper limb movement will be evaluated using the SMART-D 140® system (BTS, Milan, Italy), with eight cameras sensitive to infrared light, a sampling frequency of 100 Hz and video system synchronized with the SMART-D system. Passive markers will be positioned at anatomic references points directly on the skin with specific adhesive tape, following the protocol of the *SMARTup: The experimental setup*.[40-42] A total of 18 markers measuring 15 mm in diameter will be used to identify the position of the head, trunk and upper limbs (upper arm, forearm and hand).

The movement will be divided into three phases: going phase (upper limb moving toward the target), adjusting phase (adjustment of arm to locate target precisely) and returning phase (return to initial position). At least six complete movements will be performed to obtain three adequate cycles for analysis. The biomechanical model, filtering

of the data, and processing of the variables will be performed using the *SMART analyser* software program (BTS, Milan, Italy). The variables will be identified and calculated for each movement cycle to evaluate any changes that occur after the intervention. The following variables will be considered, with the mean of the results used in the statistical analyses:

- Total duration of movement: total time required to perform the complete reaching task.
- Mean movement velocity: computed during the going phase and determined using the marker positioned on the index finger.
- Adjusting sway index: Defined as the length of the three-dimensional path described by the marker on the index finger during the adjusting phase.
- Range of motion of elbow and shoulder: calculated as the difference between the maximum and minimum angles of the elbow and shoulder on the sagittal (elbow and shoulder) and frontal (shoulder) planes during the going phase, as described in the literature. [41,42]

Figure 2: Placement of markers for three-dimensional analysis using *SMARTup: The experimental setup* [40]

Figure 3: Phases of reaching cycle [40]

3.4.2 Electromyographic (EMG) analysis: Muscle activity during the reaching movement will be determined using EMG. The electrical activity resulting from the activation of the elbow flexors and extensors will be collected using an eight-channel electromyograph (FREEEMG®, BTS Engineering) with a bioelectrical signal amplifier, wireless data transmission and bipolar electrodes with a total gain of 2000 fold and frequency ranging from 20 to 450 Hz. Impedance and the common rejection mode ratio of the equipment are > 10^{15} Ω//0.2 pF and 60/10Hz 92 dB, respectively. The motor point of the muscles will be identified for the placement of the electrodes and the skin will be cleaned with 70% alcohol to reduce bioimpedance, following the recommendations of Surface Electromyography for the Non-Invasive Assessment of Muscles.[43] All EMG data will be digitized at 1000 frames per second using the BTS MYOLAB® software program. The data will be collected simultaneously to the kinematic data and both will be managed using the BTS® system and *Smart Capture*® software program.[44]

3.4.3 Electroencephalographic analysis: Cerebral activity will be investigated using electroencephalography (EEG), which will be performed during both the three-dimensional analysis of the reaching task and the evaluation of muscle activation using EMG. For such, the volunteer will be seated in an erect position on a chair in front of the table on which the reaching task will be performed. The BrainNet BNT36 device with 36 configurable channels (32 AC and four DC) and a 16-bit analog-digital converter will be used for the acquisition of the EEG signal. The analysis of the signal will be performed with the aid of the EEGLab tool implemented on Matlab, which is also capable of furnishing a topographic map of cerebral activity as a function of time. The electrodes will be positioned following the guidelines of the 10/20 electroencephalogram system.[45,46]

Figure 4 – Phase relationships. (A) synchronized signals – differences in phases between both signals are stable (constant); (B) non-synchronized signals – a differences in phases are variable [47]

Figure 5 – Positioning of EEG electrodes following 10-20 standard [45]

3.4.5 Pediatric Evaluation of Disability Inventory (PEDI): The children's functional performance will be assessed quantitatively using the PEDI, which is a questionnaire administered in interview format to a caregiver who can provide information regarding the child's performance on typical activities and routine tasks. The PEDI is composed of three parts, the first of which is used to evaluate skills grouped into three functional domains: self-care (73 items), mobility (59 items) and social function (65 items). Each item is scored either zero (not part of the child's repertoire) or 1 (part of the child's repertoire). The scores are then summed per domain.[48,49]

3.4.6 Wechsler Intelligence Scale for Children: The Wechsler Intelligence Scale (WIS) was developed for the assessment of the intellectual performance of adults. The WISC was developed as a version for children, which was followed by the revised version, WISC-R. The WISC III is the third version of the scale for children and is used to assess intellectual capacity using 13 subtests, 12 of which were from earlier versions and one was new. The subtests are organized into two groups (verbal and perceptive-motor or execution)and are administrated in alternating order. The verbal subtests are Information, Similarities, Arithmetic, Vocabulary, Comprehension and Digits. The execution group is composed of Matrix Reasoning, Coding, Figure Weights, Block Design, Picture Concepts, Symbol Search and Mazes. Many studies have been conducted and, although improvements have

been made with the addition of new items, the fundamental characteristics of the WISC and WISC-R remained the same in WISC III.[50]

4. Procedures

4.1 Transcranial direct current stimulation

Stimulation will be administered using a TDCS device (*DC-Stimulator NeuroCo nn*, Germany), with three sponge (non-metallic) surface electrodes measuring 5 x 7 cm soaked in saline solution. The children will be randomly allocated to two types of treatment: 1) active anodal stimulation over the primary cortex bilaterally; and 2) sham transcranial stimulation. The two anodal electrodes will be positioned over C3 and C4 of the 10-20 international electroencephalogram system [37] and the cathode will be positioned over the right deltoid muscle. This montage will enable the child to receive anodal stimulation of the primary motor cortex, specifically the area that manages upper limb motor control, and minimize the effect of cathodal stimulation in the brain. Sham stimulation will consist of the same electrode montage and the stimulator will be switched on for 30 seconds, giving the child the initial sensation of stimulation, but no current will be administered during the remainder of the session. This is considered a valid control procedure in studies involving TDCS. A current of 1mA will be administered over the primary motor cortex for 20 minutes during the upper limb motor training activity. The stimulator has a knob that allows the operator to control the intensity of the current. At the beginning of the session, stimulation will be increased gradually until reaching 1mA and gradually diminished during the final ten seconds of the session.

Adverse effects: Potential adverse effects of TDCS will be evaluated at the end of each session using a questionnaire administered to the child. The questionnaire will address the perception of symptoms having occurred during the session, such as tingling, a burning sensation, headache, pain at the electrode sites, sleepiness and altered mood. The children will be instructed to answer using a three-point scale. The caregivers and children will also be asked open-ended questions at the beginning of each session regarding the occurrence of headache, scalp pain, burning sensation, redness of the skin, sleepiness, difficulty concentrating and mood swings during periods between sessions.

4.2 Virtual reality training protocol

Training sessions will be held three times per week on non-consecutive days. Each session will last 20 minutes and will involve the use of the XBOX 360™ with the Kinect™ motion detector. The game entitled "Bursting Bubbles" of the Adventure set of games was chosen based on the potential to stimulate cognitive skills and enhance execution time, motor coordination, attention, concentration, reasoning, memory, persistence and precise movement,. The activity will be held in a specific room of the Integrated Movement Analysis Laboratory measuring 2.5 x 4.0 m, with a projection screen (200 x 150 cm) attached to the wall and stereo speakers to provide adequate visual and auditory stimuli. Initially, the child will be instructed to remain standing at a distance of two to three meters in front of the motion detector to capture the movements better as well as for the estimation of height and calculation of the body mass index. Two mobility training sessions with the use of the XBOX 360 exercises will be performed prior to the onset of the intervention protocol. Records will be made of the number of sessions attended and duration of each session.

5. Analysis of results

The Shapiro-Wilk test will be used to determine whether the data adhere to the Gaussian curve. Parametric variables will be expressed as mean and standard deviation. Nonparametric variables will be expressed as median and interquartile range. Effect sizes will be calculated from the differences in means between the pre-intervention and post-intervention evaluations. The effect size values will be expressed with respective 95% confidence intervals. Either two-way ANOVA (parametric variables) or the Kruskal-Wallis test (non-parametric variables) will be used for the analysis of the effects of the upper limb motor training activity with active and sham TDCS. Logistic regression models will be created to determine factors predictive of the response to the intervention. For such, movement velocity and total duration of movement will be considered. The response capacity will be defined as a clinically significant increase in performance in comparison to baseline. The independent variables will be age (years), sex (male/female), activity of elbow flexors and extensors, cerebral activity (C3 and C4) and functional independence (aspects of self-care). Univariate regressions will be performed for each variable. Based on the initial analyses, the predictors associated with the outcome with a p-value ≤ 0.05 will be incorporated into the multivariate model. Moreover, Pearson's correlation coefficients will be calculated to determine correlations among the variables analyzed. A p-value < 0.05 will be considered indicative of statistical significance. The data will be

organized and tabulated with the aid of the Statistical Package for the Social Sciences (SPSS v.19.0).

6. Discussion

As upper limb motor control enables individuals to perform functional activities, virtual reality will be used as a therapeutic tool to enhance motor control. Moreover, a noninvasive brain stimulation method TDCS will be employed to facilitate motor cortical excitability in the areas subjacent to stimulation to enhance the effects of motor control and learning. This document offers a detailed description of a randomized, controlled, double-blind, clinical trial designed to determine the effectiveness of VR training combined with TDCS on upper limb movements in individuals with DS.

7. Ethical aspects and divulgation

The present study is in compliance with the guidelines regulating studies involving human subjects established by the Brazilian National Board of Health in October 1996 and updated in Resolution 466 in 2012. The study will be developed at the Integrated Movement Analysis Laboratory of University Nove de Julho (Sao Paulo, Brazil) and has received approval from the Human Research Ethics Committee of the university under process number 1.517.470 (APPENDIX 1). The protocol has been registered with Clinical Trials. All legal guardians will receive clarifications regarding the procedures and will be aware that participation is voluntary, free of cost and experimental. Those who agree to their child's participation will sign a statement of informed consent (APPENDIX 2). The guardians will be assured of access to all information and will be informed of the possibility of dropping out of the study or withdrawing consent at any time with no negative consequences. The anonymity of the children and the confidentiality of their information will be ensured, following the ethical principles of privacy. The findings will be published and will contribute evidence regarding the use of transcranial direct current stimulation combined with upper limb motor training in this population.

8. Conflict of Interest Statement

The authors have no financial or competing interests

9. Acknowledgments and funding

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10. Abbreviations

DS: Down Syndrome; TDCS: Transcranial direct-current stimulation; EMG: Electromyography; PEDI: Pediatric Evaluation of Disability Inventory; WISC III: Wechsler Intelligence Scale for Children; VR:Virtual Reality

11. List of Figures

REFERENCES

1. Moreira LMA, El-Hani CN, Gusmão FAF. **A syndrome de down e sua patogênese: considerações sobre o determinismo genético**. Rev Bras Psiquiatr. 2000; 22(2): 96-9.
2. Lewada AF, Matsonff A, Revenis M, Futtermam C, Nino G, Greenberg J, et al. **Preoperative evaluation and comprehensive risk assessment for children with Down syndrome. Pediatric Anesthesia**. 2016; 26: 356–362.
3. Moreira LMA, Gusmão FAF. **Aspectos genéticos e sociais da sexualidade em pessoas com síndrome de Down**. Rev Bras Psiquiatria. 2002; 24(2): 94-9.
4. Silva MFMC, Kleinahas ACS. **Cognitive processes and brain plasticity in Down Syndrome**. Revis. Bras. Edc. Esp. 2006; 12: 123-138.
5. Luria, AR, Tskvetkova, LS. **The programing of constructive activety in local brain injuries**. Neuropsychologia .1964; 95-107.
6. Flórez BJ, Troncoso VM. **Síndrome de Down y educacíon**. 3. reimp. Barcelona: Masson – Salvat Medicina y Santander, 1997.
7. Santos APM, Weiss LI, Almeida GMF**. Assessment and intervention in the motor development of a child with Down syndrome**. Rev Bras Ed Esp Marília. 2010; 16: 19-30.
8. Bomono LMM, Rosseti CB. **Aspects in perceptual-motor development and sensory-motor intelligence in Down syndrome**. Rev bras crescimento desenvolv hum. 2010; 3: 723-734.
9. Polastri PV, Barela JA **Percepção-ação no desenvolvimento motor de crianças portadoras de Síndrome de Down**. Revista da Sobama. 2002; 7: 1-8.
10. Dessen MA, Silva NLP. **Deficiência mental e família: uma análise da produção científica**. Cadernos de Psicologia e Educação Paidéia. 2000; 10: 12-23.
11. Latash ML, Corcos DM. **Kinematic and electromyographic characteristics of single-joint movements of individuals with Down syndrome**. Am J Ment Retard. 1991; 96: 189-201.
12. Almeida GL, Corcos DM, Latash ML. **Practice and transfer effects during fast single-joint elbow movements in individuals with Down syndrome**. Phys Ther. 1994; 74: 1000-1016.
13. Almeida GL, Hasan Z, Corcos DM. **Horizontal-plane arm movements with direction reversals performed by normal individuals and individuals with Down syndrome**. J Neurophysiol. 2000; 84: 1949-1960.
14. Aruin AS, Almeida GL, Latash ML. **Organization of a simple two-joint synergy in individuals with Down syndrome**. Am J Ment Retard. 1996; 101: 256-268.
15. Aruin, AS, Almeida GL. **A co-activation strategy in antecipatory postural adjustments in persons with Down syndrome**. Motor Control 1997; 01: 178-191.
16. Latash ML, Anson JG. **What are "normal movements" in atypical populations**. Behav Brain Sci. 1996; 19: 55-68.
17. Marconi NF. **Controle motor de movimentos de reversão em indivíduos neurologicamente normais e portadores da síndrome de Down: O efeito do feedback intrínseco** [dissertação]. Campinas (SP): Universidade Estadual de Campinas; 2000.

18. Ferreira SMS. **Modulação da Latência da Musculatura Antagonista em Indivíduos "neurologicamente normais" e portadores da Síndrome de Down** [dissertação]. Rio Claro (SP): Universidade Estadual Paulista; 2000.

19. Marconi NF. **Controle motor de movimentos de reversão do braço em indivíduos neurologicamente normais e portadores da síndrome de Down: O efeito do treinamento** [tese]. Campinas (SP): Universidade Estadual de Campinas; 2005.

20. Anson JG. **Neuromotor control and Down syndrome. In: JJ Summers, editores. Approaches to the study of motor control and learning.** Amsterdam; 1992: 387-41.

21. Lawrence GP, Reilly NE, Mottram TM, Khan MA, Eliott. **Sequential aiming movements and the one-target advantage in individuals with Down** Syndrome. Res in Dev Disabil 2013; 34: 3858–3866.

22. Vimercatti SL, Galli M, Stella G, Ancilão A, Albertini G. **Motor strategies and motor programs during an arm tapping task in adults with Down**. Ex Brain Res. 2013; 225: 333-8.

23. Lorenzo SM, Braccialli LMP, Araújo RCT. **Realidade virtual como intervenção na Síndrome de down: uma perpectiva de ação na interface educação e saúde**. Rev Bras Educ Espc. 2015; 21: 392-396.

24. Wuang YP, Chiang CS, Su CY, Wang CC. **Effectiveness of virtual reality using Wii gaming technology in children with Down syndrome**. Res Dev Disabil. 2011; 32: 312–321.

25. Lin HC, WuangYP. Strength and agility training in adolescents with Down syndrome: A randomized controlled trial. Res Dev Disabil. 2012; 33: 2236–2244.

26. Mello BCC, Ramalho TF. **Use of virtual reality in the physical therapeutic treatment of individuals with Down syndrome**. Rev Neurocienc. 2015; 23: 143-149.

27. Stagg CJ, Bachtiar V, O'Shea J, Allman C, Bosnell RA, Kischka U, Matthews PM, Johansen-Berg H. **Cortical activation changes underlying stimulation induced behavioral gains in chronic stroke**. Brain. 2012; 135:276-84.

28. Miranda PC, Lomarev M, Hallett M. **Modeling the current distribution during transcranial direct current stimulation**. Clin Neurophysiol. 2006; 117(7):1623-9.

29. Wagner T, Fregni F, Fecteau S, Grodzinsky A, Zahn M, Pascual-Leone A. **Transcranial direct current stimulation: A computer-based human model study**. Neuroimage 2007; 35:1113-24.

30. Liebetanz D, Nitsche MA, Teragau F, Paulus W. **Pharmacological approacha to the mechanisms of transcranial DC- stimulation- induced after-effects of humam motor cortex excitability**. Brain 2002; 125: 2238-47.

31. Kuo MF, Unger M, Liebetanz D, Lang N, Tergau F, Paulus W, Nittshe MA. **Limited impact of homeostatic plasticity on motor learning in humans**. Neuropsychologia. 2008; 46: 2122-8.

32. Monte-SilvaK,Kuo M-F, Thirugnanasambandam N,Liebetanz D, Paulus W, Nitsche MA. **Dose-dependente inverted U-shaped effect of dopamine (D2-like) receptor activation on focal and nonfocal plasticity in humans**. The Journal of Neuroscience. 2009; 29 (19):6124-31.

33. Nitsche MA, Liebetanz D, Schlitterlau a, Henschke U, Friche K, Frommann K, et al**. GABAergic modulation of DC stimulation-induced motor cortex excitability shifts in humans**. Eur J Neurosco. 2004;19(10): 2720-6.

34. Mendonça ME, Fregni F.**Neuromodulação com estimulação cerebral não invasiva: aplicação no acidente vascular encefálico, doença de Parkinson e dor crônica**. In.:ASSIS, R.D. Condutas práticas em fisioterapia neurológica. Manole. São Paulo, p. 307-39, 2012.

35. Fregni F, Gimenes R, Valle AC, Ferreira MJ, Rocha RR, Natalle L, Bravo R, Rigonatti SP, Freedman S, Nitsche M, Pascual-Leone A, Boggio PS. **A randomized, sham-controlled, proof of principle study of transcranial direct current stimulation for the treatment of pain in fibromyalgia. Arthritis and Rheumatism**, 2006; 54:3988-98.

36. Fregni F, Bossio PS, Brunoni AR. **Neuromodulação terapêutica: Princípios e avanços da estimulação cerebral não invasiva em neurologia, reabilitação, psiquiatria e neuropsicologia**. Sarvier. São Paulo, 2012.

37. Fritsch B, Reis J, Martinowich K, Schambra HM, Ji Y, Cohen LG, et al. **Direct current stimulation promotes BDNF-dependent synaptic plasticity: potential implications for motor learning**. Neuron. 2010;66(2): 198-204.

38. Reis J, Robertson EM, Krakauer JW, Rothwell J, Marshall L, Gerloff C, et al. **Consensus: Can transcranial direct current stimulation and transcranial magnetic stimulation enhance motor learning and memory formation. Brain stimulation.** 2008;1(4): 363-9.

39. Antal A, Lang N, Boros K, Nitsche M, Siebner HR, Paulus W. **Homeostatic metaplasticity of the motor cortex is altered during headache-free intervals in migraine with aura.** Cerebral cortex. 2008;18(11):2701-5.

40. Cimolin V, Beretta E, Piccinini L, Turconi AC, Galli M, Strazzer S. **Constraint-induced movement therapy for children with hemiplegia after traumatic brain injury: a quantitative study**. J Head Trauma Rehabil. 2012; 27(3): 177-87.

41. Menegoni F, Milano E, Trotti C, Galli M, Bigoni M, Baudo S, Mauro A. **Quantitative evaluation of functional limitation of upper limb movements in subjects affected by ataxia**. Eur J Neurol. 2009; 16(2): 232-9.

42. Petuskey K, Bagley A, Abdala E, James MA, Rab G. **Upper extremity kinematics during functional activities: three-dimensional studies in a normal pediatric population**. Gait Posture. 2007; 25(4): 573-9.

43. Hermes JH, Freriks B, Merletti R, Steggeman D, Blok J, Rau G, Disselhorst-Klug C, Hagg G: **SENIAM 8: Surface Electromyography for the Non-Invasive Assessment of Muscles**. Roessingh Research and Development 1999.

44. Rab G, Petuskey K, Bagley A. **A method for determination of upper extremity kinematics**. Gait Posture. 2002; 15(2): 113-9.

45. Jasper HH. **The ten-twenty electrode system of the International Federation electroencephalogria**. Clin Neurophysiol. 1958; 10: 371-375.

46. Homan RW, Herman J, Purdy P. **Cerebral location of international 10-20 system electrode placement.** Electroencephalogr Clin Neurophysiol. 1987; 66(4): 376-82.

