Accepted Manuscript

Effects of aerobic training, resistance training, or both on brain-derived neurotrophic factor in adolescents with obesity: The hearty randomized controlled trial

Gary S. Goldfield, Glen P. Kenny, Denis Prud'Homme, Martin Holcik, Angela S. Alberga, Margaret Fahnestock, Jameason D. Cameron, Steve Doucette, Stasia Hadjiyannakis, Heather Tulloch, Mark S. Tremblay, Jeremy Walsh, Eva Guerin, Katie E. Gunnell, Amedeo D'Angiulli, Ronald J. Sigal

PII: S0031-9384(18)30207-5
Reference: PHB 12177
To appear in: Physiology & Behavior
Received date: 12 December 2017
Revised date: 3 April 2018
Accepted date: 17 April 2018

Please cite this article as: Gary S. Goldfield, Glen P. Kenny, Denis Prud'Homme, Martin Holcik, Angela S. Alberga, Margaret Fahnestock, Jameason D. Cameron, Steve Doucette, Stasia Hadjiyannakis, Heather Tulloch, Mark S. Tremblay, Jeremy Walsh, Eva Guerin, Katie E. Gunnell, Amedeo D'Angiulli, Ronald J. Sigal, Effects of aerobic training, resistance training, or both on brain-derived neurotrophic factor in adolescents with obesity: The hearty randomized controlled trial. The address for the corresponding author was captured as affiliation for all authors. Please check if appropriate. Phb(2018), doi:10.1016/j.physbeh.2018.04.026

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.
Effects of Aerobic Training, Resistance Training, or Both on Brain-Derived Neurotrophic Factor in Adolescents with Obesity: the HEARTY randomized controlled trial

Gary S. Goldfield, PhD1,2,3,4; Glen P. Kenny, PhD3,6; Denis Prud’homme, MD, MSc5; Martin Holcik, Ph.D7; Angela S. Alberga, Ph.D8; Margaret Fahnestock, Ph.D9; Jameason D. Cameron, Ph.D1; Steve Doucette, MSc10; Stasia Hadjiyannakis, MD11; Heather Tulloch, Ph.D12; Mark S. Tremblay, Ph.D1,2; Jeremy Walsh, Ph.D1; Eva Guerin, Ph.D5; Katie E. Gunnell, Ph.D13; Amedeo D’Angiulli, Ph.D14; Ronald J. Sigal, MD, MPH1,6,15

1 Healthy Active Living and Obesity Research Group, Children’s Hospital of Eastern Ontario Research Institute, Ottawa, ON, Canada

2 Department of Pediatrics, University of Ottawa, Ottawa, ON, Canada

3 School of Human Kinetics, University of Ottawa, Ottawa, ON Canada

4 School of Psychology, University of Ottawa, Ottawa, ON Canada

5 Institut du Savoir Montfort, Ottawa, ON, Canada

6 Clinical Epidemiology Program, Ottawa Hospital Research Institute, Ottawa, ON, Canada

7 Department of Health Sciences, Carleton University, Ottawa, ON, Canada

8 Department of Kinesiology, Concordia University, Montreal, QC, Canada

9 Department of Psychiatry & Behavioural Neurosciences, McMaster University, Hamilton, ON, Canada

10 Department of Community Health & Epidemiology, Dalhousie University, Halifax, NS, Canada

11 Centre for Healthy Active Living, Children’s Hospital of Eastern Ontario Research Institute Ottawa, ON, Canada

12 Prevention & Rehabilitation Centre, University of Ottawa Heart Institute, Ottawa, ON, Canada

13 Department of Psychology, Carleton University, Ottawa, ON Canada

14 Department of Neuroscience, Carleton University, Ottawa, ON Canada

15 Departments of Medicine, Cardiac Sciences and Community Health Sciences Cumming School of Medicine, University of Calgary, Calgary, AB, Canada

Trial Registration: ClinicalTrials.Gov NCT00195858 http://clinicaltrials.gov/show/NCT00195858,

September 12, 2005 ( Funded by the Canadian Institutes of Health Research).
Address Correspondence to:

Gary Goldfield, Ph.D. C. Psych.
Associate Professor, Department of Pediatrics, University of Ottawa
Senior Scientist, Healthy Active Living & Obesity (HALO) Research Group
CHEO Research Institute
401 Smyth Rd.
Ottawa, ON K1H 8L1
Phone: 613-737-7600, Ext. 3288
Fax: 613-738-4800
Email: ggoldfield@cheo.on.ca
ABSTRACT

Brain derived neurotrophic factor (BDNF) is a protein that plays a critical role in modulating cognition in animals and humans. Aerobic exercise often increases BDNF in adults, but effects of this exercise modality and others among adolescents remain uncertain. This study examined the effects of aerobic training, resistance training, and combined training on resting serum BDNF levels in adolescents with overweight and obesity. After a 4-week pre-randomization treatment, 304 post-pubertal, adolescents with overweight or obesity (70% females) aged 14-18 years were randomized to one of four groups for 22 weeks: aerobic training (N=75), resistance training (N=78), combined aerobic and resistance training (N=75), or non-exercising control (N=76). All participants received dietary counseling targeting a daily energy deficit of 250 kcal. The exercise prescription was 4 times per week, progressing to 45 minutes/session for the aerobic and resistance groups and 90 minutes/session for the combined group. Resting serum BDNF levels were measured at baseline and 6-months. Results showed that in both intention-to-treat (ITT) and per protocol (>70% adherence to prescribed sessions) analyses, there were no significant within- or between-group changes in BDNF. Findings indicate that aerobic training, resistance training or their combination did change serum BDNF levels in adolescents with overweight and obesity.

