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# Effects of pediatric adiposity on heart rate variability

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EFFECTS OF PEDIATRIC ADIPOSITY  
ON HEART RATE VARIABILITY

By

Angela K. Lucas

A THESIS

Submitted in partial fulfillment of the requirements

for the degree of

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2009

This thesis, "Effects of Pediatric Adiposity on Heart Rate Variability" is hereby approved in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE in the field of Biological Sciences.

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## **Abstract**

The relationship between obesity and heart rate variability (HRV) has been studied in adults and adolescents, but is not determined in young pediatrics. The purpose of this study was to assess autonomic activity using HRV in a pediatric population. We hypothesized that obese children would have reduced parasympathetic and increased sympathetic activity compared to age-matched subjects. 42 pediatric subjects (ages 3-5) were classified into 3 groups based on body mass index-for-age; normal, overweight and obese. HRV and respiratory rate were recorded during 3 minute baseline, 2 minute isometric handgrip and 3 minute recovery. HRV was analyzed in the time domain [heart rate (HR), RR interval (RRI) and RRI standard deviation (RRISD)] and frequency domain [low frequency (LF), high frequency (HF) and LF/HF ratio] using repeated measures ANOVA. Spearman's correlations were used to examine the relations between BMI and HRV at rest. Significant condition effects were found between baseline, exercise and recovery, but these responses were not significantly different between the normal, overweight and obese children. BMI was negatively correlated with LF/HF, while BMI was positively correlated with RRISD, LF, HF and nHF. Our data demonstrate that higher BMI in the pediatric population is correlated with higher parasympathetic and lower sympathetic activity. These findings are contrary to HRV responses observed in adults and adolescents, suggesting complex relationships between age, obesity and autonomic control of the heart. The data supports the concept of an age reliance of HRV and a novel relationship between adiposity and body mass index in 3-5 year olds.

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## List of Abbreviations

BMI	Body mass index
bpm	Beats per minute
CDC	Centers for disease control and prevention
DAP	Diastolic arterial blood pressure
ECG	Electrocardiogram
FFT	Fast Fourier transform
HF	High frequency
HR	Heart rate
HRV	Heart rate variability
LF	Low frequency
LF/HF	Low frequency to high frequency ratio
MAP	Mean arterial pressure
MVC	Maximal voluntary contraction`
nHF	Normalized high frequency
nLF	Normalized low frequency
Resp	Respiration
RRI	RR intervals
RRISD	RR interval standard deviation
SAP	Systolic arterial blood pressure
TP	Total spectral power

## *Effects of Pediatric Adiposity on Heart Rate Variability*

### **INTRODUCTION**

Heart rate variability (HRV) provides a noninvasive index of the neural activity of the heart and is commonly used to examine the autonomic nervous system. Time domain analysis and frequency domain analysis are the two main HRV analytical approaches. The power spectrum of HRV characterizes the frequency domain properties of heart beat variation by applying spectrum assessment techniques to R-R interval data measured by an electrocardiogram. This analysis produces frequency components including high frequency (HF) which reflects mainly parasympathetic activity and low frequency (LF) which reflects both parasympathetic and sympathetic activity. HRV is considered a reliable, noninvasive technique for assessing neural control of the cardiovascular system (Pagani and Malliani 2000).

The incidence of obesity is rapidly rising to epic proportions, especially in younger populations. According to the 2003-2004 National Health and Nutrition Examination Survey, 17% of children and adolescents ages 2-19 years are overweight (Ogden et al. 2006). Previous researchers have shown that overweight children have a much greater chance of becoming overweight or obese adults (Kotani et al. 1997 and Whitaker et al. 1997). Obesity has been shown to increase the risk of many diseases, including diabetes, cardiovascular disease, osteoarthritis, urinary incontinence, gallbladder disease, sleep disorders, cancer, psychological problems and mental disorders (Kotani et al. 1997 and Yakinci et al. 2000). Previous studies in obese adolescents show an association with increased morbidity (Folsom et al. 1996 and Liu et al. 1996) and mortality rates (Evrengul et al. 2006 and Finley et al. 1995). Obesity is also characterized



by autonomic nervous system dysfunction. Low HRV, which represents a greater prevalence of sympathetic tone, has been associated with a high cardiovascular risk (Kleiger et al. 1987).

Body Mass Index (BMI) is a common method utilized to screen for obesity in children due to its ease of measurement and reliability. Although BMI does not directly measure body fat, it is a reliable indicator of adiposity for most children. Studies have shown that BMI correlates to underwater weighing and dual energy x-ray absorptiometry (Mei et al. 2002). BMI for children is calculated on a separate scale than adults and is referred to as BMI-for-age. Percentile rankings are established based on The Centers for Disease Control and Prevention BMI-for-age growth charts based on sex. Percentile ranges are as follows: 1) underweight is less than the 5<sup>th</sup> percentile, 2) healthy weight includes the 5<sup>th</sup> percentile to less than the 85<sup>th</sup> percentile, 3) at risk of overweight includes the 85<sup>th</sup> to less than the 95<sup>th</sup> percentile, and 4) overweight is equal to or greater than the 95<sup>th</sup> percentile. The American Medical Association recommends classifying children with BMI's greater than the 95<sup>th</sup> percentile for age and sex as obese and those with BMI's greater than the 85<sup>th</sup> but less than the 95<sup>th</sup> percentile as overweight (Goutham 2008). For the purpose of this thesis, we will classify our subjects based on Goutham (2008).