47. FELL, J.; AXMACHER, N. The role of phase synchronization in memory processes. **Nature Reviews Neuroscience**, v. 12, n. 2, p. 105–118, fev. 2011

48. Haley S, Coster W, Ludlow L. **Inventário de avaliação pediátrica de disfunção: versão brasileira.** Tradução e adaptação cultural: Mancini M C. Belo Horizonte: Laboratórios de Atividade e Desenvolvimento Infantil, Departamento de Terapia Ocupacional, Universidade Federal de Minas Gerais. 2000.
49. Feldman AB, Haley SM, Corvell J. **Concurrent and construct validity of the Pediatric Evaluation of Disability Inventory**. Phys. Ther. 1990; 70(10): 602-10.
50. Cruz MBZ, WISC III: **Escala de Inteligência Wechsler para crianças: Manual**. Periodicos eletrônicos em psicologia. 2005; 4: 309.

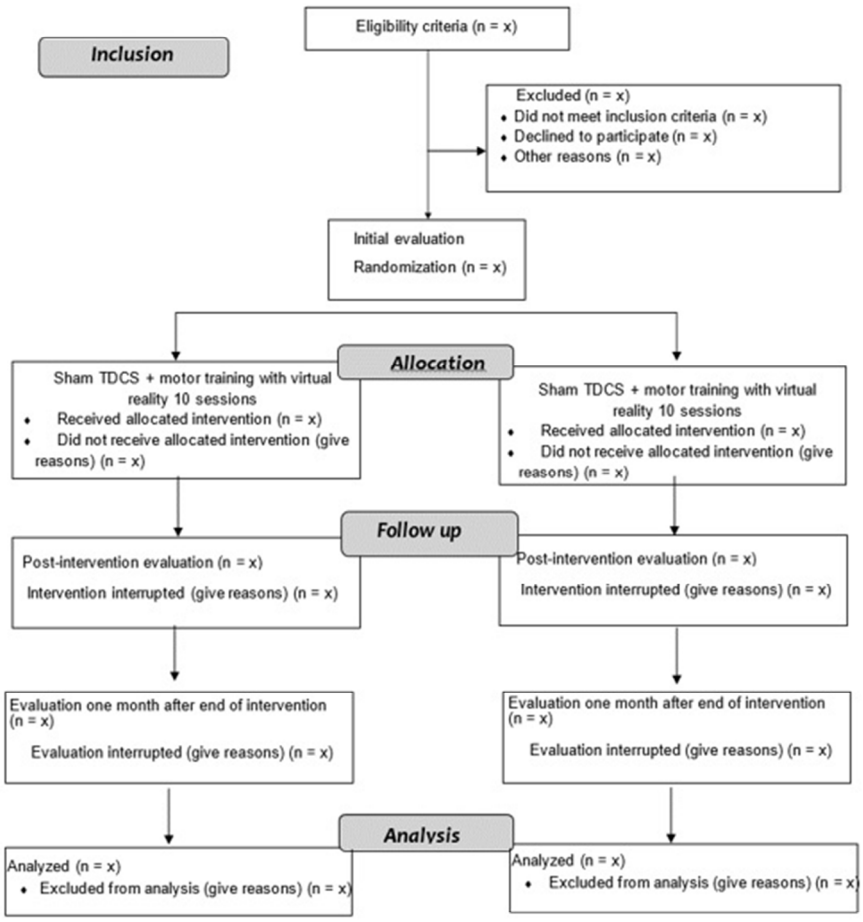

Figure 1: Flowchart of study following CONSORT statement

Legend: TDCS = transcranial direct current stimulation

Figure 1: Flowchart of study following CONSORT statement
Legend: TDCS = transcranial direct current stimulation

163x199mm (96 x 96 DPI)

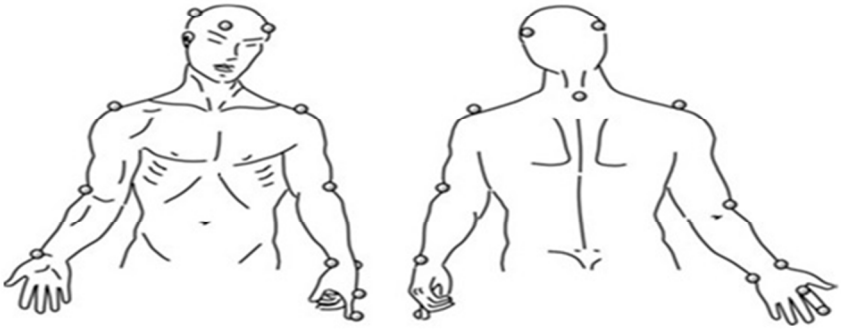

Figure 2: Placement of markers for three-dimensional analysis using *SMARTup: The experimental setup*[40]

Figure 2: Placement of markers for three-dimensional analysis using SMARTup: The experimental setup

178x110mm (96 x 96 DPI)

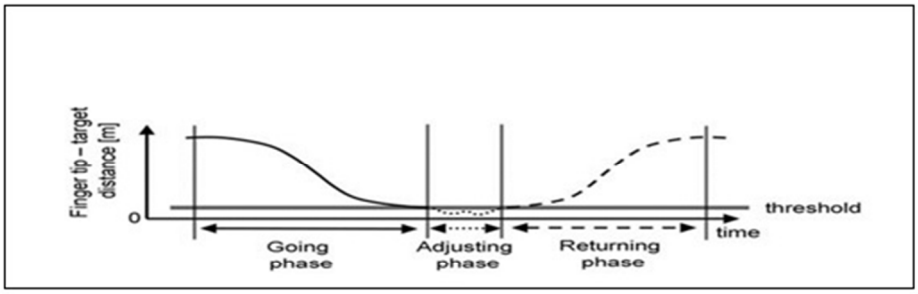

Figure 3: Phases of reaching cycle[40]

Figure 3: Phases of reaching cycle

165x98mm (96 x 96 DPI)

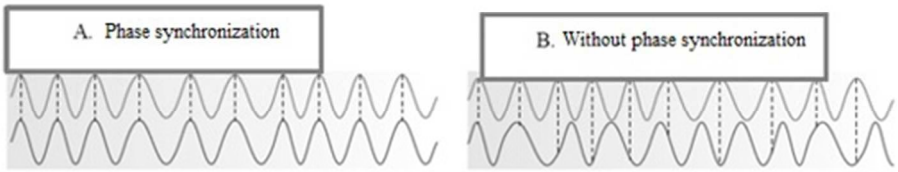

Figure 4 – Phase relationships. (A) synchronized signals – differences in phases between both signals are stable (constant); (B) non-synchronized signals – a differences in phases are variable[47]

Figure 4 – Phase relationships. (A) synchronized signals – differences in phases between both signals are stable (constant); (B) non-synchronized signals – a differences in phases are variable

161x79mm (96 x 96 DPI)

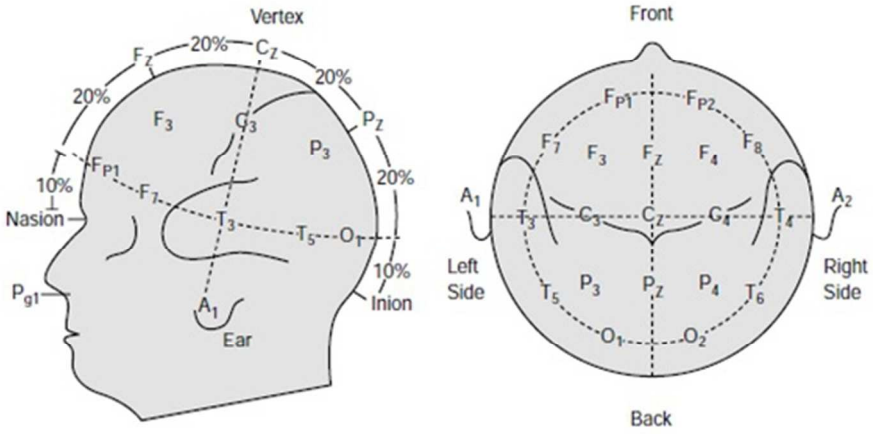

Figure 5 – Positioning of EEG electrodes following 10-20 standard[45]

Figure 5 – Positioning of EEG electrodes following 10-20 standard

169x128mm (96 x 96 DPI)

APPENDIX 1

Termo de Consentimento para Participação em Pesquisa Clínica

Nome do Voluntário: _____

Endereço:_____

Telefone para contato:_____Cidade:_____CEP:_____

E mail: _____

1. As informações contidas neste prontuário foram fornecidas pela aluna Jamile Benite Palma Lopes (Mestranda da Universidade Nove de Julho), Profª. Claudia Santos Oliveira, objetivando firmar acordo escrito mediante o qual, o voluntário da pesquisa autoriza sua participação com pleno conhecimento da natureza dos procedimentos e riscos a que se submeterá, com a capacidade de livre arbítrio e sem qualquer coação.

2. Título do Trabalho Experimental: Realidade virtual e estimulação transcraniana por corrente contínua anódica para melhora da função motora de membros superiores em crianças com síndrome de down: ensaio clínico controlado aleatorizado e duplo cego.

3. Objetivo: Examinar os efeitos da estimulação por corrente sobre o controle motor, atividade dos músculos, atividade do cérebro e independência funcional de crianças com Síndrome de Down.

4. Justificativa: acredita-se que ao aplicar a estimulação por corrente, especificamente, durante o treino motor com uso de um vídeo game, será possível, otimizar a atividade do cérebro e a melhora motora.

5. Procedimentos da Fase Experimental: Será selecionas crianças diagnosticadas com Síndrome de Down, com capacidade de entendimento e colaboração para realização dos procedimentos envolvidos no estudo, crianças com idade entre seis e 12 anos, crianças com queixas de comprometimento Na coordenação motora dos braços. O processo de avaliação (antes, após e um mês após o treino, será realizado em três dias não consecutivos, mas na mesma semana, com período máximo de uma hora e 30 minutos por dia. A avaliação será constituída dos seguintes itens: (1) Analise de movimento dos braços durante uma tarefa: avaliado pela cinemática, eletromiografia e eletroencefalograma, a criança realizara uma tarefa com os braços e ao mesmo tempo será avaliada pelos aparelhos, sendo acompanhada pelo fisioterapeuta responsável e pelos assistentes (2) PEDI o PEDI é um questionário aplicado no formato de entrevista estruturada com um dos cuidadores da criança, que possa informar sobre seu desempenho em atividades e tarefas típicas da rotina diária. O teste é composto de três partes: a primeira avalia habilidades de repertório da criança agrupadas segundo três áreas

funcionais: autocuidado (73 itens), mobilidade (59 itens) e função social (65 itens). Cada item dessa parte é pontuado com escore 0 (zero) se a criança não é capaz de desempenhar a atividade, ou 1 (um), se a atividade fizer parte de seu repertório de habilidades. O Grupo 1 terá o movimento do braço analisado após realizar treino com o vídeo game junto com a estimulação desligada (placebo). O Grupo 2 terá o movimento do braço analisado após realizar treino com o vídeo game junto com a estimulação ligada. A estimulação por corrente é uma técnica não invasiva que será realizada colocando eletrodos de superfície conectados a um aparelho de corrente galvânica (corrente elétrica de baixa intensidade) sobre o crânio (cabeça) da criança, durante 20 minutos por 15 dias. A estimulação é indolor.

6. Desconforto ou Risco Esperado: Embora os procedimentos adotados no estudo sejam não-invasivos os voluntários serão submetidos a risco como por exemplo, quedas, fadiga muscular, câimbras durante o treino motor de realidade virtual. Para que estes riscos sejam minimizados ao máximo serão adotadas as seguintes medidas protetoras: A estimulação será realizada por uma fisioterapeuta com experiência na técnica. No treino de realidade virtual serão realizados por uma fisioterapeuta com experiência em treino motor que será acompanhada por ao menos um voluntário ambos permanecerão posicionados do lado do paciente por todo o treino.

7. Informações: o voluntário tem garantia que receberá respostas a qualquer pergunta ou esclarecimento de qualquer dúvida quanto aos procedimentos, riscos benefícios e outros assuntos relacionados com pesquisa. Também os pesquisadores supracitados assumem o compromisso de proporcionar informação atualizada obtida durante o estudo, ainda que esta possa afetar a vontade do indivíduo em continuar participando.

8. Retirada do Consentimento: o voluntário tem a liberdade de retirar seu consentimento a qualquer momento e deixar de participar do estudo, sem que isto lhe traga qualquer prejuízo.

9. Aspecto Legal: Elaborados de acordo com as diretrizes e normas regulamentadas de pesquisa envolvendo seres humanos atendendo à Resolução nº. 466/12 do Conselho Nacional de Saúde do Ministério de Saúde – Brasília – DF.

10. Garantia de Sigilo: Os pesquisadores asseguram a privacidade dos voluntários quanto aos dados confidenciais envolvidos na pesquisa.

11. Formas de ressarcimento das despesas decorrentes da participação na pesquisa: Se necessário, será dado aos pesquisados auxilio transporte de ida e volta ao local da pesquisa. Não será dada ao pesquisado qualquer tipo de remuneração e auxilio de custo pela participação na pesquisa. Pelo curto tempo das avaliações e intervenções não haverá fornecimento de alimentação ao pesquisado.

12. Local da Pesquisa: A pesquisa será desenvolvida no Laboratório Integrado de Análise do Movimento Humano - LIAMH e Núcleo de Apoio a Pesquisa na Analise do Movimento - NAPAM, Universidade Nove de Julho UNINOVE, localizada na rua Vergueiro, no 235/249, 2º subsolo, Vergueiro, São Paulo - SP.

13. Comitê de Ética em Pesquisa (CEP) é um colegiado interdisciplinar e independente, que deve existir nas instituições que realizam pesquisas envolvendo seres humanos no Brasil, criado para defender os interesses dos participantes de pesquisas em sua integridade e dignidade e para contribuir no desenvolvimento das pesquisas dentro dos padrões éticos (Normas e Diretrizes Regulamentadoras da Pesquisa envolvendo Seres Humanos – Res. CNS nº 466/12). O Comitê de Ética é responsável pela avaliação e acompanhamento dos protocolos de pesquisa no que corresponde aos aspectos éticos.

Endereço do Comitê de Ética da Uninove: Rua. Vergueiro nº 235/249 – 3º subsolo - Liberdade – São Paulo – SP CEP. 01504-001 Fone: 3385-9197 . comitedeetica@uninove.br

14. Nome Completo e telefones dos pesquisadores para contato: Orientadora: Claudia Santos Oliveira (11 3665 9344) e aluno de pós graduação: Jamile Benite Palma Lopes (11) 975123549.

15. Eventuais intercorrências que vierem a surgir no decorrer da pesquisa poderão ser discutidas pelos meios próprios.

16. Consentimento Pós-Informação:

Eu,_____, após leitura e compreensão deste termo de informação e consentimento, entendo que minha participação é voluntária, e que posso sair a qualquer momento do estudo, sem prejuízo algum. Confirmo que recebi cópia deste termo de consentimento, e autorizo a execução do trabalho de pesquisa e a divulgação dos dados obtidos neste estudo no meio científico.

* Não assine este termo se ainda tiver alguma dúvida a respeito. São Paulo, de de 2016.

Nome (por extenso) do pesquisado:

_____

Assinatura pesquisado:

_____

Nome (por extenso) do pesquisado:

_____

Assinatura pesquisado:

_____

APPENDIX 2

Approval of the Ethics Committee

UNINOVE
Universidade Nove de Julho

UNIVERSIDADE NOVE DE JULHO - UNINOVE

Plataforma Brasil

PARECER CONSUBSTANCIADO DO CEP

DADOS DO PROJETO DE PESQUISA

Título da Pesquisa: REALIDADE VIRTUAL E ESTIMULAÇÃO TRANSCRANIANA POR CORRENTE CONTÍNUA PARA MELHORA DA FUNÇÃO MOTORA DE MEMBROS SUPERIROES EM CRIANÇAS COM SÍNDROME DE DOWN: ENSAIO CLÍNICO CONTROLADO ALEATORIZADO E DUPLO CEGO

Pesquisador: Jamile Benite Palma Lopes

Área Temática:

Versão: 1

CAAE: 55196516.0.0000.5511

Instituição Proponente: ASSOCIACAO EDUCACIONAL NOVE DE JULHO

Patrocinador Principal: Financiamento Proprio

DADOS DO PARECER

Número do Parecer: 1.517.470

Apresentação do Projeto:

O projeto chama a atenção para as dificuldades motoras apresentadas por crianças com síndrome de down, e sugere uma intervenção que possibilite uma melhora no controle motor dessa população.

Objetivo da Pesquisa:

Verificar os efeitos da estimulação transcraniana por corrente contínua (tDCS) associado a um treino neuromotor em membro superior com realidade virtual sobre a atividade cerebral e controle motor em crianças com Síndrome de Down (SD).

Avaliação dos Riscos e Benefícios:

Embora os procedimentos adotados no estudo sejam não-invasivos os voluntários serão submetidos a risco como por exemplo, quedas, fadiga muscular, câimbras durante o treino motor de realidade virtual. Para que estes riscos sejam minimizados ao máximo serão adotadas as seguintes medidas protetoras: A estimulação transcraniana será realizada por uma fisioterapeuta com experiência na técnica. No treino de realidade virtual serão realizados por uma fisioterapeuta com experiência em treino motor que será acompanhada por ao menos um voluntário ambos

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Telefone: (11)3385-9197 **E-mail:** comitedeetica@uninove.br

UNIVERSIDADE NOVE DE JULHO - UNINOVE

Continuação do Parecer: 1.517.470

permanecerão posicionados do lado do paciente por todo o treino.

Comentários e Considerações sobre a Pesquisa:

O projeto apresenta as características éticas necessárias para realização da pesquisa.

Considerações sobre os Termos de apresentação obrigatória:

Foram apresentados os termos exigidos para aprovação da pesquisa.

Considerando que tratam-se de crianças com Síndrome de Down, não é necessário o TCLE específico para menores.

Recomendações:

O TCLE, embora apresente os aspectos exigidos, ele deve ser reescrito, uma vez que se encontra com uma linguagem estritamente técnica sobre os procedimentos que serão realizados na pesquisas, assim como constitui-se num documento muito longo (com 6 páginas).

Conclusões ou Pendências e Lista de Inadequações:

Recomenda-se reescrever o TCLE, adequando a linguagem (mais simples e acessível ao responsável pela criança participante)

Resumir as informações tornando o TCLE mais simplificado e objetivo.

Este parecer foi elaborado baseado nos documentos abaixo relacionados:

Tipo Documento	Arquivo	Postagem	Autor	Situação
Informações Básicas do Projeto	PB_INFORMAÇÕES_BÁSICAS_DO_PROJETO_693482.pdf	12/04/2016 15:41:42		Aceito
Folha de Rosto	DOC120416.pdf	12/04/2016 15:40:49	Jamile Benite Palma Lopes	Aceito
Projeto Detalhado / Brochura Investigador	projetofinal.pdf	06/04/2016 16:57:04	Jamile Benite Palma Lopes	Aceito
TCLE / Termos de Assentimento / Justificativa de Ausência	Termo.pdf	06/04/2016 16:46:17	Jamile Benite Palma Lopes	Aceito
Cronograma	cronograma.pdf	06/04/2016 16:45:48	Jamile Benite Palma Lopes	Aceito

Situação do Parecer:

Pendente

Necessita Apreciação da CONEP:

Endereço: VERGUEIRO nº 235/249
Bairro: LIBERDADE **CEP:** 01.504-001
UF: SP **Município:** SAO PAULO
Telefone: (11)3385-9197 **E-mail:** comitedeetica@uninove.br

UNIVERSIDADE NOVE DE JULHO - UNINOVE

Continuação do Parecer: 1.517.470

Não

SAO PAULO, 27 de Abril de 2016

Assinado por:
Stella Regina Zamuner
(Coordenador)

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	01
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	03-08-14
	2b	All items from the World Health Organization Trial Registration Data Set	14
Protocol version	3	Date and version identifier	NA
Funding	4	Sources and types of financial, material, and other support	15
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	01-02-03
	5b	Name and contact information for the trial sponsor	01-02-14
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	08-14
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	14

Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	05-06
	6b	Explanation for choice of comparators	05-06
Objectives	7	Specific objectives or hypotheses	07
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	07-08
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	08
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	07
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	12-13-14
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	NA
n	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	NA
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	NA
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	12-13
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	08-09 -10

Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	08
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	08
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	08
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	08
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	08
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	08
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	08
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	08-14
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	08-09

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	14
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	14
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	NA
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	NA
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	NA
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	NA
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	09
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	NA
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	13
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	NA

Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	13
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	NA
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	13
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	03-14
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	14
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	NA
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	13
	31b	Authorship eligibility guidelines and any intended use of professional writers	14
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	NA
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	13
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	NA

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

BMJ Open

Protocol study for a randomized, controlled, double-blind, clinical trial involving virtual reality and anodal transcranial direct current stimulation for the improvement of upper limb motor function in children with Down syndrome

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2017-016260.R1
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Protocol study for a randomized, controlled, double-blind, clinical trial involving virtual reality and anodal transcranial direct current stimulation for the improvement of upper limb motor function in children with Down syndrome

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Authors' contributions

All authors made substantial contributions.