Keywords: Exercise, physical activity, obesity, BDNF, adolescents, strength training, cognition
INTRODUCTION

Obesity in childhood and adolescence has increased dramatically in recent decades.\textsuperscript{1, 2} This raises public health concern given the well documented social, psychological and medical comorbidities manifesting during childhood,\textsuperscript{3, 4} that also track into adulthood.\textsuperscript{5} Obesity in youth is also associated with diminished academic achievement\textsuperscript{6} and cognitive function, most notably executive functions such as attention and inhibitory control,\textsuperscript{7} with evidence indicating these cognitive deficits are both determinants and consequences of obesity.\textsuperscript{7} Given that adolescence is a stage of life in which the brain is still developing, especially the prefrontal cortical regions known to govern executive functioning, identifying methods of enhancing brain health during this developmental stage is important for optimizing the trajectory of cognitive development, academic achievement and vocational attainment.\textsuperscript{8}

Brain-derived neurotrophic factor (BDNF) is a protein that exerts pleiotropic effects on the brain and peripheral tissue.\textsuperscript{9} BDNF is the most abundant neurotrophin in the brain, with large concentrations found in the hippocampus, cerebral cortex, hypothalamus and cerebellum.\textsuperscript{10, 11} BDNF is well known to play a critical role in modulating cognition and activity-dependent neuroplasticity in animals\textsuperscript{12} and humans.\textsuperscript{13} BDNF is also essential for angiogenesis and strengthening neural connectivity during childhood, biological activities important for learning and memory and required for enhanced academic performance and brain health.\textsuperscript{14} The action of BDNF extends beyond the brain, as BDNF is also involved in regulating metabolic functions, such as fat oxidation and glucose uptake\textsuperscript{9, 15, 16} and is believed to be downregulated in those with obesity and type 2 diabetes.\textsuperscript{17} Importantly, BDNF can cross the blood-brain barrier in a bidirectional manner\textsuperscript{18} and can be measured peripherally in serum and plasma.

Regular physical activity or exercise is well known to play a key role in the management of pediatric obesity.\textsuperscript{19} Physical activity has also be shown to enhance cognitive development and academic
achievement in children and adolescents, including youth with obesity, as well as in adults. Although several physiological mechanisms may exist, one hypothesized mechanism for these cognitive benefits is that exercise increases BDNF levels in the brain. There is consistent evidence supporting that acute bouts of moderate to vigorous intensity aerobic exercise increase circulating BDNF in adults, with meta-analyses showing moderate effect sizes, and with longer sessions (more than 30-minutes) and greater intensity producing stronger effects. Programmed aerobic exercise of at least two-weeks in duration has been shown to lead to increases in resting peripheral BDNF levels in young (20-39 years), middle aged (30-64) and older (> 65 years) adults, although effect sizes were smaller than from acute aerobic training. Approximately 60% (11 of 18) of these aerobic exercise interventions showed no effects, highlighting the considerable variability in BDNF responses to aerobic exercise. Although the vast majority of the exercise intervention studies with measurements of peripheral BDNF have examined aerobic training in adults, a recent meta-analysis showed that the acute effects of resistance training in primarily middle- and older-aged adults were comparable to those of aerobic training. Moreover, the meta-analytic data of laboratory studies show that effects of both modalities are strengthened in individuals with higher levels of physical activity and aerobic fitness.

Comparatively less literature exists in youth, with only two studies examining effects of exercise on BDNF in adolescents. Lee et al. found that 12-weeks of thrice weekly aerobic training produced a significant (2.5 fold) increase in resting serum BDNF levels in the adolescents with obesity, but no changes were observed in those with Type-2 Diabetes Mellitus (T2DM) or in healthy weight controls. Jeon and Ha found that 12-weeks of aerobic exercise in adolescent males led to greater increases in peripheral BDNF levels and cognitive performance (working memory) than controls, with evidence of intensity-dependent effects, findings consistent with aerobic exercise trials in adults. Although encouraging, these studies in adolescents are limited by small samples, which consisted of males only,
and neither of the interventions examined resistance training, which has been shown to acutely increase serum BDNF levels in middle-aged and older adults.\textsuperscript{29} Although the studies of resistance training interventions on BDNF in adults show weaker and more equivocal effects when compared to aerobic training,\textsuperscript{10,25} most interventions were of brief duration and limited by small sample sizes. A paucity of data exists on the possible additive effects of combining aerobic and resistance training on BDNF in adolescents, although evidence for stronger effects of combined training on cognition has recently been shown in youth.\textsuperscript{33} Taken together, the effects of aerobic training, resistance training, or combined aerobic and resistance training on resting BDNF in adolescents with obesity remain uninvestigated.

