BMJ Open

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Journal:	*BMJ Open*
Manuscript ID	bmjopen-2018-025219
Article Type:	Research
Date Submitted by the Author:	06-Jul-2018
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Keywords:	Magnetic resonance imaging < RADIOLOGY & IMAGING, Spinal manipulation, Neck pain, Vertebral artery dissection, Blood flow

A Cross-over Randomized Controlled Trial of the Effect of Cervical Manipulation on Vertebral Artery and Cerebral Hemodynamics in Patients with Chronic Neck Pain

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Keywords: fMRI$_1$, BOLD fMRI$_2$, Neck Pain$_3$, Vertebral Artery$_4$, Spinal Manipulation$_5$, Chronic neck pain$_6$, Vertebral Artery Dissection$_7$, Stroke$_8$

Summary

Objective

It is hypothesized that cervical manipulation may increase the risk of cerebrovascular accidents. We aimed to determine whether cervical spine manipulation is associated with changes in vertebral artery and cerebrovascular hemodynamics measured with magnetic resonance imaging compared to neutral neck position and maximum neck rotation in patients with chronic neck pain.

Setting

The Imaging Research Centre at St. Joseph's Hospital in Hamilton, Ontario, Canada.

Participants

Twenty patients were included. The mean age was 32 years (SD ± 12.5), mean neck pain duration was 5.3 years (SD ± 5.7) and mean Neck Disability Index score was 13/50 (SD ± 6.4).

Interventions

Following baseline measurement of cerebrovascular hemodynamics, we randomized participants to: 1) maximal neck rotation followed by cervical manipulation; or 2) cervical manipulation followed by maximal neck rotation. The primary outcome, vertebral arteries and cerebral hemodynamics, was measured after each intervention and was obtained by measuring 3D T1-weighted high resolution anatomical images, arterial spin labeling (ASL) and phase contrast flow encoded MRI. Our secondary outcome was functional connectivity within the default mode network (DMN) measured with resting state functional MRI (fMRI).

Results

Compared to the neck in neutral position, we found no significant change in blood flow within the cerebrum nor in blood flow and velocity within the vertebral arteries following cervical manipulation or maximal neck rotation. However, we observed significant increases in functional connectivity in the posterior cerebrum and cerebellum (resting state MRI) after manipulation and maximum rotation.

Conclusion

Our study suggests that cervical manipulation does not result in vertebral artery blood flow or brain perfusion changes compared to a neutral neck position or maximal neck rotation. This finding suggests that cervical manipulation may not increase the risk of cerebrovascular events through a hemodynamic mechanism.

Trial Registration

US National Institute of Health # NCT02667821.

Word Count: 4,104

Strengths and limitations of this study

- A strength of the study was the design, which insured control of confounders and provided statistical efficiency.
- We used what is considered the criterion standard for both diagnosis of vertebrobasilar artery stroke and quantifying blood flow (phase-contrast MRI) because of its greater sensitivity compared to ultrasonography.
- A limitation of the study was the restriction of analysis to a time following the test maneuvers. Real time measures currently are technically not feasible and transient effects of various neck positions on vertebral artery and cerebrovascular hemodynamics may have been missed.

INTRODUCTION

Anecdotal evidence from case reports and case-series suggest that chiropractic neck manipulation increases the risk of vertebrobasilar artery stroke.[1-3] However, the epidemiological evidence does not support this hypothesis.[4, 5] In their case-crossover study, Cassidy *et al.* found that the risk of vertebrobasilar artery stroke was similar for patients with neck pain or headaches who consult physicians and those who consult chiropractors.[4] This suggests that the hypothesized association is due to protopathic bias.

Understanding whether chiropractic neck manipulation increases the risk of stroke is important because patients with neck pain frequently consult chiropractors.[6-10] Dissection of the vertebral artery is involved in most cases that implicate cervical manipulation.[11] However, when damage to the vertebral artery is absent, vasospasm[12-15] and 'subclinical' endothelial injury have also been hypothesized to be causes of stroke. According to the vasospasm hypothesis, placing the head in rotation and hyperextension during a manipulation leads to considerable stress and stretch forces in the vertebral artery, specifically in the suboccipital portion.[16, 17] This mechanical compression or stretching of the vertebral artery may lead to changes in blood flow and the 'subclinical' injury to the vertebral artery can lead to thrombosis.[18]

Several studies have investigated changes in blood flow during cervical spine motion.[19-27] Most studies report a decrease in vertebral artery flow contralateral to the side of rotation.[19-22, 24-27] Less is understood about blood flow during and after a cervical manipulation, but two studies found no significant changes in vertebral artery blood flow or blood velocity following cervical manipulation in healthy individuals.[28, 29] However, the impact of cervical manipulation on vertebral artery blood flow in the population likely to undergo this maneuver for therapeutic purposes is unknown. Neck pain patients may differ from that of healthy populations because moderate to severe perceived neck disability, as measured with the Neck Disability Index (NDI) is correlated with cortical hypoperfusion.[30]

Therefore, our primary aim was to determine whether cervical manipulation leads to a clinically meaningful change in vertebral and cerebral hemodynamics compared to neutral neck position or neck rotation in adult patients with chronic neck pain. Our secondary aim was to compare the functional connectivity within the default mode network (DMN) between neutral neck position, neck rotation and cervical manipulation.

METHODS

Study Design

We conducted a case-crossover randomized controlled study to compare the effects various head positions, including cervical manipulation on cerebrovascular hemodynamics. No washout period was used between each intervention. It was assumed that the time needed to measure the blood hemodynamics allowed enough time to return to their baseline status.[31] The study was registered with www.clinicaltrials.gov, NCT02667821. The McMaster University Hamilton Integrated Research Ethics Board (HiREB) (REB#1303) and Canadian Memorial Chiropractic College Research Ethics Board approved the study (REB#1604X01).

Participants

Patients who were eligible for the study were attending a teaching clinic of the Canadian Memorial Chiropractic College, Toronto, Canada between September 2016 and April 2017. We recruited participants via poster advertising displayed at the teaching clinic, word of mouth, and referrals from the supervising clinicians at the clinic. To be included, patients had to meet the following criteria: 1) at least 18 years old; 2) chronic neck pain (≥ 3 months' duration); 3) grade I-II neck pain[32], defined as neck pain with no signs or symptoms of major structural pathology, which may or may not interfere with activities of daily living; 4) prescribed cervical manipulation by the clinician at the teaching clinic supervising their care; and 5) provide written informed consent. Exclusion criteria were: a history of neck pain with associated arm pain within the last 6 months; any current or history of neurologic symptoms including facial or extremity weakness, abnormal sensation to the face, body, or extremities, uncontrolled movements, abnormal gait, dizziness, unexplained nausea/vomiting, difficulty with speaking or swallowing; history of new or severe (Visual Analogue Scale >6/10) headaches in the last 3 months; any contraindications to magnetic resonance imaging (MRI); or any history of using drugs that affect blood flow such as Warfarin, or anti-coagulants. In addition, all participants refrained from vigorous physical activity and ingesting alcohol and caffeine one day before their scheduled participation.

Randomization and masking

We used simple randomization to allocate participants to one of two sequences of interventions: 1) maximal neck rotation followed by cervical manipulation; or 2) cervical manipulation followed by maximal neck rotation. The study coordinator (NM) conducted the randomization using a randomized table generator (GraphPad Software Inc, La Jolla, CA). The random allocation was communicated verbally to the study clinician (SM) on the day of the study protocol. Randomization was concealed, no other study personnel or participants were aware of the intervention assignments.

Procedures

Prior to commencement of the study protocol, participants underwent a cervical spine examination by the clinician (SM) performing the test manoeuvres to identify the site of manipulation. Baseline information on each participant was collected and included: age, gender, height, weight, NDI score, neck pain intensity, duration of neck complaint and headache pain intensity (Table 1).

Initial intervention was a baseline MRI of the upper cervical spine and brain with the neck in the neutral position. Neutral neck position was defined by alignment of the Frankfort plane in a vertical orientation. For continuity of neutral alignment during imaging between test conditions, the MRI laser land-marking tool was used to triangulate between three oil-based markers (Vitamin E capsules) taped to the nasion (bridge of the nose) and immediately in front of the tragus of the ears, bilaterally.

Following random allocation, maximal neck rotation or cervical manipulation were performed to the side of clinical symptoms as elicited during the cervical spine examination. Maximal neck rotation was achieved by instructing participants to rotate their head as far as comfortably possible. The clinician performing the interventions assisted the rotation via a soft hand contact on the patient's head. The degree of maximal neck rotation was measured by an inclinometer and the position was held for one minute before returning to neutral neck position for MRI sequencing. The cervical manipulation procedure was a high velocity, low amplitude (HVLA) impulse, with the participant's head in combined axial rotation, flexion and lateral flexion postures. Variations of head positions between operators for this procedure have been demonstrated to be relatively small.[33, 34] A practitioner with more than 30 years of practice experience conducted the cervical manipulation (SM).[35-37] The manipulation procedure was performed on the adjustable and pivotal MRI bed in the MRI room with the participant in the supine position. The clinician performed the procedure by first establishing the end range of motion to determine appropriate preload position for the manipulation before applying a clinical force impulse in the coronal plane with minimal traction component.

Before each maneuver, the participants were queried on their comfort, condition, and willingness to continue. The participant's head was repositioned to neutral immediately after each maneuver, and then retracted into the MRI bore. Each maneuver was carried out on the scanner bed in the MRI room.

Outcomes

The primary outcome measure was cerebrovascular hemodynamics within the vertebral arteries and posterior cerebrum measured with MRI. The MRI data was acquired using a 3Telsa MR750 scanner and 20-channel neurovascular array radiofrequency coil (General Electric (GE) Healthcare, Milwaukee WI).

The primary outcome was measured following a standardized protocol. First, the baseline MRI was performed. The head was immobilized with sponges in a neutral neck position and a localizer scan was completed. Next, high-resolution anatomical images were collected using a 3D inversion recovery (IR) prepped fast spoiled gradient recalled echo (fSPGR) T1-weighted scan (repetition time [TR]/echo time [TE]=11.4/4.3ms, TI = 450ms, flip angle = 12°, 512 x 256 matrix, 140 slices, 24 cm field of view [FOV], reconstructed to $1mm^3$ isotropic voxels). Following the anatomical scan, microvascular perfusion was assessed using a 3D spiral-based fast spin echo pseudo-continuous arterial spin labeling (pCASL) (TE/TR/TI=10.5/4629/1525ms, FOV=24cm, 512x8spiral interleaves, 3NEX, reconstructed to give an in-plane resolution of 1.875x1.875 (128x128) with 4mm thick slices, scan time =30s). Following arterial spin labeling, a phase contrast scan for vertebral artery flow quantification was performed. At the upper cervical level C1-2, the contralateral and ipsilateral vertebral arteries, defined to the direction of head motion, were assessed and anatomical images were established to localize the vertebral artery circulation. As previously published by Ho *et al.*[38] the method for obtaining flow quantification of the vertebral artery was a 2-dimensional phase-contrast pulse sequence. To capture accurate vertebral artery flow, the imaging plane is ideally perpendicular to the central axis of the blood vessel. This imaging plane was selected on the vessel of interest at the C1-2 intervertebral level based on arterial visualization on a maximum intensity projection (MIP) of a 3D time-of flight MRI angiogram. Acquisition parameters were as follows: fast gradient recalled echo (fGRE); echo time/repeat time=3.9/8.9ms, flip angle = 20^o, 20cm field of view; 512 × 512 matrix; 244-Hz/pixel receiver bandwidth; 1 average; 4 mm thick; and velocity encoding (venc) of 50 cm/s encoded over 30 phases per cardiac cycle. All image measurements were obtained by manually selecting the optimal anatomical site between the base of the odontoid process and approximately 1cm above the tip of the dens. Data acquisition was triggered by peripheral gating using a pulse oximeter, with sequence acquisition time for each flow measurement being approximately 1.5 minutes, depending on heart rate.[38] According to Lotz *et al.* vessel obliquity is tolerable to ± 15°, above which will cause a significant deviation from true flow.[39]

The secondary outcome measure was functional connectivity within the default mode network (DMN). For baseline measurement this was performed immediately following the phase contrast MRI, but prior to manipulation. Resting state functional MRI data (i.e. blood oxygen level dependent, BOLD signal) was acquired using a gradient echo, echo planar imaging (GE-EPI) sequence (64x64 matrix, 28 axial slices (5mm thick, no skip), 24cm FOV, TE/TR/flip angle = 35ms/2000ms/90^o, 180 temporal points, total scan time=6 minutes). During the resting state scan, participants were asked to keep their eyes open, stay awake, and not think of anything in particular.

Following the baseline MRI scan, the image sequence to obtain the primary and secondary outcomes changed to the following: 3-plane localizer, ASL, phase contrast, resting state MRI and 3D anatomical scan. The latter three were always performed with the 3D anatomical last seeing as microvascular changes (i.e. blood flow) were more likely to be detected early after the cervical manipulation and maximal neck rotation, while structural changes (i.e. 3D scan) wouldn't be expected to occur. Participants were scanned a total of 3 times for each session: baseline, and two procedures in random order.

After testing, each participant was observed for one hour and contacted by a study coordinator (NM) within 24 hours of release for follow-up on status. Adverse events, defined as side effects that are harmful, were assessed via open-ended questions.[40] These included local soreness and pain in the area of the applied test maneuvers (minor adverse events), as well as signs of stroke or transient ischemic attack (major adverse events).

Statistical Analysis

Previous research examining reproducibility of cerebral perfusion measurements using ASL suggested that the mean percent perfusion difference was 7.1 (SD ± 12).[41] To date, no study has provided information on cerebral blood flow and perfusion after mechanical challenges to the cervical spine; therefore, we assumed that a change of at least 2 SD from the normal mean flow would be clinically meaningful. Based on this effect size, a power of 80%, a significance level at $p < 0.05$ and two-sided t-test, we calculated that a sample size of 16 participants was necessary.[42] We increased our sample size by 20% (n=20 participants) to account for possible dropouts.