Conceived and designed the experiments: JL LG RC HL ND IM GM AD MG CO. Acquisition of data: JL LG RC HL ND IM GM AD MG CO. Interpretation of data: JL LG RC HL ND IM GM AD MG CO. Contributed analysis tools: JL LG RC HL ND IM GM AD MG CO Wrote the paper: JL LG RC HL ND IM GM AD MG CO. Final approval of the version: JL LG RC HL ND IM GM AD MG CO

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The study received approval from the ethics committee of University Nove de Julho (Sao Paulo, Brazil) under protocol number 1.540.113 and is registered with the Brazilian Registry of Clinical Trials (RBR3PHPXB)

The authors declare that there are no additional unpublished data from the study.

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Abstract

Introduction: Down syndrome results in neuromotor impairment that affects selective motor control, compromising the acquisition of motor skills and functional independence. The aim of the proposed study is to evaluate and compare the effects of multiple-monopolar anodal transcranial direct current stimulation and sham stimulation over the primary motor cortex during upper limb motor training involving virtual reality on motor control, muscle activity, cerebral activity, and functional independence. **Methods and Analysis:** A randomized, controlled, double-blind, clinical trial is proposed. The calculation of the sample size will be defined based on the results of a pilot study involving the same methods. The participants will be randomly allocated to two groups. Evaluations will be conducted before and after the intervention as well as one month after the end of the intervention process. At each evaluation, three-dimensional, analysis of upper limb movement muscle activity will be measured using electromyography, cerebral activity will be measured using an electroencephalogram system, and intellectual capacity will be assessed using the Wechsler Intelligence Scale for Children. Virtual reality training will be performed three times a week (one 20-minute session per day) for a total of ten sessions. During the protocol, transcranial stimulation will be administered concomitantly to upper limb motor training. The results will be analyzed statistically, with a p-value ≤ 0.05 considered indicative of statistical significance. **Ethical aspects and publicity**: The present study received approval from the Institutional Review Board of *Universidade Nove de Julho* (Sao Paulo,Brazil) under process number 1.540.113 and is registered with the Brazilian Registry of Clinical Trials (N° RBR3PHPXB). The participating institutions have presented a declaration of participation. The volunteers will be permitted to drop out of the study at any time with no negative repercussions. The results will be published and will contribute evidence regarding the use of this type of intervention on children.

Keywords: Down syndrome; transcranial direct current stimulation; upper limb.

Strengths and limitations of this study

The proposed project involves the combination of virtual reality (RV) activities for upper limb motor training and multiple-monopolar anodal transcranial direct current stimulation (tDCS) over the primary motor cortex with the aim of optimizing motor control and upper limb function in children with Down syndrome (DS).

1. Adequate upper limb motor control enables individuals to perform daily, functional, and academic activities in an independent fashion.
2. The use of RV activities to improve motor control is a promising therapeutic resource that has demonstrated satisfactory results in the scientific literature, including for individuals with DS.
3. Non-invasive brain stimulation techniques, specifically tDCS, are currently considered effective means to facilitate motor cortical excitability of brain regions underlying the stimulation electrode, leading to improvements in motor control and motor learning. Despite the lack of reports on the effects of transcranial stimulation in children with DS, studies involving pediatric patients have demonstrated that the technique is safe, with little or no adverse effects.
4. We believe that the administration of multiple-monopolar anodal transcranial direct current stimulation over the primary motor cortex, specifically the areas that correspond to upper limb motor control (C3 and C4 of the 10-20 electroencephalogram system) during upper limb motor training with the use of VR activities will enhance the cortical excitability of motor regions and optimize cerebral activity, thereby potentiating the effects of upper limb motor therapy.
5. The literature reports positive effects with the use of tDCS on upper limb movements in children with cerebral palsy. Optimizing such movements has a direct impact on improving one's performance of activities of daily living and functional independence. However, no scientific data were found regarding the use of tDCS during upper limb training in the population of the proposed study

(children with DS).

6. The literature also reports promising results with the use of VR regarding improvements in cognitive aspects of the population in question, as this intervention constitutes multisensory therapy that optimizes one's concentration and assists in the anticipation of movements, thereby exerting an impact on learning aspects in children submitted to this intervention.
7. The limitations of the proposed study regard the lack of scientific data from previous studies involving children with DS for the purposes of comparison with the findings obtained in the proposed study. However, this aspect also demonstrates the importance of the data that will be generated in the proposed study.

1. INTRODUCTION

Down syndrome (DS) is a highly prevalent genetic disease caused by the inheritance of an additional chromosome 21 and is one of the most frequent causes of mental impairment, affecting approximately 20% of the total number of individuals with mental disability.[1] The incidence in the United States is one out of every 700 births and it is estimated that at least 100 thousand individuals in Brazil are diagnosed with the syndrome.[2-4]

Structural and functional abnormalities are found in the nervous system of children with DS. Diffuse brain damage and peculiar electrical functioning during cognitive development result in poor analysis, synthesis, and speech skills. Moreover, such children demonstrate difficulties in selecting and directing a stimulus due to the fatigue of the connections. These abnormalities result in neurological disorders that vary in terms of manifestation and intensity.[5]

According to Flórez and Troncoso (1997), the brain of individuals with DS is smaller in volume in comparison to individuals without this condition. Hypoplasia of the frontal and occipital lobes is a common finding. A unilateral or bilateral reduction in the temporal lobe occurs in up to 50% of cases and reductions in the corpus callosum, anterior commissure, and hippocampus are found.[6-7] Such individuals also have a smaller number of secondary sulci in comparison to individuals without this syndrome, the temporal gyri are underdeveloped and differences in nerve cells are also reported. For instance, Pandilla (1976) reports differences in the axons and dendrites of pyramidal neurons in the motor cortex.[8] Such differences are highly correlated with fragmentation problems and necrosis of these branches as well as differences in the electrical activity of the brain.[9] This problem leads to limitations with regard to synaptic connections and the neural transmission of nerve impulses.

The literature also reports atrophied nerve cells, which are likely associated with lags during the integration of visual and spatial information. According to Block (1991),

individuals with DS also have a smaller cerebellum and base ganglia, which are related to the control of coordination, timing, and balance. Such problems imply limitations with regard to the acquisition of motor skills.[10] According to Bomono & Rosetti (2010), neuromotor abnormalities in DS include hypotonia, diminished primitive reflexes, delayed motor and cognitive development, and lower levels of learning.[11].

Seaman and DePauw (1982) propose a model in which reaching phases of fundamental movements and culturally determined movements is conditioned by the achievement of previous development phases.[9] As this population exhibits problems with regard to systems of early-onset and late-onset maturation, children with DS could encounter difficulties reaching the phase of sensory-motor responses and even acquiring motor skills. According to Connolly (1970), the mechanisms or systems that offer support to development and the acquisition of motor skills can be understood using the concepts of "hardware" and "software", in which changes in "hardware" regard structure, such as the myelinization that occurs in axons, whereas changes in "software" regard function, such as a gain in information processing speed as a result of myelinization; thus, individuals with DS have problems with their "hardware" that have repercussions on their "software".[12] "Hardware" problems lead to limitations with regard to physical and motor aspects, which is an important problem, as both physical proficiency and perceptive-motor proficiency contribute to the acquisition and performance of motor skills. In other words, it is possible that problems with balance, timing, and agility constitute a hindrance to the acquisition of fundamental patterns or specialized skills.[13]

The population with DS exhibits abnormal muscle coordination, difficulty processing sensory information and functional limitations. The upper limb dysfunctions in this population (muscle weakness and hypotonus, slow reflexes, abnormal biomechanics, sensory deficiency) exert a negative impact on the performance of activities of daily living, independence and quality of life.[14]

Studies have been conducted to understand why individuals with DS have slow, unharmonious movements.[15,25] The investigation of electromyographic activity and

muscle torque demonstrates this deficiency, which can be corrected by the repetition of a given movement during motor training activities. Motor control strategies used in the execution of complex activities, such as a reaching task, have been investigated in this population.[15-25]

The positive results achieved with virtual reality (VR) are believed to be related to training in an interactive environment that provides a broad range of activities and scenarios with multiple sensory channels, enabling the creation of exercises at an intensity that is promising for the needs of individuals with DS.[26-28] VR can be used as an auxiliary tool involving a playful, motivational objective that can facilitate the development of perceptions and motor skills through the training of planning skills and motor control as well as stimulation of the plasticity of the central nervous system.[27-28]

Non-invasive brain stimulation methods have been employed in physical rehabilitation protocols due to the promising results achieved with regard to motor learning.[29-30] Transcranial direct current stimulation (tDCS) is a relatively low-cost, noninvasive brain stimulation technique that is easy to administer and offers minimal adverse effects. This method is known to produce lasting changes in motor cortical excitability.[31] Cortical modulation depends on the polarity of the current: anodal stimulation increases cortical excitability, favoring the depolarization of the neuronal membrane, whereas cathodal stimulation has an inhibitory effect due to the hyperpolarization of the neuronal membrane.[31-36]

TDCS has advantages over other transcranial stimulation techniques, such as providing a longer lasting modulatory effect on cortical function as well as its ease of use and lower cost. The results of clinical trials have demonstrated its considerable potential in the treatment of neurological disorders and the investigation of processes of cortical excitability modulation. Moreover, this type of intervention offers a better condition for sham stimulation, which confers greater specificity to the findings.[37-40] In the rehabilitation process, the aim of neuromodulating techniques is to enhance local synaptic efficiency and alter the maladaptive plasticity pattern that emerges after a cortical injury.[41-45].

Although DS is one of the most prevalent diseases in the pediatric population, no studies were found on the effects of tDCS on children with this syndrome. Thus, the lack of investigations on anodal tDCS over the primary motor cortex during motor training for children with DS constitutes a gap in the scientific literature.[46] Considering the high prevalence of DS, the motor limitations stemming from this disease, which exert a negative impact on functionality and independence, and the fact that tDCS is not contraindicated in most cases of this syndrome, the investigation of the effects of this noninvasive brain stimulation technique on children with DS is relevant.[43-45]

The proposed study could be used as the basis for the development of further projects conducted to broaden knowledge on this technique, enabling a novel intervention option for the optimization of motor training in individuals with DS.

2. OBJECTIVES

2.1 Primary objective

The aim of the proposed study is to evaluate and compare the effect of multiple-monopolar anodal tDCS and sham stimulation over the primary motor cortex during upper limb motor training involving virtual reality on motor control (spatiotemporal variables and kinematics of a reaching task), activity of the elbow flexors and extensors, cerebral activity and functional independence in children with DS.

2.1.1 HYPOTHESES

Null hypothesis: Ten sessions of anodal transcranial direct current stimulation over the motor cortex concomitantly to upper limb motor training involving the use of virtual reality activities will result in the same effects as motor training with the use of virtual reality combined with sham transcranial stimulation in children with Down syndrome.

Alternative hypothesis: Ten sessions of anodal transcranial direct current stimulation over the motor cortex concomitantly to upper limb motor training involving the use of virtual reality activities will result in the better effects than motor training with the use of virtual reality combined with sham transcranial stimulation in children with Down syndrome.

2.2 Secondary objectives

- Determine possible correlations between upper limb motor control (movement velocity and total duration of movement) and muscle activity (elbow flexors and extensors), cerebral activity (activity of the parietal lobe, specifically regions C3 and C4) and functional independence with regard to self-care.

- Identify possible prediction factors for the response of upper limb motor control (movement velocity and total duration of movement) in children with DS. Muscle activity of elbow flexors and extensors, cerebral activity (areas C3 and C4 of the 10-20 electroencephalogram system) and transcranial direct current stimulation (active and sham) will be the factors investigated.

3. METHODS AND ANALYSIS

The sample will be composed of children with DS recruited from the physical therapy clinics of *Universidade Nove de Julho*, São Paulo, Brazil. Letters and emails will be sent to pediatricians, physiotherapists and pediatric neurologists to divulge the study. The following will be the inclusion criteria: 1) a diagnosis of DS; 2) adequate comprehension and cooperation during the procedures; 3) age six to 12 years; 4) compromised upper limb motor coordination; and 5) statement of informed consent signed by a legal guardian. The exclusion criteria will be 1) having undergone surgical procedures in the 12 months prior to the onset of the training sessions, 2) orthopedic deformity of the lower limbs or spinal column with an indication for surgery, 3) epilepsy, 4) metal implant in skull or hearing aids, 5) associated neurological disorder, and 6) use of a pacemaker.

3.1 Study Design

A Phase I-II study will be conducted (figure 1): analytical, paired, randomized, controlled, double- blind, clinical trial.

3.2 Sample size

The sample size will be calculated based on the results of a pilot study with the same methods as those of the main study. The pilot study will involve ten children randomly allocated to the experimental and control groups (five children in each group). The sample size will be calculated based on the mean of both groups considering total duration of movement as the primary outcome, with a unidirectional alpha of 0.05 and an 80% power. The sample will be increased by 20% to compensate for possible dropouts.

3.3 Randomization

Patients with DS who meet the eligibility criteria and agree to participate in the study will be submitted to an initial evaluation and will then be randomly allocated to two groups using a randomization method available at the site www.randomizacion.com. This process will be performed by a member of the research team who is not involved in the recruitment or development of the study. During the protocol, the blinding of the main researcher will be ensured with the use of the DC-Stimulator (NeuroConn, Germany), which has active and sham modes that function based on encrypted code, with three configurations to choose so that the more complex conditions of the study can be achieved. The parameters are adjusted individually and the activated mode can only be altered by the programmer.

Experimental group: multiple-monopolar anodal tDCS over the primary motor cortex bilaterally combined with upper limb motor training involving the use of VR;

Control group: sham tDCS over the primary motor cortex bilaterally combined with upper limb motor training involving the use of VR.

3.4 Evaluations

The participants will be submitted to three evaluations: Pre-intervention, post-evaluation (after ten training sessions), and follow up (one month after last training session).

3.4.1 Three-dimensional movement analysis:

Three-dimensional analysis of upper limb movement: the kinematics of upper limb movement will be evaluated using the SMART-D 140® system (BTS, Milan, Italy), with eight cameras sensitive to infrared light, a sampling frequency of 100 Hz and video system synchronized with the SMART-D system. Passive markers will be positioned at anatomic references points directly on the skin with specific adhesive tape, following the protocol of the *SMARTup: The experimental setup* (figure 2). [47-49] A total of 18 markers measuring 15 mm in diameter will be used to identify the position of the head, trunk and upper limbs (upper arm, forearm and hand).

The movement will be divided into three phases: going phase (upper limb moving toward the target), adjusting phase (adjustment of arm to locate target precisely) and returning phase (return to initial position). At least six complete movements will be performed to obtain three adequate cycles for analysis (figure 3). The biomechanical model, filtering of the data, and processing of the variables will be performed using the *SMART analyser* software program (BTS, Milan, Italy). The variables will be identified and calculated for each movement cycle to evaluate any changes that occur after the intervention. The following variables will be considered, with the mean of the results used in the statistical analyses:

- Total duration of movement: total time required to perform the complete reaching task.
- Mean movement velocity: computed during the going phase and determined using the marker positioned on the index finger.
- Adjusting sway index: Defined as the length of the three-dimensional path described by the marker on the index finger during the adjusting phase.
- Range of motion of elbow and shoulder: calculated as the difference between the maximum and minimum angles of the elbow and shoulder on the sagittal (elbow and shoulder) and frontal (shoulder) planes during the going phase, as described in the

literature. [47-48]

3.4.2 Electromyographic (EMG) analysis: Muscle activity during the reaching movement will be determined using EMG. The electrical activity resulting from the activation of the elbow flexors and extensors will be collected using an eight-channel electromyograph (FREEEMG®, BTS Engineering) with a bioelectrical signal amplifier, wireless data transmission and bipolar electrodes with a total gain of 2000 fold and frequency ranging from 20 to 450 Hz. Impedance and the common rejection mode ratio of the equipment are > 10^{15} Ω//0.2 pF and 60/10Hz 92 dB, respectively. The motor point of the muscles will be identified for the placement of the electrodes and the skin will be cleaned with 70% alcohol to reduce bioimpedance, following the recommendations of Surface Electromyography for the Non-Invasive Assessment of Muscles.[50] All EMG data will be digitized at 1000 frames per second using the BTS MYOLAB® software program. The data will be collected simultaneously to the kinematic data and both will be managed using the BTS® system and *Smart Capture*® software program.[51]

3.4.3 Electroencephalographic analysis: Brain activity will be investigated using electroencephalography (EEG), which will be performed during both the three-dimensional analysis of the reaching task and the evaluation of muscle activation using EMG. For such, the volunteer will be seated in an erect position on a chair in front of the table on which the reaching task will be performed. The BrainNet BNT36 device with 36 configurable channels (32 AC and four DC) and a 16-bit analog-digital converter will be used for the acquisition of the EEG signal (figure 4). The analysis of the signal will be performed with the aid of the EEGLab tool implemented on Matlab, which is also capable of furnishing a topographic map of cerebral activity as a function of time. The electrodes will be positioned following the guidelines of the 10/20 electroencephalogram system (figure 5).[52,53]

3.4.5 Pediatric Evaluation of Disability Inventory (PEDI): The children's functional performance will be assessed quantitatively using the PEDI, which is a questionnaire administered in interview format to a caregiver who can provide information regarding the child's performance on typical activities and routine tasks. The PEDI is composed of three parts, the first of which is used to evaluate skills grouped into three functional domains: self-care (73 items), mobility (59 items) and social function (65 items). Each item is scored either zero (not part of the child's repertoire) or 1 (part of the child's repertoire). The scores

are then totaled per domain.[55,56]

3.4.6 Wechsler Intelligence Scale for Children: The Wechsler Intelligence Scale (WIS) was developed for the assessment of the intellectual performance of adults. The WISC was developed as a version for children, which was followed by the revised version, WISC-R. The WISC III is the third version of the scale for children and is used to assess intellectual capacity using 13 subtests, 12 from earlier versions and one additional subtest. The subtests are organized into two groups (verbal and perceptive-motor or execution) and are administrated in alternating order. The verbal subtests are Information, Similarities, Arithmetic, Vocabulary, Comprehension and Digits. The execution group is composed of Matrix Reasoning, Coding, Figure Weights, Block Design, Picture Concepts, Symbol Search and Mazes. Many studies have been conducted and, although improvements have been made with the addition of new items, the fundamental characteristics of the WISC and WISC-R remained the same in WISC III.[57]

4. Procedures

4.1 Intervention protocol

The therapeutic intervention will consist of a combination of tDCS and VR during reaching movements. The protocol will follow safety procedures described in the literature for the use of tDCS on the pediatric population.[29,58,59] Three 20-minute sessions of combined therapy (tDCS concomitantly to upper limb motor training) will be held for a total of ten sessions.[29,30,39,40]

4.2 Transcranial direct current stimulation

Stimulation will be administered using a tDCS device (*DC-Stimulator NeuroCo nn*, Germany), with three sponge (non-metallic) surface electrodes measuring 25 cm^2 (5 x 5 cm) soaked in saline solution.[60] The children will be randomly allocated to two types of treatment: 1) active anodal stimulation over the primary cortex bilaterally; and 2) sham transcranial stimulation. The two anodal electrodes will be positioned over C3 and C4 of the 10-20 international electroencephalogram system [] and the cathode will be positioned over the right deltoid muscle. This montage will enable the child to receive multiple-monopolar anodal tDCS over the primary motor cortex, specifically the area that manages upper limb motor control, while minimizing the effect of cathodal stimulation in the brain.[61-63] A current of 1 mA (current density: 0.029 mA/cm^2) will be administered over the primary motor cortex for 20 minutes during upper limb training.[29,30,39,40] The stimulator has a button that allows the operator to control the intensity of the current. At the beginning of the session, stimulation will be increased gradually until reaching 1 mA and gradually diminished during the final ten seconds of the session. Sham stimulation will consist of the same electrode montage and the stimulator will be switched on for 30 seconds, giving the child the initial sensation of stimulation, but no current will be administered during the remainder of the session. This is considered a valid control procedure in studies involving tDCS.