The Healthy Eating Aerobic and Resistance Training in Youth (HEARTY) trial examined the effects of aerobic training, resistance exercise training, or their combination on body composition, and found that all exercise modalities reduced body fat in adolescents living with overweight and obesity.\textsuperscript{34} Using data from the HEARTY trial, the purpose of the current study was to fill a research gap by examining the effects of aerobic, resistance or combined training on resting serum BDNF levels. In the context of a limited knowledge-base, we hypothesized that 6-months of combined aerobic and resistance exercise training would elicit greater increases in resting serum BDNF levels than either exercise training modality alone or than non-exercising (diet only) controls.

**METHODS**

*Design*

This was a single blind, randomized controlled trial with a parallel group design conducted at a single centre. Once baseline testing was completed, participants entered a run-in period including supervised moderate-intensity exercise training four times weekly for four weeks, during which participants were exposed to both aerobic and resistance exercise training. To qualify for randomization, participants needed to attend at least 13 of 16 prescribed exercise sessions (\textgreater 80\%)
adherence) during this run-in phase. Participants (n=304) were randomized using a computer generated program into one of 4 groups for 22 weeks: aerobic training (aerobic, n=75), resistance training (resistance, n=78), combined aerobic and resistance training (combined, n=75), or non-exercising control (control, n=76). Outcome assessors were blinded to group assignment, ensuring unbiased measurement.

**Participants**

Of the 1285 adolescents assessed, 358 met eligibility criteria and entered the run-in phase. Subsequently, 304 were randomized with baseline resting serum BDNF data only available for 282 participants (see Figure 1). Inclusion criteria included being post-pubertal (Tanner stage IV-V)\(^{35,36}\) adolescents aged 14-18 years with body mass index (BMI) \(\geq 95^{th}\) percentile for age and sex (http://www.cdc.gov/growthcharts) and/or \(\geq 85^{th}\) percentile with an additional diabetes or cardiovascular risk factor. The vast majority (93%; 262 of 282) of participants met criteria for obesity. Exclusion criteria included free-living regular exercise more than twice weekly for over 20 minutes per session, diabetes mellitus, or any illness or disability rendering study exercise programs inadvisable or unfeasible. Physical activity accrued from school physical education classes was not an exclusion criterion. For participants taking medication that could impact body composition (metformin, oral contraceptives, stimulants), the dose was required to have been stable over the previous two months and to remain unchanged throughout the trial. More detail on rationale, design and methods can be found elsewhere.\(^{37}\) The study inclusion/exclusion criteria were reviewed by the research coordinator and informed consent of participants was obtained based on the Canadian Tri-Council Policy Statement Guidelines.\(^{38}\) For participants younger than 16 years, a parent or guardian was asked to co-sign the consent form. The HEARTY study and this report of secondary data analysis were reviewed and
approved by the Research Ethics Boards at the Children’s Hospital of Eastern Ontario and the Ottawa Hospital. The trial began in March 2005 and was completed in June 2011.

**Intervention**

All participants received counseling by a registered dietitian that was designed to promote healthy eating with a daily energy deficit of 250 kcal. In addition, participants in the three exercise groups were asked to attend gyms 4 times weekly. The Aerobic group exercised on treadmills, elliptical machines and/or bicycle ergometers. Heart rate monitors (Polar Electro Oy, Kempele, Finland) were used to adjust workloads to achieve target heart rates. Participants in the Aerobic group gradually increased duration of exercise each session, starting with 20 minutes and progressing to 45 minutes. Intensity of exercise was also progressive, whereby participants began at 65% and progressed to 85% of their pre-determined maximum heart rate. The Resistance group also progressed from 20 to 45 minutes per session, performing 7 exercises using weight machines or free weights, and progressing from 2 sets of 15 repetitions at moderate intensity to 3 sets of 8 repetitions at the maximum resistance that could be moved 8 times (8-RM). The Combined exercise group did the full aerobic training program plus the resistance training program during each session. The control group received only dietary counselling with no exercise prescription. Further details on the intervention can be obtained elsewhere.\(^{37}\)

**Setting**

Exercise training was conducted in six community-based gymnasiums. Exercise sessions were supervised by personal trainers twice weekly during the run-in phase, weekly from randomization to 3 months, and biweekly from 3 to 6 months. Personal trainers monitored attendance and exercise progression by reviewing sign-in sheets and exercise logs. Adherence to the intervention was assessed by the total number of exercise sessions attended divided by the total number of sessions prescribed.

**Measurements**
Demographic and Anthropometric Variables: A structured history and physical examinations were conducted at baseline, including demographics (age, race, ethnicity, parental education), pubertal status, measured height and weight for body mass index (BMI) calculations, and self-reported physical activity, family medical history and current medical status and medications. Waist circumference was measured at the end of normal expiration midway between the lowest rib and the top of the iliac crest using a retractable anthropometric measuring tape (Seca GmbH & Co Kg, Hamburg, Germany). Hip circumference was measured at the maximal protuberance of the buttocks. Waist-to-hip ratio was calculated as waist circumference (cm) divided by hip circumference (cm).

Blood analysis: Serum BDNF

Twelve-hour (overnight-fasting) blood samples of approximately 20 mL of venous blood were taken in the morning, from a forearm or antecubital vein and were stored in a freezer at -80°C. Samples were obtained at baseline before the run-in period and between 2 to 10 days after the last exercise session at 6-months post-intervention to avoid potentially confounding acute effects of exercise, in accordance with the main objectives of the HEARTY design. Serum samples of BDNF were diluted 75 fold into Calibrator Diluent RD6P prior to starting the assay. The ELISA was performed following the manufacturers protocol (Human Free BDNF Quantikine ELISA kit, R&D systems, Cat# DBD00) and was performed in duplicate. The sensitivity of this assay product is 20 pg/mL with a range of 62.5-4,000 pg/mL. All standard curves were linear within the range used for the analysis with a strong corresponding correlation of coefficient (r-square >0.99).