HRV has been used in several studies with infants and children because it is easily applicable and noninvasive. Although many studies have utilized HRV with infants and children, few have specifically examined the relationship between HRV and childhood obesity. In those that have, it appears that obesity does influence HRV in obese children (Martini et al. 2001; Nagai et al. 2003; Rabbia et al. 2003; Riva et al. 2001; Yakinci et al. 2000). Previous studies have focused primarily on the adolescent population and the link

between reduced HRV and obesity. Specifically, HRV studies suggest increased obesity is associated with a reduction in parasympathetic activity (Martini et al. 2001; Nagai et al. 2003; Rabbia et al. 2003; Riva et al. 2001 and Yakinci et al. 2000) and either an increase (Martini et al. 2001; Rabbia et al. 2003 and Riva et al. 2001), decrease (Nagai et al. 2003) or no change (Yakinci et al. 2000) in sympathetic activity. Since reduced HRV has been found to be a significant predictor of cardiac mortality and morbidity, the altered HRV response to obesity is of great importance to the pediatric population. To date, no studies have examined the relationship between HRV and obesity in the young pediatric population, specifically 3 to 5 year olds.

The purpose of this study was to determine whether the autonomic nervous system, specifically sympathetic and parasympathetic activity assessed via HRV, is reduced in a pediatric obese population. Based on HRV studies performed in adults and adolescents, we hypothesized that obese children will have reduced parasympathetic activity. Examining this relationship in the younger population will provide new insights into the effects of early obesity on the autonomic nervous system.

## **METHODS**

**Subjects.** Forty two children (age  $4.0 \pm 0.1$  years) were recruited from a local preschool program (BHK Child Development, Houghton, MI) to participate in this study. Three experimental groups were studied based on BMI-for-age: 1) normal weight (9 males, 5 females), 2) overweight (9 males, 5 females), and 3) obese (2 males, 12 females). Subject baseline characteristics are outlined in Table 1. All subjects abstained from exercise and caffeine for  $\geq 12$  h before laboratory testing. The Michigan

Technological University Human Subjects Research Committee granted approval for testing. All children and their parents were instructed on the study and provided informed assent and consent to participate.

**Table 1.** Baseline characteristics of normal weight, overweight and obese children.

<b>Variable</b>	<b>Normal</b>	<b>Overweight</b>	<b>Obese</b>
<b>HT</b> (cm)	103 ± 1	106 ± 1	111 ± 3*
<b>WT</b> (kg)	17 ± 0	20 ± 1*	24 ± 1*†
<b>BMI</b> (kg/m <sup>2</sup> )	16 ± 0	18 ± 0*	19 ± 0*†
<b>SAP</b> (mmHg)	88 ± 3	87 ± 3	92 ± 2
<b>DAP</b> (mmHg)	59 ± 1	59 ± 1	59 ± 1
<b>MAP</b> (mmHg)	69 ± 2	68 ± 2	69 ± 2
<b>MVC</b> (p.s.i.)	2 ± 0	2 ± 0	2 ± 0

\* Significantly different from normal, † significantly different from overweight,  $p < 0.05$ .

HT, height (cm); WT, weight (kg); BMI, body mass index (kg/m<sup>2</sup>); SAP, systolic arterial blood pressure (mmHg); DAP, diastolic arterial blood pressure (mmHg); MAP, mean arterial blood pressure (mmHg); MVC, maximal voluntary contraction (p.s.i.).

**Study design.** Participants were prepared for electrode placement for measurement of heart rate via a 3-lead electrocardiogram. Resting arterial blood pressure was obtained using sphygmomanometer after the subject had been sitting quietly for 5 minutes. Three measurements were taken and the average was reported for resting arterial blood pressure. Subjects were then asked to grip a pediatric bulb dynamometer to obtain a maximal voluntary contraction (MVC). An average MVC was determined from 3 recordings. A colorful sticker was placed on the dynamometer to illustrate 30% MVC. Subjects then demonstrated they could perform the task by maintaining 30% MVC. Subjects were positioned supine on a table for five minutes to obtain a resting measurement. Following a 3 minute baseline, subjects performed isometric handgrip at 30% of their MVC for 2 minutes. A 3 minute recovery was recorded after the isometric

handgrip. All subjects watched the movie “Stuart Little” during the baseline and recovery stages.

**Heart rate variability analyses.** An electrocardiogram (ECG) detects electrical activity occurring in the heart through electrodes placed on the skin. An ECG is used as a means of measuring electrical potential with impulses appearing as a series of positive and negative deflections. Cardiac events are correlated to the ECG and produce distinct waves. Specifically, the SA node depolarizes to produce a P wave followed by depolarization of the ventricles which brings about ventricular contraction and the QRS complex. The T wave is produced from ventricular repolarization. Figure 1 depicts a representative tracing of an ECG from the current study.