Adverse effects: Potential adverse effects of tDCS will be evaluated at the end of each session using a questionnaire administered to the child. The questionnaire will address the perception of symptoms having occurred during the session, such as tingling, a burning sensation, headache, pain at the electrode sites, sleepiness, and altered mood. The children will be instructed to answer using a three-point scale. The caregivers and children will also be asked open-ended questions at the beginning of each session regarding the occurrence of headache, scalp pain, burning sensations, redness of the skin, sleepiness, difficulty concentrating, and mood swings during periods between sessions.

4.3 Virtual reality training protocol

Training sessions will be held three times per week on non-consecutive days. Each session will last 20 minutes and will involve the use of the XBOX 360™ with the Kinect™ motion detector.[64] The game entitled "Bursting Bubbles" of the Adventure set of games was chosen based on the potential to stimulate cognitive skills and enhance execution time, motor coordination, attention, concentration, reasoning, memory, persistence, and precise movement. The activity will be held in a specific room of the Integrated Human Movement Analysis Laboratory measuring 2.5 x 4.0 m, with a projection screen (200 x 150 cm) attached to the wall and stereo speakers to provide adequate visual and auditory stimuli. Initially, the child will be instructed to remain standing at a distance of two to three meters in front of the motion detector to capture the movements better as well as for the estimation of height and calculation of the body mass index. Two mobility training sessions with the use of the XBOX 360 exercises will be performed prior to the onset of the intervention protocol. Records will be made of the number of sessions attended and duration of each session.[64-66]

5. Analysis of results

The Shapiro-Wilk test will be used to determine whether the data adhere to the Gaussian curve. Parametric variables will be expressed as mean and standard deviation. Nonparametric variables will be expressed as median and interquartile range. Effect sizes will be calculated from the differences in means between the pre-intervention and post-intervention evaluations. The effect size values will be expressed with respective 95% confidence intervals. Either two-way ANOVA (parametric variables) or the Kruskal-Wallis test (non-parametric variables) will be used for the analysis of the effects of the upper limb motor training activity with active and sham tDCS. Logistic regression models will be created to determine factors predictive of the response to the intervention. For such, movement velocity and total duration of movement will be considered. The response capacity will be defined as a clinically significant increase in performance in comparison to baseline. The independent variables will be age (years), sex (male/female), activity of elbow flexors and extensors, cerebral activity (C3 and C4) and functional independence (aspects of self-care). Univariate regressions will be performed for each variable. Based on the initial analyses, the predictors associated with the outcome with a p-value ≤ 0.05 will be incorporated into the multivariate model. Moreover, Pearson's correlation coefficients will be calculated to determine correlations among the variables analyzed. A p-value < 0.05 will be considered indicative of statistical significance. The data will be organized and tabulated with the aid of the Statistical Package for the Social Sciences (SPSS v.19.0).

6. Discussion

Upper limb motor control enables individuals to perform functional activities. VR will be used as a therapeutic tool to enhance motor control.[29-30] Moreover, a noninvasive brain stimulation method (tDCS) will be employed to facilitate motor cortical excitability in the areas subjacent to stimulation to enhance the effects of motor control and learning.[67] Lazzari et al. (2016) demonstrated the efficacy of the combination of tDCS and VR in potentiating motor effects on balance and functional mobility in children with cerebral palsy.[30]

This document offers a detailed description of a randomized, controlled, double-blind, clinical trial designed to determine the effectiveness of VR training combined with tDCS on upper limb movements in individuals with DS

7. Ethical aspects and divulgation

The present study is in compliance with the guidelines regulating studies involving human subjects established by the Brazilian National Board of Health in October 1996 and updated in Resolution 466 in 2012. The study will be developed at the Integrated Human Movement Analysis Laboratory of University *Nove de Julho* (Sao Paulo, Brazil) and has received approval from the Human Research Ethics Committee of the university under process number 1.517.470 (APPENDIX 1). The protocol has been registered with Clinical Trials. All legal guardians will receive clarifications regarding the procedures and will be aware that participation is voluntary, free of cost and experimental. Those who agree to their child's participation will sign a statement of informed consent (APPENDIX 2). The guardians will be assured of access to all information and will be informed of the possibility of dropping out of the study or withdrawing consent at any time with no negative consequences. The anonymity of the children and the confidentiality of their information will be ensured, following the ethical principles of privacy. The findings will be published and will contribute evidence regarding the use of transcranial direct current stimulation combined with upper limb motor training in this population.

8. Conflict of Interest Statement

The authors have no financial or competing interests

9. Acknowledgments

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11. Abbreviations

DS: Down Syndrome; tDCS: Transcranial direct-current stimulation; EMG: Electromyography; PEDI: Pediatric Evaluation of Disability Inventory; WISC III: Wechsler Intelligence Scale for Children; VR: Virtual Reality

12. List of Figures

REFERENCES

1. Moreira LMA, El-Hani CN, Gusmão FAF. **A syndrome de down e sua patogênese: considerações sobre o determinismo genético**. Rev Bras Psiquiatr. 2000; 22(2): 96-9.
2. Lewada AF, Matsonff A, Revenis M, Futtermam C, Nino G, Greenberg J, et al. **Preoperative evaluation and comprehensive risk assessment for children with Down syndrome. Pediatric Anesthesia**. 2016; 26: 356–362.
3. Moreira LMA, Gusmão FAF. **Aspectos genéticos e sociais da sexualidade em pessoas com síndrome de Down**. Rev Bras Psiquiatria. 2002; 24(2): 94-9.
4. Silva MFMC, Kleinahas ACS. **Cognitive processes and brain plasticity in Down Syndrome**. Revis. Bras. Edc. Esp. 2006; 12: 123-138.
5. Luria, AR, Tskvetkova, LS. **The programing of constructive activety in local brain injuries**. Neuropsychological .1964; 95-107
6. Flórez BJ, Troncoso VM. **Síndrome de Down y educacíon**. 3. reimp. Barcelona: Masson – Salvat Medicina y Santander, 1997.
7. Santos APM, Weiss LI, Almeida GMF**. Assessment and intervention in the motor development of a child with Down syndrome**. Rev Bras Ed Esp Marília. 2010; 16: 19-30
8. Pandilla, M.M. **Pyramidal cell abnormalities in the motor cortex of a child with Down's Syndrome: a golgi study.** Journal of Comparative Neurology. 197:667;63-81.
9. Seaman, J.; Depauw, K.P. **The new adapted physical education**. California, Mayfield, 1982.
10. Block, M.E. **Motor development in children with Down Syndrome: a review of the literature.** Adapted Physical Actvity Quaterly. 1991. 8, 79-209.
11. Bomono LMM, Rosseti CB. **Aspects in perceptual-motor development and sensory-motor intelligence in Down syndrome.** Rev bras crescimento desenvolv hum. 2010; 3: 723-734.
12. Connolly, K.J. **Skill development: Problems and Plans**. In: CONNOLLY, K.J. (ed.) **Mechanisms of motor skill development**. London, Academic Press, 1970
13. Gimenez, R; Stefanoni, F.F & Farias, P.B. **Relação entre a capacidade de sincronização temporal e as habilidades motoras rebater e receber em indivíduos portadores da síndrome de Down.** Revista Brasileira de Ciência e Movimento.
14. Schwartzman, J.S. **Síndrome de Down**. São Paulo, Editora Mackenzie, 1999.

15. Marconi NF, Almeida GL **Principles for learning horizontal-planar arm movements with reversal**. Journal electromygraphic and Kinesiology. 2008. 18;771-779

16. Marconi NF, Almeida GL, Gottlieb GL. **Electromyographic and kinetic strategies to control movements.** Brazilian journal of physical therapy. 2006 10;1-8.

17. Latash ML, Corcos DM. **Kinematic and electromyographic characteristics of single-joint movements of individuals with Down syndrome.** Am J Ment Retard. 1991; 96: 189-201.

18. Almeida GL, Corcos DM, Latash ML. **Practice and transfer effects during fast single-joint elbow movements in individuals with Down syndrome.** Phys Ther. 1994; 74: 1000-1016.

19. Almeida GL, Hasan Z, Corcos DM. **Horizontal-plane arm movements with direction reversals performed by normal individuals and individuals with Down syndrome.** J Neurophysiol. 2000; 84: 1949-1960.

20. Aruin AS, Almeida GL, Latash ML. **Organization of a simple two-joint synergy in individuals with Down syndrome.** Am J Ment Retard. 1996; 101: 256-268.

21. Aruin, AS, Almeida GL. **A co-activation strategy in antecipatory postural adjustments in persons with Down syndrome. Motor Control** 1997; 01: 178-191.

22. Latash ML, Anson JG. **What are "normal movements" in atypical populations**. Behav Brain Sci. 1996; 19: 55-68.

23. Marconi NF. **Controle motor de movimentos de reversão em indivíduos neurologicamente normais e portadores da síndrome de Down:** O efeito do feedback intrínseco [dissertação]. Campinas (SP): Universidade Estadual de Campinas; 2000.

24. Ferreira SMS. **Modulação da Latência da Musculatura Antagonista em Indivíduos "neurologicamente normais" e portadores da Síndrome de Down** [dissertação]. Rio Claro (SP): Universidade Estadual Paulista; 2000.

25. Lorenzo SM, Braccialli LMP, Araújo RCT. **Realidade virtual como intervenção na Síndrome de down: uma perpectiva de ação na interface educação e saúde**. Rev Bras Educ Espc. 2015; 21: 392-396.

26. Wuang YP, Chiang CS, Su CY, Wang CC. **Effectiveness of virtual reality using Wii gaming technology in children with Down syndrome.** Res Dev Disabil. 2011; 32: 312–321.

27. Lin HC, WuangYP. **Strength and agility training in adolescents with Down syndrome: A randomized controlled trial**. Res Dev Disabil. 2012; 33: 2236–2244.

28. Mello BCC, Ramalho TF. **Use of virtual reality in the physical therapeutic treatment of individuals with Down syndrome.** Rev Neurocienc. 2015; 23: 143-149.

29. Moura RFC, Santos C, Grecco LCA, Albertini G, Cimolin V, Galli M, Oliveira C **Effects of a single session of transcranial direct current stimulation on upper limb movements in children with cerebral palsy: A randomized, sham controlled study**. Developmental Neurorehabilitation. 2017. 19:18.

30. Lazzari RD, Politti F, Belina ST, Grecco LAC, Santos CA, Dumont AJL et al. **Effect of Transcranial Direct Current Stimulation Combined With Virtual Reality Training on Balance in Children With Cerebral Palsy: A Randomized, Controlled, Double-Blind, Clinical Trial.** Journal of Motor Behavior. 2016. 1940-1027.

31. Stagg CJ, Bachtiar V, O'Shea J, Allman C, Bosnell RA, Kischka U, Matthews PM, Johansen-Berg H. **Cortical activation changes underlying stimulation induced behavioral gains in chronic stroke. Brain**. 2012; 135:276-84.

32. Miranda PC, Lomarev M, Hallett M. **Modeling the current distribution during transcranial direct current stimulation.** Clin Neurophysiol. 2006; 117(7):1623-9.

33. Wagner T, Fregni F, Fecteau S, Grodzinsky A, Zahn M, Pascual-Leone **A. Transcranial direct current stimulation: A computer-based human model study.** Neuroimage 2007; 35:1113-24.

34. Liebetanz D, Nitsche MA, Teragau F, Paulus W. **Pharmacological approacha to the mechanisms of transcranial DC- stimulation- induced after-effects of humam motor cortex excitability.** Brain 2002; 125: 2238-47.

35. Kuo MF, Unger M, Liebetanz D, Lang N, Tergau F, Paulus W, Nittshe MA. **Limited impact of homeostatic plasticity on motor learning in humans.** Neuropsychologia. 2008; 46: 2122-8.

36. Monte-SilvaK,Kuo M-F, Thirugnanasambandam N,Liebetanz D, Paulus W, Nitsche MA. **Dose- dependente inverted U-shaped effect of dopamine (D2-like) receptor activation on focal and nonfocal plasticity in humans**. The Journal of Neuroscience. 2009; 29 (19):6124-31.

37. Nitsche MA, Liebetanz D, Schlitterlau a, Henschke U, Friche K, Frommann K, et al. **GABAergic modulation of DC stimulation-induced motor cortex excitability shifts in humans.** Eur J Neurosco. 2004;19(10): 2720-6.

38. Dumont AJL, Araujo M, Lazzari RD, Santos CA, Carvalho DB, Moura RCF. **Effects of a single session of transcranial direct current stimulation on static balance in a patient with hemiparesis: a case study**. J. Phys. Ther. Sci. 2015. 27: 955–958.

39. Duarte NDA, Grecco LAC, Galli M, Fregni F, Oliveira C. **Effect of Transcranial Direct-Current Stimulation Combined with Treadmill Training on Balance and Functional Performance in Children with Cerebral Palsy: A Double-Blind Randomized Controlled Trial.** PLOS ONE. 2014. 9;8

40. Grecco LAC, Duarte NDA, Zanon N, Galli M, Fregni F, Oliveira C. **Effect of a single session of transcranial direct-current stimulation on balance and spatiotemporal gait variables in children with cerebral palsy: A randomized sham-controlled study**. Braz J Phys Ther. 2014; 18: 419-427.

41. Mendonça ME, Fregni F**.Neuromodulação com estimulação cerebral não invasiva: aplicação no acidente vascular encefálico, doença de Parkinson e dor crônica. In.:ASSIS, R.D. Condutas práticas em fisioterapia neurológica**. Manole. São Paulo, p. 307-39, 2012.

42. Fregni F, Gimenes R, Valle AC, Ferreira MJ, Rocha RR, Natalle L, Bravo R, Rigonatti SP, Freedman S, Nitsche M, Pascual-Leone A, Boggio PS. **A randomized, sham-controlled, proof of principle study of transcranial direct current stimulation for the treatment of pain in fibromyalgia.** Arthritis and Rheumatism, 2006; 54:3988-98.

43. Fregni F, Bossio PS, Brunoni AR. **Neuromodulação terapêutica: Princípios e avanços da estimulação cerebral não invasiva em neurologia, reabilitação, psiquiatria e neuropsicologia. Sarvier.** São Paulo, 2012.

44. Fritsch B, Reis J, Martinowich K, Schambra HM, Ji Y, Cohen LG, et al. **Direct current stimulation promotes BDNF-dependent synaptic plasticity: potential implications for motor learning. Neuron.** 2010;66(2): 198-204.

45. Reis J, Robertson EM, Krakauer JW, Rothwell J, Marshall L, Gerloff C, et al. **Consensus: Can transcranial direct current stimulation and transcranial magnetic stimulation enhance motor learning and memory formation**. Brain stimulation. 2008;1(4): 363-9.

46. Antal A, Lang N, Boros K, Nitsche M, Siebner HR, Paulus W. **Homeostatic metaplasticity of the motor cortex is altered during headache-free intervals in migraine with aura. Cerebral cortex.** 2008;18(11):2701-5.

47. Cimolin V, Beretta E, Piccinini L, Turconi AC, Galli M, Strazzer S. **Constraint-induced movement therapy for children with hemiplegia after traumatic brain injury: a quantitative study.** J Head Trauma Rehabil. 2012; 27(3): 177-87.

48. Menegoni F, Milano E, Trotti C, Galli M, Bigoni M, Baudo S, Mauro A. **Quantitative evaluation of functional limitation of upper limb movements in subjects affected by ataxia.** Eur J Neurol. 2009; 16(2): 232-9.

49. Petuskey K, Bagley A, Abdala E, James MA, Rab G**. Upper extremity kinematics during functional activities: three-dimensional studies in a normal pediatric population.** Gait Posture. 2007; 25(4): 573-9.

50. Hermes JH, Freriks B, Merletti R, Steggeman D, Blok J, Rau G, Disselhorst-Klug C, Hagg G: **SENIAM 8: Surface Electromyography for the Non-Invasive Assessment of Muscles.** Roessingh Research and Development 1999.

51. Rab G, Petuskey K, Bagley A. **A method for determination of upper extremity kinematics.** Gait Posture. 2002; 15(2): 113-9.

52. Jasper HH. **The ten-twenty electrode system of the International Federation electroencephalogria.** Clin Neurophysiol. 1958; 10: 371-375.

53. Homan RW, Herman J, Purdy P. **Cerebral location of international 10-20 system electrode placement.** Electroencephalogr Clin Neurophysiol. 1987; 66(4): 376-82.

54. FELL, J.; AXMACHER, N. **The role of phase synchronization in memory processes.** Nature Reviews Neuroscience, v. 12, n. 2, p. 105–118, fev. 2011

55. Haley S, Coster W, Ludlow L. **Inventário de avaliação pediátrica de disfunção: versão brasileira. Tradução e adaptação cultural**: Mancini M C. Belo Horizonte: Laboratórios de Atividade e Desenvolvimento Infantil, Departamento de Terapia Ocupacional, Universidade Federal de Minas Gerais. 2000.

56. Feldman AB, Haley SM, Corvell J**. Concurrent and construct validity of the Pediatric Evaluation of Disability Inventory**. Phys. Ther. 1990; 70(10): 602-10.

57. Cruz MBZ, WISC III: **Escala de Inteligência Wechsler para crianças: Manual. Periodicos eletrônicos em psicologia**. 2005; 4: 309.

58. Grecco LA, de Almeida Carvalho Duarte N, Mendonça ME, Cimolin V, Galli M, Fregni F, Santos Oliveira C. **Transcranial direct current stimulation during treadmill training in children with cerebral palsy: a randomized controlled double-blind clinical trial.** Research in Developmental Disabilities 2014;3 (11):2840–2848.

59. Gillick T, Feyma T, Menk J, Usset M, Vaith A, Wood J, Worthing R et al., **Safety and feasibility of transcranial direct current stimulationin pediatric hemiparesis: randomized controlled preliminarystudy.** Physical Therapy 2015;95(3):337–349.

60. Grecco et al. **Cerebellar transcranial direct current stimulation in children with ataxic cerebral palsy: a sham-controlled, crossover,pilot study.** Developmental Neurorehabilitation 2016;22:1–7

61. Naseri P, Nitsche MA e Ekhtiar H. **A framework for categorizing electrode montages in transcranial direct current stimulation.** Front Hum Neurosci. 2015, 6;9:54.

62. Boggio P. S., Ferruci R., Mameli F., Martins D., Martins O., Vergari M., et al. . (2012). **Prolonged visual memory enhancement after direct current stimulation in Alzheimer's disease. Brain Stimul**. 5, 223–230. 10.1016/j.brs.2011.06.006 [PubMed] [Cross Ref]

63. Lapenta O. M., Fregni F., Oberman L. M., Boggio P. S. (2012). **Bilateral temporal cortex transcranial direct current stimulation worsens male performance in a multisensory integration task.** Neurosci. Lett. 527, 105–109. 10.1016/j.neulet.2012.08.076 [PubMed] [Cross Ref]

64. Chamovitz YS e Weiss PL **Virtual reality as a leisure activity for young adults with physical and intellectual disabilities** Science Direct. 2008;29:273- 287

65. Wuang PY, Chiang SC, Su YC et al**., Effectiveness of virtual reality using Wii gaming technology in children with Down syndrome Science Direct.** 2011; 32: 312-321

66. Lin HC e Wuang PY **Strength and agility training in adolescents with Down syndrome: A randomized controlled trial**. Science Direct 2012;33:2236-2244.