An experienced analyst (MB), who remained blinded to the sequence of test maneuvers, performed the data quantification of the primary and secondary outcomes. Microvascular perfusion and resting state MRI data were analyzed using analysis of functional neuroimaging (AFNI).[43] For each participant all ASL and resting state MRI data were spatially registered to the initial (neutral condition) position. Anatomical, blood flow and functional data were transformed automatically to the Colin27 atlas, using the AFNI command @auto_tlrc, with functional data resampled to a 2mm isometric grid.[44] Temporal band-pass filtering with cut-offs of $0.009\text{Hz} < f < 0.08\text{Hz}$ was performed in order to suppress unwanted physiological signals and some hardware noise.[45] Functional connectivity within the DMN was assessed using the AFNI plugin *InstaCorr*, a seed-based approach, which uses the Pearson method of correlation to compare time signals.[46] The DMN is the most dominant temporally correlated resting network in the awake brain, defined as regions positively correlated in time with the posterior cingulate cortex (PCC) seed voxel. The PCC was defined automatically using the AFNI Talairach method 'Talairach to' and selecting a single voxel from each the left and right PCC for one analysis (ColinN27 coordinates: 10, 54, 14, and -10, 54, 14). These were both subsequently fused in the post-processing. A 5mm FWHM Gaussian spatial smoothing filter was applied for maximize likelihood of overlap with inter-participant group analysis. Also, temporal outliers determined with the AFNI function *3dToutcount* were censored out. Finally, the AFNI plugin 3d*ClustSim* was used to threshold any clusters with fewer than 20 voxels. The ASL cerebral blood flow data was analyzed similar to resting state MRI in that following spatial co-registration to the neutral condition and spatial blurring with a 5mm FWHM Gaussian convolution kernel, ASL data was warped to the N27 atlas. Group analysis was accomplished using a 1-way within-participant 3D-ANOVA, with the one factor being neck position. Post hoc testing included contrasts between neutral, maximum voluntary rotation and cervical manipulation, and also a contrast between maximum voluntary rotation and cervical manipulation. Statistical significance was defined as anything lower than an alpha value of 0.05, with prior cluster thresholding dealing with multiple comparisons.

Flow analysis was performed using Segment v1.9 software 47 (Medviso, Lund, Sweden). Dynamic regions of interest were drawn on the left and right vertebral arteries to quantify the mean, as well as peak velocity and flow. Data in the trigger window portion of the cardiac cycle were derived by spline interpolation using Matlab (Mathworks, Natick, MA). Mean and SDs were calculated for vertebral artery blood velocity, flow, peak velocity, and peak flow for each of the head conditions and vertebral artery side. Differences between task maneuvers and vertebral artery flow and velocity were evaluated using a repeated-measures analysis of variance with factors of head position and vertebral artery side, and a level of significance was set at 0.05, using R-project version 2.12.1 (R Development Core Team, 2010. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org/). Vertebral artery blood flow variability was additionally calculated over the 30 phases per cardiac cycle by examining the flow errors in the left and right vertebral arteries for each of the conditions.

Role of the funding source

The funders of the study had no role in the study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

RESULTS

Between September 2016, and April 2017, a total of 936 participants were screened for the study; 916 failed pre-screening (Figure 1). Most did not meet the inclusion criteria (n= 890); three participants

declined to participate; 23 participants were either unable to acquire transportation to the study location or unable to attend the scheduled time. Twenty participants (14 female, 6 male) aged 23 to 66 years (mean [SD], 32 [± 12.5] years) were enrolled in the study. The average NDI score was 13/50 (SD ± 6.4) with an average neck pain intensity of 5/10 (SD ± 2.1) and a mean duration of neck pain of 5.3 years (SD ± 5.7) (Table 1).

The total time elapsed for each participant testing protocol was 60 minutes. The total time elapsed for each test maneuver was approximately 20 minutes. This included applying the test maneuver (maximum of 1 minute), replacing the participant back into the MRI bore (approximately 1 minute), and image sequencing as noted above (approximately 18 minutes).

When compared to the neutral neck position, maximal neck rotation and cervical manipulation did not significantly alter cerebral perfusion within the posterior cerebrum or cerebellum (Table 2). We found no statistically significant changes in bulk blood flow. Similarly, there were no significant changes in mean vertebral artery blood flow or blood velocity between interventions (Table 3). In addition, no significant changes in mean blood flow or velocity were observed between the ipsilateral and contralateral vertebral artery between the three head positions (Table 3). Finally, we found no significant differences in flow variability within the vertebral arteries between the interventions (Figures I and II in the online only Data Supplement).

We measured a significant ($p<0.05$) increased functional connectivity post-manipulation. More specifically, when compared to the resting position, the following areas showed significantly increased functional connectivity: uvula, cerebellar tonsils, left fusiform gyrus and left middle temporal gyrus, right middle temporal gyrus, right middle occipital gyrus, bilateral cuneus, left precuneus, and left middle occipital gyrus (Table I in the online only Data Supplement).

No major adverse events were reported. One minor adverse event, mild neck soreness, was reported immediately post-procedure by one participant. The participant attributed the soreness to having to lay motionless on the hard scanner bed during study protocol.

Discussion

Our primary objective was to assess the cerebrovascular and vertebral artery blood flow and velocity changes between various head positions including cervical manipulation in patients with chronic neck pain. We found no significant cerebral perfusion changes within the posterior cerebrum or cerebellum nor significant changes in blood flow and velocity within the vertebral arteries at the level of C1-2 between various neck positions. In addition, we found no significant change in blood flow variability between the vertebral arteries as well as between the various head positions. Our findings support previous studies[28, 29] reporting that cervical manipulation does not have a significant effect on these variables when compared to a resting neutral neck position.

No reference values for minimally clinically important differences in the vertebral artery blood flow velocities have been established. However, hemodynamic stenosis has been long considered as a diameter reduction of greater than 50%, which in the vertebral artery has been associated with a peak and end-diastolic flow greater than 108-cm/s and 36-cm/s, respectively.[47] As suggested by Licht *et al.*[48], a change in peak velocity of greater than 25% from baseline would be necessary for clinically relevant decrease in the vertebral artery. No such reductions were observed.

Vertebrobasilar artery stroke secondary to vertebral artery dissection is a rare but devastating occurrence.[49] A unique characteristic of these strokes is that they can develop in healthy adults and they frequently occur in close temporal relationship to benign neck movements[50, 51], cervical manipulation[52] or trivial trauma.[53, 54] Due to the rarity of the condition, very little is known about the risk factors for vertebrobasilar artery stroke. Our study extends the understanding on the effects of cervical manipulation on vertebral artery and cerebrovascular hemodynamics. It is also the first study to directly measure the impact of cervical spinal manipulation on intracranial and extracranial blood flow in a chronic neck pain population. Together with

previous work[28, 29] our results support the position that the association between cervical manipulation and stroke is due to protopathic bias.[4]

When compared to a neutral neck position, both maximum voluntary neck rotation and cervical manipulation resulted in significantly increased functional connectivity throughout the DMN as seen on the BOLD signal. Changes in the BOLD signal arise from complex interactive modulation of blood flow, blood volume and local metabolic rate, all leading to change in the local ratio of oxy- to deoxyhaemoglobin. This ratio drives the change in BOLD signal through magnetic susceptibility differences that exist between oxidation states of the haemoglobin complex. However, when examining the aforementioned areas specifically for changes in blood flow using ASL, no significant changes were observed between any of the test head conditions. This strongly suggests that the observed increased functional connectivity were not a result of altered blood flow but secondary to changes in either blood volume or metabolic activity.

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Our study had strengths. First, our design insured control of confounders and provided statistical efficiency. Second, we used phase-contrast MRI blood flow measurement because of its greater sensitivity compared to ultrasonography (which would not have been able to adequately penetrate through bone to probe the vessels we wanted to measure), and because it is considered the criterion standard for both diagnosis of vertebrobasilar artery stroke and quantifying blood flow.[38, 55-58] Estimates of repeatability of flow measures were made in preliminary work by quantifying vertebral artery flow in a single healthy participant twice over a two-month interval.[28]

A limitation of the study was the restriction of analysis to a time (average 115 seconds) following the test maneuvers. Real time measures currently are technically not feasible and transient effects of various neck positions on vertebral artery and cerebrovascular hemodynamics may have been missed. Post-maneuver analysis makes comparisons with other real-time studies challenging. Thus, the results only describe post-procedural effects and cannot be generalized to the possible effects occurring during the test maneuvers. However, it is notable that to be clinically relevant, sustained changes would likely be required that would extend into the sampled time interval of this study. Moreover, the possibility that neck pain participants exist who exhibit idiosyncratic responses cannot be excluded. To date, no such mechanisms have been measured and reported in the literature.

In conclusion, we found no significant change in blood flow in the posterior cerebrum or cerebellum in chronic neck pain participants after maximum head rotation and cervical manipulation. In addition, we found no significant nor clinically meaningful changes in the blood flow or velocity in the vertebral arteries before-after head positional change and spinal manipulation. Our study adds to a growing body of knowledge regarding the impact of head position and cervical manipulation on vascular and neural activity in patients with neck pain. It is the first study to measure cerebral blood flow, vertebral artery blood flow and velocity in patients undergoing neck manipulation for neck pain. Our study does not support the hypothesis that neck manipulation or neck rotation are associated with vasospasm of the vertebral artery.

Patient and Public Involvement

Patients and public were not involved in the development, design, recruitment and randomization of this study.

Author Contributions

Concept development (provided idea for the research): NM, JT, GW, SM. Design (planned the methods to generate the results): NM, JT, MN, GW. Supervision (provided oversight, responsible for organization and implementation, writing of the manuscript): NM, SM, JT, GW, PC. Data collection/processing (responsible for experiments, patient management, organization, or reporting data): NM, SM, MB, MN. Analysis/interpretation (responsible for statistical analysis, evaluation, and presentation of the results): NM, MB, SM, JT, GW, MN. Literature search (performed the literature search): NM. Writing (responsible for writing a substantive part of the manuscript): NM, SM. Critical review (revised manuscript for intellectual content, this does not relate to spelling and grammar checking): NM, SM, PC, MN, GW, JT.

Competing Interest Statement

Conflicts of interest were reported for this study include the following: Dr. Triano is an occasional lecturer on behalf of NCMIC and CCPA, and Dr. Noseworthy received an honorarium for lecture on behalf of Bayer. No other conflicts were reported.

Funding Statement

Partial financial support was provided by the Canadian Chiropractic Protective Association and NCMIC Research Foundation and Canadian Memorial Chiropractic College.

Data Sharing Statement

No additional data are available.

References

1. Albuquerque FC, Hu YC, Dashti SR, Abla AA, Clark JC, Alkire B, et al. Craniocervical arterial dissections as sequelae of chiropractic manipulation: patterns of injury and management. *J Neurosurg* 2011; **115**(6):1197-205.
2. Ernst E. Manipulation of the cervical spine: a systematic review of case reports of serious adverse events, 1995-2001. *Med J Aust* 2002; **176**(8):376-80.
3. Norris JW, Beletsky V, Nadareishvili ZG. Sudden neck movement and cervical artery dissection. The Canadian Stroke Consortium. *CMAJ* 2000; **163**(1):38-40.
4. Cassidy JD, Boyle E, Côté P, He Y, Hogg-Johnson S, Silver FL, Bondy SJ. Risk of vertebrobasilar stroke and chiropractic care: results of a population-based case-control and case-crossover study. *Spine* 2008; **33**(4 Suppl):S176-83.
5. Boyle E, Côté P, Grier AR, Cassidy J. D. Examining vertebrobasilar artery stroke in two Canadian provinces. *Spine* 2008; **33**(4 Suppl):S170-5.
6. Côté P, Wong JJ, Sutton D, Shearer HM, Mior S, Randhawa K, et al. Management of neck pain and associated disorders: A clinical practice guideline from the Ontario Protocol for Traffic Injury Management (OPTIMa) Collaboration. *Eur Spine J* 2016; **25**(7):2000-22.
7. Kjaer P, Kongsted A, Hartvigsen J, Isenberg-Jørgensen A, Schiøttz-Christensen B, Søborg B, et al. National clinical guidelines for non-surgical treatment of patients with recent onset neck pain or cervical radiculopathy. *Eur Spine J* 2017; **26**(9):2242-57.
8. Hurwitz EL, Coulter ID, Adams AH, Genovese BJ, Shekelle PG. Use of chiropractic services from 1985 through 1991 in the United States and Canada. *Am J Public Health* 1998; **88**(5):771-6.
9. Hurwitz EL, Chiang LM. A comparative analysis of chiropractic and general practitioner patients in North America: findings from the joint Canada/United States Survey of Health, 2002-03. *BMC Health Serv Res* 2006; **6**:49.
10. Haldeman S, Carey P, Townsend M, Papadopoulos C. Arterial dissections following cervical manipulation: the chiropractic experience. *CMAJ* 2001; **165**(7):905-6.
11. Patijn J. Complications in manual medicine: A review of the literature. *J Manual Med* 1991; **6**:89-92.
12. Smith RA, Estridge MN. Neurological complications of head and neck manipulations: A report of two cases. *JAMA* 1962; **182**(5):528-31.
13. Frisoni GB, Anzola GP. Vertebrobasilar ischemia after neck motion. *Stroke* 1991; **22**(11):1452-60.
14. Mann T, Refshauge KM. Causes of complications from cervical spine manipulation. *Aust J Physiother* 2001; **47**(4):255-66.
15. Schmitt HP. Anatomical structure of the cervical spine with reference to pathology of manipulation complications. *Man Med* 1991; **6**:93-101.
16. Mitchell J. Vertebral Artery Blood flow Velocity Changes Associated with Cervical Spine rotation: A Meta-Analysis of the Evidence with implications for Professional Practice. *J Manipulative Physiol Ther* 2009; **17**(1):46-57.
17. Kawchuk GN, Jhangri GS, Hurwitz EL, Wynd S, Haldeman S, Hill MD. The relation between the spatial distribution of vertebral artery compromise and exposure to cervical manipulation. *J Neurol* 2008; **255**(3):371-7.
18. Dunne JW, Conacher GN, Khangure M, Harper CG. Dissecting aneurysms of the vertebral arteries following manipulation: a case report. *Neuro Neurosurg Psych* 1987; **50**:349-353.
19. Arnold C, Bourassa T, Langer T, Stoneham G. Doppler studies evaluating the effect of a physical therapy screening protocol on vertebral artery blood flow. *Man Ther* 2004; **9**:13–21.
20. Licht PB, Christensen HW, Højgaard P, Marving J. Vertebral artery flow and spinal manipulation: a randomized, controlled and observer-blinded study. *J Manipulative Physiol Ther* 1998; **21**(3):141-4.
21. Mitchell J. Changes in vertebral artery blood flow following rotation of the cervical spine. *J Manipulative Physiol Ther* 2003; **26**:347– 351.
22. Mitchell J, Keene D, Dyson C, Harvey L, Pruvey C, Phillips R. Is cervical spine rotation, as used in the standard vertebrobasilar insufficiency test, associated with a measurable change in intracranial vertebral artery blood flow? *Man Ther* 2004; **9**:220–227.