Statistical Analysis

Baseline demographics were summarized as means and standard deviations, and between group differences were examined using independent t-tests for continuous data. Categorical data were summarized as frequencies and percentages, and group differences at baseline were examined using chi-
square tests. Using an intention-to-treat analytic approach, linear mixed-effects regression modeling for repeated measures was used to assess the effects of exercise training modality on changes in resting serum BDNF over time, with BDNF as the dependent variable, group (aerobic, resistance, combined, control), time (baseline and 6-months) and group by time interaction as the independent variables, with adjustment for age and sex using an unstructured covariance matrix for the repeated measures. Within the mixed model, we used 95% CIs and p-values (<0.05) for the intergroup comparisons (aerobic vs control, resistance vs control, combined vs control, aerobic vs resistance, combined vs aerobic, combined vs resistance), and for changes over time in BDNF concentrations within each group. Effect sizes were computed as Cohen’s d where the greater the number, the stronger the effect. We also examined the effects of the exercise interventions with per-protocol analyses in participants who had complete baseline and follow-up data and maintained at least 70% exercise adherence (average of at least 2.8 sessions per week) throughout the intervention following the same procedures as in the intention-to-treat analyses. We also examined the relationships between adherence (i.e number of exercise sessions attended) and changes in BDNF values in both the intention-to-treat analysis and per-protocol analysis using Pearson correlation coefficients. Analyses were conducted using SAS, version 9.4 (SAS Institute, Cary, North Carolina).

RESULTS

Table 1 shows baseline characteristics of the sample. There were no intergroup baseline differences. Most participants (263 of 282 or 93%) had obesity (BMI≥95th percentile for age and sex). The sample was about 70% Caucasian and most parents had some post-secondary education. There were no statistically significant group differences on socio-demographic variables, free-living physical activity, body composition, aerobic fitness or psychological functioning at baseline as reported elsewhere. 34, 39, 40
Adherence and Attrition

From baseline to 26 weeks, median exercise training adherence was 62% (interquartile range, 36% to 81%) in aerobic, 56% (interquartile range, 37% to 75%) in resistance and 64% (interquartile range, 39% to 75%) in combined with no significant differences between groups. Twenty-five percent (71 participants) withdrew between randomization and 6 months: 18 (24%) from Aerobic, 21 (28%) from Resistance, 17 (23%) from Combined, and 19 (25%) from Control. The main reasons for withdrawal were lack of interest and lack of time.

**BDNF**

Table 2 presents the effects of exercise interventions on resting serum BDNF levels from the intention to treat analyses. There was no significant group x time interaction for BDNF ($F (3,227) = 0.89$, $p=0.45$), indicating changes between groups over time did not differ significantly. Similarly, there were no main effects of time $F (1,227)=0.01$, $p=0.91$, indicating that BDNF levels within exercising and control groups did not change significantly over time. Moreover, the effect of group was not significant $F (3,250)=0.74$, $p=0.53$). The sex by exercise group by time interaction on BDNF values was not significant, ($F(3,219)=0.14$, $p=0.93$), indicating the effects of exercise on changes in BDNF levels did not differ between males and females in the intention to treat analysis.

Table 3 presents the effects of exercise interventions on resting BDNF levels from the per protocol analyses. The group x time interaction was not significant, $F (3,128)=0.32$, $p=0.81$), indicating that changes in BDNF levels between groups did not differ significantly over time. Likewise, the main effect of time was not significant, $F(1,128)=0.08$, $p=0.78$), indicating BDNF levels within exercise and control groups did not increase significantly over the 6-month intervention. The main effect of group did not reach significance, $F(3,126)=0.99$, $p=.40$). The sex by exercise group by time interaction on
BDNF values was not significant, (F(3,130)=1.12, p=0.34), indicating the effects of exercise on changes in BDNF levels did not differ between males and females in the per-protocol analysis.

The correlations between number of sessions attended and changes in BDNF from aerobic training (r=-0.07), resistance training (r=0.11), combined training (r=0.04) and diet-only control (r=-0.13) were small and did not reach significance (all p-values >0.05) in the intention-to-treat analysis. Similarly, the correlations between the number of sessions attended and changes in BDNF from aerobic training (r=-0.06), resistance training (r=0.15), combined training (r=-0.05) and diet-only controls (r=-0.13) were small and not statistically significant (i.e. p>0.05). These results suggest that the null intervention effects are unlikely to be related to adherence.

**DISCUSSION**

To our knowledge, this was the first randomized controlled trial comparing effects of prolonged aerobic exercise training, resistance exercise training and their combination on resting serum BDNF levels in a large sample of adolescents living with overweight and obesity. The primary findings were that none of the exercise interventions, or the non-exercising diet-only control group produced a significant change increase in BDNF levels.