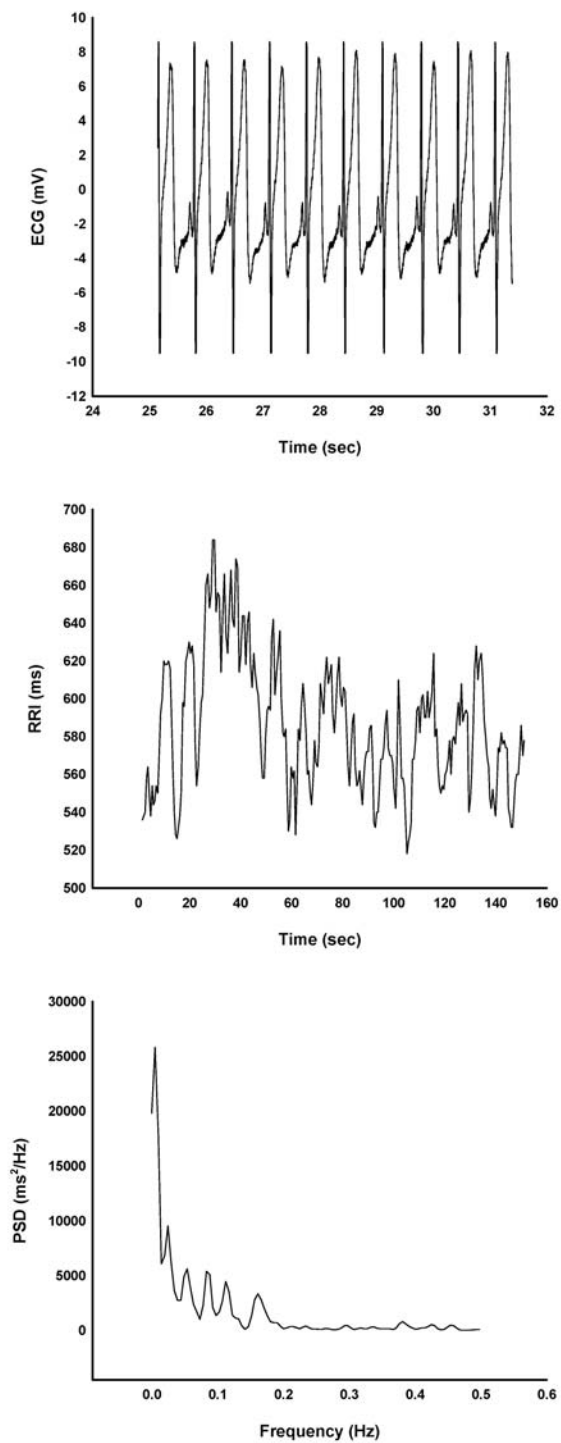
Heart rate variability (HRV) refers to the beat to beat variations in heart rate and can be measured using an ECG. The ECG exhibits periodic variation in R-R intervals which can be associated with the phase of respiration. The analysis of HRV offers a non-invasive method of evaluating vagal input into cardiac rhythm and only requires ECG equipment, microprocessors and relevant software for carrying out Fourier analyses. There are 2 major components of HRV. High frequency is synchronous with respiration and reflects parasympathetic activity. Low frequency is mediated by both the vagus and cardiac sympathetic nerves and reflects both sympathetic and parasympathetic activity.

HRV can be measured using time or frequency domain analysis. Time domain is measured manually from an ECG by calculating the mean R-R interval and its standard deviation. Small standard deviations in R-R intervals represented low HRV. Nearly 30 different types of arithmetic manipulations of R-R intervals in the time domain exist today, all seeming to be largely equal with no one superior method. Time domain is

easily performed using the heart rate at any point in time or the intervals between successive normal complexes.

HRV can also be expressed in the frequency domain as the sum of oscillations based on their frequency and amplitudes. The frequency domain is represented by the sum of sinusoidal components of different amplitude, frequency and phase values. More specifically, the R-R signal is transformed from time to frequency domain by representing the signal as a combination of sine and cosine waves, with different amplitudes and frequencies. The oscillatory components are evaluated using many different algorithms (Task Force on HRV 1996), including the fast Fourier transform (FFT). The FFT is frequently used since it is easier to implement than many of the other methods.

Fast Fourier Transform converts the R-R interval time domain into sine and cosine waves, with the goal of ending up with something easier to deal with than the original signal. The frequency of each sinusoid is fixed and only the amplitude is changed depending on the shape of the waveform being decomposed. The frequency domain contains similar information as the time domain, just in a different form. This thesis will report the effects of pediatric adiposity on heart rate variability expressed in both the time domain and frequency domain. Both are well established techniques that provide insight into autonomic neural control of the heart.



**Figure 1.** Representative tracings of electrocardiogram (ECG), RR interval (RRI) and power spectral density (PSD) during baseline in one subject.

**Measurements.** Height and weight were measured on each subject. Body mass index was calculated from each subject's weight and height by plotting on the BMI-for-age growth chart dependent on sex to obtain a percentile ranking. Resting blood pressure was measured with a manual sphygmomanometer. A three-lead electrocardiogram was used to record continuous heart rate. Respiratory rate was measured using a pneumograph. Data from the electrocardiogram was sampled at 500 Hz and stored on a computer (WINDAQ, Dataq Instruments, Akron, OH). A pediatric bulb dynamometer was used for the isometric handgrip.

**Data analysis.** Data were imported into a software program for data analysis (WinCPRS, Absolute Aliens Ay, Turku, Finland). R-waves were detected and marked in the time series. Data were analyzed by using SPSS statistical software (SPSS, version 15.0, Chicago, IL). Analyses of HRV variables between groups were performed using repeated measures analysis of variance. The least-significant difference pairwise comparison method was used for post-hoc analyses. The relationship between BMI and HRV variables was examined by Spearman correlation analysis. Significant differences were determined when  $p < 0.05$ . All results are presented as means  $\pm$  standard errors.

## RESULTS

Table 2 displays HRV variables for normal, overweight and obese children. There were significant differences during baseline, exercise and recovery for all 9 HRV variables (condition effect,  $p < 0.05$ ). Figures 2 and 3 demonstrate that there was a significant difference between baseline and exercise for HR, RRI, LF/HF, nLF, nHF and respiration. Figures 2 and 3 also demonstrate that there was a significant difference