23. Mitchell J, Kramschuster K. Real-time ultrasound measurements of changes in suboccipital vertebral artery diameter and blood flow velocity associated with cervical spine rotation. *Physiother Res Internat* 2009; **13**:241–254.
24. Rivett DA, Sharples KJ, Milburn PD. Effect of pre-manipulative tests on vertebral artery and internal carotid artery blood flow: A pilot study. *J Manipulative Physiol Ther* 1999; **22**:368–375.
25. Rivett DA, Sharples KJ, Milburn PD. Reliability of ultrasonograpahic measurement of vertebral artery blood flow. *NZ J Physiother* 2003; **31**:119–128.
26. Rossitti S, Volkmann R, Lofgren J. Changes of blood flow velocity in the vertebro-basilar circulation during rotation of the head in the normal human. *Biomech Sem* 1992; **6**:92–99.
27. Stevens A. Functional Doppler sonography of the vertebral artery and some considerations about manual techniques. *Man Med* 1991; **6**:102–105.
28. Quesnele JJ, Triano JJ, Noseworthy MD, Wells GD. Changes in vertebral artery blood flow following various head positions and cervical spine manipulation. *J Manipulative Physiol Ther* 2014; **37**(1):22-31.
29. Erhardt JW, Windsor BA, Kerry R, Hoekstra C, Powell DW, Porter-Hoke A, Taylor A. The immediate effect of atlanto-axial high velocity thrust techniques on blood flow in the vertebral artery: A randomized controlled trial. *Man Ther* 2015; **20**(4):614-22.
30. Bakhtadze MA, Vernon H, Karalkin AV, Pasha SP, Tomashevskiy IO, Soave D. Cerebral perfusion in patients with chronic neck and upper back pain: preliminary observations. *J Manipulative Physiol Ther* 2012; **35**:76-85.
31. Barrett KE, Ganong WF. Ganong's Review of Medical Physiology 25th ed. New York, NY: McGraw-Hill Medical, 2015.
32. Guzman J, Haldeman S, Carroll LJ, Carragee EJ, Hurwitz EL, Peloso P, et al. Bone and Joint Decade 2000-2010 Task Force on Neck Pain and Its Associated Disorders. Clinical practice implications of the Bone and Joint Decade 2000-2010 Task Force on Neck Pain and Its Associated Disorders: from concepts and findings to recommendations. *Spine* 2008; **33**(4 Suppl):S199-213.
33. Cambridge E, Triano J, Ross K, Abbott M. Comparison of force development strategies of spinal manipulation used for thoracic pain. *Man Ther* 2012; **17**:241-245.
34. Descarreaux M, Dugas C, Raymond J, Normand M. Kinetic analysis of expertise in spinal manipulative therapy using an instrumented manikin. *J Chiropr Med* 2005; **4**:53–60.
35. Cohen E, Triano JJ, McGregor M, Papakyriakou M. Biomechanical performance of spinal manipulation therapy by newly trained vs. practicing providers: does experience transfer to unfamiliar procedures? *J Manipulative Physiol Ther* 1995; **18**:347-52.
36. Triano J, Rogers C, Combs S, Potts D, Sorrels K. Developing skilled performance of lumbar spine manipulation. *J Manipulative Physiol Ther* 2002; **25**:353-61.
37. Triano J, Rogers C, Combs S, Potts D, Sorrels K. Quantitative feedback versus standard training for cervical and thoracic manipulation. *J Manipulative Physiol Ther* 2003; **26**:131-8.
38. Ho SS, Chan YL, Yeung DK, Metreweli C. Blood flow volume quantification of cerebral ischemia: comparison of three noninvasive imaging techniques of carotid and vertebral arteries. *Am J Roentgenol* 2002; **178**(3):551-6.
39. Lotz J, Meier C, Leppert A, Galanski M. Cardiovascular flow measurement with phase-contrast MR imaging: basic facts and implementation. *Radiographics* 2002; **22**(3):651-71.
40. Ioannidis JP, Evans SJ, Gøtzsche PC, O'Neill RT, Altman DG, Schulz K, Moher D.; CONSORT Group. Better reporting of harms in randomized trials: an extension of the CONSORT statement. *Ann Intern Med* 2004; **141**(10):781-8.
41. Parkes LM, Rashid W, Chard DT, Tofts PS. Normal cerebral perfusion measurements using arterial spin labeling: reproducibility, stability, and age and gender effects. *Magn Reson Med* 2004; **51**(4):736-43.
42. Cohen J. Statistical power analysis for the behavioral sciences 2nd ed. New Jersey: Lawrence Erlbaum Associates, Publishers, 1988.
43. Cox RW. AFNI: software for analysis and visualization of functional magnetic resonance neuroimages. *Comput Biomed Res* 1996; **29**(3):162-73.
44. Holmes CJ, Hoge R, Collins L, Woods R, Toga AW, Evans AC. Enhancement of MR images using registration for signal averaging. *J Comput Assist Tomogr* 1998; **22**(2):324-33.

45. Jang JH, Kim JH, Jung WH, Choi JS, Jung MH, Lee JM, et al. Functional connectivity in fronto-subcortical circuitry during the resting state in obsessive-compulsive disorder. *Neurosci Lett* 2010; **474**(3):158-62.
46. Wang J, Wang L, Zang Y, Yang H, Tang H, Gong Q, et al. Parcellation-dependent small-world brain functional networks: a resting-state fMRI study. *Hum Brain Mapp* 2009; **30**(5):1511-23.
47. Yurdakul M, Tola M. Doppler criteria for identifying proximal vertebral artery stenosis of 50% or more. *J Ultrasound Med* 2011; **30**(2):163-8.
48. Licht PB, Christensen HW, Højgaard P, Marving J. Vertebral artery flow and spinal manipulation: a randomized, controlled and observer-blinded study. *J Manipulative Physiol Ther* 1998; **21**(3):141-4.
49. Debette S, Leys D. Cervical-artery dissections: predisposing factors, diagnosis, and outcome. *Lancet Neurol* 2009; **8**(7):668-78.
50. Senter HJ, Sarwar M. Nontraumatic dissecting aneurysm of the vertebral artery. Case report. *J Neurosurg* 1982; **56**(1):128-30.
51. Sherman DG, Hart RG, Easton JD. Abrupt change in head position and cerebral infarction. *Stroke* 1981; **12**(1):2-6.
52. Haldeman S, Kohlbeck FJ, McGregor M. Unpredictability of cerebrovascular ischemia associated with cervical spine manipulation therapy: a review of sixty-four cases after cervical spine manipulation. *Spine* 2002; **27**(1):49-55.
53. Josien E. Extracranial vertebral artery dissection: nine cases. *J Neurol* 1992; **239**(6):327-30.
54. Okawara S, Nibbelink D. Vertebral artery occlusion following hyperextension and rotation of the head. *Stroke* 1974; **5**(5):640-2.
55. Sengelhoff C, Nebelsieck J, Nassenstein I, Maintz D, Nabavi DG, Kuhlenbaeumer G, et al. Neurosonographical follow-up in patients with spontaneous cervical artery dissection. *Neurol Res* 2008; **30**(7):687-9.
56. Dittrich R. Dziewas R, Ritter MA, Kloska SP, Bachmann R, Nassenstein I, et al. Negative ultrasound findings in patients with cervical artery dissection. Negative ultrasound in CAD. *J Neurol* 2006; **253**(4):424-33.
57. Nebelsieck J, Sengelhoff C, Nassenstein I, Maintz D, Kuhlenbäumer G, Nabavi DG, et al. Sensitivity of neurovascular ultrasound for the detection of spontaneous cervical artery dissection. *J Clin Neurosci* 2009; **16**(1):79-82.
58. Oktar SO, Yücel C, Karaosmanoglu D, Akkan K, Ozdemir H, Tokgoz N, et al. Blood-flow volume quantification in internal carotid and vertebral arteries: comparison of 3 different ultrasound techniques with phase-contrast MR imaging. *Am J Neuroradiol* 2006; **27**(2):363-9.

Table 1. *Baseline participant characteristics*

Participant characteristics	Mean (SD)
Age (years)	32.05 (12.5)
Sex (n, %)	
Female	14 (70%)
Height (cm)	169 (8.9)
Weight (kg)	69.6 (18.7)
NDI	13 (6.4)
NRS neck	4.7 (2.1)
Duration of complaint (years)	5.3 (5.7)
NRS headache	3 (2.6)

SD= standard deviation, cm= centimeters, kg- kilograms, NDI= Neck Disability Index, NRS= numerical rating scale

Table 2. *Mean cerebral and cerebellar perfusion (in milliliters per 100 grams of tissue per minute) for each condition as determined by arterial spin labeling technique*

Cerebrum and cerebellum regions of interest (mL/100g/min), mean (SD)	Neutral	Manipulation	Maximum rotation
Uvula	79.2 (25)	80.1 (26.7)	79.1 (25.6)
Cerebellar Tonsils	69.2 (19.7)	66.8 (16.6)	68.2 (17.4)
L Fusiform Gyrus/L Middle Temporal Gyrus	80.7 (27.7)	78.0 (25.8)	79.0 (23.4)
R Middle Temporal Gyrus	37.4 (12.8)	35.7 (11.9)	36.1 (13.7)
R Middle Occipital Gyrus	58.2 (21.5)	52.8 (18.4)	53.2 (19.4)
Right and Left Cuneus	66.5 (30.6)	62.2 (25.8)	61.0 (28.5)
Left Precuneus	75.7 (31.4)	74.9 (27.3)	64.1 (23.4)
Left Cuneus, Left Middle Occipital Gyrus	49.4 (17.8)	47.6 (16.9)	46.9 (18.4)

Table 3. *Ipsilateral and contralateral mean and peak vertebral artery blood flow and velocity at the level of C1-2 for each condition.*

	Velocity (cm/s)				Flow (mL/s)			
Test maneuver	**Ipsilateral**		**Contralateral**		**Ipsilateral**		**Contralateral**	
	Mean(SD)	Peak(SD)	Mean(SD)	Peak(SD)	Mean(SD)	Peak(SD)	Mean(SD)	Peak(SD)
Neutral	15.5(3.5)	33.75(11.7)	15.6(3.2)	33.1(8.3)	1.8 (0.7)	2.1(1.2)	1.8 (0.8)	2.8(1.1)
Cervical manipulation	14.7(3.6)	35.4(17.2)	14.4(2.6)	35.9(15.8)	1.8 (0.8)	2.2(1.4)	1.8 (0.8)	2.8(1.04)
Max rotation	15.1(3.9)	33.02(18.3)	14.4(3.2)	31.6(8.8)	1.7 (0.6)	2.0(1.1)	1.6 (0.7)	2.6(1.1)

Table 4. *Analysis of resting state fMRI indicating coordinates from areas of significantly increased functional connectivity, post-procedures.*

Number of Voxels	**CM_X**	**CM_Y**	**CM_Z**	**PV_X**	**PV_Y**	**PV_Z**	**Side**	**Location**	**P Values**
54	-56.2	-32.2	+7.2	-47.2	-46.5	+17.0	L	TG	0.0027
53	-5.1	-59.4	-28.0	-5.2	-60.5	-32.0	L, R	Uvula	0.0003
52	+23.8	+10.8	+30.4	+26.2	+2.5	+34.5	R	FG	0.000035
48	-6.2	-38.6	-38.1	-5.2	-36.0	-39.0	L, R	CT	0.0014
47	-45.3	-57.7	-14.4	-43.8	-64.0	-28.5	L	FfG, MTG	0.0001
43	+33.4	-46.2	+25.6	+36.8	-39.5	+27.5	R	MTG	0.00235
37	+31.6	-75.2	+6.2	+36.8	-67.5	+3.0	R	MOG	0.0047
32	-17.2	-19.3	+51.0	-12.2	-22.0	+48.5	L	PG, MCG	0.00165
31	+22.0	-56.1	-35.4	+22.8	-57.0	-39.0	R	CT	0.00047
31	-28.0	-39.2	+30.7	-29.8	-36.0	+31.0	L	IPL	0.0017
25	+55.8	-12.7	-3.2	+57.8	-8.0	-4.0	R	STG	0.0031
24	+1.4	-91.3	+8.9	+5.2	-95.5	+3.0	L, R	Cuneus	0.0003
24	-8.5	-76.1	+44.3	-5.2	-78.0	+48.5	L	Precuneus	0.0127
20	-10.7	+59.2	+2.4	-12.2	+65.5	-0.5	L	MSFG	0.00345
20	-21.0	-77.0	+7.5	-22.8	-74.5	+6.5	L	Cuneus, MOG	0.00205

CM: centre of mass. PV: peak velocity. X, Y, Z: coordinates in the Colin N_27 atlas. L, R: left and right. TG: temporal gyrus. FG: frontal gyrus. CT: cerebellar tonsil. FfG: fusiform gyrus. MTG: middle temporal gyrus. MOG: middle occipital gyrus. PG: precentral gyrus. MCG: middle cingulate gyrus. IPL: inferior parietal lobe. STG: superior temporal gyrus. MSFG: middle, superior frontal gyrus.

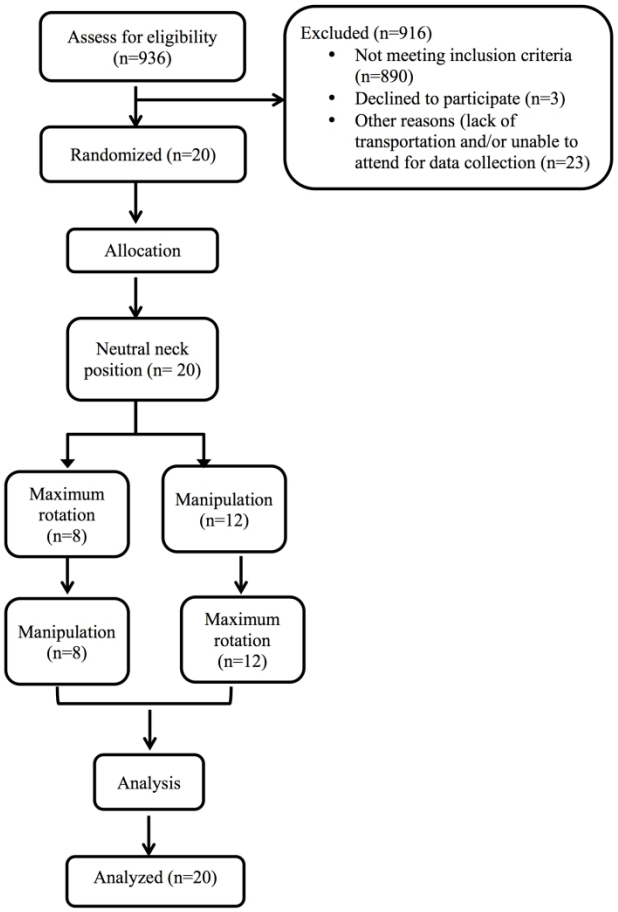

Figure 1. *Protocol flow and method process*

Figure 1. Flow chart

215x279mm (300 x 300 DPI)

ONLINE SUPPLEMENT

SUPPLEMENTAL MATERIAL

A Cross-over Randomized Trial of the Effect of Cervical Manipulation on Vertebral Artery and Cerebral Hemodynamics in Neck Pain Patients