Our null findings are not consistent with previous studies with comparable frequency and intensity of aerobic exercise but shorter duration (i.e. 12 weeks vs our 22 weeks) which reported an increase in resting serum BDNF levels and concomitant improvements in cognitive functioning in adolescents with obesity and without. It is possible that the discrepant findings could be due, in part, to previous studies being comprised of adolescent males only, whereas the current study was largely comprised (70%) of adolescent females. It is important to note, however, that although the trend for increased BDNF resulting from programmed aerobic training found in adolescent males is generally supported by meta-analytic data in adults, the overall effect size of aerobic exercise interventions
was small and more than 60% of trials showed null findings, including those conducted with clinical populations such as participants with obesity and T2DM or neurological conditions. This pattern of results highlights the considerable variability in BDNF response to programmed aerobic exercise interventions.

We hypothesized that the combined training group, which performed up to 90 minutes of aerobic and resistance training, would elicit greater increases in serum BDNF than that of aerobic or resistance training inly (45 min/each) given there is some evidence, albeit limited and focusing primarily on aerobic training, for dose-response effects of exercise on BDNF in adults. However, our null findings are consistent with studies in adults that found the combined effects of programmed aerobic and resistance training failed to increase both plasma and serum BDNF. Indeed, in the most comparably designed exercise trial to the current study, Swift et al. found in a large randomized controlled trial in adults with T2DM that 9 months of 3-4 times/wk supervised aerobic training (50-80% of maximal oxygen consumption), resistance training or combined training produced no changes in resting serum BDNF levels despite improvements in adiposity, aerobic fitness and cardiometabolic health induced by exercise, especially in the aerobic and combined groups. Similarly, the HEARTY trial demonstrated that the exercise interventions, especially combined training, reduced adiposity and improved musculoskeletal and cardiorespiratory fitness in adolescents with obesity, but with no concomitant increase in peripheral BDNF levels. Relatedly, studies examining the cross-sectional and prospective relationships between aerobic fitness and BDNF show mixed findings, underscoring these relationships are complex and further inquiry is needed.

Our study was the first to our knowledge to examine the effects of resistance training alone on resting serum BDNF levels in adolescents. Although a few studies in adults have shown that this modality increases BDNF levels, primarily in the elderly, our null findings are consistent with the
majority of studies in a meta-analytic review in adults. Taken together, these findings indicate that while resistance training may confer important physiological and psychological health benefits in adolescents with obesity, it does not appear to be effective in increasing resting peripheral BDNF.

We designed our aerobic training prescription, either alone or in the combined group, to progress to 45 minutes of moderate to vigorous intensity (70-85% of VO\textsubscript{2} peak) exercise. The resistance training program, either alone, or in the combined intervention, allowed at least 1 minute breaks between sets, thereby allowing heart rates to recover toward resting. There is evidence that increases in BDNF levels may be intensity dependent, with data showing greater increases relative to controls are obtained from high intensity exercise (90% of maximal oxygen consumption) than from moderate intensity, (50-70% of maximal oxygen consumption). However, most of these effects were observed after an acute bout of exercise. In the lone intervention study in adolescents to examine intensity-dependent effects, Jeon and Ha found that 12 weeks of high intensity aerobic training, performed 4-times/week, led to greater increases in serum BDNF levels and cognitive performance (working memory) than low intensity aerobic training or sedentary controls. Similarly, moderate intensity aerobic exercise but not low intensity exercise led to greater increases in BDNF levels than controls. Thus, the role that intensity plays in the effects of chronic aerobic or resistance exercise interventions have on BDNF in youth remains understudied and poorly understood. Given the evidence that short, high-intensity interval training (HIIT) may be preferred over the continuous moderate to high intensity exercise prescriptions in adult clinical populations, including those with obesity and chronic disease, the efficacy and tolerability of HIIT interventions of varying frequency and duration need to be evaluated to determine if they have a positive impact on BDNF in youth with obesity. Moreover, the effects of exercise on cognition in children appear greater when the physical activity is cognitively engaging rather than
repetitious activity.\textsuperscript{54, 55} In this context, future research should explore the role of cognitive engagement when examining effects of exercise on BDNF in adolescents with, as well as other pediatric populations. Considering there is evidence that exercising outside in the fresh air and a connection to nature may potentiate cognitive processes and well-being compared to exercising indoors,\textsuperscript{56, 57} it would also be important to evaluate the effects that exercising outdoors has on BDNF levels in youth and adults.

Given that our results are somewhat consistent with comparable trials in adults, it is entirely possible that regular aerobic, resistance or combined training may not have a reliably measureable effect on resting serum BDNF in adolescents with obesity. Indeed, there are data to support many other mechanisms linking exercise to improved cognitive functioning and brain health, such as increased cerebral blood flow (perfusion), changes in neuro-endocrine function, changes in neurotransmitter and endocannabinoid release/reuptake, as well as structural changes in the central nervous system.\textsuperscript{25-27} BDNF exerts its cascade of effects via the high-affinity tropomyosin receptor kinase B (TrkB) in the brain and periphery.\textsuperscript{58} Interestingly, activation of TrkB via BDNF upregulates the expression of TrkB, which subsequently increases the sensitivity of a target cell to future BDNF signaling.\textsuperscript{59, 60} Accordingly, it is possible that the transient increases in serum BDNF that would be expected to occur with acute exercise\textsuperscript{29} over the 6-month training period would increase the neuronal expression of TrkB and sensitivity to BDNF.\textsuperscript{12} Therefore, a seemingly negligible change in resting serum BDNF following exercise training does not preclude a lack of effect on brain function via BDNF with exercise training.