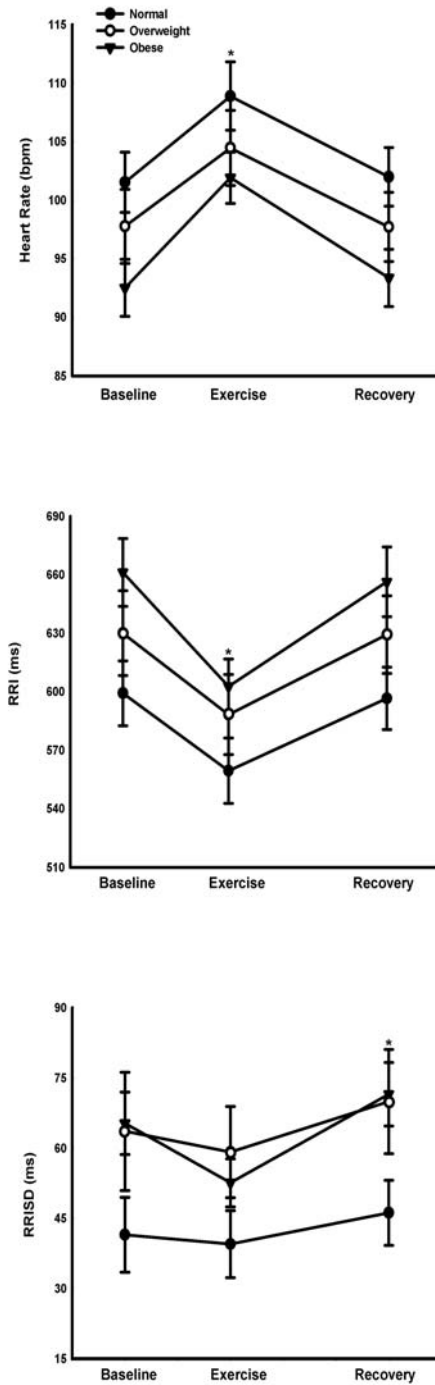
between baseline and recovery for RRISD, LF, nLF and respiration. The HRV responses were not significantly different between the normal, overweight and obese children (condition x group effect,  $p > 0.05$ ). Therefore, adiposity did not have an effect on HRV.

**Table 2.** Difference in HRV parameters of normal weight, overweight and obese children.

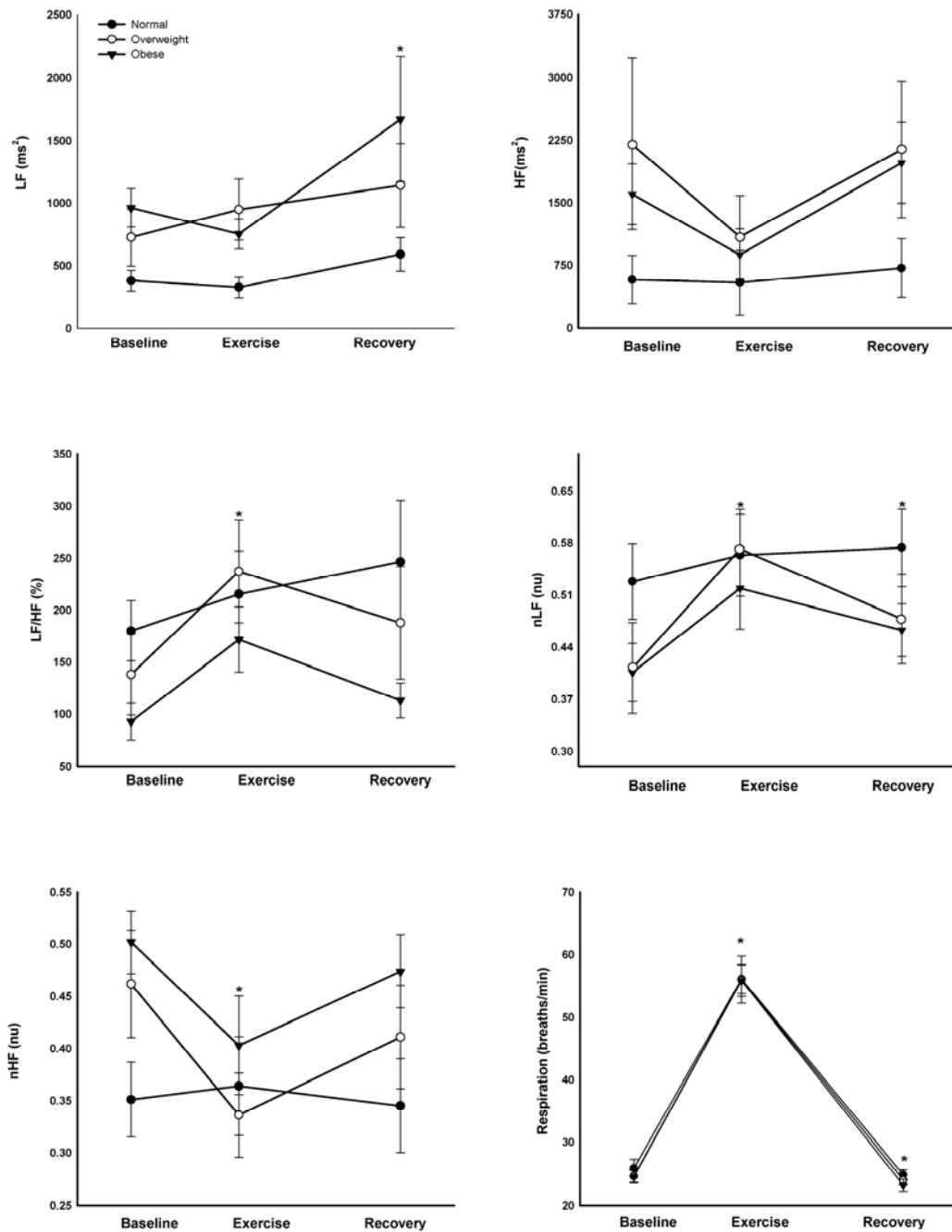
Variable	Group	N	Mean Base		Mean Exercise		Mean Recovery	
HR (bpm)	Normal	13	102	± 3	109	± 3	102	± 2
	Overweight	14	98	± 3	104	± 3	98	± 3
	Obese	14	93	± 2	102	± 2	93	± 2
RRI (ms)	Normal	13	599	± 17	559	± 17	597	± 16
	Overweight	14	630	± 22	588	± 21	629	± 20
	Obese	14	661	± 17	602	± 14	656	± 18
RRISD (ms)	Normal	13	41	± 8	39	± 7	46	± 7
	Overweight	14	64	± 13	59	± 10	70	± 11
	Obese	14	65	± 7	53	± 5	72	± 7
LF (ms <sup>2</sup> )	Normal	13	378	± 85	324	± 85	588	± 133
	Overweight	14	723	± 229	947	± 244	1140	± 336
	Obese	14	961	± 154	751	± 120	1667	± 497
HF (ms <sup>2</sup> )	Normal	13	575	± 290	539	± 387	713	± 352
	Overweight	14	2198	± 1025	1085	± 496	2136	± 820
	Obese	14	1599	± 364	874	± 309	1977	± 484
LF/HF	Normal	13	182	± 29	215	± 42	246	± 59
	Overweight	14	138	± 39	237	± 49	188	± 54
	Obese	14	93	± 18	172	± 33	113	± 17
nLF (nu)	Normal	13	0.53	± 0.05	0.56	± 0.06	0.57	± 0.05
	Overweight	14	0.41	± 0.06	0.57	± 0.05	0.48	± 0.06
	Obese	14	0.41	± 0.04	0.52	± 0.06	0.46	± 0.04
nHF (nu)	Normal	13	0.35	± 0.04	0.36	± 0.17	0.34	± 0.05
	Overweight	14	0.46	± 0.05	0.34	± 0.04	0.41	± 0.05
	Obese	14	0.50	± 0.03	0.40	± 0.05	0.47	± 0.04
Resp (breaths/min)	Normal	13	26	± 1	28	± 1	25	± 1
	Overweight	14	25	± 1	28	± 2	24	± 1
	Obese	14	24	± 1	28	± 1	23	± 1