67. Minhas P, Bikson M, Woods AJ, Rosen AR, Kessler SK. **Transcranial direct current stimulation in pediatric brain: a computational modeling study**. Conf Proc IEEE Eng Med Biol Soc. 2012;2012:859–62

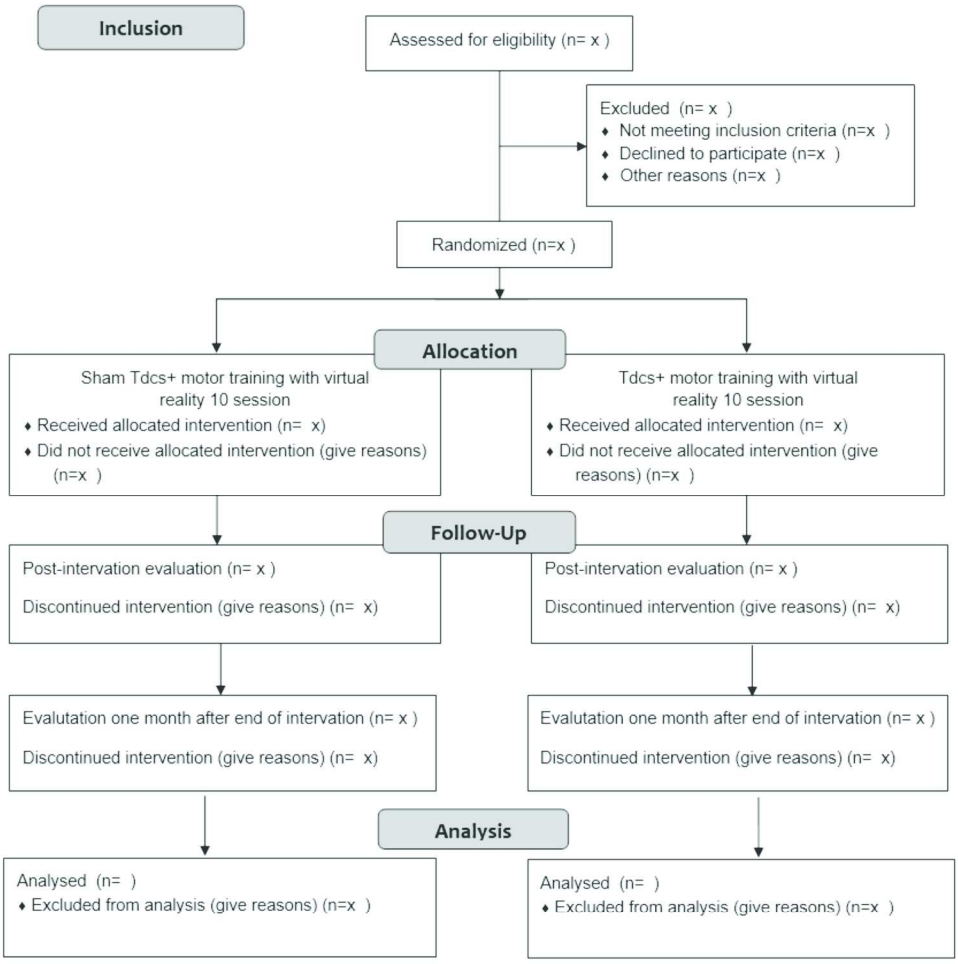

Figure 1: Flowchart of study following CONSORT statement

205x213mm (300 x 300 DPI)

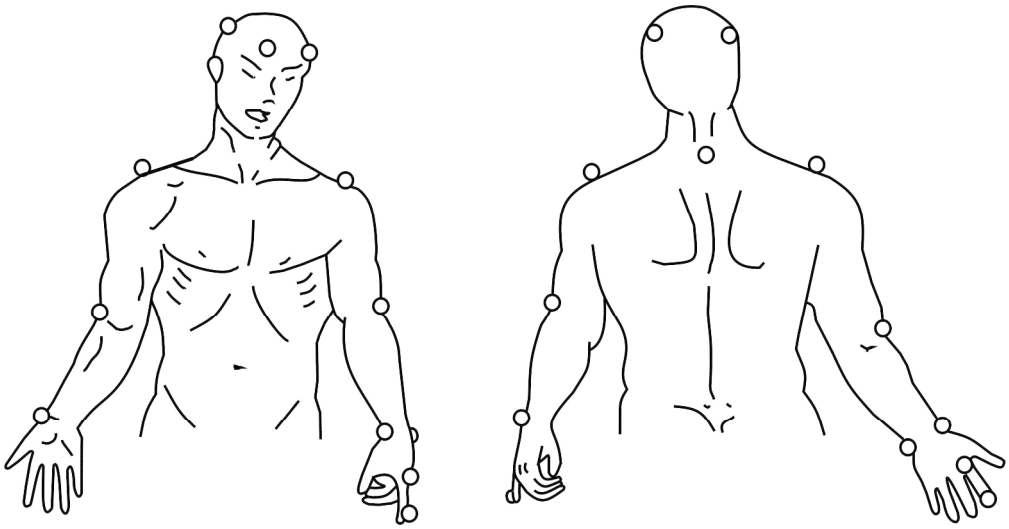

Figure 2: Placement of markers for three-dimensional analysis using SMARTup: The experimental setup

287x149mm (300 x 300 DPI)

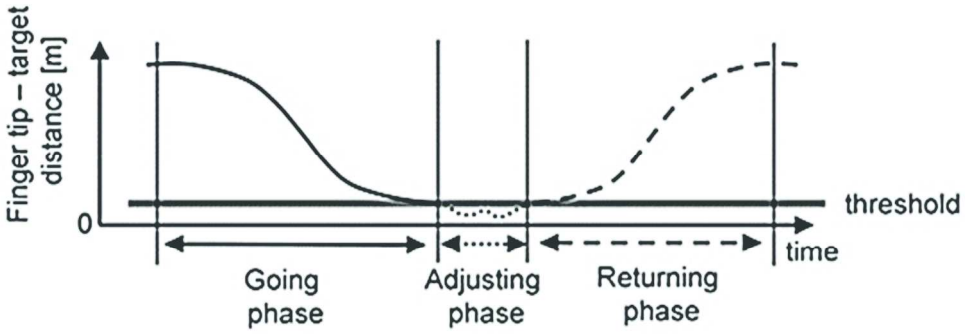

Figure 3: Phases of reaching cycle

294x137mm (300 x 300 DPI)

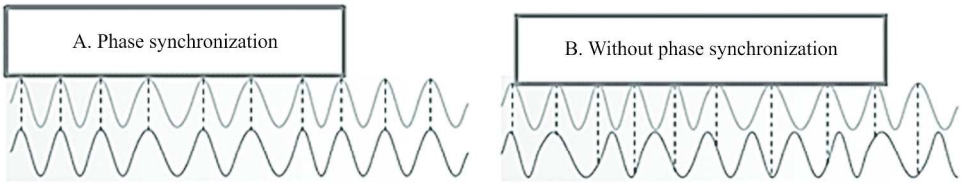

Figure 4 – Phase relationships. (A) synchronized signals – differences in phases between both signals are stable (constant); (B) non-synchronized signals – differences in phases are variable

280x95mm (300 x 300 DPI)

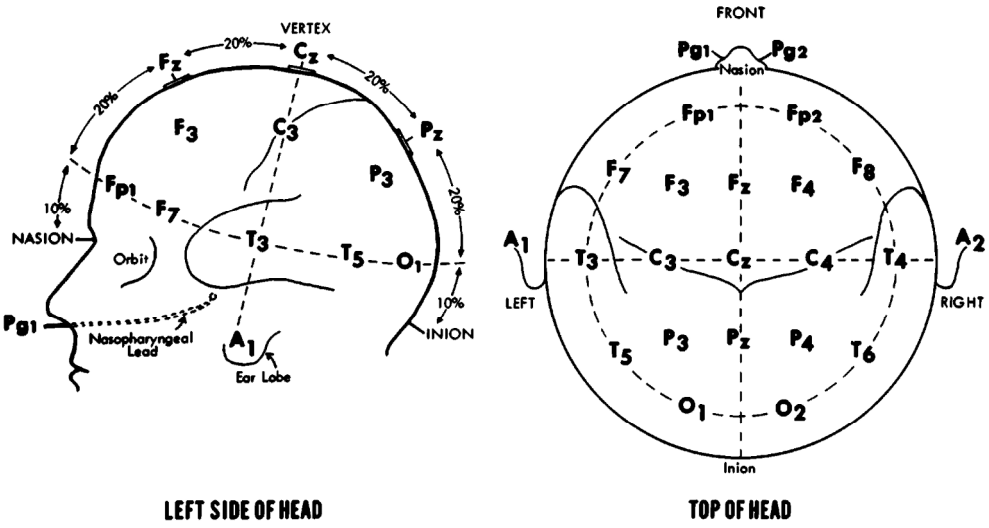

Figure 5 – Positioning of EEG electrodes following 10-20 standard

355x188mm (300 x 300 DPI)

APPENDIX 1

Termo de Consentimento para Participação em Pesquisa Clínica

Nome do Voluntário: _____

Endereço:_____

Telefone para contato:_____Cidade:_____CEP:_____

E mail: _____

1. As informações contidas neste prontuário foram fornecidas pela aluna Jamile Benite Palma Lopes (Mestranda da Universidade Nove de Julho), Profª. Claudia Santos Oliveira, objetivando firmar acordo escrito mediante o qual, o voluntário da pesquisa autoriza sua participação com pleno conhecimento da natureza dos procedimentos e riscos a que se submeterá, com a capacidade de livre arbítrio e sem qualquer coação.

2. Título do Trabalho Experimental: Realidade virtual e estimulação transcraniana por corrente contínua anódica para melhora da função motora de membros superiores em crianças com síndrome de down: ensaio clínico controlado aleatorizado e duplo cego.

3. Objetivo: Examinar os efeitos da estimulação por corrente sobre o controle motor, atividade dos músculos, atividade do cérebro e independência funcional de crianças com Síndrome de Down.

4. Justificativa: acredita-se que ao aplicar a estimulação por corrente, especificamente, durante o treino motor com uso de um vídeo game, será possível, otimizar a atividade do cérebro e a melhora motora.

5. Procedimentos da Fase Experimental: Será selecionas crianças diagnosticadas com Síndrome de Down, com capacidade de entendimento e colaboração para realização dos procedimentos envolvidos no estudo, crianças com idade entre seis e 12 anos, crianças com queixas de comprometimento Na coordenação motora dos braços. O processo de avaliação (antes, após e um mês após o treino, será realizado em três dias não consecutivos, mas na mesma semana, com período máximo de uma hora e 30 minutos por dia. A avaliação será constituída dos seguintes itens: (1) Analise de movimento dos braços durante uma tarefa: avaliado pela cinemática, eletromiografia e eletroencefalograma, a criança realizara uma tarefa com os braços e ao mesmo tempo será avaliada pelos aparelhos, sendo acompanhada pelo fisioterapeuta responsável e pelos assistentes (2) PEDI o PEDI é um questionário aplicado no formato de entrevista estruturada com um dos cuidadores da criança, que possa informar sobre seu desempenho em atividades e tarefas típicas da rotina diária. O teste é composto de três partes: a primeira avalia habilidades de repertório da criança agrupadas segundo três áreas

funcionais: autocuidado (73 itens), mobilidade (59 itens) e função social (65 itens). Cada item dessa parte é pontuado com escore 0 (zero) se a criança não é capaz de desempenhar a atividade, ou 1 (um), se a atividade fizer parte de seu repertório de habilidades. O Grupo 1 terá o movimento do braço analisado após realizar treino com o vídeo game junto com a estimulação desligada (placebo). O Grupo 2 terá o movimento do braço analisado após realizar treino com o vídeo game junto com a estimulação ligada. A estimulação por corrente é uma técnica não invasiva que será realizada colocando eletrodos de superfície conectados a um aparelho de corrente galvânica (corrente elétrica de baixa intensidade) sobre o crânio (cabeça) da criança, durante 20 minutos por 15 dias. A estimulação é indolor.

6. Desconforto ou Risco Esperado: Embora os procedimentos adotados no estudo sejam não-invasivos os voluntários serão submetidos a risco como por exemplo, quedas, fadiga muscular, câimbras durante o treino motor de realidade virtual. Para que estes riscos sejam minimizados ao máximo serão adotadas as seguintes medidas protetoras: A estimulação será realizada por uma fisioterapeuta com experiência na técnica. No treino de realidade virtual serão realizados por uma fisioterapeuta com experiência em treino motor que será acompanhada por ao menos um voluntário ambos permanecerão posicionados do lado do paciente por todo o treino.

7. Informações: o voluntário tem garantia que receberá respostas a qualquer pergunta ou esclarecimento de qualquer dúvida quanto aos procedimentos, riscos benefícios e outros assuntos relacionados com pesquisa. Também os pesquisadores supracitados assumem o compromisso de proporcionar informação atualizada obtida durante o estudo, ainda que esta possa afetar a vontade do indivíduo em continuar participando.

8. Retirada do Consentimento: o voluntário tem a liberdade de retirar seu consentimento a qualquer momento e deixar de participar do estudo, sem que isto lhe traga qualquer prejuízo.

9. Aspecto Legal: Elaborados de acordo com as diretrizes e normas regulamentadas de pesquisa envolvendo seres humanos atendendo à Resolução nº. 466/12 do Conselho Nacional de Saúde do Ministério de Saúde – Brasília – DF.

10. Garantia de Sigilo: Os pesquisadores asseguram a privacidade dos voluntários quanto aos dados confidenciais envolvidos na pesquisa.

11. Formas de ressarcimento das despesas decorrentes da participação na pesquisa: Se necessário, será dado aos pesquisados auxilio transporte de ida e volta ao local da pesquisa. Não será dada ao pesquisado qualquer tipo de remuneração e auxilio de custo pela participação na pesquisa. Pelo curto tempo das avaliações e intervenções não haverá fornecimento de alimentação ao pesquisado.

12. Local da Pesquisa: A pesquisa será desenvolvida no Laboratório Integrado de Análise do Movimento Humano - LIAMH e Núcleo de Apoio a Pesquisa na Analise do Movimento - NAPAM, Universidade Nove de Julho UNINOVE, localizada na rua Vergueiro, no 235/249, 2º subsolo, Vergueiro, São Paulo - SP.

13. Comitê de Ética em Pesquisa (CEP) é um colegiado interdisciplinar e independente, que deve existir nas instituições que realizam pesquisas envolvendo seres humanos no Brasil, criado para defender os interesses dos participantes de pesquisas em sua integridade e dignidade e para contribuir no desenvolvimento das pesquisas dentro dos padrões éticos (Normas e Diretrizes Regulamentadoras da Pesquisa envolvendo Seres Humanos – Res. CNS nº 466/12). O Comitê de Ética é responsável pela avaliação e acompanhamento dos protocolos de pesquisa no que corresponde aos aspectos éticos.

Endereço do Comitê de Ética da Uninove: Rua. Vergueiro nº 235/249 – 3º subsolo - Liberdade – São Paulo – SP CEP. 01504-001 Fone: 3385-9197 . comitedeetica@uninove.br

14. Nome Completo e telefones dos pesquisadores para contato: Orientadora: Claudia Santos Oliveira (11 3665 9344) e aluno de pós graduação: Jamile Benite Palma Lopes (11) 975123549.

15. Eventuais intercorrências que vierem a surgir no decorrer da pesquisa poderão ser discutidas pelos meios próprios.

16. Consentimento Pós-Informação:

Eu,_____, após leitura e compreensão deste termo de informação e consentimento, entendo que minha participação é voluntária, e que posso sair a qualquer momento do estudo, sem prejuízo algum. Confirmo que recebi cópia deste termo de consentimento, e autorizo a execução do trabalho de pesquisa e a divulgação dos dados obtidos neste estudo no meio científico.

* Não assine este termo se ainda tiver alguma dúvida a respeito. São Paulo, de de 2016.

Nome (por extenso) do pesquisado:

_____

Assinatura pesquisado:

_____

Nome (por extenso) do pesquisado:

_____

Assinatura pesquisado:

_____

APPENDIX 2

Approval of the Ethics Committee

UNINOVE
Universidade Nove de Julho

UNIVERSIDADE NOVE DE JULHO - UNINOVE

Plataforma Brasil

PARECER CONSUBSTANCIADO DO CEP

DADOS DO PROJETO DE PESQUISA

Título da Pesquisa: REALIDADE VIRTUAL E ESTIMULAÇÃO TRANSCRANIANA POR CORRENTE CONTÍNUA PARA MELHORA DA FUNÇÃO MOTORA DE MEMBROS SUPERIROES EM CRIANÇAS COM SÍNDROME DE DOWN: ENSAIO CLÍNICO CONTROLADO ALEATORIZADO E DUPLO CEGO

Pesquisador: Jamile Benite Palma Lopes

Área Temática:

Versão: 1

CAAE: 55196516.0.0000.5511

Instituição Proponente: ASSOCIACAO EDUCACIONAL NOVE DE JULHO

Patrocinador Principal: Financiamento Próprio

DADOS DO PARECER

Número do Parecer: 1.517.470

Apresentação do Projeto:
O projeto chama a atenção para as dificuldades motoras apresentadas por crianças com síndrome de down, e sugere uma intervenção que possibilite uma melhora no controle motor dessa população.

Objetivo da Pesquisa:
Verificar os efeitos da estimulação transcraniana por corrente contínua (tDCS) associado a um treino neuromotor em membro superior com realidade virtual sobre a atividade cerebral e controle motor em crianças com Síndrome de Down (SD).

Avaliação dos Riscos e Benefícios:
Embora os procedimentos adotados no estudo sejam não-invasivos os voluntários serão submetidos a risco como por exemplo, quedas, fadiga muscular, câimbras durante o treino motor de realidade virtual. Para que estes riscos sejam minimizados ao máximo serão adotadas as seguintes medidas protetoras: A estimulação transcraniana será realizada por uma fisioterapeuta com experiência na técnica. No treino de realidade virtual serão realizados por uma fisioterapeuta com experiência em treino motor que será acompanhada por ao menos um voluntário ambos

Endereço: VERGUEIRO nº 235/249
Bairro: LIBERDADE **CEP:** 01.504-001
UF: SP **Município:** SAO PAULO
Telefone: (11)3385-9197 **E-mail:** comitedeetica@uninove.br

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Continuação do Parecer: 1.517.470

permanecerão posicionados do lado do paciente por todo o treino.

Comentários e Considerações sobre a Pesquisa:

O projeto apresenta as características éticas necessárias para realização da pesquisa.

Considerações sobre os Termos de apresentação obrigatória:

Foram apresentados os termos exigidos para aprovação da pesquisa.

Considerando que tratam-se de crianças com Síndrome de Down, não é necessário o TCLE específico para menores.

Recomendações:

O TCLE, embora apresente os aspectos exigidos, ele deve ser reescrito, uma vez que se encontra com uma linguagem estritamente técnica sobre os procedimentos que serão realizados na pesquisas, assim como constitui-se num documento muito longo (com 6 páginas).

Conclusões ou Pendências e Lista de Inadequações:

Recomenda-se reescrever o TCLE, adequando a linguagem (mais simples e acessível ao responsável pela criança participante)

Resumir as informações tornando o TCLE mais simplificado e objetivo.

Este parecer foi elaborado baseado nos documentos abaixo relacionados:

Tipo Documento	Arquivo	Postagem	Autor	Situação
Informações Básicas do Projeto	PB_INFORMAÇÕES_BASICAS_DO_PROJETO_693482.pdf	12/04/2016 15:41:42		Aceito
Folha de Rosto	DOC120416.pdf	12/04/2016 15:40:49	Jamile Benite Palma Lopes	Aceito
Projeto Detalhado / Brochura Investigador	projetofinal.pdf	06/04/2016 16:57:04	Jamile Benite Palma Lopes	Aceito
TCLE / Termos de Assentimento / Justificativa de Ausência	Termo.pdf	06/04/2016 16:46:17	Jamile Benite Palma Lopes	Aceito
Cronograma	cronograma.pdf	06/04/2016 16:45:48	Jamile Benite Palma Lopes	Aceito

Situação do Parecer:

Pendente

Necessita Apreciação da CONEP:

Endereço: VERGUEIRO nº 235/249
Bairro: LIBERDADE **CEP:** 01.504-001
UF: SP **Município:** SAO PAULO
Telefone: (11)3385-9197 **E-mail:** comitedeetica@uninove.br

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Continuação do Parecer: 1.517.470

Não

SAO PAULO, 27 de Abril de 2016

Assinado por:
Stella Regina Zamuner
(Coordenador)

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	01
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	03-08-14
	2b	All items from the World Health Organization Trial Registration Data Set	14
Protocol version	3	Date and version identifier	NA
Funding	4	Sources and types of financial, material, and other support	15
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	01-02-03
	5b	Name and contact information for the trial sponsor	01-02-14
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	08-14
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	14

Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	05-06
	6b	Explanation for choice of comparators	05-06
Objectives	7	Specific objectives or hypotheses	07
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	07-08
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	08
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	07
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	12-13-14
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	NA
n	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	NA
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	NA
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	12-13
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	08-09 -10

Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	08
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	08
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	08
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	08
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	08
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	08
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	08
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	08-14
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	08-09

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	14
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	14
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	NA
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	NA
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	NA
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	NA
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	09
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	NA
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	13
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	NA

Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	13
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	NA
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	13
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	03-14
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	14
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	NA
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	13
	31b	Authorship eligibility guidelines and any intended use of professional writers	14
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	NA
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	13
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	NA

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated.