The effects of exercise on resting serum BDNF levels are complex and highly variable. Although research suggests that some of this variability, especially from acute bouts of exercise, may be due to the potential moderating effects of study population (healthy vs clinical samples), age, sex (stronger effects in males), method of measurement (serum vs plasma), and programmatic factors such as frequency, intensity, and duration,\textsuperscript{29} a more recent meta-analytic review on exercise interventions greater than two
weeks did not show these factors moderated effects on BDNF levels.\textsuperscript{25} Nevertheless, it would be prudent to consider these potential moderating factors when designing future interventions and interpreting outcomes given the heterogeneity inherent in pooling data in meta-analytic reviews, with the understanding that studies rarely experimentally examined these factors to determine their influence.

The current study has several strengths and limitations. The strengths include a randomized controlled trial design, which provides a high quality of evidence to base conclusions. In addition, this was the first study to comparatively examine the effects of resistance training, with or without aerobic training, on BDNF levels in adolescents with obesity, a population known to be at high risk of medical and psychosocial comorbidities,\textsuperscript{3,4} and diminished executive functioning.\textsuperscript{7} Moreover, the current study utilized the largest sample size of any study examining the effects of aerobic and resistance training and their combination on BDNF, thereby achieving strong statistical power. Furthermore, the volume of training and duration of the intervention prescribed to participants matched or exceeded the majority of previous studies.

Although this was the largest randomized controlled trial to date assessing the effects of aerobic and resistance training and their combination in adolescents, the current study did not account for the potential role that sex may have on BDNF. Our sample was predominantly (70\%) comprised of females, and the two studies in adolescents that found aerobic exercise increased BDNF relative to controls were comprised solely of males,\textsuperscript{31,32} thus it is possible our results would have differed if a greater proportion of males participated. Also, similar to many exercise trials in persons with and without obesity,\textsuperscript{61} adherence to the exercise prescription was a problem for some participants, which may have contributed to the null findings, although the pattern of results from the more adherent participants (\textit{\geq} 70\% of attendance at exercise sessions) was similar to the intention-to-treat population. Given that not all participants approached consented to participate, results may not generalize to all
youth with obesity in the community. Although the diet-only control group that involved a 250 kcal (about 10%) restriction showed no change in BDNF levels, evidence from research in adult humans suggest that a more aggressive diet with 20-40% caloric restriction could impact BDNF. Thus, future research should examine the independent effect of dietary restriction or diet composition on BDNF values given this aim was beyond the scope and design of the HEARTY study. Since blood samples were collected at baseline and 6-months, we did not have the opportunity to evaluate the trajectory of changes in resting serum BDNF levels that may have occurred over longer periods of time. Similar to the vast majority of other intervention studies, we did not systematically track or report the length of time between the last bout of exercise and the timing of the 6-month BDNF measurement that occurred between 2 and 10 days post-intervention. Future research should address this pervasive reporting omission in the literature to gain a better understanding of how the timing of post-intervention BDNF measurement may impact treatment effects. Lastly, we did not collect data on cognitive function or its relationship to changes in BDNF. Future research addressing these limitations is needed to gain a better understanding of the effects of exercise on peripheral BDNF in adolescents with and without obesity.

In conclusion, the HEARTY trial, which previously showed that 6-months of aerobic, resistance or combined training improved physiological and psychosocial health indicators, did not increase resting serum BDNF levels beyond that of diet-only controls in a large sample of adolescents living with overweight and obesity. This pattern of results remained even among participants with good adherence to the exercise interventions. As with most intervention studies, BDNF responses from exercise are heterogeneous and highly variable. Thus, future RCTs are needed to verify these initial findings, and further elucidate the effects that various exercise modalities, doses, and environments have on resting peripheral BDNF levels and concomitant cognitive function in attempt to optimize exercise prescriptions in high risk populations such as youth with obesity.
Acknowledgments

The HEARTY trial was supported by grant MCT-71979 from the Canadian Institutes of Health Research. Dr. Goldfield was supported by a New Investigator Award from the Canadian Institutes of Health Research for part of this trial and subsequently by an Endowed Research Chair from the Children’s Hospital of Eastern Ontario Volunteer Association Board. Dr. Sigal was supported by a Health Senior Scholar award from the Alberta Innovates-Health Solutions and previously supported by a Research Chair from the Ottawa Hospital Research Institute during part of this trial. Dr. Kenny was supported by a University Research Chair from the University of Ottawa. Dr. Alberga was supported by a Doctoral Student Research Award from the Canadian Diabetes Association. We would like to thank the HEARTY trial participants, as well as Krista Hind, Bsc (deceased), Bruno Lemire, Ph.D., Marta Wein BSc, Kim Robertson, BSc., Kim Fetch, BSc., Brittany Hanlon, MHA, Jane Yardley, Ph.D., Nadia Balaa, BSc., Karen Lopez, BSc., Pamela Martino, MSc., Kim Morin, BSc., Colleen Gilchrist, BSc., RD., Pascale Messier, BSc., RD., Kelley Phillips, MA, and students in the School of Human Kinetics, University of Ottawa, who contributed to study coordination, exercise training, and evaluation of study participants. Robert Ross, Ph.D. (Queens, University, Kingston, Ontario, Canada), Alison Bradshaw, MSc., and Jennifer Kuk, Ph.D., (York University, Toronto, Ontario, Canada), and Yves Martel, Ph.D., (Tomovision, Magog, Quebec, Canada) assisted with training and provided ongoing advice on body composition analysis. The Ottawa-Carleton Regional YMCA/YWCA (Ottawa, Ontario, Canada), RA Centre (Ottawa, Ontario, Canada), Children’s Hospital of Eastern Ontario, and Nautilus Plus and MRI Plus (both in Gatineau, Quebec, Canada) collaborated throughout the trial.
Reference List