Nicholas Moser DC, Silvano Mior PhD, Michael D. Noseworthy PhD, Pierre Côté PhD, Greg Wells PhD, Michael Behr MSc, John J. Triano PhD

Table of Contents

Supplemental Table

Table I. *Analysis of resting state fMRI indicating coordinates from areas of significantly increased functional connectivity, post-procedures.*

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53	-5.1	-59.4	-28.0	-5.2	-60.5	-32.0	L, R	Uvula	0.0003
52	+23.8	+10.8	+30.4	+26.2	+2.5	+34.5	R	FG	0.000035
48	-6.2	-38.6	-38.1	-5.2	-36.0	-39.0	L, R	CT	0.0014
47	-45.3	-57.7	-14.4	-43.8	-64.0	-28.5	L	FfG, MTG	0.0001
43	+33.4	-46.2	+25.6	+36.8	-39.5	+27.5	R	MTG	0.00235
37	+31.6	-75.2	+6.2	+36.8	-67.5	+3.0	R	MOG	0.0047
32	-17.2	-19.3	+51.0	-12.2	-22.0	+48.5	L	PG, MCG	0.00165
31	+22.0	-56.1	-35.4	+22.8	-57.0	-39.0	R	CT	0.00047
31	-28.0	-39.2	+30.7	-29.8	-36.0	+31.0	L	IPL	0.0017
25	+55.8	-12.7	-3.2	+57.8	-8.0	-4.0	R	STG	0.0031
24	+1.4	-91.3	+8.9	+5.2	-95.5	+3.0	L, R	Cuneus	0.0003
24	-8.5	-76.1	+44.3	-5.2	-78.0	+48.5	L	Precuneus	0.0127
20	-10.7	+59.2	+2.4	-12.2	+65.5	-0.5	L	MSFG	0.00345
20	-21.0	-77.0	+7.5	-22.8	-74.5	+6.5	L	Cuneus, MOG	0.00205

CM: centre of mass. PV: peak velocity. X, Y, Z: coordinates in the Colin N_27 atlas. L, R: left and right. TG: temporal gyrus. FG: frontal gyrus. CT: cerebellar tonsil. FfG: fusiform gyrus. MTG: middle temporal gyrus. MOG: middle occipital gyrus. PG: precentral gyrus. MCG: middle cingulate gyrus. IPL: inferior parietal lobe. STG: superior temporal gyrus. MSFG: middle, superior frontal gyrus.

Supplemental Figures

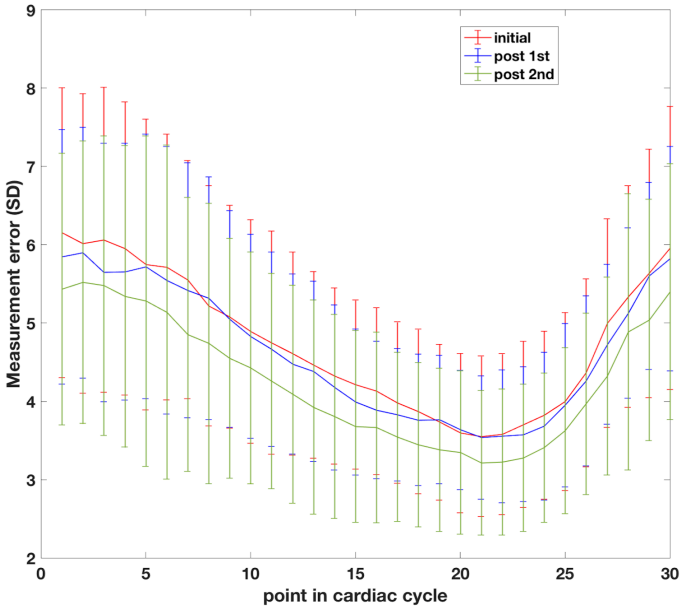

Figure I. *Left vertebral artery mean blood flow variability in the cardiac cycle between neutral (initial), cervical manipulation (post 1st) and maximum rotation (post 2nd). Bars represent the standard deviation of the mean errors at each point.*

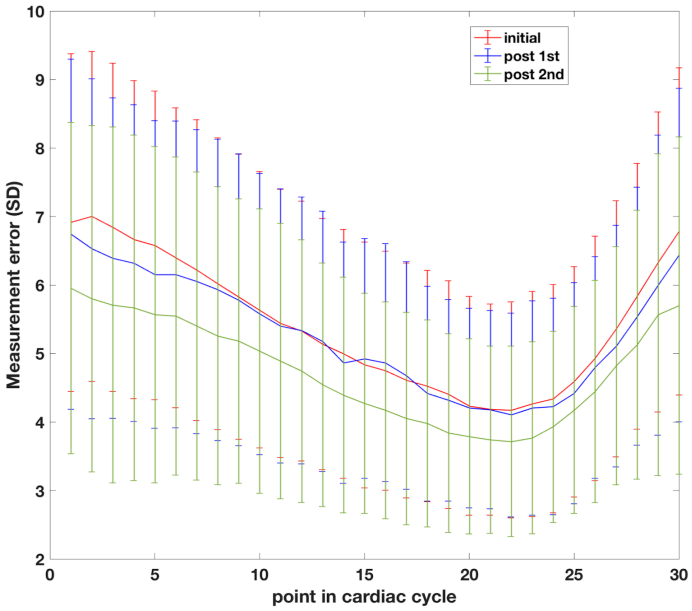

Figure II. *Right vertebral artery mean blood flow variability in the cardiac cycle between neutral (initial), cervical manipulation (post 1st) and maximum rotation (post 2nd). Bars represent the standard deviation of the mean errors at each point.*

1

Table 1: CONSORT 2010 checklist of information to include when reporting a within-person randomised trial. For within-person trials, a group is the set of participants' body sites that was allocated a particular intervention.

Section/Topic	Item no.	Standard CONSORT Checklist item	Extension for within-person trials	Page no.
Title and abstract				
	1a	Identification as a randomised trial in the title	Identification as a within-person randomised trial in the title	1
	1b	Structured summary of trial design, methods, results, and conclusions (for specific guidance see CONSORT for abstracts[3])	Specify a within-person design and report all information outlined in table 2	2
Introduction				
Background and objectives	2a	Scientific background and explanation of rationale		3
	2b	Specific objectives or hypotheses		3
Methods				
Trial design	3a	Description of trial design (such as parallel, factorial) including allocation ratio	Rationale for using a within-person design and identification of body sites	3
	3b	Important changes to methods after trial commencement (such as eligibility criteria), with reasons		3
Participants	4a	Eligibility criteria for participants	Eligibility criteria for body sites	4
	4b	Settings and locations where the data were collected		4
Interventions	5	The interventions for each group with sufficient details to allow replication, including how and when they were actually administered	Whether interventions were given sequentially or concurrently	4
Outcomes	6a	Completely defined pre-specified primary and secondary outcome measures, including how and when	Outcomes should be clearly defined as per-site or per-	5

From: Pandis N, Chung B, Scherer RW, Elbourne D, Altman DG. CONSORT 2010 statement: extension checklist for reporting within person randomised trials. BMJ. 2017;357.

Section/Topic	Item no.	Standard CONSORT Checklist item	Extension for within-person trials	Page no.
		they were assessed	person	
	6b	Any changes to trial outcomes after the trial commenced, with reasons		5
Sample size	7a	How sample size was determined	Report the correlation between body sites	6
	7b	When applicable, explanation of any interim analyses and stopping guidelines		N/A
Randomisation:				
Sequence generation	8a	Method used to generate the random allocation sequence		4
	8b	Type of randomisation; details of any restriction (such as blocking and block size)	Methods used to determine the allocation sequence of body sites and treatments within an individual (e.g. how first site to be treated was decided)	4
Allocation concealment mechanism	9	Mechanism used to implement the random allocation sequence (such as sequentially numbered containers), describing any steps taken to conceal the sequence until interventions were assigned		4
Implement-ation	10	Who generated the random allocation sequence, who enrolled participants, and who assigned participants to interventions	Replaced by 10a	4
	10a		Who generated the random allocation sequence, who enrolled participants, and who assigned body sites to interventions	4
Blinding (masking)	11a	If done, who was blinded after assignment to interventions (for example, participants, care providers, those assessing outcomes) and how		4
	11b	If relevant, description of the similarity of interventions		N/A

From: Pandis N, Chung B, Scherer RW, Elbourne D, Altman DG. CONSORT 2010 statement: extension checklist for reporting within person randomised trials. BMJ. 2017;357.

Section/Topic	Item no.	Standard CONSORT Checklist item	Extension for within-person trials	Page no.
Statistical methods	12a	Statistical methods used to compare groups for primary and secondary outcomes	Statistical methods appropriate for within-person design	6
	12b	Methods for additional analyses, such as subgroup analyses and adjusted analyses		6
Results				
Participant flow (a diagram is strongly recommended)	13a	For each group, the numbers of participants who were randomly assigned, received intended treatment, and were analysed for the primary outcome	Number of participants and number of body sites at each stage [See Figure 1]	6
	13b	For each group, losses and exclusions after randomisation, together with reasons	Number of participants and number of body sites lost or excluded at each stage, with reasons	7
Recruitment	14a	Dates defining the periods of recruitment and follow-up		6
	14b	Why the trial ended or was stopped		N/A
Baseline data	15	A table showing baseline demographic and clinical characteristics for each group	Baseline characteristics for site and individual participants as applicable	13
Numbers analysed	16	For each group, number of participants (denominator) included in each analysis and whether the analysis was by original assigned groups	Number of randomised body sites in each group included in each analysis	7
Outcomes and estimation	17a	For each primary and secondary outcome, results for each group, and the estimated effect size and its precision (such as 95% confidence interval)	Observed correlation between body sites for continuous outcomes and tabulation of paired results for binary outcomes	7
	17b	For binary outcomes, presentation of both absolute and relative effect sizes is recommended		N/A
Ancillary analyses	18	Results of any other analyses performed, including subgroup analyses and adjusted analyses, distinguishing pre-specified from exploratory		14

From: Pandis N, Chung B, Scherer RW, Elbourne D, Altman DG. CONSORT 2010 statement: extension checklist for reporting within person randomised trials. BMJ. 2017;357.

Section/Topic	Item no.	Standard CONSORT Checklist item	Extension for within-person trials	Page no.
Harms	19	All important harms or unintended effects in each group (for specific guidance see CONSORT for harms)	Harms or unintended effects reported by participant and by body site	7
Discussion				
Limitations	20	Trial limitations, addressing sources of potential bias, imprecision, and, if relevant, multiplicity of analyses		8
Generalisability	21	Generalisability (external validity, applicability) of the trial findings		8
Interpretation	22	Interpretation consistent with results, balancing benefits and harms, and considering other relevant evidence		8
Other information				
Registration	23	Registration number and name of trial registry		3
Protocol	24	Where the full trial protocol can be accessed, if available		N/A
Funding	25	Sources of funding and other support (such as supply of drugs), role of funders		9

From: Pandis N, Chung B, Scherer RW, Elbourne D, Altman DG. CONSORT 2010 statement: extension checklist for reporting within person randomised trials. BMJ. 2017;357.

BMJ Open

A Cross-over Randomized Controlled Trial of the Effect of Cervical Manipulation on Vertebral Artery and Cerebral Hemodynamics in Patients with Chronic Neck Pain

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2018-025219.R1
Article Type:	Research
Date Submitted by the Author:	18-Jan-2019
Complete List of Authors:	Moser, Nicholas; Canadian Memorial Chiropractic College, Graduate studies Mior, Silvano ; University of Ontario Institute of Technology; Canadian Memorial Chiropractic College Noseworthy , Michael; McMaster University Faculty of Engineering Côté, Pierre ; University of Ontario Institute of Technology, Faculty of Health Sciences Wells, Greg; University of Toronto Faculty of Kinesiology and Physical Education Behr, Michael; University Health Network Triano, John; Canadian Memorial Chiropractic College, Graduate studies
Primary Subject Heading:	Complementary medicine
Secondary Subject Heading:	Rehabilitation medicine, Neurology, Radiology and imaging
Keywords:	Magnetic resonance imaging < RADIOLOGY & IMAGING, Spinal manipulation, Neck pain, Vertebral artery dissection, Blood flow

A Cross-over Randomized Controlled Trial of the Effect of Cervical Manipulation on Vertebral Artery and Cerebral Hemodynamics in Patients with Chronic Neck Pain

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Keywords: fMRI$_1$, BOLD fMRI$_2$, Neck Pain$_3$, Vertebral Artery$_4$, Spinal Manipulation$_5$, Chronic neck pain$_6$, Vertebral Artery Dissection$_7$, Stroke$_8$

Summary

Objective

It is hypothesized that cervical manipulation may increase the risk of cerebrovascular accidents. We aimed to determine whether cervical spine manipulation is associated with changes in vertebral artery and cerebrovascular hemodynamics measured with magnetic resonance imaging compared to neutral neck position and maximum neck rotation in patients with chronic neck pain.

Setting

The Imaging Research Centre at St. Joseph's Hospital in Hamilton, Ontario, Canada.

Participants

Twenty patients were included. The mean age was 32 years (SD ± 12.5), mean neck pain duration was 5.3 years (SD ± 5.7) and mean Neck Disability Index score was 13/50 (SD ± 6.4).

Interventions

Following baseline measurement of cerebrovascular hemodynamics, we randomized participants to: 1) maximal neck rotation followed by cervical manipulation; or 2) cervical manipulation followed by maximal neck rotation. The primary outcome, vertebral arteries and cerebral hemodynamics, was measured after each intervention and was obtained by measuring 3D T1-weighted high resolution anatomical images, arterial spin labeling (ASL) and phase contrast flow encoded MRI. Our secondary outcome was functional connectivity within the default mode network (DMN) measured with resting state functional MRI (fMRI).

Results

Compared to neutral neck position, we found a significant change in contralateral blood flow following maximal neck rotation. There was also a significant change in contralateral vertebral artery blood velocity following maximal neck rotation and cervical manipulation. We found no significant changes within the cerebral hemodynamics following cervical manipulation or maximal neck rotation. However, we observed significant increases in functional connectivity in the posterior cerebrum and cerebellum (resting state MRI) after manipulation and maximum rotation.

Conclusion

Our results are in accordance with previous work, which has shown a decrease in blood flow and velocity in the contralateral vertebral artery with head rotation. This may explain why we also observed a decrease in blood velocity with manipulation because it involves neck rotation. Our work is the first to show that cervical manipulation does not result in brain perfusion changes compared to a neutral neck position or maximal neck rotation. The changes observed were found to not be clinically meaningful and suggests that cervical manipulation may not increase the risk of cerebrovascular events through a hemodynamic mechanism.

Trial Registration

US National Institute of Health # NCT02667821.