BMJ Open

Protocol study for a randomized, controlled, double-blind, clinical trial involving virtual reality and anodal transcranial direct current stimulation for the improvement of upper limb motor function in children with Down syndrome

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Protocol study for a randomized, controlled, double-blind, clinical trial involving virtual reality and anodal transcranial direct current stimulation for the improvement of upper limb motor function in children with Down syndrome

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Authors' contributions

All authors made substantial contributions.

Conceived and designed: JL LG RC HL ND IM GM AD MG CO. Contributed clinical expertise to the study design: JL LG RC HL ND IM GM AD MG CO. Contributed health systems expertise to the study design.: JL LG RC HL ND IM GM AD MG CO. Developed this study protocol.: JL LG RC HL ND IM GM AD MG CO Wrote the paper: JL LG RC HL ND IM GM AD MG CO. Final approval of the version: JL LG RC HL ND IM GM AD MG CO

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The study received approval from the ethics committee of University Nove de Julho (Sao Paulo, Brazil) under protocol number 1.540.113 and is registered with the Brazilian Registry of Clinical Trials (RBR3PHPXB)

The authors declare that there are no additional unpublished data from the study.

Word count: 5600

Abstract

Introduction: Down syndrome results in neuromotor impairment that affects selective motor control, compromising the acquisition of motor skills and functional independence. The aim of the proposed study is to evaluate and compare the effects of multiple-monopolar anodal transcranial direct current stimulation and sham stimulation over the primary motor cortex during upper limb motor training involving virtual reality on motor control, muscle activity, cerebral activity, and functional independence. **Methods and Analysis:** A randomized, controlled, double-blind, clinical trial is proposed. The calculation of the sample size will be defined based on the results of a pilot study involving the same methods. The participants will be randomly allocated to two groups. Evaluations will be conducted before and after the intervention as well as one month after the end of the intervention process. At each evaluation, three-dimensional, analysis of upper limb movement muscle activity will be measured using electromyography, cerebral activity will be measured using an electroencephalogram system, and intellectual capacity will be assessed using the Wechsler Intelligence Scale for Children. Virtual reality training will be performed three times a week (one 20-minute session per day) for a total of ten sessions. During the protocol, transcranial stimulation will be administered concomitantly to upper limb motor training. The results will be analyzed statistically, with a p-value ≤ 0.05 considered indicative of statistical significance. **Ethical aspects and publicity**: The present study received approval from the Institutional Review Board of *Universidade Nove de Julho* (Sao Paulo,Brazil) under process number 1.540.113 and is registered with the Brazilian Registry of Clinical Trials (N° RBR3PHPXB). The participating institutions have presented a declaration of participation. The volunteers will be permitted to drop out of the study at any time with no negative repercussions. The results will be published and will contribute evidence regarding the use of this type of intervention on children.

Keywords: Down syndrome; transcranial direct current stimulation; upper limb.

Strengths and limitations of this study

The proposed project involves the combination of virtual reality (VR) activities for upper limb motor training and multiple-monopolar anodal transcranial direct current stimulation (tDCS) over the primary motor cortex with the aim of optimizing motor control and upper limb function in children with Down syndrome (DS).

1. Adequate upper limb motor control enables individuals to perform daily, functional, and academic activities in an independent fashion.
2. The use of RV activities to improve motor control is a promising therapeutic resource that has demonstrated satisfactory results in the scientific literature, including for individuals with DS.
3. Non-invasive brain stimulation techniques, specifically tDCS, are currently considered effective means to facilitate motor cortical excitability of brain regions underlying the stimulation electrode, leading to improvements in motor control and motor learning. Despite the lack of reports on the effects of transcranial stimulation in children with DS, studies involving pediatric patients have demonstrated that the technique is safe, with little or no adverse effects.
4. We believe that the administration of multiple-monopolar anodal transcranial direct current stimulation over the primary motor cortex, specifically the areas that correspond to upper limb motor control (C3 and C4 of the 10-20 electroencephalogram system) during upper limb motor training with the use of VR activities will enhance the cortical excitability of motor regions and optimize cerebral activity, thereby potentiating the effects of upper limb motor therapy.
5. The literature reports positive effects with the use of tDCS on upper limb movements in children with cerebral palsy. Optimizing such movements has a direct impact on improving one's performance of activities of daily living and functional independence. However, no scientific data were found regarding the use of tDCS during upper limb training in the population of the proposed study

(children with DS).

6. The literature also reports promising results with the use of VR regarding improvements in cognitive aspects of the population in question, as this intervention constitutes multisensory therapy that optimizes one's concentration and assists in the anticipation of movements, thereby exerting an impact on learning aspects in children submitted to this intervention.
7. The limitations of the proposed study regard the lack of scientific data from previous studies involving children with DS for the purposes of comparison with the findings obtained in the proposed study. However, this aspect also demonstrates the importance of the data that will be generated in the proposed study.

1. INTRODUCTION

Down syndrome (DS) is a highly prevalent genetic disease caused by the inheritance of an additional chromosome 21 and is one of the most frequent causes of mental impairment, affecting approximately 20% of the total number of individuals with mental disability.[1] The incidence in the United States is one out of every 700 births and it is estimated that at least 100 thousand individuals in Brazil are diagnosed with the syndrome.[2-4]

Structural and functional abnormalities are found in the nervous system of children with DS. Diffuse brain damage and peculiar electrical functioning during cognitive development result in poor analysis, synthesis, and speech skills. Moreover, such children demonstrate difficulties in selecting and directing a stimulus due to the fatigue of the connections. These abnormalities result in neurological disorders that vary in terms of manifestation and intensity.[5]

According to Flórez and Troncoso (1997), the brain of individuals with DS is smaller in volume in comparison to individuals without this condition. Hypoplasia of the frontal and occipital lobes is a common finding. A unilateral or bilateral reduction in the temporal lobe occurs in up to 50% of cases and reductions in the corpus callosum, anterior commissure, and hippocampus are found.[6-7] Such individuals also have a smaller number of secondary sulci in comparison to individuals without this syndrome, the temporal gyri are underdeveloped and differences in nerve cells are also reported. For instance, Pandilla (1976) reports differences in the axons and dendrites of pyramidal neurons in the motor cortex.[8] Such differences are highly correlated with fragmentation problems and necrosis of these branches as well as differences in the electrical activity of the brain.[9] This problem leads to limitations with regard to synaptic connections and the neural transmission of nerve impulses.

The literature also reports atrophied nerve cells, which are likely associated with lags during the integration of visual and spatial information. According to Block (1991),

individuals with DS also have a smaller cerebellum and base ganglia, which are related to the control of coordination, timing, and balance. Such problems imply limitations with regard to the acquisition of motor skills.[10] According to Bomono & Rosetti (2010), neuromotor abnormalities in DS include hypotonia, diminished primitive reflexes, delayed motor and cognitive development, and lower levels of learning.[11]

Seaman and DePauw (1982) propose a model in which reaching phases of fundamental movements and culturally determined movements is conditioned by the achievement of previous development phases.[9] As this population exhibits problems with regard to systems of early-onset and late-onset maturation, children with DS could encounter difficulties reaching the phase of sensory-motor responses and even acquiring motor skills. According to Connolly (1970), the mechanisms or systems that offer support to development and the acquisition of motor skills can be understood using the concepts of "hardware" and "software", in which changes in "hardware" regard structure, such as the myelinization that occurs in axons, whereas changes in "software" regard function, such as a gain in information processing speed as a result of myelinization; thus, individuals with DS have problems with their "hardware" that have repercussions on their "software".[12] "Hardware" problems lead to limitations with regard to physical and motor aspects, which is an important problem, as both physical proficiency and perceptive-motor proficiency contribute to the acquisition and performance of motor skills. In other words, it is possible that problems with balance, timing, and agility constitute a hindrance to the acquisition of fundamental patterns or specialized skills.[13]

The population with DS exhibits abnormal muscle coordination, difficulty processing sensory information and functional limitations. The upper limb dysfunctions in this population (muscle weakness and hypotonus, slow reflexes, abnormal biomechanics, sensory deficiency) exert a negative impact on the performance of activities of daily living, independence and quality of life.[14]

Studies have been conducted to understand why individuals with DS have slow, unharmonious movements.[15,25] The investigation of electromyographic activity and

muscle torque demonstrates this deficiency, which can be corrected by the repetition of a given movement during motor training activities. Motor control strategies used in the execution of complex activities, such as a reaching task, have been investigated in this population.[15-25]

The positive results achieved with virtual reality (VR) are believed to be related to training in an interactive environment that provides a broad range of activities and scenarios with multiple sensory channels, enabling the creation of exercises at an intensity that is promising for the needs of individuals with DS.[26-28] VR can be used as an auxiliary tool involving a playful, motivational objective that can facilitate the development of perceptions and motor skills through the training of planning skills and motor control as well as stimulation of the plasticity of the central nervous system.[27-28]

Non-invasive brain stimulation methods have been employed in physical rehabilitation protocols due to the promising results achieved with regard to motor learning, in the pediatric population with cerebral palsy since it was never used in DS.[29-30] Transcranial direct current stimulation (tDCS) is a relatively low-cost, noninvasive brain stimulation technique that is easy to administer and offers minimal adverse effects. This method is known to produce lasting changes in motor cortical excitability.[31] Cortical modulation depends on the polarity of the current: anodal stimulation increases cortical excitability, favoring the depolarization of the neuronal membrane, whereas cathodal stimulation has an inhibitory effect due to the hyperpolarization of the neuronal membrane.[31-36]

TDCS has advantages over other transcranial stimulation techniques, such as providing a long-lasting modulating effect on cortical function as well as its ease of use because its device is portable, so it is possible to becaused simultaneously with rehabilitation techniques and has Lower cost. The results of clinical trials have demonstrated its considerable potential in the treatment of neurological disorders and the investigation of processes of cortical excitability modulation [37-42] . Moreover, this type of intervention offers a better condition for sham stimulation, which confers greater specificity to the findings.[39-40] In the rehabilitation process, the aim of neuromodulating techniques is to enhance local

synaptic efficiency and alter the maladaptive plasticity pattern that emerges after a cortical injury.[41-45].

Although DS is one of the most prevalent diseases in the pediatric population, no studies were found on the effects of tDCS on children with this syndrome. Thus, the lack of investigations on anodal tDCS over the primary motor cortex during motor training for children with DS constitutes a gap in the scientific literature.[46-48] Considering the high prevalence of DS, the motor limitations stemming from this disease, which exert a negative impact on functionality and independence, and the fact that tDCS is not contraindicated in most cases of this syndrome, the investigation of the effects of this noninvasive brain stimulation technique on children with DS is relevant.[43-45]

The proposed study could be used as the basis for the development of further projects conducted to broaden knowledge on this technique, enabling a novel intervention option for the optimization of motor training in individuals with DS.

2. OBJECTIVES

2.1 Primary objective

The aim of the proposed study is to evaluate and compare the effect of multiple-monopolar anodal tDCS and sham stimulation over the primary motor cortex during upper limb motor training involving VR on motor control (spatiotemporal variables and kinematics of a reaching task), activity of the elbow flexors and extensors, cerebral activity and functional independence in children with DS.

2.1.1 HYPOTHESES

Null hypothesis: Ten sessions of tDCS over the motor cortex concomitantly to upper limb motor training involving the use of VR activities will result in the same effects as motor training with the use of virtual reality combined with sham transcranial stimulation in children with DS

Alternative hypothesis: Ten sessions of tDCS over the motor cortex concomitantly to upper limb motor training involving the use of virtual reality activities will result in the better effects than motor training with the use of VR combined with sham tDCS in children with DS.

2.2 Secondary objectives

- Determine possible correlations between upper limb motor control (movement velocity and total duration of movement) and muscle activity (elbow flexors and extensors), cerebral activity (activity of the parietal lobe, specifically regions C3 and C4) and functional independence with regard to self-care.

- Identify possible prediction factors for the response of upper limb motor control (movement velocity and total duration of movement) in children with DS. Muscle activity of elbow flexors and extensors, cerebral activity (areas C3 and C4 of the 10-20 electroencephalogram system) and tDCS (active and sham) will be the factors investigated.

3. METHODS AND ANALYSIS

The sample will be composed of children with DS recruited from the physical therapy clinics of *Universidade Nove de Julho*, São Paulo, Brazil. Letters and emails will be sent to pediatricians, physiotherapists and pediatric neurologists to divulge the study. The following will be the inclusion criteria: 1) a diagnosis of DS; 2) adequate comprehension and cooperation during the procedures; 3) age six to 12 years; 4) compromised upper limb motor coordination; and 5) statement of informed consent signed by a legal guardian. The exclusion criteria will be 1) having undergone surgical procedures in the 12 months prior to the onset of the training sessions, 2) orthopedic deformity of the lower limbs or spinal column with an indication for surgery, 3) epilepsy, 4) metal implant in skull or hearing aids, 5) associated neurological disorder, and 6) use of a pacemaker.

3.1 Study Design

A Phase I-II study will be conducted (figure 1): analytical, paired, randomized, controlled, double-blind, clinical trial

3.2 Sample size

The sample size will be calculated based on the results of a pilot study with the same methods as those of the main study. The pilot study will involve ten children randomly allocated to the experimental and control groups (five children in each group). The sample size will be calculated based on the mean of both groups considering total duration of movement as the primary outcome, with a unidirectional alpha of 0.05 and an 80% power. The sample will be increased by 20% to compensate for possible dropouts.

3.3 Randomization

Patients with DS who meet the eligibility criteria and agree to participate in the study will be submitted to an initial evaluation and will then be randomly allocated to two groups using a randomization method available at the site www.randomizacion.com. This process will be performed by a member of the research team who is not involved in the recruitment or development of the study. During the protocol, the blinding of the main researcher will be ensured with the use of the DC-Stimulator (NeuroConn, Germany), which has active and sham modes that function based on encrypted code, with three configurations to choose so that the more complex conditions of the study can be achieved. The parameters are adjusted individually and the activated mode can only be altered by the programmer.

Experimental group: multiple-monopolar anodal tDCS over the primary motor cortex bilaterally combined with upper limb motor training involving the use of VR;

Control group: sham tDCS over the primary motor cortex bilaterally combined with upper limb motor training involving the use of VR.

3.4 Evaluations

The participants will be submitted to three evaluations: Pre-intervention, post-evaluation (after ten training sessions), and follow up (one month after last training session).

3.4.1 Three-dimensional movement analysis:

Three-dimensional analysis of upper limb movement: the kinematics of upper limb movement will be evaluated using the SMART-D 140® system (BTS, Milan, Italy), with eight cameras sensitive to infrared light, a sampling frequency of 100 Hz and video system synchronized with the SMART-D system. Passive markers will be positioned at anatomic references points directly on the skin with specific adhesive tape, following the protocol of the *SMARTup: The experimental setup* (figure 2).[49-51] A total of 18 markers measuring 15 mm in diameter will be used to identify the position of the head, trunk and upper limbs (upper arm, forearm and hand).

The movement will be divided into three phases: going phase (upper limb moving toward the target), adjusting phase (adjustment of arm to locate target precisely) and returning phase (return to initial position). At least six complete movements will be performed to obtain three adequate cycles for analysis (figure 3). The biomechanical model, filtering of the data, and processing of the variables will be performed using the *SMART analyser* software program (BTS, Milan, Italy). The variables will be identified and calculated for each movement cycle to evaluate any changes that occur after the intervention. The following variables will be considered, with the mean of the results used in the statistical analyses:

- Total duration of movement: total time required to perform the complete reaching task.
- Mean movement velocity: computed during the going phase and determined using the marker positioned on the index finger.
- Adjusting sway index: Defined as the length of the three-dimensional path described by the marker on the index finger during the adjusting phase.
- Range of motion of elbow and shoulder: calculated as the difference between the maximum and minimum angles of the elbow and shoulder on the sagittal (elbow and shoulder) and frontal (shoulder) planes during the going phase, as described in the literature. [49-51]

Figure 2: Placement of markers for three-dimensional analysis using *SMARTup: The experimental setup* [49]

Figure 3: Phases of reaching cycle [49]

3.4.2 Electromyographic (EMG) analysis: Muscle activity during the reaching movement will be determined using EMG. The electrical activity resulting from the activation of the elbow flexors and extensors will be collected using an eight-channel electromyograph (FREEEMG®, BTS Engineering) with a bioelectrical signal amplifier, wireless data transmission and bipolar electrodes with a total gain of 2000 fold and frequency ranging from 20 to 450 Hz. Impedance and the common rejection mode ratio of the equipment are > 10^{15} Ω//0.2 pF and 60/10Hz 92 dB, respectively. The motor point of the muscles will be identified for the placement of the electrodes and the skin will be cleaned with 70% alcohol to reduce bioimpedance, following the recommendations of Surface Electromyography for the Non-Invasive Assessment of Muscles.[52] All EMG data will be digitized at 1000 frames per second using the BTS MYOLAB® software program. The data will be collected simultaneously to the kinematic data and both will be managed using the BTS® system and *Smart Capture*® software program.[52-53]

3.4.3 Electroencephalographic analysis: Brain activity will be investigated using electroencephalography (EEG), which will be performed during both the three-dimensional analysis of the reaching task and the evaluation of muscle activation using EMG. For such, the volunteer will be seated in an erect position on a chair in front of the table on which the reaching task will be performed. The BrainNet BNT36 device with 36 configurable channels (32 AC and four DC) and a 16-bit analog-digital converter will be used for the acquisition of the EEG signal (figure 4). The analysis of the signal will be performed with the aid of the EEGLab tool implemented on Matlab, which is also capable of furnishing a topographic map of cerebral activity as a function of time. The electrodes will be positioned following the guidelines of the 10/20 electroencephalogram system (figure 5).[54-55]

3.4.5 Pediatric Evaluation of Disability Inventory (PEDI): The children's functional performance will be assessed quantitatively using the PEDI, which is a questionnaire administered in interview format to a caregiver who can provide information regarding the child's performance on typical activities and routine tasks. The PEDI is composed of three parts, the first of which is used to evaluate skills grouped into three functional domains: self-care (73 items), mobility (59 items) and social function (65 items). Each item is scored either zero (not part of the child's repertoire) or 1 (part of the child's repertoire). The scores are then totaled per domain.[56-58]

3.4.6 Wechsler Intelligence Scale for Children: The Wechsler Intelligence Scale (WIS) was developed for the assessment of the intellectual performance of adults. The WISC was developed as a version for children, which was followed by the revised version, WISC-R. The WISC III is the third version of the scale for children and is used to assess intellectual capacity using 13 subtests, 12 from earlier versions and one additional subtest. The subtests are organized into two groups (verbal and perceptive-motor or execution) and are administrated in alternating order. The verbal subtests are Information, Similarities, Arithmetic, Vocabulary, Comprehension and Digits. The execution group is composed of Matrix Reasoning, Coding, Figure Weights, Block Design, Picture Concepts, Symbol Search and Mazes. Many studies have been conducted and, although improvements have been made with the addition of new items, the fundamental characteristics of the WISC and WISC-R remained the same in WISC III.[59]

4. Procedures

4.1 Intervention protocol

The therapeutic intervention will consist of a combination of tDCS and VR during reaching movements. The protocol will follow safety procedures described in the literature for the use of tDCS on the pediatric population.[29,60,51] Three 20-minute sessions of combined therapy (tDCS concomitantly to upper limb motor training) will be held for a total of ten sessions.[29,30,39,40]

4.2 Transcranial direct current stimulation

Stimulation will be administered using a tDCS device (*DC-Stimulator NeuroCo nn*, Germany), with three sponge (non-metallic) surface electrodes measuring 25 cm^2 (5 x 5 cm) soaked in saline solution.[61-62] The children will be randomly allocated to two types of treatment: 1) active anodal stimulation over the primary cortex bilaterally; and 2) sham transcranial stimulation. The two anodal electrodes will be positioned over C3 and C4 of the 10-20 international electroencephalogram system [62] and the cathode will be positioned over the right deltoid muscle. This montage will enable the child to receive multiple-monopolar anodal tDCS over the primary motor cortex, specifically the area that manages upper limb motor control, while minimizing the effect of cathodal stimulation in the brain.[61-63] A current of 1 mA (current density: 0.029 mA/cm^2) will be administered over the primary motor cortex for 20 minutes during upper limb training.[29,30,39,41] The stimulator has a button that allows the operator to control the intensity of the current. At the beginning of the session, stimulation will be increased gradually until reaching 1 mA and gradually diminished during the final ten seconds of the session. Sham stimulation will consist of the same electrode montage and the stimulator will be switched on for 30 seconds, giving the child the initial sensation of stimulation, but no current will be administered during the remainder of the session. This is considered a valid control procedure in studies involving tDCS .[64-65].