HR, heart rate (bpm); RRI, RR intervals (ms); RRISD, RR interval standard deviation (ms); LF, low frequency (ms<sup>2</sup>); HF, high frequency (ms<sup>2</sup>); LF/HF, low to high frequency ratio (ms<sup>2</sup>); nLF, normalized low frequency (nu); nHF, normalized high frequency (nu); Resp, Respiration breaths per minute. No group x condition effects were found. Condition effects are shown in Figures 1.1 and 1.2.



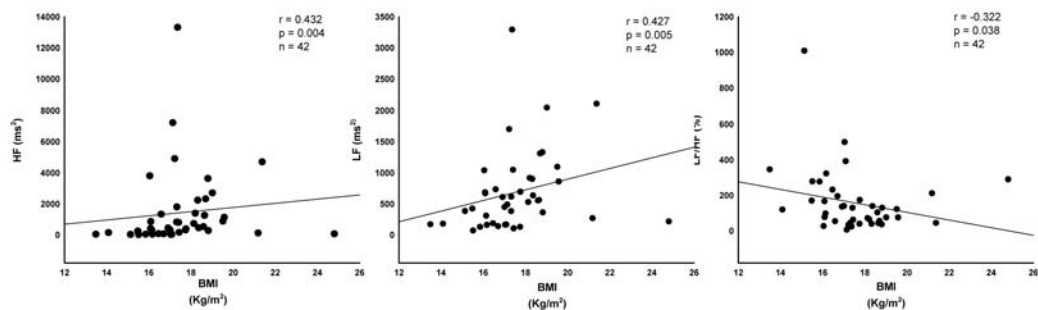


**Figure 2.** Heart rate (HR), RR interval (RRI) and RRI standard deviation (RRISD) during baseline, exercise and recovery for normal, overweight and obese children. HR and RRI were significantly different from baseline during exercise. RRISD was significantly different from baseline during recovery. \* Significantly different from baseline ( $p < 0.05$ ).

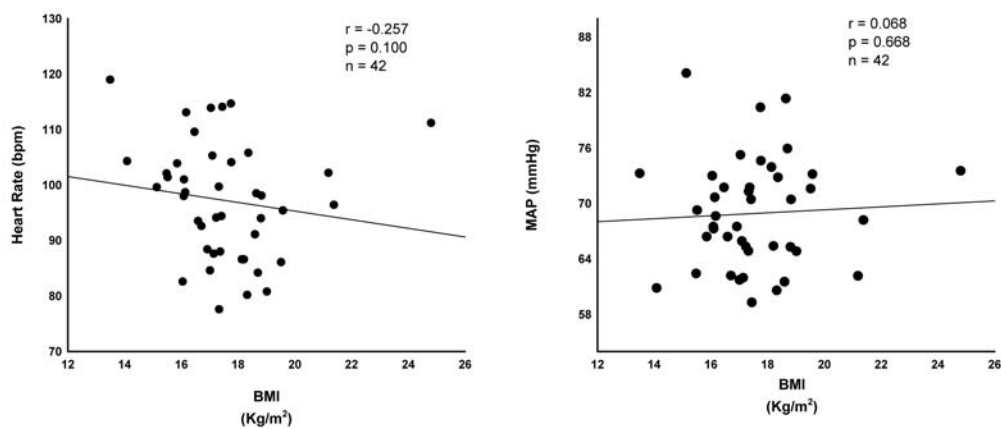


**Figure 3.** Low frequency (LF), high frequency (HF), LF/HF, normalized LF (nLF), normalized HF (nHF) and Respiration during baseline, exercise and recovery for normal, overweight and obese children. LF/HF, nLF, nHF and respiration were significantly different from baseline during exercise. LF, nLF and respiration were significantly different from baseline during recovery. \* Significantly different from baseline (p<0.05).

Figure 4 demonstrates that BMI was significantly correlated to LF, HF and LF/HF. Figure 5 shows that BMI was not correlated to HR and MAP.