Strengths and limitations of this study

- A strength of the study was the design, which ensured control of confounders and provided statistical efficiency.
- We used what is considered the criterion standard for both diagnosis of vertebrobasilar artery stroke and quantifying blood flow (phase-contrast MRI) because of its greater sensitivity compared to ultrasonography.
- A limitation of the study was the restriction of analysis to a time following the test maneuvers. Real time measures currently are technically not feasible and transient effects of various neck positions on vertebral artery and cerebrovascular hemodynamics may have been missed.

INTRODUCTION

Anecdotal evidence from case reports and case-series suggest that neck manipulation increases the risk of vertebrobasilar artery stroke.[1-3] However, the epidemiological evidence does not support this hypothesis.[4, 5] In their case-crossover study, Cassidy *et al.* found that the risk of vertebrobasilar artery stroke was similar for patients with neck pain or headaches who consult physicians and those who consult chiropractors.[4] This suggests that the hypothesized association is due to protopathic bias.

Understanding whether neck manipulation increases the risk of stroke is important because patients with neck pain frequently consult chiropractors and manipulation of the cervical spine is commonly performed for symptomatic relief.[6-10] Dissection of the vertebral artery is involved in most cases that implicate cervical manipulation.[11] However, when damage to the vertebral artery is absent, vasospasm[12-15] and 'subclinical' endothelial injury have also been hypothesized to be causes of stroke. According to the vasospasm hypothesis, placing the head in rotation and hyperextension during a manipulation leads to considerable stress and stretch forces in the vertebral artery, specifically at the C2/C1 and cephalad/distal portion of the vertebral artery.[16-18] This mechanical compression or stretching of the vertebral artery may lead to changes in blood flow and the 'subclinical' injury to the vertebral artery can lead to thrombosis.[19]

Several studies have investigated changes in blood flow during cervical spine motion.[20-28] Most studies report a decrease in vertebral artery flow contralateral to the side of rotation.[20-23, 25-28] Less is understood about blood flow during and after a cervical manipulation, but two studies found no significant changes in vertebral artery blood flow or blood velocity following cervical manipulation in healthy individuals.[29, 30] However, the impact of cervical manipulation on vertebral artery blood flow in the population likely to undergo this maneuver for therapeutic purposes is unknown. Neck pain patients may differ from that of healthy populations because moderate to severe perceived neck disability, as measured with the Neck Disability Index (NDI) is correlated with cortical hypoperfusion.[31]

Therefore, our primary aim was to determine whether cervical manipulation leads to a meaningful change in vertebral and cerebral hemodynamics compared to neutral neck position or neck rotation in adult patients with chronic neck pain. Our secondary aim was to compare the functional connectivity within the default mode network (DMN) between neutral neck position, neck rotation and cervical manipulation. We hypothesized that cervical manipulation or maximal neck rotation is associated with significant change in vertebral and cerebral hemodynamics compared to neutral neck position in adult patients with chronic neck pain.

METHODS

Study Design

We conducted a crossover randomized controlled study. In each subject, we randomized the sequence of cervical manipulation and maximal neck rotation and compared their effects on cerebrovascular hemodynamics. No washout period was used between each intervention. It was assumed that the time needed to measure the blood hemodynamics allowed enough time to return to their baseline status.[32] The study was registered with www.clinicaltrials.gov, NCT02667821. The McMaster University Hamilton Integrated Research Ethics Board (HiREB) (REB#1303) and Canadian Memorial Chiropractic College Research Ethics Board approved the study (REB#1604X01).

Participants

Patients who were eligible for the study were attending a teaching clinic of the Canadian Memorial Chiropractic College, Toronto, Canada between September 2016 and April 2017. We recruited participants via poster advertising displayed at the teaching clinic, word of mouth, and referrals from the supervising clinicians at the clinic. To be included, patients had to meet the following criteria: 1) at least 18 years old; 2) chronic neck pain (≥ 3 months' duration) defined as either neck pain-associated disorder or whiplash associated-disorder; 3) grade I-II neck pain[33], defined as neck pain with no signs or symptoms of major structural pathology, which may or may not interfere with activities of daily living; 4) prescribed cervical manipulation by the clinician at the teaching clinic supervising their care; and 5) provide written informed consent. Exclusion criteria were: a history of neck pain with associated arm pain within the last 6 months; any current or history of neurologic symptoms including facial or extremity weakness, abnormal sensation to the face, body, or extremities, uncontrolled movements, abnormal gait, dizziness, unexplained nausea/vomiting, difficulty with speaking or swallowing; history of new or severe (Visual Analogue Scale >6/10) headaches in the last 3 months; any contraindications to magnetic resonance imaging (MRI); or any history of using drugs that affect blood flow such as Warfarin, or anti-coagulants. In addition, all participants refrained from vigorous physical activity and ingesting alcohol and caffeine one day before their scheduled participation.

Randomization and masking

We used simple randomization to allocate participants to one of two sequences of interventions: 1) maximal neck rotation followed by upper cervical manipulation; or 2) upper cervical manipulation followed by maximal neck rotation. The study coordinator (NM) conducted the randomization using a randomized table generator (GraphPad Software Inc, La Jolla, CA). The random allocation was communicated verbally to the study clinician (SM) on the day of the study protocol. Randomization was concealed, no other study personnel or participants were aware of the intervention assignments.

Procedures

Prior to commencement of the study protocol, participants underwent a cervical spine examination by the clinician (SM) performing the test manoeuvres to identify the site of manipulation. Baseline information on each participant was collected and included: age, gender, height, weight, NDI score, neck pain intensity, duration of neck complaint and headache pain intensity (Table 1).

Baseline MRI of the upper cervical spine and brain with the neck in the neutral position was conducted before randomization. Neutral neck position was defined by alignment of the Frankfort plane in a vertical orientation. For continuity of neutral alignment during imaging between test conditions, the MRI laser land-marking tool was used to triangulate between three oil-based markers (Vitamin E capsules) taped to the nasion (bridge of the nose) and immediately in front of the tragus of the ears, bilaterally.

Following random allocation, either maximal neck rotation or cervical manipulation was first performed, followed by the other procedure. Maximal neck rotation was achieved by instructing participants to rotate their head as far as comfortably possible in the direction opposite to the side of clinical symptoms as elicited during the cervical spine examination. The clinician performing the interventions assisted the rotation via a soft hand contact on the patient's head. The degree of maximal neck rotation was measured by an inclinometer and the position was held for one minute before returning to neutral neck position for MRI sequencing. The cervical manipulation procedure was a high velocity, low amplitude (HVLA) impulse, with targeted contact at C1-C2 on the side of most discomfort as elicited upon palpation, with the participant's head in combined axial rotation, flexion and lateral flexion postures. Variations of head positions between operators for this procedure have been demonstrated to be relatively small.[34, 35] A practitioner with more than 30 years of practice experience conducted the cervical manipulation (SM).[36-38] The manipulation procedure was performed on the adjustable and pivotal MRI bed in the MRI room with the participant in the supine position. The clinician performed the procedure by first establishing the end range of motion to determine

appropriate preload position for the manipulation before applying a clinical force impulse in the coronal plane with minimal traction component.

Before each maneuver, the participants were queried on their comfort, condition, and willingness to continue. The participant's head was repositioned to neutral immediately after each maneuver, and then retracted into the MRI bore. Each maneuver was carried out on the scanner bed in the MRI room.

Outcomes

The primary outcome measure was cerebrovascular hemodynamics within the vertebral arteries and posterior cerebrum measured with MRI. The MRI data was acquired using a 3Telsa MR750 scanner and 20-channel neurovascular array radiofrequency coil (General Electric (GE) Healthcare, Milwaukee WI).

The primary outcome was measured following a standardized protocol. First, the baseline MRI was performed. The head was immobilized with sponges in a neutral neck position and a localizer scan was completed. Next, high-resolution anatomical images were collected using a 3D inversion recovery (IR) prepped fast spoiled gradient recalled echo (fSPGR) T1-weighted scan (repetition time [TR]/echo time [TE]=11.4/4.3ms, TI = 450ms, flip angle = 12°, 512 x 256 matrix, 140 slices, 24 cm field of view [FOV], reconstructed to 1mm^3 isotropic voxels). Following the anatomical scan, microvascular perfusion was assessed using a 3D spiral-based fast spin echo pseudo-continuous arterial spin labeling (pCASL) (TE/TR/TI=10.5/4629/1525ms, FOV=24cm, 512x8spiral interleaves, 3NEX, reconstructed to give an in-plane resolution of 1.875x1.875 (128x128) with 4mm thick slices, scan time =30s). Following arterial spin labeling, a phase contrast scan for vertebral artery flow quantification was performed. At the upper cervical level C1-2, the contralateral and ipsilateral vertebral arteries, defined to the direction of head motion, were assessed and anatomical images were established to localize the vertebral artery circulation. As previously published by Ho *et al.*[38] the method for obtaining flow quantification of the vertebral artery was a 2-dimensional phase-contrast pulse sequence. To capture accurate vertebral artery flow, the imaging plane is ideally perpendicular to the central axis of the blood vessel. This imaging plane was selected on the vessel of interest at the C1-2 intervertebral level based on arterial visualization on a maximum intensity projection (MIP) of a 3D time-of flight MRI angiogram. Acquisition parameters were as follows: fast gradient recalled echo (fGRE); echo time/repeat time=3.9/8.9ms, flip angle = 20^o, 20cm field of view; 512 × 512 matrix; 244-Hz/pixel receiver bandwidth; 1 average; 4 mm thick; and velocity encoding (venc) of 50 cm/s encoded over 30 phases per cardiac cycle. All image measurements were obtained by manually selecting the optimal anatomical site between the base of the odontoid process and approximately 1cm above the tip of the dens. Data acquisition was triggered by peripheral gating using a pulse oximeter, with sequence acquisition time for each flow measurement being approximately 1.5 minutes, depending on heart rate.[39] According to Lotz *et al.* vessel obliquity is tolerable to ± 15°, above which will cause a significant deviation from true flow.[40]

The secondary outcome measure was functional connectivity within the default mode network (DMN). For baseline measurement this was performed immediately following the phase contrast MRI, but prior to manipulation. Resting state functional MRI data (i.e. blood oxygen level dependent, BOLD signal) was acquired using a gradient echo, echo planar imaging (GE-EPI) sequence (64x64 matrix, 28 axial slices (5mm thick, no skip), 24cm FOV, TE/TR/flip angle = 35ms/2000ms/90^o, 180 temporal points, total scan time=6 minutes). During the resting state scan, participants were asked to keep their eyes open, stay awake, and not think of anything in particular.

Following the baseline MRI scan, the image sequence to obtain the primary and secondary outcomes changed to the following: 3-plane localizer, ASL, phase contrast, resting state MRI and 3D anatomical scan. The latter three were always performed with the 3D anatomical last seeing as microvascular changes (i.e. blood flow) were more likely to be detected early after the cervical manipulation and maximal neck rotation, while structural changes (i.e. 3D scan) wouldn't be expected to occur. Participants were scanned a total of 3 times for each session: baseline, and two procedures in random order.

After testing, each participant was observed for one hour and contacted by a study coordinator (NM) within 24 hours of release for follow-up on status. Adverse events, defined as side effects that are harmful, were assessed via open-ended questions.[41] These included local soreness and pain in the area of the applied test

maneuvers (minor adverse events), as well as signs of stroke or transient ischemic attack (major adverse events).

Statistical Analysis

Previous research examining reproducibility of cerebral perfusion measurements using ASL suggested that the mean percent perfusion difference was 7.1 (SD ± 12).[42] To date, no study has provided information on cerebral blood flow and perfusion after mechanical challenges to the cervical spine.[43] Since minimal clinically important differences have not yet been established, we chose to calculate the study sample size by assuming an effect size. We assumed that a change of at least 2 standard deviation from the normal mean flow would indicate a significant variability of the hemodynamics to the mechanical challenges performed to the cervical spine. Based on this effect size, a power of 80%, a significance level at $p< 0.05$ and two-sided t-test, we calculated that a sample size of 16 participants was necessary.[43] We increased our sample size by 20% (n=20 participants) to account for possible dropouts.

An experienced analyst (MB), who remained blinded to the sequence of test maneuvers, performed the data quantification of the primary and secondary outcomes. Microvascular perfusion and resting state MRI data were analyzed using analysis of functional neuroimaging (AFNI).[44] For each participant all ASL and resting state MRI data were spatially registered to the initial (neutral condition) position. Anatomical, blood flow and functional data were transformed automatically to the Colin27 atlas, using the AFNI command @auto_tlrc, with functional data resampled to a 2mm isometric grid.[45] Temporal band-pass filtering with cut-offs of $0.009Hz < f < 0.08Hz$ was performed in order to suppress unwanted physiological signals and some hardware noise.[46] Functional connectivity within the DMN was assessed using the AFNI plugin *InstaCorr*, a seed-based approach, which uses the Pearson method of correlation to compare time signals.[47] The DMN is the most dominant temporally correlated resting network in the awake brain, defined as regions positively correlated in time with the posterior cingulate cortex (PCC) seed voxel. The PCC was defined automatically using the AFNI Talairach method 'Talairach to' and selecting a single voxel from each the left and right PCC for one analysis (ColinN27 coordinates: 10, 54, 14, and -10, 54, 14). These were both subsequently fused in the post-processing. A 5mm FWHM Gaussian spatial smoothing filter was applied for maximize likelihood of overlap with inter-participant group analysis. Also, temporal outliers determined with the AFNI function *3dToutcount* were censored out. Finally, the AFNI plugin 3d*ClustSim* was used to threshold any clusters with fewer than 20 voxels. The ASL cerebral blood flow data was analyzed similar to resting state MRI in that following spatial co-registration to the neutral condition and spatial blurring with a 5mm FWHM Gaussian convolution kernel, ASL data was warped to the N27 atlas. Group analysis was accomplished using a repeat one-way within-participant 3D-ANOVA, with the one factor being neck position. Post hoc testing included contrasts between neutral, maximum voluntary rotation and cervical manipulation, and also a contrast between maximum voluntary rotation and cervical manipulation. Statistical significance was defined as anything lower than an alpha value of 0.05, with prior cluster thresholding dealing with multiple comparisons.