### Table 1. Baseline characteristics of the sample

<table>
<thead>
<tr>
<th>Variable</th>
<th>Total sample (n=282) Mean (SD)</th>
<th>Aerobic (n=69) Mean (SD)</th>
<th>Resistance (n=70) Mean (SD)</th>
<th>Combined (n=74) Mean (SD)</th>
<th>Control (n=69) Mean (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>15.6 (1.4)</td>
<td>15.5 (1.3)</td>
<td>15.8 (1.5)</td>
<td>15.5 (1.3)</td>
<td>15.6 (1.3)</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>97.9 (17.2)</td>
<td>96.5 (15.2)</td>
<td>99.9 (18)</td>
<td>97.3 (16.4)</td>
<td>97.9 (19.3)</td>
</tr>
<tr>
<td>Height, cm</td>
<td>167.7 (7.5)</td>
<td>166.8 (7.2)</td>
<td>167.9 (7.5)</td>
<td>167.6 (7.6)</td>
<td>168.5 (7.6)</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>34.7 (4.5)</td>
<td>34.6 (4.2)</td>
<td>35.3 (4.8)</td>
<td>34.5 (4.1)</td>
<td>34.3 (5)</td>
</tr>
<tr>
<td>Percent Body Fat</td>
<td>49.6 (5.4)</td>
<td>49.4 (5.7)</td>
<td>50 (5.6)</td>
<td>50.1 (5.1)</td>
<td>48.8 (5.3)</td>
</tr>
<tr>
<td>Waist circumference, cm</td>
<td>96.7 (10.9)</td>
<td>96.2 (9.8)</td>
<td>98.8 (11.6)</td>
<td>96.6 (10.4)</td>
<td>95.1 (11.6)</td>
</tr>
<tr>
<td>Males, N (%)</td>
<td>84 (29.8%)</td>
<td>19 (27.5%)</td>
<td>21 (30.0%)</td>
<td>22 (29.7%)</td>
<td>22 (31.9%)</td>
</tr>
<tr>
<td>Females, N (%)</td>
<td>198 (70.2%)</td>
<td>50 (72.5%)</td>
<td>49 (70.0%)</td>
<td>52 (70.3%)</td>
<td>47 (68.1%)</td>
</tr>
<tr>
<td>Ethnicity, N (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caucasian</td>
<td>200 (70.9%)</td>
<td>49 (71%)</td>
<td>48 (68.6%)</td>
<td>46 (62.2%)</td>
<td>57 (82.6%)</td>
</tr>
<tr>
<td>Black</td>
<td>30 (10.6%)</td>
<td>6 (8.7%)</td>
<td>10 (14.3%)</td>
<td>12 (16.2%)</td>
<td>2 (2.9%)</td>
</tr>
<tr>
<td>Mixed racial</td>
<td>12 (4.3%)</td>
<td>4 (5.8%)</td>
<td>2 (2.9%)</td>
<td>3 (4.1%)</td>
<td>3 (4.3%)</td>
</tr>
<tr>
<td>Arabic</td>
<td>11 (3.9%)</td>
<td>4 (5.8%)</td>
<td>1 (1.4%)</td>
<td>4 (5.4%)</td>
<td>2 (2.9%)</td>
</tr>
<tr>
<td>Hispanic</td>
<td>9 (3.2%)</td>
<td>4 (5.8%)</td>
<td>2 (2.9%)</td>
<td>3 (4.1%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>Asian</td>
<td>8 (2.8%)</td>
<td>0 (0%)</td>
<td>4 (5.7%)</td>
<td>2 (2.7%)</td>
<td>2 (2.9%)</td>
</tr>
<tr>
<td>Native Canadian</td>
<td>4 (1.4%)</td>
<td>0 (0%)</td>
<td>0 (0%)</td>
<td>2 (2.7%)</td>
<td>2 (2.9%)</td>
</tr>
<tr>
<td>Indonesian/Asian</td>
<td>2 (0.7%)</td>
<td>1 (1.4%)</td>
<td>1 (1.4%)</td>
<td>0 (0%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>Other</td>
<td>6 (2.1%)</td>
<td>1 (1.4%)</td>
<td>2 (2.9%)</td>
<td>2 (2.7%)</td>
<td>1 (1.4%)</td>
</tr>
<tr>
<td>Parental Education, N (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maternal</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>High school or less</td>
<td>72 (25.8%)</td>
<td>19 (27.5%)</td>
<td>18 (25.7%)</td>
<td>16 (21.6%)</td>
<td>19 (27.5%)</td>
</tr>
<tr>
<td>College / University</td>
<td>207 (74.2%)</td>
<td>48 (69.6%)</td>
<td>52 (74.3%)</td>
<td>57 (77%)</td>
<td>50 (72.5%)</td>
</tr>
<tr>
<td>Unknown</td>
<td>3 (1.1%)</td>
<td>2 (2.9%)</td>
<td>0 (0%)</td>
<td>1 (1.4%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>Paternal</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>High school or less</td>
<td>88 (34.5%)</td>
<td>23 (33.3%)</td>
<td>18 (25.7%)</td>
<td>22 (29.7%)</td>
<td>25 (36.2%)</td>
</tr>
<tr>
<td>College / University</td>
<td>167 (65.5%)</td>
<td>43 (62.3%)</td>
<td>45 (64.3%)</td>
<td>43 (58.1%)</td>
<td>36 (52.2%)</td>
</tr>
<tr>
<td>Unknown</td>
<td>27 (9.6%)</td>
<td>3 (4.4%)</td>
<td>7 (10%)</td>
<td>9 (12.2%)</td>
<td>8 (11.6%)</td>
</tr>
</tbody>
</table>