**Figure 4.** Relations between BMI (kg/m<sup>2</sup>) and HF, LF and LF/HF during baseline.



**Figure 5.** Relations between BMI (kg/m<sup>2</sup>) and heart rate (HR) and mean arterial pressure (MAP) during baseline.

## DISCUSSION

This study is the first to report the influence of adiposity on HRV in a pediatric population of 3-5 year olds. Primary findings indicate significant differences in HRV during exercise, but no differences were found between normal, overweight and obese children. Contrary to our original hypothesis, higher BMI in the pediatric population was associated with higher parasympathetic and lower sympathetic activity. These findings are contrary to HRV responses observed in adults and adolescents, which suggests complex relationships between age, obesity and autonomic control of the heart.

Although differences in HRV variables were not found between normal, overweight and obese children, significant condition effects were observed. Specifically, HR, LF/HF and nLF increased during exercise while RRI, nHF and respiration decreased. RRISD, LF and HF were not altered during exercise. Studies have examined the pattern of HRV variables during exercise, but never in children ages 3-5 years old. Chen et al. (2007) looked at HRV variables in children (ages 8-13 years) with Type I diabetes during 10 minutes of exercise on a fixed rhythm stair-stepper. Both LF and HF significantly decreased during exercise, demonstrating a reduction in both sympathetic and parasympathetic activity. A normal cardiovascular response to exercise in healthy subjects is parasympathetic withdrawal, which results in increased HR and greatly reduced HRV (Yoshiharu et al. 1991 and Macor et al. 1996). This exercise response was observed in all 3 groups of the current study, including the overweight and obese groups. The reduction in parasympathetic activity, determined by HF, tended to decrease during exercise in all groups, but did not quite reach statistical significance ( $p < 0.09$ ).

Therefore, our exercise findings confirm previous work of Chen et al. (2007), that parasympathetic activity decreases during exercise.

Previous studies on children ages 6 to 14 years old have reported increased adiposity in children is associated with reduced HRV (Martini et al. 2001; Nagai et al. 2003; Rabbia et al. 2003; Riva et al. 2001 and Yakinci et al. 2000). Increased levels of adiposity were associated with decreased parasympathetic activity and either an increase (Martini et al. 2001; Rabbia et al. 2003 and Riva et al. 2001), decrease (Nagai et al. 2003) or no change (Yakinci et al. 2000) in sympathetic activity. To date, no studies have examined the influence of adiposity on HRV in young pediatric patients. The current study demonstrated no change in HRV between normal, overweight and obese 3-5 year olds. In fact, higher BMI was associated with higher parasympathetic and lower sympathetic activity. This is in stark contrast to what has been observed in previous studies, which is lower parasympathetic activity and variable sympathetic responses to obesity.

Reduced sympathetic activity in obese children has been previously reported by Nagai et al. (2004). Reductions of sympathetic activity have been shown to lead to lower energy expenditure levels, and consequently, a positive energy balance and weight gain (Peterson et al. 1988). However, lower sympathetic activity induced by obesity is not a consistent finding, as Martini et al. (2001); Rabbia et al. (2003) and Riva et al. (2001) have reported increased sympathetic activity with obesity. Controversial findings regarding sympathetic activity are also present in adult obesity. Bray et al. (1986) and Matsumoto et al. (1999) reported decreased sympathetic activity and associated it with the duration of obesity, leading to the conclusion that reduced sympathetic activity leads

to the onset or development of obesity. Discrepancies may result from the use of different methods in analyzing the autonomic nervous system. Thus, the role of adiposity on sympathetic activity remains controversial. Potential explanations for this controversy may be due to difficulty controlling for variables such as age, gender, duration of obesity, medical complications, nutrition, activity habits and emotional stress.

Whereas the influence of adiposity on sympathetic activity remains controversial, parasympathetic responses to obesity are remarkably consistent. All previous literature on parasympathetic activity and obese children aged 6 years and older demonstrate reduced parasympathetic activity (Martini et al. 2001; Nagai et al. 2003; Rabbia et al. 2003; Riva et al. 2001 and Yakinci et al. 2000). Specifically, Nagai et al. (2003) reported significantly decreased HF in obese children during 5 minute recordings. Martini et al. (2001); Rabbia et al. (2003); and Riva et al. (2001) observed significantly reduced HF using 24 hour Holter monitors. Furthermore, Yakinci et al. (2000) observed decreased HF measured via Valsalva's maneuver. Thus, evidence suggests increased adiposity is associated with decreased parasympathetic activity in children and adolescents.

In the current study, power spectral analyses detected several correlations between BMI and HRV variables. RRISD, LF, HF and nHF were all positively correlated with BMI while LF/HF was negatively correlated with BMI. The increase of HF with increasing levels of BMI suggest an increase in parasympathetic activity as one becomes obese. Thus, in contrast to previous studies (Martini et al. 2001; Nagai et al. 2003; Rabbia et al. 2003; Riva et al. 2001 and Yakinci et al. 2000), the current study demonstrated higher adiposity in children ages 3-5 years old was associated with higher

parasympathetic activity. The differences between our study and others are likely due to the influence of age on the developing heart.

Massin et al. (1997) demonstrated that heart rate variability and mean RR intervals are dependant on age in normal subjects, and that both contribute to the differences in overall HRV. More specifically, cholinergic increases and adrenergic decreases of HRV with age lead to the conclusion that the autonomic nervous system matures progressively. Finley et al. (1995) further showed an age reliance of heart rate variability with LF and HF components increasing from one month to six years and decreasing there after until 24 years. It was concluded that the changes in parasympathetic and sympathetic activity of heart rate during development are dependent upon age. The greatest variability in the study by Finley et al. (1995) was found in the infant group, which was thought to be due to several mechanisms producing the heart rate variations, including development of the parasympathetic innervations of the sinoatrial node, changes in cardiac volume occurring with growth and respiratory factors affecting HF and LF variability. Other studies have found that the maturation state in children affects autonomic nervous activity with greater vagal tone than adults (Lenard et al. 2004 and Silvetti et al. 2001). It is believed that both mechanical and neural alterations with age account for these differences (Lenard et al. 2004). The data presented in this thesis supports a novel relation between adiposity and BMI in 3-5 year olds. The tight age range of our subjects (3-5 years old) and the variability associated with HRV did not allow us to probe more closely for the influence of age on our data. Future studies should be conducted to further observe the combined influence of age and adiposity on HRV.