Flow analysis was performed using Segment v1.9 software 47 (Medviso, Lund, Sweden). Dynamic regions of interest were drawn on the left and right vertebral arteries to quantify the mean, as well as peak velocity and flow. Data in the trigger window portion of the cardiac cycle were derived by spline interpolation using Matlab (Mathworks, Natick, MA). Mean and SDs were calculated for vertebral artery blood velocity, flow, peak velocity, and peak flow for each of the head conditions and vertebral artery side. Differences between task maneuvers and vertebral artery flow and velocity were evaluated using a two-way analysis of variance with factors for participants and head position. The level of significance was set at 0.05. Analyses were conducted using R-project version 2.12.1 (R Development Core Team, 2010. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org/) and SAS software. Vertebral artery blood flow variability was calculated over the 30 phases per cardiac cycle by examining the flow errors in the left and right vertebral arteries for each of the conditions. Additionally, we analyzed the impact of order of procedures by examining the interaction between order and head position in the two-way analysis of variance.[48]

Role of the funding source

The funders of the study had no role in the study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

RESULTS

Between September 2016, and April 2017, a total of 936 participants were screened for the study; 916 failed pre-screening (Figure 1). Most did not meet the inclusion criteria (n=890); 446 participants were found to have acute neck pain (<3 months duration); 119 participants had pain in the upper extremity and/or were identified to have radicular symptoms; 216 participants were not receiving spinal manipulative therapy to the cervical spine as part of their ongoing treatment; 68 participants reported >6/10 headache intensity on average; 15 participants were using anti-coagulants; three participants declined to participate; 23 participants were either unable to acquire transportation to the study location or unable to attend the scheduled time. Twenty participants (14 female, 6 male) aged 23 to 66 years (mean [SD], 32 [± 12.5] years) were enrolled in the study. The average NDI score was 13/50 (SD ± 6.4) with an average neck pain intensity of 5/10 (SD ± 2.1) and a mean duration of neck pain of 5.3 years (SD ± 5.7) (Table 1).

The total time elapsed for each participant testing protocol was 60 minutes. The total time elapsed for each test maneuver was approximately 20 minutes. This included applying the test maneuver (maximum of 1 minute), replacing the participant back into the MRI bore (approximately 1 minute), and image sequencing as noted above (approximately 18 minutes).

When compared to neutral neck position, maximal neck rotation and cervical manipulation did not significantly alter cerebral perfusion within the posterior cerebrum or cerebellum (Table 2). A significant change was found in both contralateral vertebral artery blood flow and blood velocity between the three procedures (Table 3). When comparing interventions to discern which interventions were different, we found a significant difference in the contralateral vertebral artery blood flow between neutral and maximal neck rotation (0.26 mL/min, 95% CI 0.11-0.41) as well as between cervical manipulation and maximal neck rotation (0.23, 95% CI 0.04-0.42). We also found a significant difference in contralateral vertebral artery blood velocity between neutral and cervical manipulation (1.15, 95% CI 0.4-1.9) and between neutral and maximal neck rotation (1.18, 95% CI 0.77-1.59) (Table 4). Finally, we found no significant differences in flow variability within the vertebral arteries between the interventions (Figures I and II in the online only Data Supplement).

We measured a significant increased functional connectivity post-manipulation ($p<0.05$). More specifically, when compared to the resting position, the following areas showed significantly increased functional connectivity: uvula, cerebellar tonsils, left fusiform gyrus and left middle temporal gyrus, right middle temporal gyrus, right middle occipital gyrus, bilateral cuneus, left precuneus, and left middle occipital gyrus (Table I in the online only Data Supplement).

When examining for an order effect of experimental procedures on outcomes, we found a significant order effect only for contralateral vertebral artery blood velocity (Table 5, $p=0.02$). For subjects receiving the manipulation first, the difference in contralateral velocity between manipulation and maximal rotation conditions is on average 0.50 cm/s (95%CI -0.15, 1.16), whereas it is on average -0.60 cm/s (95%CI -1.54, 0.34) if they received the manipulation second. That is, velocity is higher for which ever condition comes first.

No major adverse events were reported. One minor adverse event, mild neck soreness, was reported immediately post-procedure by one participant. The participant attributed the soreness to having to lay motionless on the hard scanner bed during study protocol.

Discussion

Our primary objective was to assess the cerebrovascular and vertebral artery blood flow and velocity changes between various head positions including cervical manipulation in patients with chronic neck pain. We found no significant cerebral perfusion changes within the posterior cerebrum or cerebellum. There was however, a significant change in the contralateral vertebral artery blood flow following maximal neck rotation. We found similar changes in the contralateral vertebral artery blood velocity following both cervical manipulation and maximal neck rotation. We found no significant change in blood flow variability between the vertebral arteries, nor between the various head positions. The decrease in contralateral blood flow to the side of maximal rotation supports previous studies.[16] The finding of non-significant change in blood flow ipsilateral and contralateral to the side of cervical manipulation is also consistent with previous works.[29, 30] Unlike previous work, our results suggest a significant change in contralateral blood velocity following cervical manipulation and maximal head rotation. Given the changes in vertebral artery hemodynamics are more pronounced following maximal head rotation compared to cervical manipulation, specifically in contralateral flow, the changes may be the result of the head turning rather than the effect of the thrust associated with cervical manipulation. This assumption is supported by Herzog et al. who suggest that cervical manipulation imposes less stretch to the vertebral artery than the turning of the head.[18]

No reference values for minimally clinically important differences in the vertebral artery blood flow and velocities have been established. However, hemodynamic stenosis has been long considered as a diameter reduction of greater than 50%, which in the vertebral artery has been associated with a peak and end-diastolic flow greater than 108-cm/s and 36-cm/s, respectively.[49] As suggested by Licht *et al.*[50], a change in peak velocity of greater than 25% from baseline would be necessary for clinically relevant decrease in the vertebral artery. We observed no such reductions in our study. Furthermore, an arbitrary threshold of 200 mL/min net vertebral artery flow volume was originally proposed and below this value patients were said to be at risk of becoming symptomatic with vertebrobasilar ischemia.[51] Seidel *et al.*, however reported that net vertebral artery blood flow volume of less than approximately 100 mL/min can be considered as an indicator of low blood volume.[52] In our study, the net vertebral artery blood flow volume showed that in both experimental procedures, values remained above 200 mL/min (222 mL/min for cervical manipulation and 203 mL/min for maximal neck rotation). When examining flow changes, the largest change was 14%, which occurred in the contralateral vertebral artery following maximal rotation. When we examined the vertebral artery blood velocities, we found a 7% change for both cervical manipulation and maximal rotation compared to neutral. Therefore, the relative blood flow and velocity changes observed are small and not considered clinically relevant. Continuing, none of the participants during any of the experimental procedures reported, or were observed by the investigators, to have any signs or symptoms of neurological compromise. Although vertebral artery blood flow and velocity reductions can occur with head positional changes, the individual typically remains asymptomatic due to several factors, including the presence of collateral circulation.[53, 54] In the present work, this was illustrated by the preservation of cerebral perfusion despite the changes in contralateral vertebral artery hemodynamics.

Vertebrobasilar artery stroke secondary to vertebral artery dissection is a rare but devastating occurrence.[55] A unique characteristic of these strokes is that they can develop in healthy adults and they frequently occur in close temporal relationship to benign neck movements[56, 57], cervical manipulation[58] or trivial trauma.[59, 60] Due to the rarity of the condition, very little is known about the risk factors for vertebrobasilar artery stroke. Our study extends the understanding on the effects of cervical manipulation on vertebral artery and cerebrovascular hemodynamics. It is also the first study to directly measure the impact of cervical spinal manipulation on intracranial and extracranial blood flow in a chronic neck pain population. Together with previous work[29, 30] our results support the position that the association between cervical manipulation and stroke is due to protopathic bias.[4]

When compared to a neutral neck position, both maximum voluntary neck rotation and cervical manipulation resulted in significantly increased functional connectivity throughout the DMN as seen on the BOLD signal. Changes in the BOLD signal arise from complex interactive modulation of blood flow, blood volume and local metabolic rate, all leading to change in the local ratio of oxy- to deoxyhaemoglobin. This ratio drives the change in BOLD signal through magnetic susceptibility differences that exist between oxidation states of the haemoglobin complex. The areas affected within the DMN have been identified as being involved with functions of visually guided eye movements[61], facial and word recognition[62], visuospatial processing[63], episodic memory, reflection upon self and consciousness[64]. The observed increase in functional connectivity

may be a consequence of sensory stimulation and patient self-awareness from handling of a body region such as the neck. When examining the aforementioned areas specifically for changes in blood flow using ASL, no significant changes were observed between any of the test head conditions. This strongly suggests that the observed increased functional connectivity were not a result of altered blood flow but secondary to changes in either blood volume or metabolic activity.

Our study had strengths. First, our design ensured control of confounders and provided statistical efficiency. Second, we used phase-contrast MRI blood flow measurement because of its greater sensitivity compared to ultrasonography (which would not have been able to adequately penetrate through bone to probe the vessels we wanted to measure), and because it is considered the criterion standard for both diagnosis of vertebrobasilar artery stroke and quantifying blood flow.[39, 65-68] Estimates of repeatability of flow measures were made in preliminary work by quantifying vertebral artery flow in a single healthy participant twice over a two-month interval.[29]

A limitation of the study was the restriction of analysis to a time (average 115 seconds) following the test maneuvers. Real time measures currently are technically not feasible and transient effects of various neck positions on vertebral artery and cerebrovascular hemodynamics may have been missed. Post-maneuver analysis makes comparisons with other real-time studies challenging. Thus, the results only describe post-procedural effects and cannot be generalized to the possible effects occurring during the test maneuvers. However, it is notable that to be clinically relevant, sustained changes would likely be required that would extend into the sampled time interval of this study. Moreover, the possibility that neck pain participants exist who exhibit idiosyncratic responses cannot be excluded. To date, no such mechanisms have been measured and reported in the literature.

In conclusion, we found no significant change in blood flow in the posterior cerebrum or cerebellum in chronic neck pain participants after maximum head rotation and cervical manipulation. In addition, we found no ~~significant nor~~ clinically meaningful changes in the blood flow or velocity in the vertebral arteries before-after head positional change and spinal manipulation. Our study adds to a growing body of knowledge regarding the impact of head position and cervical manipulation on vascular and neural activity in patients with neck pain. It is the first study to measure cerebral blood flow, vertebral artery blood flow and velocity in patients undergoing neck manipulation for neck pain. Our study does not support the hypothesis that neck manipulation or neck rotation are associated with vasospasm of the vertebral artery.

Patient and Public Involvement

Patients and public were not involved in the development, design, recruitment and randomization of this study.

Author Contributions

Concept development (provided idea for the research): NM, JT, GW, SM. Design (planned the methods to generate the results): NM, JT, MN, GW. Supervision (provided oversight, responsible for organization and implementation, writing of the manuscript): NM, SM, JT, GW, PC. Data collection/processing (responsible for experiments, patient management, organization, or reporting data): NM, SM, MB, MN. Analysis/interpretation (responsible for statistical analysis, evaluation, and presentation of the results): NM, MB, SM, JT, GW, MN. Literature search (performed the literature search): NM. Writing (responsible for writing a substantive part of the manuscript): NM, SM. Critical review (revised manuscript for intellectual content, this does not relate to spelling and grammar checking): NM, SM, PC, MN, GW, JT.

Competing Interest Statement

Conflicts of interest were reported for this study include the following: Dr. Triano is an occasional lecturer on behalf of NCMIC and CCPA, and Dr. Noseworthy received an honorarium for lecture on behalf of Bayer. No other conflicts were reported.

Funding Statement

Partial financial support was provided by the Canadian Chiropractic Protective Association and NCMIC Research Foundation and Canadian Memorial Chiropractic College.

Data Sharing Statement

No additional data are available.

Acknowledgments

Appreciation is extended to MRI technologists at the Imaging Research Center for their assistance in data capture and for all of their support and effort with this study. Similarly, thanks are due to the Imaging Research Center at St Joseph's Healthcare and Dr. Shanker Nesathurai and the Department of Physical Medicine & Rehabilitation at St Joseph's Hospital for their ongoing support in this project. Special thanks are given to Dr. Sheilah Hogg-Johnson for her timely help with this work's statistical analysis.

References

1. Albuquerque FC, Hu YC, Dashti SR, Abla AA, Clark JC, Alkire B, et al. Craniocervical arterial dissections as sequelae of chiropractic manipulation: patterns of injury and management. *J Neurosurg* 2011; **115**(6):1197-205.
2. Ernst E. Manipulation of the cervical spine: a systematic review of case reports of serious adverse events, 1995-2001. *Med J Aust* 2002; **176**(8):376-80.
3. Norris JW, Beletsky V, Nadareishvili ZG. Sudden neck movement and cervical artery dissection. The Canadian Stroke Consortium. *CMAJ* 2000; **163**(1):38-40.
4. Cassidy JD, Boyle E, Côté P, He Y, Hogg-Johnson S, Silver FL, Bondy SJ. Risk of vertebrobasilar stroke and chiropractic care: results of a population-based case-control and case-crossover study. *Spine* 2008; **33**(4 Suppl):S176-83.
5. Boyle E, Côté P, Grier AR, Cassidy J. D. Examining vertebrobasilar artery stroke in two Canadian provinces. *Spine* 2008; **33**(4 Suppl):S170-5.
6. Côté P, Wong JJ, Sutton D, Shearer HM, Mior S, Randhawa K, et al. Management of neck pain and associated disorders: A clinical practice guideline from the Ontario Protocol for Traffic Injury Management (OPTIMa) Collaboration. *Eur Spine J* 2016; **25**(7):2000-22.
7. Kjaer P, Kongsted A, Hartvigsen J, Isenberg-Jørgensen A, Schiøttz-Christensen B, Søborg B, et al. National clinical guidelines for non-surgical treatment of patients with recent onset neck pain or cervical radiculopathy. *Eur Spine J* 2017; **26**(9):2242-57.
8. Hurwitz EL, Coulter ID, Adams AH, Genovese BJ, Shekelle PG. Use of chiropractic services from 1985 through 1991 in the United States and Canada. *Am J Public Health* 1998; **88**(5):771-6.
9. Hurwitz EL, Chiang LM. A comparative analysis of chiropractic and general practitioner patients in North America: findings from the joint Canada/United States Survey of Health, 2002-03. *BMC Health Serv Res* 2006; **6**:49.
10. Haldeman S, Carey P, Townsend M, Papadopoulos C. Arterial dissections following cervical manipulation: the chiropractic experience. *CMAJ* 2001; **165**(7):905-6.
11. Patijn J. Complications in manual medicine: A review of the literature. *J Manual Med* 1991; **6**:89-92.
12. Smith RA, Estridge MN. Neurological complications of head and neck manipulations: A report of two cases. *JAMA* 1962; **182**(5):528-31.
13. Frisoni GB, Anzola GP. Vertebrobasilar ischemia after neck motion. *Stroke* 1991; **22**(11):1452-60.
14. Mann T, Refshauge KM. Causes of complications from cervical spine manipulation. *Aust J Physiother* 2001; **47**(4):255-66.
15. Schmitt HP. Anatomical structure of the cervical spine with reference to pathology of manipulation complications. *Man Med* 1991; **6**:93-101.
16. Mitchell J. Vertebral Artery Blood flow Velocity Changes Associated with Cervical Spine rotation: A Meta-Analysis of the Evidence with implications for Professional Practice. *J Manipulative Physiol Ther* 2009; **17**(1):46-57.
17. Kawchuk GN, Jhangri GS, Hurwitz EL, Wynd S, Haldeman S, Hill MD. The relation between the spatial distribution of vertebral artery compromise and exposure to cervical manipulation. *J Neurol* 2008; **255**(3):371-7.
18. Herzog W, Leonard TR, Symons B, Tang C, Wuest S. Vertebral artery strains during high-speed, low amplitude cervical spinal manipulation. J Electromyogr Kinesiol 2012;**22**(5):740-6.
19. Dunne JW, Conacher GN, Khangure M, Harper CG. Dissecting aneurysms of the vertebral arteries following manipulation: a case report. *Neuro Neurosurg Psych* 1987; **50**:349-353.
20. Arnold C, Bourassa T, Langer T, Stoneham G. Doppler studies evaluating the effect of a physical therapy screening protocol on vertebral artery blood flow. *Man Ther* 2004; **9**:13–21.
21. Licht PB, Christensen HW, Højgaard P, Marving J. Vertebral artery flow and spinal manipulation: a randomized, controlled and observer-blinded study. *J Manipulative Physiol Ther* 1998; **21**(3):141-4.
22. Mitchell J. Changes in vertebral artery blood flow following rotation of the cervical spine. *J Manipulative Physiol Ther* 2003; **26**:347– 351.
23. Mitchell J, Keene D, Dyson C, Harvey L, Pruvey C, Phillips R. Is cervical spine rotation, as used in the standard vertebrobasilar insufficiency test, associated with a measurable change in intracranial vertebral artery blood flow? *Man Ther* 2004; **9**:220–227.