Adverse effects: Potential adverse effects of tDCS will be evaluated at the end of each session using a questionnaire administered to the child. The questionnaire will address the perception of symptoms having occurred during the session, such as tingling, a burning sensation, headache, pain at the electrode sites, sleepiness, and altered mood. The children will be instructed to answer using a three-point scale. The caregivers and children will also be asked open-ended questions at the beginning of each session regarding the occurrence of headache, scalp pain, burning sensations, redness of the skin, sleepiness, difficulty concentrating, and mood swings during periods between sessions.

4.3 Virtual reality training protocol

Training sessions will be held three times per week on non-consecutive days. Each session will last 20 minutes and will involve the use of the XBOX 360™ with the Kinect™ motion detector.[66] The game entitled "Bursting Bubbles" of the Adventure set of games was chosen based on the potential to stimulate cognitive skills and enhance execution time, motor coordination, attention, concentration, reasoning, memory, persistence, and precise movement. The activity will be held in a specific room of the Integrated Human Movement Analysis Laboratory measuring 2.5 x 4.0 m, with a projection screen (200 x 150 cm) attached to the wall and stereo speakers to provide adequate visual and auditory stimuli. Initially, the child will be instructed to remain standing at a distance of two to three meters in front of the motion detector to capture the movements better as well as for the estimation of height and calculation of the body mass index. Two mobility training sessions with the use of the XBOX 360 exercises will be performed prior to the onset of the intervention protocol. Records will be made of the number of sessions attended and duration of each session.[66-68]

5. Analysis of results

The Shapiro-Wilk test will be used to determine whether the data adhere to the Gaussian curve. Parametric variables will be expressed as mean and standard deviation. Nonparametric variables will be expressed as median and interquartile range. Effect sizes will be calculated from the differences in means between the pre-intervention and post-intervention evaluations. The effect size values will be expressed with respective 95% confidence intervals. Either two-way ANOVA (parametric variables) or the Kruskal-Wallis test (non-parametric variables) will be used for the analysis of the effects of the upper limb motor training activity with active and sham tDCS. Logistic regression models will be created to determine factors predictive of the response to the intervention. For such, movement velocity and total duration of movement will be considered. The response capacity will be defined as a clinically significant increase in performance in comparison to baseline. The independent variables will be age (years), sex (male/female), activity of elbow flexors and extensors, cerebral activity (C3 and C4) and functional independence (aspects of self-care). Univariate regressions will be performed for each variable. Based on the initial analyses, the predictors associated with the outcome with a p-value ≤ 0.05 will be incorporated into the multivariate model. Moreover, Pearson's correlation coefficients will be calculated to determine correlations among the variables analyzed. A p-value < 0.05 will be considered indicative of statistical significance. The data will be organized and tabulated with the aid of the Statistical Package for the Social Sciences (SPSS v.19.0).

6. Discussion

Upper limb motor control enables individuals to perform functional activities. VR will be used as a therapeutic tool to enhance motor control.[29-30] Moreover, a noninvasive brain stimulation method (tDCS) will be employed to facilitate motor cortical excitability in the areas subjacent to stimulation to enhance the effects of motor control and learning.[37-42] Lazzari et al. (2016) demonstrated the efficacy of the combination of tDCS and VR in potentiating motor effects on balance and functional mobility in children with cerebral palsy.[37]

This document offers a detailed description of a randomized, controlled, double-blind, clinical trial designed to determine the effectiveness of VR training combined with tDCS on upper limb movements in individuals with DS

7. Ethical aspects and divulgation

The present study is in compliance with the guidelines regulating studies involving human subjects established by the Brazilian National Board of Health in October 1996 and updated in Resolution 466 in 2012. The study will be developed at the Integrated Human Movement Analysis Laboratory of University *Nove de Julho* (Sao Paulo, Brazil) and has received approval from the Human Research Ethics Committee of the university under process number 1.517.470 (APPENDIX 1). The protocol has been registered with Clinical Trials. All legal guardians will receive clarifications regarding the procedures and will be aware that participation is voluntary, free of cost and experimental. Those who agree to their child's participation will sign a statement of informed consent (APPENDIX 2). The guardians will be assured of access to all information and will be informed of the possibility of dropping out of the study or withdrawing consent at any time with no negative consequences. The anonymity of the children and the confidentiality of their information will be ensured, following the ethical principles of privacy. The findings will be published and will contribute evidence regarding the use of transcranial direct current stimulation combined with upper limb motor training in this population.

8. Conflict of Interest Statement

The authors have no financial or competing interests

9. Acknowledgments

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10. Funding

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11. Abbreviations

DS: Down Syndrome; tDCS: Transcranial direct-current stimulation; EMG: Electromyography; PEDI: Pediatric Evaluation of Disability Inventory; WISC III: Wechsler Intelligence Scale for Children; VR: Virtual Reality

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Figure 4: Phase relationships. (A) synchronized signals – differences in phases between both signals are stable (constant); (B) non-synchronized signals – differences in phases are variable

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REFERENCES

1. Moreira LMA, El-Hani CN, Gusmão FAF. **A syndrome de down e sua patogênese: considerações sobre o determinismo genético**. Rev Bras Psiquiatr. 2000; 22(2): 96-9.
2. Lewada AF, Matsonff A, Revenis M, Futtermam C, Nino G, Greenberg J, et al. **Preoperative evaluation and comprehensive risk assessment for children with Down syndrome. Pediatric Anesthesia**. 2016; 26: 356–362.
3. Moreira LMA, Gusmão FAF. **Aspectos genéticos e sociais da sexualidade em pessoas com síndrome de Down**. Rev Bras Psiquiatria. 2002; 24(2): 94-9.
4. Silva MFMC, Kleinahas ACS. **Cognitive processes and brain plasticity in Down Syndrome**. Revis. Bras. Edc. Esp. 2006; 12: 123-138.
5. Luria, AR, Tskvetkova, LS. **The programing of constructive activety in local brain injuries**. Neuropsychological .1964; 95-107
6. Flórez BJ, Troncoso VM. **Síndrome de Down y educacíon**. 3. reimp. Barcelona: Masson – Salvat Medicina y Santander, 1997.
7. Santos APM, Weiss LI, Almeida GMF**. Assessment and intervention in the motor development of a child with Down syndrome**. Rev Bras Ed Esp Marília. 2010; 16: 19-30
8. Pandilla, M.M. **Pyramidal cell abnormalities in the motor cortex of a child with Down's Syndrome: a golgi study.** Journal of Comparative Neurology. 197:667;63-81.
9. Seaman, J.; Depauw, K.P. **The new adapted physical education**. California, Mayfield, 1982.
10. Block, M.E. **Motor development in children with Down Syndrome: a review of the literature.** Adapted Physical Actvity Quaterly. 1991. 8, 79-209.
11. Bomono LMM, Rosseti CB. **Aspects in perceptual-motor development and sensory-motor intelligence in Down syndrome.** Rev bras crescimento desenvolv hum. 2010; 3: 723-734.
12. Connolly, K.J. **Skill development: Problems and Plans**. In: CONNOLLY, K.J. (ed.) **Mechanisms of motor skill development**. London, Academic Press, 1970
13. Gimenez, R; Stefanoni, F.F & Farias, P.B. **Relação entre a capacidade de sincronização temporal e as habilidades motoras rebater e receber em indivíduos portadores da síndrome de Down.** Revista Brasileira de Ciência e Movimento.
14. Schwartzman, J.S. **Síndrome de Down**. São Paulo, Editora Mackenzie, 1999.

15. Marconi NF, Almeida GL **Principles for learning horizontal-planar arm movements with reversal**. Journal electromygraphic and Kinesiology. 2008. 18;771-779

16. Marconi NF, Almeida GL, Gottlieb GL. **Electromyographic and kinetic strategies to control movements.** Brazilian journal of physical therapy. 2006 10;1-8.

17. Latash ML, Corcos DM. **Kinematic and electromyographic characteristics of single-joint movements of individuals with Down syndrome.** Am J Ment Retard. 1991; 96: 189-201.

18. Almeida GL, Corcos DM, Latash ML. **Practice and transfer effects during fast single-joint elbow movements in individuals with Down syndrome.** Phys Ther. 1994; 74: 1000-1016.

19. Almeida GL, Hasan Z, Corcos DM. **Horizontal-plane arm movements with direction reversals performed by normal individuals and individuals with Down syndrome.** J Neurophysiol. 2000; 84: 1949-1960.

20. Aruin AS, Almeida GL, Latash ML. **Organization of a simple two-joint synergy in individuals with Down syndrome.** Am J Ment Retard. 1996; 101: 256-268.

21. Aruin, AS, Almeida GL. **A co-activation strategy in antecipatory postural adjustments in persons with Down syndrome. Motor Control** 1997; 01: 178-191.

22. Latash ML, Anson JG. **What are "normal movements" in atypical populations**. Behav Brain Sci. 1996; 19: 55-68.

23. Marconi NF. **Controle motor de movimentos de reversão em indivíduos neurologicamente normais e portadores da síndrome de Down:** O efeito do feedback intrínseco [dissertação]. Campinas (SP): Universidade Estadual de Campinas; 2000.

24. Ferreira SMS. **Modulação da Latência da Musculatura Antagonista em Indivíduos "neurologicamente normais" e portadores da Síndrome de Down** [dissertação]. Rio Claro (SP): Universidade Estadual Paulista; 2000.

25. Lorenzo SM, Braccialli LMP, Araújo RCT. **Realidade virtual como intervenção na Síndrome de down: uma perpectiva de ação na interface educação e saúde**. Rev Bras Educ Espc. 2015; 21: 392-396.

26. Wuang YP, Chiang CS, Su CY, Wang CC. **Effectiveness of virtual reality using Wii gaming technology in children with Down syndrome.** Res Dev Disabil. 2011; 32: 312–321.

27. Lin HC, WuangYP. **Strength and agility training in adolescents with Down syndrome: A randomized controlled trial**. Res Dev Disabil. 2012; 33: 2236–2244.

28. Mello BCC, Ramalho TF. **Use of virtual reality in the physical therapeutic treatment of individuals with Down syndrome.** Rev Neurocienc. 2015; 23: 143-149.

29. Moura RFC, Santos C, Grecco LCA, Albertini G, Cimolin V, Galli M, Oliveira C **Effects of a single session of transcranial direct current stimulation on upper limb movements in children with cerebral palsy: A randomized, sham controlled study**. Developmental Neurorehabilitation. 2017. 19:18.

30. Lazzari, Roberta Delasta; Politti, Fabiano; Santos, Cibele Almeida; et al. **Effect of a single session of transcranial direct-current stimulation combined with virtual reality training on the balance of children with cerebral palsy: a randomized, controlled, double-blind trial**. Journal of Physical Therapy Science. 2015.763-768.

31. Stagg CJ, Bachtiar V, O'Shea J, Allman C, Bosnell RA, Kischka U, Matthews PM, Johansen-Berg H. **Cortical activation changes underlying stimulation induced behavioral gains in chronic stroke. Brain**. 2012; 135:276-84.

32. Miranda PC, Lomarev M, Hallett M. **Modeling the current distribution during transcranial direct current stimulation.** Clin Neurophysiol. 2006; 117(7):1623-9.

33. Wagner T, Fregni F, Fecteau S, Grodzinsky A, Zahn M, Pascual-Leone **A. Transcranial direct current stimulation: A computer-based human model study.** Neuroimage 2007; 35:1113-24.

34. Liebetanz D, Nitsche MA, Teragau F, Paulus W. **Pharmacological approacha to the mechanisms of transcranial DC- stimulation- induced after-effects of humam motor cortex excitability.** Brain 2002; 125: 2238-47.

35. Kuo MF, Unger M, Liebetanz D, Lang N, Tergau F, Paulus W, Nittshe MA. **Limited impact of homeostatic plasticity on motor learning in humans.** Neuropsychologia. 2008; 46: 2122-8.

36. Monte-SilvaK,Kuo M-F, Thirugnanasambandam N,Liebetanz D, Paulus W, Nitsche MA. **Dose- dependente inverted U-shaped effect of dopamine (D2-like) receptor activation on focal and nonfocal plasticity in humans**. The Journal of Neuroscience. 2009; 29 (19):6124-31.

37. Lazzari RD, Politti F, Belina ST, Grecco LAC, Santos CA, Dumont AJL et al. **Effect of Transcranial Direct Current Stimulation Combined With Virtual Reality Training on Balance in Children With Cerebral Palsy: A Randomized, Controlled, Double-Blind, Clinical Trial.** Journal of Motor Behavior. 2016. 1940-1027.

38. Grecco LA1, Duarte Nde A, de Mendonça ME, Pasini H, Lima VL, Franco RC, de Oliveira LV, de Carvalho Pde T, Corrêa JC, Collange NZ, Sampaio LM, Galli M, Fregni F, Oliveira CS. et al. **Effect of transcranial direct current stimulation combined with gait and mobility training on functionality in children with cerebral palsy: study protocol for a double-blind randomized controlled clinical trial**. BMC Pediatr. 2013:168.

39. Dumont AJL, Araujo M, Lazzari RD, Santos CA, Carvalho DB, Moura RCF. **Effects of a single session of transcranial direct current stimulation on static balance in a patient with hemiparesis: a case study**. J. Phys. Ther. Sci. 2015. 27: 955–958.

40. Nitsche MA, Liebetanz D, Schlitterlau a, Henschke U, Friche K, Frommann K, et al. **GABAergic modulation of DC stimulation-induced motor cortex excitability shifts in humans.** Eur J Neurosco. 2004;19(10): 2720-6.

41. Duarte NDA, Grecco LAC, Galli M, Fregni F, Oliveira C. **Effect of Transcranial Direct-Current Stimulation Combined with Treadmill Training on Balance and Functional Performance in Children with Cerebral Palsy: A Double-Blind Randomized Controlled Trial.** PLOS ONE. 2014. 9;8

42. Grecco LAC, Duarte NDA, Zanon N, Galli M, Fregni F, Oliveira C. **Effect of a single session of transcranial direct-current stimulation on balance and spatiotemporal gait variables in children with cerebral palsy: A randomized sham-controlled study**. Braz J Phys Ther. 2014; 18: 419-427.

43. Mendonça ME, Fregni F**.Neuromodulação com estimulação cerebral não invasiva: aplicação no acidente vascular encefálico, doença de Parkinson e dor crônica. In.:ASSIS, R.D. Condutas práticas em fisioterapia neurológica**. Manole. São Paulo, p. 307-39, 2012.

44. Fregni F, Gimenes R, Valle AC, Ferreira MJ, Rocha RR, Natalle L, Bravo R, Rigonatti SP, Freedman S, Nitsche M, Pascual-Leone A, Boggio PS. **A randomized, sham-controlled, proof of principle study of transcranial direct current stimulation for the treatment of pain in fibromyalgia.** Arthritis and Rheumatism, 2006; 54:3988-98.

45. Fregni F, Bossio PS, Brunoni AR. **Neuromodulação terapêutica: Princípios e avanços da estimulação cerebral não invasiva em neurologia, reabilitação, psiquiatria e neuropsicologia. Sarvier.** São Paulo, 2012.

46. Fritsch B, Reis J, Martinowich K, Schambra HM, Ji Y, Cohen LG, et al. **Direct current stimulation promotes BDNF-dependent synaptic plasticity: potential implications for motor learning. Neuron.** 2010;66(2): 198-204.

47. Reis J, Robertson EM, Krakauer JW, Rothwell J, Marshall L, Gerloff C, et al. **Consensus: Can transcranial direct current stimulation and transcranial magnetic stimulation enhance motor learning and memory formation**. Brain stimulation. 2008;1(4): 363-9.

48. Antal A, Lang N, Boros K, Nitsche M, Siebner HR, Paulus W. **Homeostatic metaplasticity of the motor cortex is altered during headache-free intervals in migraine with aura. Cerebral cortex.** 2008;18(11):2701-5.

49. Cimolin V, Beretta E, Piccinini L, Turconi AC, Galli M, Strazzer S. **Constraint-induced movement therapy for children with hemiplegia after traumatic brain injury: a quantitative study.** J Head Trauma Rehabil. 2012; 27(3): 177-87.

50. Menegoni F, Milano E, Trotti C, Galli M, Bigoni M, Baudo S, Mauro A. **Quantitative evaluation of functional limitation of upper limb movements in subjects affected by ataxia.** Eur J Neurol. 2009; 16(2): 232-9.

51. Petuskey K, Bagley A, Abdala E, James MA, Rab G**. Upper extremity kinematics during functional activities: three-dimensional studies in a normal pediatric population.** Gait Posture. 2007; 25(4): 573-9.

52. Hermes JH, Freriks B, Merletti R, Steggeman D, Blok J, Rau G, Disselhorst-Klug C, Hagg G: **SENIAM 8: Surface Electromyography for the Non-Invasive Assessment of Muscles.** Roessingh Research and Development 1999.

53. Rab G, Petuskey K, Bagley A. **A method for determination of upper extremity kinematics.** Gait Posture. 2002; 15(2): 113-9.

54. Jasper HH. **The ten-twenty electrode system of the International Federation electroencephalogria.** Clin Neurophysiol. 1958; 10: 371-375.

55. Homan RW, Herman J, Purdy P. **Cerebral location of international 10-20 system electrode placement.** Electroencephalogr Clin Neurophysiol. 1987; 66(4): 376-82.

56. FELL, J.; AXMACHER, N. **The role of phase synchronization in memory processes.** Nature Reviews Neuroscience, v. 12, n. 2, p. 105–118, fev. 2011

57. Haley S, Coster W, Ludlow L. **Inventário de avaliação pediátrica de disfunção: versão brasileira. Tradução e adaptação cultural**: Mancini M C. Belo Horizonte: Laboratórios de Atividade e Desenvolvimento Infantil, Departamento de Terapia Ocupacional, Universidade Federal de Minas Gerais. 2000.

58. Feldman AB, Haley SM, Corvell J**. Concurrent and construct validity of the Pediatric Evaluation of Disability Inventory**. Phys. Ther. 1990; 70(10): 602-10.

59. Cruz MBZ, WISC III: **Escala de Inteligência Wechsler para crianças: Manual. Periodicos eletrônicos em psicologia**. 2005; 4: 309.

60. Grecco LA, de Almeida Carvalho Duarte N, Mendonça ME, Cimolin V, Galli M, Fregni F, Santos Oliveira C. **Transcranial direct current stimulation during treadmill training in children with cerebral palsy: a randomized controlled double-blind clinical trial.** Research in Developmental Disabilities 2014;3 (11):2840–2848.

61. Gillick T, Feyma T, Menk J, Usset M, Vaith A, Wood J, Worthing R et al., **Safety and feasibility of transcranial direct current stimulationin pediatric hemiparesis: randomized controlled preliminarystudy.** Physical Therapy 2015;95(3):337–349.

62. Grecco et al. **Cerebellar transcranial direct current stimulation in children with ataxic cerebral palsy: a sham-controlled, crossover,pilot study.** Developmental Neurorehabilitation 2016;22:1–7

63. Naseri P, Nitsche MA e Ekhtiar H. **A framework for categorizing electrode montages in transcranial direct current stimulation.** Front Hum Neurosci. 2015, 6;9:54.

64. Boggio P. S., Ferruci R., Mameli F., Martins D., Martins O., Vergari M., et al. . (2012). **Prolonged visual memory enhancement after direct current stimulation in Alzheimer's disease. Brain Stimul**. 5, 223–230. 10.1016/j.brs.2011.06.006 [PubMed] [Cross Ref]

65. Lapenta O. M., Fregni F., Oberman L. M., Boggio P. S. (2012). **Bilateral temporal cortex transcranial direct current stimulation worsens male performance in a multisensory integration task.** Neurosci. Lett. 527, 105–109. 10.1016/j.neulet.2012.08.076 [PubMed] [Cross Ref]

66. Chamovitz YS e Weiss PL **Virtual reality as a leisure activity for young adults with physical and intellectual disabilities** Science Direct. 2008;29:273- 287

67. Wuang PY, Chiang SC, Su YC et al**., Effectiveness of virtual reality using Wii gaming technology in children with Down syndrome Science Direct.** 2011; 32: 312-321

68. Lin HC e Wuang PY **Strength and agility training in adolescents with Down syndrome: A randomized controlled trial**. Science Direct 2012;33:2236-2244.