Increased physical activity is associated with higher levels of parasympathetic and sympathetic activity in adults (Davy et al. 1998; Goldsmith et al. 1992; and Amano et al. 2001) and children (Gutin et al. 2000). Physical activity levels were not considered or recorded for the subject's who participated in this study. One possible explanation for the increased parasympathetic activity in the obese children is that they were potentially more physically active than the normal and overweight subjects, although this does not agree with the assumptions that leaner children are naturally more active than their overweight and obese counterparts. Another possibility is that with short onset of obesity, as with a pediatric population, the autonomic nervous system has yet to adapt fully to the alterations imposed by obesity.

Autonomic nervous system activity was measured via HRV for this study, which is a well accepted method in the research, but was only measured for a limited amount of time. HRV was recorded for 3 minutes at baseline, 2 minutes during exercise and 3 minutes at recovery. A pilot study was conducted which included a 5 minute baseline, but it was found to be too long to keep a child's focus in a resting position. Several studies report HRV during short-term recordings between 2 and 5 minutes (Pomeranze et al. 1985; Pagani et al. 1986; Sandercock et al. 2005 and Kleiger et al. 1991). Additionally, Finley utilized 4 minute segments of continuous electrocardiogram recordings when examining HRV in subjects 5 to 24 years of age. Therefore, we are confident that our 8 minute protocol was appropriate and valid.

One conclusion that can be drawn from this study is that the relations between HRV and adiposity in 3-5 year olds may not correlate the same in adults and adolescents. HRV in adults has been reported to be highly reproducible in both short term (2-10 min)



and long-term (24 h) studies (Pagani et al. 1986; Amara et al. 1998; Schroeder et al. 2004; Guijt et al. 2007 and Pitzalis et al. 1996). In contrast, Leicht et al. (2008) compared HRV during light to moderate exercise after 8 weeks and reported moderate to poor reproducibility in children. HRV is a valuable and useful tool for analyzing the autonomic nervous system, but future studies need to confirm the reproducibility of HRV in children and adolescents.

In addition to HRV, we also examined resting heart rate and mean arterial pressure in the 3 groups. Our findings demonstrate no differences in resting HR and MAP between normal, overweight, and obese pediatric subjects. Furthermore, HR and MAP were not correlated with BMI. Yakinci et al. (2000) found similar results, demonstrating no correlation between BMI and differences of systolic or diastolic blood pressure in obese 7-13 year olds. In contrast, Riva et al. (2001) reported higher blood pressure and Nagai et al. (2003) reported significantly higher HR in obese children. Furthermore, Rabbia et al. (2003) and Martini et al. (2001) observed both higher 24-hour diastolic blood pressure and HR in obese children. These discrepancies demonstrate the complex relationships between heart rate, blood pressure and the pediatric population.

In summary, this study is the first to report the influence of adiposity on HRV in 3-5 year olds. Contrary to HRV responses in adolescents and adults, higher BMI in the pediatric population appears to be associated with higher parasympathetic and lower sympathetic activity. These discrepancies suggest a high level of complexity between the autonomic nervous system, obesity and the pediatric population.

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**Appendix A-1: Raw Data for subject descriptive characteristics****Normal**

<b>Subject #</b>	<b>Sex</b>	<b>Age (yrs)</b>	<b>Height (cm)</b>	<b>Weight (kg)</b>	<b>BMI (kg/m<sup>2</sup>)</b>
1	M	5	112	20.3	16.1
2	F	4	101	13.7	13.5
3	M	4	104	18.0	16.7
4	M	3	101	16.8	16.5
5	M	4	102	17.1	16.6
6	M	3	104	15.1	14.1
7	F	4	104	16.7	15.5
8	F	4	105	16.9	15.5
9	M	4	110	18.4	15.1
10	M	4	102	16.5	15.9
11	M	4	104	17.3	16.1
12	F	3	96	15.0	16.2
13	M	4	107	18.2	16.0
14	F	5	96	14.9	16.1

**Overweight**

<b>Subject #</b>	<b>Sex</b>	<b>Age (yrs)</b>	<b>Height (cm)</b>	<b>Weight (kg)</b>	<b>BMI (kg/m<sup>2</sup>)</b>
15	M	4	107	19.3	16.9
16	F	4	107	20.1	17.7
17	M	3	107	20.4	17.7
18	M	5	109	20.1	17.0
19	M	3	100	17.3	17.1
20	F	4	99	17.0	17.4
21	F	4	106	19.2	17.2
22	F	3	99	16.7	17.1
23	M	4	109	20.2	17.0
24	M	4	104	18.7	17.4
25	M	4	111	21.9	17.8
26	M	4	106	19.6	17.3
27	F	4	110	20.9	17.4
38	M	5	116	23.4	17.3

**Obese**

<b>Subject</b>		<b>Age</b>	<b>Height</b>	<b>Weight</b>	<b>BMI</b>
<b>#</b>	<b>Sex</b>	<b>(yrs)</b>	<b>(cm)</b>	<b>(kg)</b>	<b>(kg/m<sup>2</sup>)</b>
28	F	4	111	26.1	21.4
29	F	4	111	23.4	19.0
30	F	4	101	19.1	18.8
31	F	3	95	16.4	18.3
32	F	3	98	18.1	18.8
33	M	4	112	22.7	18.1
35	M	4	109	22.1	18.6
36	F	4	103	20.6	19.6
37	F	4	113	23.6	18.4
39	F	5	129	31.1	18.7
40	F	4	123	37.7	24.8
41	F	5	112	23.0	18.2
42	F	5	118	27.2	19.5
43	F	5	114	24.0	18.6