24. Mitchell J, Kramschuster K. Real-time ultrasound measurements of changes in suboccipital vertebral artery diameter and blood flow velocity associated with cervical spine rotation. *Physiother Res Internat* 2009; **13**:241–254.
25. Rivett DA, Sharples KJ, Milburn PD. Effect of pre-manipulative tests on vertebral artery and internal carotid artery blood flow: A pilot study. *J Manipulative Physiol Ther* 1999; **22**:368–375.
26. Rivett DA, Sharples KJ, Milburn PD. Reliability of ultrasonograpahic measurement of vertebral artery blood flow. *NZ J Physiother* 2003; **31**:119–128.
27. Rossitti S, Volkmann R, Lofgren J. Changes of blood flow velocity in the vertebro-basilar circulation during rotation of the head in the normal human. *Biomech Sem* 1992; **6**:92–99.
28. Stevens A. Functional Doppler sonography of the vertebral artery and some considerations about manual techniques. *Man Med* 1991; **6**:102–105.
29. Quesnele JJ, Triano JJ, Noseworthy MD, Wells GD. Changes in vertebral artery blood flow following various head positions and cervical spine manipulation. *J Manipulative Physiol Ther* 2014; **37**(1):22-31.
30. Erhardt JW, Windsor BA, Kerry R, Hoekstra C, Powell DW, Porter-Hoke A, Taylor A. The immediate effect of atlanto-axial high velocity thrust techniques on blood flow in the vertebral artery: A randomized controlled trial. *Man Ther* 2015; **20**(4):614-22.
31. Bakhtadze MA, Vernon H, Karalkin AV, Pasha SP, Tomashevskiy IO, Soave D. Cerebral perfusion in patients with chronic neck and upper back pain: preliminary observations. *J Manipulative Physiol Ther* 2012; **35**:76-85.
32. Barrett KE, Ganong WF. Ganong's Review of Medical Physiology 25th ed. New York, NY: McGraw-Hill Medical, 2015.
33. Guzman J, Haldeman S, Carroll LJ, Carragee EJ, Hurwitz EL, Peloso P, et al. Bone and Joint Decade 2000-2010 Task Force on Neck Pain and Its Associated Disorders. Clinical practice implications of the Bone and Joint Decade 2000-2010 Task Force on Neck Pain and Its Associated Disorders: from concepts and findings to recommendations. *Spine* 2008; **33**(4 Suppl):S199-213.
34. Cambridge E, Triano J, Ross K, Abbott M. Comparison of force development strategies of spinal manipulation used for thoracic pain. *Man Ther* 2012; **17**:241-245.
35. Descarreaux M, Dugas C, Raymond J, Normand M. Kinetic analysis of expertise in spinal manipulative therapy using an instrumented manikin. *J Chiropr Med* 2005; **4**:53–60.
36. Cohen E, Triano JJ, McGregor M, Papakyriakou M. Biomechanical performance of spinal manipulation therapy by newly trained vs. practicing providers: does experience transfer to unfamiliar procedures? *J Manipulative Physiol Ther* 1995; **18**:347-52.
37. Triano J, Rogers C, Combs S, Potts D, Sorrels K. Developing skilled performance of lumbar spine manipulation. *J Manipulative Physiol Ther* 2002; **25**:353-61.
38. Triano J, Rogers C, Combs S, Potts D, Sorrels K. Quantitative feedback versus standard training for cervical and thoracic manipulation. *J Manipulative Physiol Ther* 2003; **26**:131-8.
39. Ho SS, Chan YL, Yeung DK, Metreweli C. Blood flow volume quantification of cerebral ischemia: comparison of three noninvasive imaging techniques of carotid and vertebral arteries. *Am J Roentgenol* 2002; **178**(3):551-6.
40. Lotz J, Meier C, Leppert A, Galanski M. Cardiovascular flow measurement with phase-contrast MR imaging: basic facts and implementation. *Radiographics* 2002; **22**(3):651-71.
41. Ioannidis JP, Evans SJ, Gøtzsche PC, O'Neill RT, Altman DG, Schulz K, Moher D.; CONSORT Group. Better reporting of harms in randomized trials: an extension of the CONSORT statement. *Ann Intern Med* 2004; **141**(10):781-8.
42. Parkes LM, Rashid W, Chard DT, Tofts PS. Normal cerebral perfusion measurements using arterial spin labeling: reproducibility, stability, and age and gender effects. *Magn Reson Med* 2004; **51**(4):736-43.
43. Cohen J. Statistical power analysis for the behavioral sciences 2nd ed. New Jersey: Lawrence Erlbaum Associates, Publishers, 1988.
44. Cox RW. AFNI: software for analysis and visualization of functional magnetic resonance neuroimages. *Comput Biomed Res* 1996; **29**(3):162-73.
45. Holmes CJ, Hoge R, Collins L, Woods R, Toga AW, Evans AC. Enhancement of MR images using registration for signal averaging. *J Comput Assist Tomogr* 1998; **22**(2):324-33.

46. Jang JH, Kim JH, Jung WH, Choi JS, Jung MH, Lee JM, et al. Functional connectivity in fronto-subcortical circuitry during the resting state in obsessive-compulsive disorder. *Neurosci Lett* 2010; **474**(3):158-62.
47. Wang J, Wang L, Zang Y, Yang H, Tang H, Gong Q, et al. Parcellation-dependent small-world brain functional networks: a resting-state fMRI study. *Hum Brain Mapp* 2009; **30**(5):1511-23.
48. Fleiss JL. The Design and Analysis of Clinical Experiments. Wiley:Toronto 1986.
49. Yurdakul M, Tola M. Doppler criteria for identifying proximal vertebral artery stenosis of 50% or more. *J Ultrasound Med* 2011; **30**(2):163-8.
50. Licht PB, Christensen HW, Højgaard P, Marving J. Vertebral artery flow and spinal manipulation: a randomized, controlled and observer-blinded study. *J Manipulative Physiol Ther* 1998; **21**(3):141-4.
51. Bendick PJ, Glover JL. Vertebrobasilar insufficiency: evaluation by quantitative duplex flow measurements. A preliminary report. J Vasc Surg 1987;**5**(4):594-600.
52. Seidel E, Eicke BM, Tettenborn B, Krummenauer F. Reference values for vertebral artery flow volume by duplex sonography in young and elderly adults. Stroke 1999;**30**(12):2692-6.
53. Arnetoli G, Amadori A, Stefani P, Nuzzaci G. Sonography of vertebral arteries in De Kleyn's position in subjects and in patients with vertebrobasilar transient ischemic attacks. Angiology 1989; **40**(8):716-20.
54. Sakaguchi M, Kitagawa K, Hougaku H, Hashimoto H, Nagai Y, Yamagami H, Ohtsuki T, Oku N, Hashikawa K, Matsushita K, Matsumoto M, Hori M. Mechanical compression of the extracranial vertebral artery during neck rotation. Neurology 2003; **61**(6):845-7.
55. Debette S, Leys D. Cervical-artery dissections: predisposing factors, diagnosis, and outcome. *Lancet Neurol* 2009; **8**(7):668-78.
56. Senter HJ, Sarwar M. Nontraumatic dissecting aneurysm of the vertebral artery. Case report. *J Neurosurg* 1982; **56**(1):128-30.
57. Sherman DG, Hart RG, Easton JD. Abrupt change in head position and cerebral infarction. *Stroke* 1981; **12**(1):2-6.
58. Haldeman S, Kohlbeck FJ, McGregor M. Unpredictability of cerebrovascular ischemia associated with cervical spine manipulation therapy: a review of sixty-four cases after cervical spine manipulation. *Spine* 2002; **27**(1):49-55.
59. Josien E. Extracranial vertebral artery dissection: nine cases. *J Neurol* 1992; **239**(6):327-30.
60. Okawara S, Nibbelink D. Vertebral artery occlusion following hyperextension and rotation of the head. *Stroke* 1974; **5**(5):640-2.
61. Heinen, S. J. & Keller, E. L. The function of the cerebellar uvula in monkey during optokinetic and pursuit eye movements: single-unit responses and lesion effects. *Exp Brain Res* 1996; **110**(1):1-14.
62. Kanwisher, N. & Yovel, G. The fusiform face area: a cortical region specialized for the perception of faces. *Philos Trans R Soc Lond B Biol Sci* 2006; **361**(1476):2109-28.
63. Lamm, C., Windischberger, C., Leodolter, U., Moser, E. & Bauer, H. Evidence for premotor cortex activity during dynamic visuospatial imagery from single-trial functional magnetic resonance imaging and event-related slow cortical potentials. *Neuroimage* 2001; **14**(2):268-83.
64. Cavanna, A. E. & Trimble, M. R. The precuneus: a review of its functional anatomy and behavioural correlates. *Brain* 2006; **129**(3):564-83.
65. Sengelhoff C, Nebelsieck J, Nassenstein I, Maintz D, Nabavi DG, Kuhlenbaeumer G, et al. Neurosonographical follow-up in patients with spontaneous cervical artery dissection. *Neurol Res* 2008; **30**(7):687-9.
66. Dittrich R. Dziewas R, Ritter MA, Kloska SP, Bachmann R, Nassenstein I, et al. Negative ultrasound findings in patients with cervical artery dissection. Negative ultrasound in CAD. *J Neurol* 2006; **253**(4):424-33.
67. Nebelsieck J, Sengelhoff C, Nassenstein I, Maintz D, Kuhlenbäumer G, Nabavi DG, et al. Sensitivity of neurovascular ultrasound for the detection of spontaneous cervical artery dissection. *J Clin Neurosci* 2009; **16**(1):79-82.
68. Oktar SO, Yücel C, Karaosmanoglu D, Akkan K, Ozdemir H, Tokgoz N, et al. Blood-flow volume quantification in internal carotid and vertebral arteries: comparison of 3 different ultrasound techniques with phase-contrast MR imaging. *Am J Neuroradiol* 2006; **27**(2):363-9.

Table 1. *Baseline participant characteristics*

Participant characteristics	Mean (SD)
Age (years)	32.05 (12.5)
Sex (n, %)	
Female	14 (70%)
Height (cm)	169 (8.9)
Weight (kg)	69.6 (18.7)
NDI	13 (6.4)
NRS neck	4.7 (2.1)
Duration of complaint (years)	5.3 (5.7)
NRS headache	3 (2.6)

SD= standard deviation, cm= centimeters, kg- kilograms, NDI= Neck Disability Index, NRS= numerical rating scale

Table 2. *Mean cerebral and cerebellar perfusion (in milliliters per 100 grams of tissue per minute) for each condition as determined by arterial spin labeling technique*

Cerebrum and cerebellum regions of interest (mL/100g/min), mean (95% CI)	Neutral	Manipulation	Maximum rotation	F	p-value
Uvula	79.2 (90.2 – 68.2)	80.1 (91.8 - 68.4)	79.1 (90.3 – 67.8)	0.0064	0.9364
Cerebellar Tonsils	69.2 (77.8 – 60.6)	66.8 (74.1 – 59.6)	68.2 (75.8 – 60.6)	0.0632	0.8035
L Fusiform Gyrus/L Middle Temporal Gyrus	80.7 (92.8 – 68.6)	78.0 (89.3 – 66.7)	79.0 (89.3 – 68.8)	0.0395	0.8440
R Middle Temporal Gyrus	37.4 (43.0 – 31.8)	35.7 (40.9 – 30.5)	36.1 (42.1 – 30.1)	0.0673	0.7974
R Middle Occipital Gyrus	58.2 (79.9 – 53.1)	52.8 (60.9 – 44.8)	53.2 (61.7 – 44.7)	0.323	0.5747
Right and Left Cuneus	66.5 (79.9 – 53.1)	62.3 (73.6 – 60.0)	61.0 (73.3 – 48.4)	0.1455	0.7060
Left Precuneus	75.7 (89.4 – 61.9)	74.9 (86.8 – 62.9)	64.1 (74.3 – 53.9)	0.7736	0.3872
Left Cuneus, Left Middle Occipital Gyrus	49.4 (57. 1 – 41.6)	47.6 (55.1 – 40.2)	46.9 (55.0 – 38.8)	0.0742	0.7875

CI= Confidence interval

Table 3. *ANOVA statistics comparing mean flow and velocity across head positions for ipsilateral and contralateral side, controlling for subject.*

	Neutral Mean (SD)	Cervical Manipulation Mean (SD)	Max Rotation Mean (SD)	$F_{2,26}$*	p-value
Velocity (cm/s) Ipsilateral	15.6 (3.6)	14.7 (3.6)	15.2 (3.9)	2.43	0.11

Velocity (cm/s) Contralateral	15.6 (3.3)	14.4 (2.6)	14.4 (3.2)	12.07	0.0002
Flow (mL/s) Ipsilateral	1.85 (0.76)	1.85 (0.80)	1.77 (0.62)	0.50	0.61
Flow (mL/s) Contralateral	1.88 (0.86)	1.85 (0.82)	1.62 (0.76)	6.94	0.004