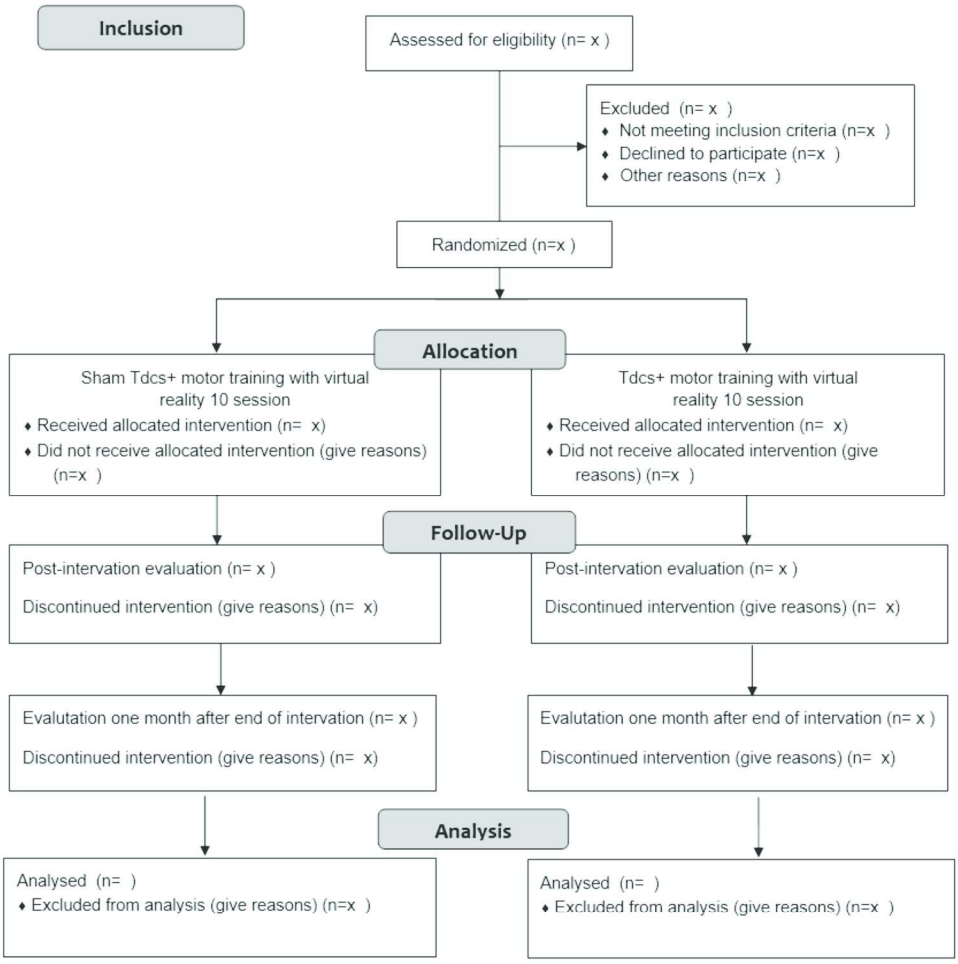

Figure 1: Flowchart of study following CONSORT statement

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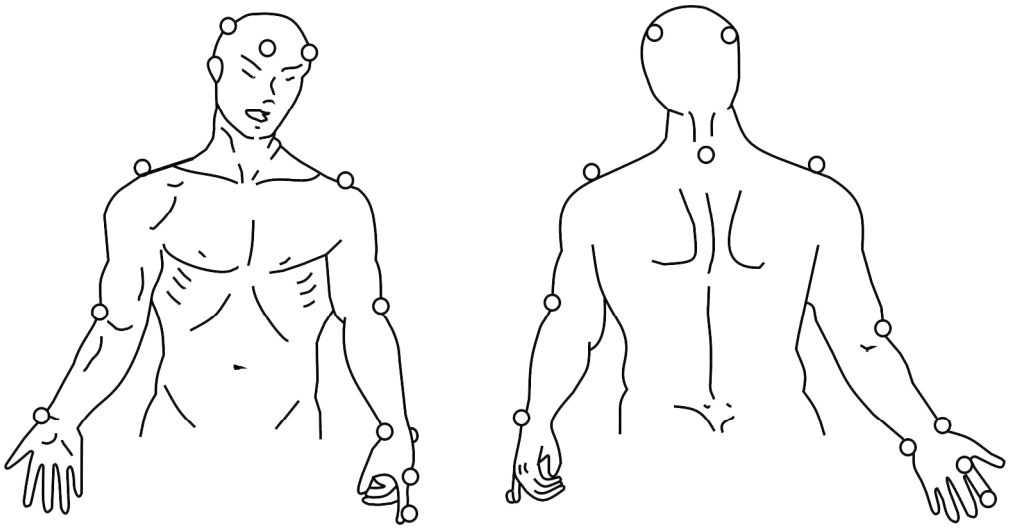

Figure 2: Placement of markers for three-dimensional analysis using SMARTup: The experimental setup

287x149mm (300 x 300 DPI)

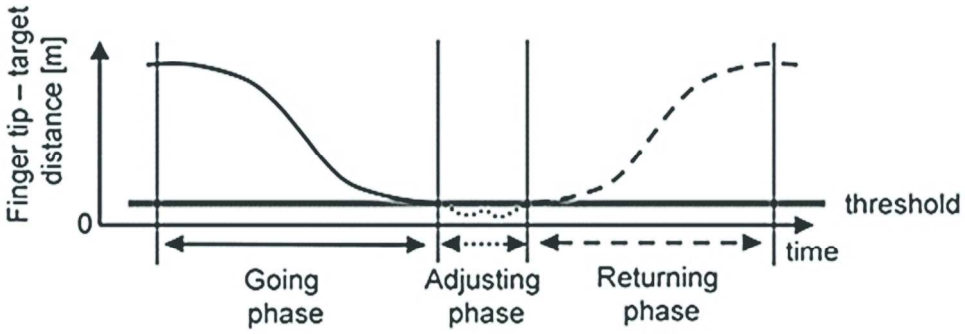

Figure 3: Phases of reaching cycle

294x137mm (300 x 300 DPI)

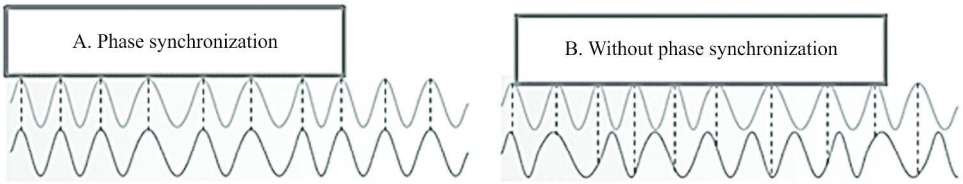

Figure 4 – Phase relationships. (A) synchronized signals – differences in phases between both signals are stable (constant); (B) non-synchronized signals – differences in phases are variable

280x95mm (300 x 300 DPI)

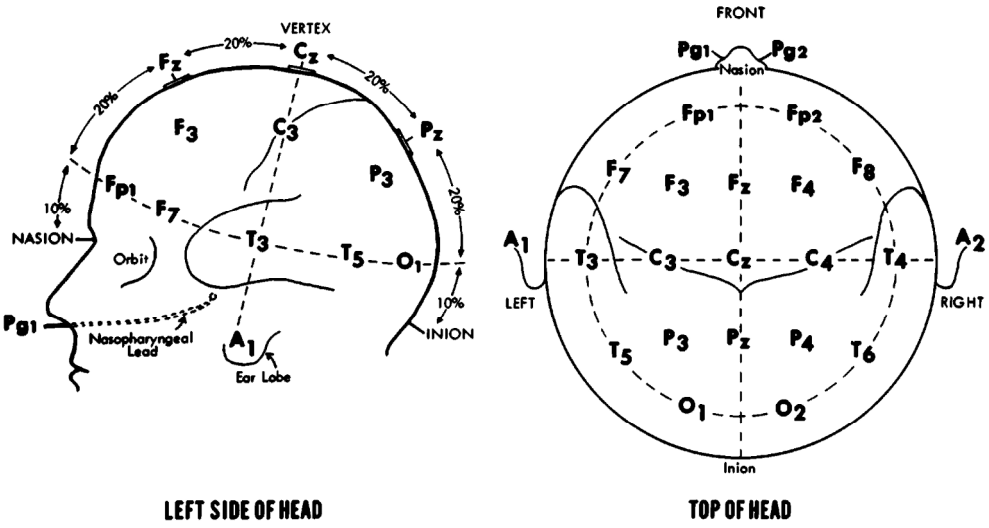

Figure 5 – Positioning of EEG electrodes following 10-20 standard

355x188mm (300 x 300 DPI)

APPENDIX 1

Termo de Consentimento para Participação em Pesquisa Clínica

Nome do Voluntário: _____

Endereço:_____

Telefone para contato:_____Cidade:_____CEP:_____

E mail: _____

1. As informações contidas neste prontuário foram fornecidas pela aluna Jamile Benite Palma Lopes (Mestranda da Universidade Nove de Julho), Profª. Claudia Santos Oliveira, objetivando firmar acordo escrito mediante o qual, o voluntário da pesquisa autoriza sua participação com pleno conhecimento da natureza dos procedimentos e riscos a que se submeterá, com a capacidade de livre arbítrio e sem qualquer coação.

2. Título do Trabalho Experimental: Realidade virtual e estimulação transcraniana por corrente contínua anódica para melhora da função motora de membros superiores em crianças com síndrome de down: ensaio clínico controlado aleatorizado e duplo cego.

3. Objetivo: Examinar os efeitos da estimulação por corrente sobre o controle motor, atividade dos músculos, atividade do cérebro e independência funcional de crianças com Síndrome de Down.

4. Justificativa: acredita-se que ao aplicar a estimulação por corrente, especificamente, durante o treino motor com uso de um vídeo game, será possível, otimizar a atividade do cérebro e a melhora motora.

5. Procedimentos da Fase Experimental: Será selecionas crianças diagnosticadas com Síndrome de Down, com capacidade de entendimento e colaboração para realização dos procedimentos envolvidos no estudo, crianças com idade entre seis e 12 anos, crianças com queixas de comprometimento Na coordenação motora dos braços. O processo de avaliação (antes, após e um mês após o treino, será realizado em três dias não consecutivos, mas na mesma semana, com período máximo de uma hora e 30 minutos por dia. A avaliação será constituída dos seguintes itens: (1) Analise de movimento dos braços durante uma tarefa: avaliado pela cinemática, eletromiografia e eletroencefalograma, a criança realizara uma tarefa com os braços e ao mesmo tempo será avaliada pelos aparelhos, sendo acompanhada pelo fisioterapeuta responsável e pelos assistentes (2) PEDI o PEDI é um questionário aplicado no formato de entrevista estruturada com um dos cuidadores da criança, que possa informar sobre seu desempenho em atividades e tarefas típicas da rotina diária. O teste é composto de três partes: a primeira avalia habilidades de repertório da criança agrupadas segundo três áreas

funcionais: autocuidado (73 itens), mobilidade (59 itens) e função social (65 itens). Cada item dessa parte é pontuado com escore 0 (zero) se a criança não é capaz de desempenhar a atividade, ou 1 (um), se a atividade fizer parte de seu repertório de habilidades. O Grupo 1 terá o movimento do braço analisado após realizar treino com o vídeo game junto com a estimulação desligada (placebo). O Grupo 2 terá o movimento do braço analisado após realizar treino com o vídeo game junto com a estimulação ligada. A estimulação por corrente é uma técnica não invasiva que será realizada colocando eletrodos de superfície conectados a um aparelho de corrente galvânica (corrente elétrica de baixa intensidade) sobre o crânio (cabeça) da criança, durante 20 minutos por 15 dias. A estimulação é indolor.

6. Desconforto ou Risco Esperado: Embora os procedimentos adotados no estudo sejam não-invasivos os voluntários serão submetidos a risco como por exemplo, quedas, fadiga muscular, câimbras durante o treino motor de realidade virtual. Para que estes riscos sejam minimizados ao máximo serão adotadas as seguintes medidas protetoras: A estimulação será realizada por uma fisioterapeuta com experiência na técnica. No treino de realidade virtual serão realizados por uma fisioterapeuta com experiência em treino motor que será acompanhada por ao menos um voluntário ambos permanecerão posicionados do lado do paciente por todo o treino.

7. Informações: o voluntário tem garantia que receberá respostas a qualquer pergunta ou esclarecimento de qualquer dúvida quanto aos procedimentos, riscos benefícios e outros assuntos relacionados com pesquisa. Também os pesquisadores supracitados assumem o compromisso de proporcionar informação atualizada obtida durante o estudo, ainda que esta possa afetar a vontade do indivíduo em continuar participando.

8. Retirada do Consentimento: o voluntário tem a liberdade de retirar seu consentimento a qualquer momento e deixar de participar do estudo, sem que isto lhe traga qualquer prejuízo.

9. Aspecto Legal: Elaborados de acordo com as diretrizes e normas regulamentadas de pesquisa envolvendo seres humanos atendendo à Resolução nº. 466/12 do Conselho Nacional de Saúde do Ministério de Saúde – Brasília – DF.

10. Garantia de Sigilo: Os pesquisadores asseguram a privacidade dos voluntários quanto aos dados confidenciais envolvidos na pesquisa.

11. Formas de ressarcimento das despesas decorrentes da participação na pesquisa: Se necessário, será dado aos pesquisados auxilio transporte de ida e volta ao local da pesquisa. Não será dada ao pesquisado qualquer tipo de remuneração e auxilio de custo pela participação na pesquisa. Pelo curto tempo das avaliações e intervenções não haverá fornecimento de alimentação ao pesquisado.

12. Local da Pesquisa: A pesquisa será desenvolvida no Laboratório Integrado de Análise do Movimento Humano - LIAMH e Núcleo de Apoio a Pesquisa na Analise do Movimento - NAPAM, Universidade Nove de Julho UNINOVE, localizada na rua Vergueiro, no 235/249, 2º subsolo, Vergueiro, São Paulo - SP.

13. Comitê de Ética em Pesquisa (CEP) é um colegiado interdisciplinar e independente, que deve existir nas instituições que realizam pesquisas envolvendo seres humanos no Brasil, criado para defender os interesses dos participantes de pesquisas em sua integridade e dignidade e para contribuir no desenvolvimento das pesquisas dentro dos padrões éticos (Normas e Diretrizes Regulamentadoras da Pesquisa envolvendo Seres Humanos – Res. CNS nº 466/12). O Comitê de Ética é responsável pela avaliação e acompanhamento dos protocolos de pesquisa no que corresponde aos aspectos éticos.

Endereço do Comitê de Ética da Uninove: Rua. Vergueiro nº 235/249 – 3º subsolo - Liberdade – São Paulo – SP CEP. 01504-001 Fone: 3385-9197 . comitedeetica@uninove.br

14. Nome Completo e telefones dos pesquisadores para contato: Orientadora: Claudia Santos Oliveira (11 3665 9344) e aluno de pós graduação: Jamile Benite Palma Lopes (11) 975123549.

15. Eventuais intercorrências que vierem a surgir no decorrer da pesquisa poderão ser discutidas pelos meios próprios.

16. Consentimento Pós-Informação:

Eu,_____, após leitura e compreensão deste termo de informação e consentimento, entendo que minha participação é voluntária, e que posso sair a qualquer momento do estudo, sem prejuízo algum. Confirmo que recebi cópia deste termo de consentimento, e autorizo a execução do trabalho de pesquisa e a divulgação dos dados obtidos neste estudo no meio científico.

* Não assine este termo se ainda tiver alguma dúvida a respeito. São Paulo, de de 2016.

Nome (por extenso) do pesquisado:

_____

Assinatura pesquisado:

_____

Nome (por extenso) do pesquisado:

_____

Assinatura pesquisado:

_____

APPENDIX 2

Approval of the Ethics Committee

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PARECER CONSUBSTANCIADO DO CEP

DADOS DO PROJETO DE PESQUISA

Título da Pesquisa: REALIDADE VIRTUAL E ESTIMULAÇÃO TRANSCRANIANA POR CORRENTE CONTÍNUA PARA MELHORA DA FUNÇÃO MOTORA DE MEMBROS SUPERIROES EM CRIANÇAS COM SÍNDROME DE DOWN: ENSAIO CLÍNICO CONTROLADO ALEATORIZADO E DUPLO CEGO

Pesquisador: Jamile Benite Palma Lopes

Área Temática:

Versão: 1

CAAE: 55196516.0.0000.5511

Instituição Proponente: ASSOCIACAO EDUCACIONAL NOVE DE JULHO

Patrocinador Principal: Financiamento Próprio

DADOS DO PARECER

Número do Parecer: 1.517.470

Apresentação do Projeto:

O projeto chama a atenção para as dificuldades motoras apresentadas por crianças com síndrome de down, e sugere uma intervenção que possibilite uma melhora no controle motor dessa população.

Objetivo da Pesquisa:

Verificar os efeitos da estimulação transcraniana por corrente contínua (tDCS) associado a um treino neuromotor em membro superior com realidade virtual sobre a atividade cerebral e controle motor em crianças com Síndrome de Down (SD).

Avaliação dos Riscos e Benefícios:

Embora os procedimentos adotados no estudo sejam não-invasivos os voluntários serão submetidos a risco como por exemplo, quedas, fadiga muscular, câimbras durante o treino motor de realidade virtual. Para que estes riscos sejam minimizados ao máximo serão adotadas as seguintes medidas protetoras: A estimulação transcraniana será realizada por uma fisioterapeuta com experiência na técnica. No treino de realidade virtual serão realizados por uma fisioterapeuta com experiência em treino motor que será acompanhada por ao menos um voluntário ambos

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permanecerão posicionados do lado do paciente por todo o treino.

Comentários e Considerações sobre a Pesquisa:

O projeto apresenta as características éticas necessárias para realização da pesquisa.

Considerações sobre os Termos de apresentação obrigatória:

Foram apresentados os termos exigidos para aprovação da pesquisa.

Considerando que tratam-se de crianças com Síndrome de Down, não é necessário o TCLE específico para menores.

Recomendações:

O TCLE, embora apresente os aspectos exigidos, ele deve ser reescrito, uma vez que se encontra com uma linguagem estritamente técnica sobre os procedimentos que serão realizados na pesquisas, assim como constitui-se num documento muito longo (com 6 páginas).

Conclusões ou Pendências e Lista de Inadequações:

Recomenda-se reescrever o TCLE, adequando a linguagem (mais simples e acessível ao responsável pela criança participante)

Resumir as informações tornando o TCLE mais simplificado e objetivo.

Este parecer foi elaborado baseado nos documentos abaixo relacionados:

Tipo Documento	Arquivo	Postagem	Autor	Situação
Informações Básicas do Projeto	PB_INFORMAÇÕES_BÁSICAS_DO_PROJETO_693482.pdf	12/04/2016 15:41:42		Aceito
Folha de Rosto	DOC120416.pdf	12/04/2016 15:40:49	Jamile Benite Palma Lopes	Aceito
Projeto Detalhado / Brochura Investigador	projetofinal.pdf	06/04/2016 16:57:04	Jamile Benite Palma Lopes	Aceito
TCLE / Termos de Assentimento / Justificativa de Ausência	Termo.pdf	06/04/2016 16:46:17	Jamile Benite Palma Lopes	Aceito
Cronograma	cronograma.pdf	06/04/2016 16:45:48	Jamile Benite Palma Lopes	Aceito

Situação do Parecer:

Pendente

Necessita Apreciação da CONEP:

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Telefone: (11)3385-9197 **E-mail:** comitedeetica@uninove.br

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Continuação do Parecer: 1.517.470

Não

SAO PAULO, 27 de Abril de 2016

Assinado por:
Stella Regina Zamuner
(Coordenador)

SPIRIT
STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	01
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	03-08-14
	2b	All items from the World Health Organization Trial Registration Data Set	14
Protocol version	3	Date and version identifier	NA
Funding	4	Sources and types of financial, material, and other support	15
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	01-02-03
	5b	Name and contact information for the trial sponsor	01-02-14
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	08-14
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	14

Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	05-06
	6b	Explanation for choice of comparators	05-06
Objectives	7	Specific objectives or hypotheses	07
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	07-08
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	08
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	07
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	12-13-14
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	NA
n	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	NA
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	NA
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	12-13
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	08-09 -10

Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	08
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	08
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	08
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	08
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	08
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	08
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	08
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	08-14
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	08-09

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	14
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	14
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	NA
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	NA
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	NA
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	NA
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	09
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	NA
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	13
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	NA

Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	13
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	NA
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	13
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	03-14
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	14
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	NA
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	13
	31b	Authorship eligibility guidelines and any intended use of professional writers	14
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	NA
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	13
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	NA

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.