Note. Continuous data are presented as means (SD=standard deviations); categorical data are presented as frequency counts and percentages. BMI= body mass index.
Table 2. Intention-to Treat Analyses on the Effects of Exercise Modality on Changes in BDNF (ng/mL)

<table>
<thead>
<tr>
<th>Variable</th>
<th>N 0</th>
<th>Baseline</th>
<th>N 6</th>
<th>6 months</th>
<th>Mean (SE)</th>
<th>Absolute Change from Baseline to 6-months</th>
<th>Adjusted Change from Baseline to 6-months</th>
<th>P-value</th>
<th>Cohen's d</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aerobic</td>
<td>69</td>
<td>24.6 (1.7)</td>
<td>52</td>
<td>26.4 (2)</td>
<td>1.8 (-2.8, 6.4)</td>
<td>0.44</td>
<td>0.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Resistance</td>
<td>70</td>
<td>29.7 (1.7)</td>
<td>44</td>
<td>27.7 (2.2)</td>
<td>-1.9 (-6.8, 3.0)</td>
<td>0.44</td>
<td>-0.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Combined</td>
<td>74</td>
<td>27.9 (1.7)</td>
<td>56</td>
<td>26.2 (2)</td>
<td>-1.7 (-6.1, 2.8)</td>
<td>0.46</td>
<td>-0.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>69</td>
<td>25.8 (1.7)</td>
<td>50</td>
<td>28.1 (2.1)</td>
<td>2.3 (-2.4, 7.0)</td>
<td>0.33</td>
<td>0.13</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aerobic vs. control</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>-0.5 (-7.1, 6.1)</td>
<td>0.88</td>
<td>-0.02</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Resistance vs. control</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>-4.3 (-11, 2.6)</td>
<td>0.22</td>
<td>-0.16</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Combined vs. aerobic</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>-3.5 (-9.9, 2.9)</td>
<td>0.29</td>
<td>-0.14</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Combined vs. resistance</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.2 (-6.4, 6.9)</td>
<td>0.94</td>
<td>0.01</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Combined vs. control</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>-4.0 (-10.4, 2.5)</td>
<td>0.23</td>
<td>-0.16</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aerobic vs. resistance</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>3.7 (-3, 10.5)</td>
<td>0.28</td>
<td>0.14</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Note: N0= sample size at baseline; N6= sample size at 6-months; Cohen’s d = effect size
### Table 3. Per Protocol (≥70% adherence) Analyses on the Effects of Exercise Modality on Changes in BDNF (ng/mL)

<table>
<thead>
<tr>
<th>Variable</th>
<th>N 0</th>
<th>Baseline (Mean (SE))</th>
<th>N 6</th>
<th>6 months (Mean (SE))</th>
<th>6 months Absolute Change from Baseline to 6-months (Mean (95% Confidence Interval))</th>
<th>Adjusted Change from Baseline to 6-months (Mean (95% Confidence Interval))</th>
<th>P-value</th>
<th>Cohen's d</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aerobic</td>
<td>33</td>
<td>23.7 (2.5)</td>
<td>33</td>
<td>23.8 (2.5)</td>
<td>0.1 (-6.3, 6.5)</td>
<td>0.98</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Resistance</td>
<td>21</td>
<td>24.4 (3.1)</td>
<td>21</td>
<td>24.9 (3.2)</td>
<td>0.4 (-7.6, 8.5)</td>
<td>0.92</td>
<td>0.02</td>
<td></td>
</tr>
<tr>
<td>Combined</td>
<td>28</td>
<td>28.8 (2.7)</td>
<td>28</td>
<td>27.5 (2.8)</td>
<td>-1.3 (-8.3, 5.6)</td>
<td>0.71</td>
<td>-0.07</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>50</td>
<td>25.4 (2)</td>
<td>50</td>
<td>28.1 (2.1)</td>
<td>2.7 (-2.5, 8)</td>
<td>0.3</td>
<td>0.18</td>
<td></td>
</tr>
</tbody>
</table>

Note: N0= sample size at baseline; N6= sample size at 6-months; Cohen’s d = effect size
Highlights:

- 6-months of diet plus aerobic or resistance training did not increase BDNF in youth
- 6-months of diet plus combined aerobic and resistance training did not increase BDNF
- Effects of exercise intervention or modality on BDNF did not differ between sex