*F test from two-way ANOVA including subject and condition as factors

Table 4. *Mean paired differences between baseline and interventions.*

	Neutral – Cervical Manipulation, Mean (95% CI)	Neutral – Max Rotation, Mean (95% CI)	Cervical Manipulation – Max Rotation, Mean (95% CI)
Velocity (cm/s) Ipsilateral	0.85 (0.05, 1.66)	0.41 (-0.37, 1.18)	-0.45 (-1.36, 0.47)
Velocity (cm/s) Contralateral	1.15 (0.40, 1.90)	1.18 (0.77, 1.59)	0.03 (-0.53, 0.59)
Flow (mL/s) Ipsilateral	-0.003 (-0.18, 0.17)	0.07 (-0.11, 0.26)	0.08 (-0.13, 0.28)
Flow (mL/s) Contralateral	0.03 (-0.13, 0.19)	0.26 (0.11, 0.41)	0.23 (0.04, 0.42)

Table 5. *Order effect on vertebral artery hemodynamics.*

	Order by Position Interaction	Neutral – Cervical Manipulation	Neutral – Max Rotation	Cervical Manipulation – Max Rotation
	$F_{2,24}$, p-value	t_{12}, p-value	t_{12}, p-value	t_{12}, p-value
Velocity (cm/s) Ipsilateral	0.70, 0.51	-0.17, 0.87	1.01, 0.33	1.01, 0.33
Velocity (cm/s) Contralateral	4.33, 0.02	-2.23, 0.045	-0.69, 0.51	2.47, 0.030
Flow (mL/s) Ipsilateral	0.11, 0.90	-0.03, 0.98	0.40, 0.70	0.39, 0.70
Flow (mL/s) Contralateral	0.06, 0.95	0.24, 0.81	0.37, 0.72	0.08, 0.93

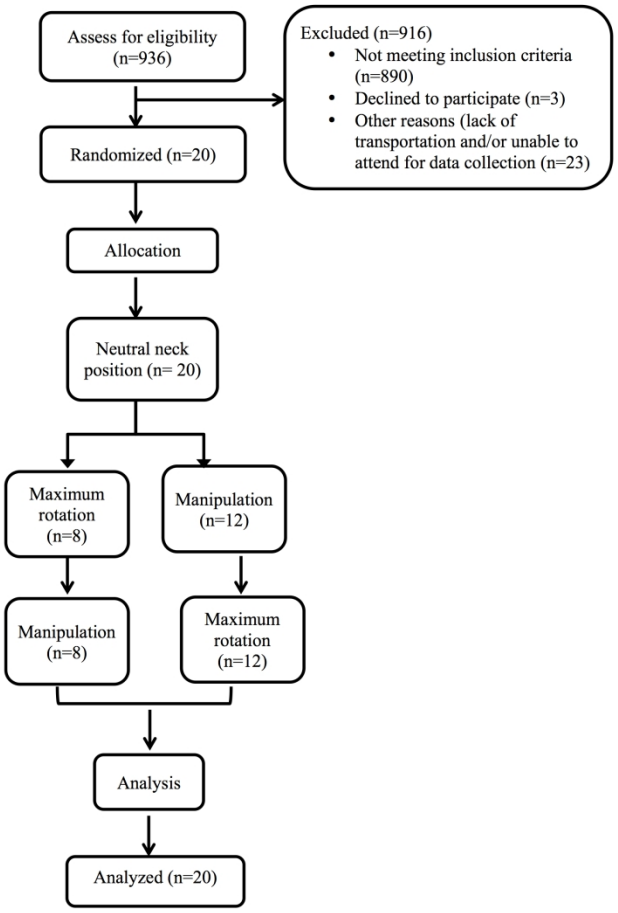

Figure 1. *Protocol flow and method process*

Figure 1. Flow chart

215x279mm (300 x 300 DPI)

ONLINE SUPPLEMENT
SUPPLEMENTAL MATERIAL

A Cross-over Randomized Trial of the Effect of Cervical Manipulation on Vertebral Artery and Cerebral Hemodynamics in Neck Pain Patients

Nicholas Moser DC, Silvano Mior PhD, Michael D. Noseworthy PhD, Pierre Côté PhD, Greg Wells PhD, Michael Behr MSc, John J. Triano PhD

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Supplemental Table

Table I. *Analysis of resting state fMRI indicating coordinates from areas of significantly increased functional connectivity, post-procedures.*

Number of Voxels	CM_X	CM_Y	CM_Z	PV_X	PV_Y	PV_Z	Side	Location	P Values
54	-56.2	-32.2	+7.2	-47.2	-46.5	+17.0	L	TG	0.0027
53	-5.1	-59.4	-28.0	-5.2	-60.5	-32.0	L, R	Uvula	0.0003
52	+23.8	+10.8	+30.4	+26.2	+2.5	+34.5	R	FG	0.000035
48	-6.2	-38.6	-38.1	-5.2	-36.0	-39.0	L, R	CT	0.0014
47	-45.3	-57.7	-14.4	-43.8	-64.0	-28.5	L	FfG, MTG	0.0001
43	+33.4	-46.2	+25.6	+36.8	-39.5	+27.5	R	MTG	0.00235
37	+31.6	-75.2	+6.2	+36.8	-67.5	+3.0	R	MOG	0.0047
32	-17.2	-19.3	+51.0	-12.2	-22.0	+48.5	L	PG, MCG	0.00165
31	+22.0	-56.1	-35.4	+22.8	-57.0	-39.0	R	CT	0.00047
31	-28.0	-39.2	+30.7	-29.8	-36.0	+31.0	L	IPL	0.0017
25	+55.8	-12.7	-3.2	+57.8	-8.0	-4.0	R	STG	0.0031
24	+1.4	-91.3	+8.9	+5.2	-95.5	+3.0	L, R	Cuneus	0.0003
24	-8.5	-76.1	+44.3	-5.2	-78.0	+48.5	L	Precuneus	0.0127
20	-10.7	+59.2	+2.4	-12.2	+65.5	-0.5	L	MSFG	0.00345
20	-21.0	-77.0	+7.5	-22.8	-74.5	+6.5	L	Cuneus, MOG	0.00205

CM: centre of mass. PV: peak velocity. X, Y, Z: coordinates in the Colin N_27 atlas. L, R: left and right. TG: temporal gyrus. FG: frontal gyrus. CT: cerebellar tonsil. FfG: fusiform gyrus. MTG: middle temporal gyrus. MOG: middle occipital gyrus. PG: precentral gyrus. MCG: middle cingulate gyrus. IPL: inferior parietal lobe. STG: superior temporal gyrus. MSFG: middle, superior frontal gyrus.

Supplemental Figures

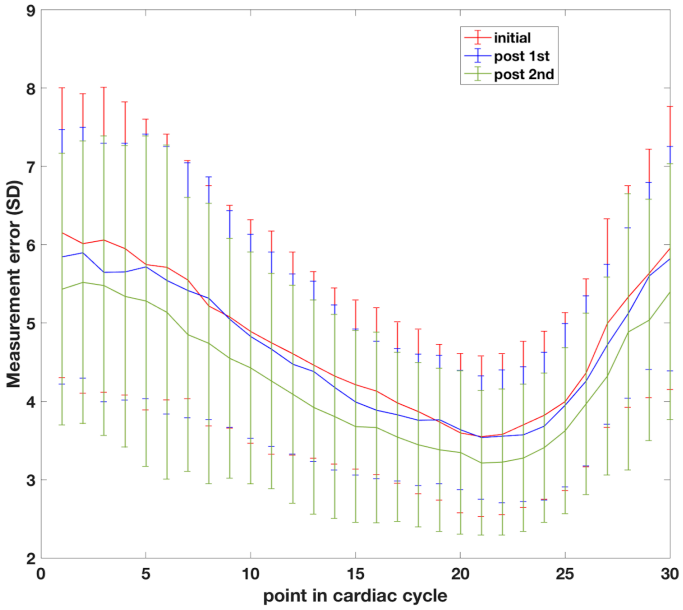

Figure I. *Left vertebral artery mean blood flow variability in the cardiac cycle between neutral (initial), cervical manipulation (post 1st) and maximum rotation (post 2nd). Bars represent the standard deviation of the mean errors at each point.*

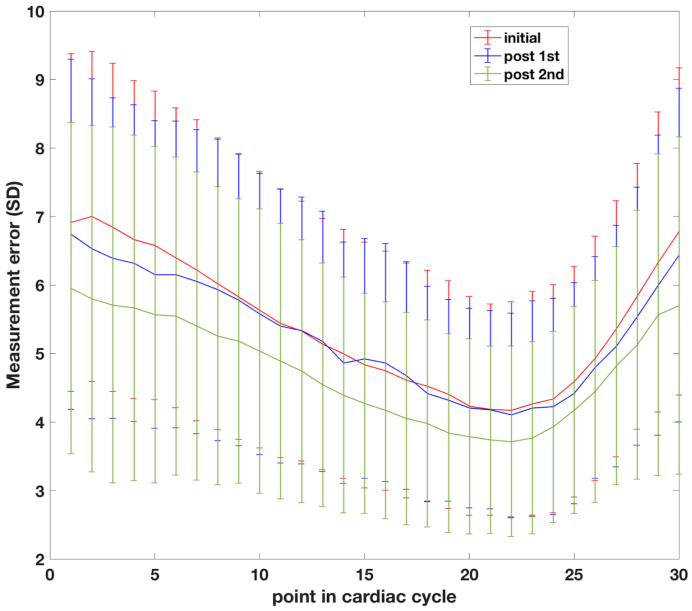

Figure II. *Right vertebral artery mean blood flow variability in the cardiac cycle between neutral (initial), cervical manipulation (post 1st) and maximum rotation (post 2nd). Bars represent the standard deviation of the mean errors at each point.*

1

Table 1: CONSORT 2010 checklist of information to include when reporting a within-person randomised trial. For within-person trials, a group is the set of participants' body sites that was allocated a particular intervention.

Section/Topic	Item no.	Standard CONSORT Checklist item	Extension for within-person trials	Page no.
Title and abstract				
	1a	Identification as a randomised trial in the title	Identification as a within-person randomised trial in the title	1
	1b	Structured summary of trial design, methods, results, and conclusions (for specific guidance see CONSORT for abstracts[3])	Specify a within-person design and report all information outlined in table 2	2
Introduction				
Background and objectives	2a	Scientific background and explanation of rationale		3
	2b	Specific objectives or hypotheses		3
Methods				
Trial design	3a	Description of trial design (such as parallel, factorial) including allocation ratio	Rationale for using a within-person design and identification of body sites	3
	3b	Important changes to methods after trial commencement (such as eligibility criteria), with reasons		3
Participants	4a	Eligibility criteria for participants	Eligibility criteria for body sites	4
	4b	Settings and locations where the data were collected		4
Interventions	5	The interventions for each group with sufficient details to allow replication, including how and when they were actually administered	Whether interventions were given sequentially or concurrently	4
Outcomes	6a	Completely defined pre-specified primary and secondary outcome measures, including how and when	Outcomes should be clearly defined as per-site or per-	5

From: Pandis N, Chung B, Scherer RW, Elbourne D, Altman DG. CONSORT 2010 statement: extension checklist for reporting within person randomised trials. BMJ. 2017;357.

Section/Topic	Item no.	Standard CONSORT Checklist item	Extension for within-person trials	Page no.
		they were assessed	person	
	6b	Any changes to trial outcomes after the trial commenced, with reasons		5
Sample size	7a	How sample size was determined	Report the correlation between body sites	6
	7b	When applicable, explanation of any interim analyses and stopping guidelines		N/A
Randomisation:				
Sequence generation	8a	Method used to generate the random allocation sequence		4
	8b	Type of randomisation; details of any restriction (such as blocking and block size)	Methods used to determine the allocation sequence of body sites and treatments within an individual (e.g. how first site to be treated was decided)	4
Allocation concealment mechanism	9	Mechanism used to implement the random allocation sequence (such as sequentially numbered containers), describing any steps taken to conceal the sequence until interventions were assigned		4
Implement-ation	10	Who generated the random allocation sequence, who enrolled participants, and who assigned participants to interventions	Replaced by 10a	4
	10a		Who generated the random allocation sequence, who enrolled participants, and who assigned body sites to interventions	4
Blinding (masking)	11a	If done, who was blinded after assignment to interventions (for example, participants, care providers, those assessing outcomes) and how		4
	11b	If relevant, description of the similarity of interventions		N/A

From: Pandis N, Chung B, Scherer RW, Elbourne D, Altman DG. CONSORT 2010 statement: extension checklist for reporting within person randomised trials. BMJ. 2017;357.

Section/Topic	Item no.	Standard CONSORT Checklist item	Extension for within-person trials	Page no.
Statistical methods	12a	Statistical methods used to compare groups for primary and secondary outcomes	Statistical methods appropriate for within-person design	6
	12b	Methods for additional analyses, such as subgroup analyses and adjusted analyses		6
Results				
Participant flow (a diagram is strongly recommended)	13a	For each group, the numbers of participants who were randomly assigned, received intended treatment, and were analysed for the primary outcome	Number of participants and number of body sites at each stage [See Figure 1]	6
	13b	For each group, losses and exclusions after randomisation, together with reasons	Number of participants and number of body sites lost or excluded at each stage, with reasons	7
Recruitment	14a	Dates defining the periods of recruitment and follow-up		6
	14b	Why the trial ended or was stopped		N/A
Baseline data	15	A table showing baseline demographic and clinical characteristics for each group	Baseline characteristics for site and individual participants as applicable	13
Numbers analysed	16	For each group, number of participants (denominator) included in each analysis and whether the analysis was by original assigned groups	Number of randomised body sites in each group included in each analysis	7
Outcomes and estimation	17a	For each primary and secondary outcome, results for each group, and the estimated effect size and its precision (such as 95% confidence interval)	Observed correlation between body sites for continuous outcomes and tabulation of paired results for binary outcomes	7
	17b	For binary outcomes, presentation of both absolute and relative effect sizes is recommended		N/A
Ancillary analyses	18	Results of any other analyses performed, including subgroup analyses and adjusted analyses, distinguishing pre-specified from exploratory		14

From: Pandis N, Chung B, Scherer RW, Elbourne D, Altman DG. CONSORT 2010 statement: extension checklist for reporting within person randomised trials. BMJ. 2017;357.

Section/Topic	Item no.	Standard CONSORT Checklist item	Extension for within-person trials	Page no.
Harms	19	All important harms or unintended effects in each group (for specific guidance see CONSORT for harms)	Harms or unintended effects reported by participant and by body site	7
Discussion				
Limitations	20	Trial limitations, addressing sources of potential bias, imprecision, and, if relevant, multiplicity of analyses		8
Generalisability	21	Generalisability (external validity, applicability) of the trial findings		8
Interpretation	22	Interpretation consistent with results, balancing benefits and harms, and considering other relevant evidence		8
Other information				
Registration	23	Registration number and name of trial registry		3
Protocol	24	Where the full trial protocol can be accessed, if available		N/A
Funding	25	Sources of funding and other support (such as supply of drugs), role of funders		9

From: Pandis N, Chung B, Scherer RW, Elbourne D, Altman DG. CONSORT 2010 statement: extension checklist for reporting within person randomised trials. BMJ. 2017;357.