**Appendix A-2: Raw Data for SAP, DAP and MAP****Normal**

<b>Subject #</b>	<b>SAP (mmHg)</b>	<b>DAP (mmHg)</b>	<b>MAP (mmHg)</b>
1	88.0	62.0	70.7
2	98.6	60.6	73.3
3	82.6	52.0	62.2
4	92.6	61.3	71.7
5	80.6	59.3	66.4
6	82.6	50.0	60.9
7	82.6	62.6	69.3
8	71.3	58.0	62.4
9	112.3	70.0	84.1
10	98.0	50.6	66.4
11	81.3	60.6	67.5
12	83.3	61.3	68.6
13	95.0	62.0	73.0
14	88.6	56.6	67.3

**Overweight**

<b>Subject #</b>	<b>SAP (mmHg)</b>	<b>DAP (mmHg)</b>	<b>MAP (mmHg)</b>
15	93.3	54.6	67.5
16	61.3	62.6	62.2
17	108.0	66.6	80.4
18	100.6	62.6	75.3
19	83.3	51.3	62.0
20	75.3	51.3	59.3
21	80.0	58.0	65.3
22	84.6	56.6	65.9
23	80.0	52.6	61.7
24	83.3	64.0	70.4
25	101.3	61.3	74.6
26	84.0	55.3	64.9
27	90.0	62.6	71.7
38	89.3	62.3	71.3



**Obese**

<b>Subject #</b>	<b>SAP (mmHg)</b>	<b>DAP (mmHg)</b>	<b>MAP (mmHg)</b>
28	84.6	60.0	68.2
29	93.3	50.6	64.8
30	85.3	55.3	65.3
31	80.6	50.6	60.6
32	87.3	62.0	70.4
33	100.6	60.6	73.9
35	108.7	67.7	81.4
36	98.3	60.6	73.2
37	97.3	60.6	72.8
39	94.6	66.6	75.9
40	98.0	61.3	73.5
41	85.6	55.3	65.4
42	93.6	60.6	71.6
43	82.0	51.3	61.5

SAP, systolic arterial blood pressure (mmHg);

DAP, diastolic arterial blood pressure (mmHg);

MAP, mean arterial blood pressure (mmHg)

**Appendix A-3: Raw Data for HR (bpm) during baseline, exercise and recovery****Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	98.7	117.9	98.5
2	119	128.6	119.3
3	92.6	103.5	92.9
4	109.6	108.2	109.6
5	93.5	101	96.7
6	104.3	117.2	105.3
7	101.4	105.7	103
8	102.1	119.3	101.6
9	99.6	-	98.4
10	103.9	109.4	107.7
11	98	102.1	99.2
12	113.1	112.9	110.8
13	82.6	86.7	83.4
14	101	103.1	97.8

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
15	88.4	98	89.9
16	102.2	119.1	99
17	114.7	119.7	116.1
18	113.9	115.5	112
19	87.6	87.7	91
20	114.1	122.5	113.8
21	94.1	107.5	96.9
22	105.3	113.8	106.2
23	84.6	93.2	83.1
24	94.4	100.2	93
25	104.1	101	102.4
26	99.7	97.3	95.1
27	88	100.6	86.8
38	77.6	86.3	82.6

**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	96.4	112.8	100.1
29	80.8	93.1	81.6
30	94	108.4	98.7
31	80.2	84.8	80.3
32	98.1	105.8	98.3
33	86.6	96.7	91.6
35	98.5	103.4	103.4
36	95.4	100.3	98.2
37	105.8	111.7	104.3
39	84.2	100.1	82.6
40	111.2	109.2	106.8
41	86.6	90.2	85.3
42	86.1	98.6	85.6
43	91.1	99.1	90

**Appendix A-4: Raw Data for RRI (ms) during baseline, exercise and recovery****Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	610	511	612
2	505	467	504
3	649	582	648
4	549	557	548
5	652	597	626
6	577	515	572
7	592	569	584
8	590	504	595
9	605	-	613
10	578	550	558
11	615	590	609
12	532	533	544
13	742	714	735
14	598	584	620

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
15	683	616	672
16	590	507	611
17	525	504	518
18	528	521	537
19	714	713	683
20	527	490	529
21	653	568	632
22	570	529	566
23	712	650	731
24	641	603	648
25	579	570	587
26	608	627	642
27	712	599	714
38	778	738	740

**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	633	539	610
29	753	647	747
30	653	557	619
31	762	724	763
32	614	570	613
33	696	626	663
35	614	584	585
36	635	602	620
37	570	541	580
39	720	606	738
40	540	551	563
41	703	670	713
42	701	611	704
43	662	609	671

**Appendix A-5: Raw Data for RRISD (ms) during baseline, exercise and recovery****Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	34	32	42
2	23	18	22
3	28	38	35
4	23	37	25
5	87	38	64
6	29	36	32
7	22	28	34
8	41	28	55
9	36	-	43
10	19	25	23
11	40	39	50
12	26	32	36
13	117	123	113
14	50	39	69

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
15	54	52	54
16	38	40	52
17	29	34	29
18	18	23	19
19	158	155	143
20	21	20	29
21	107	82	98
22	19	27	30
23	47	61	83
24	61	55	46
25	55	51	58
26	61	85	89
27	164	43	148
38	58	100	101

**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	89	67	92
29	87	42	93
30	108	46	90
31	101	111	109
32	37	41	42
33	52	60	76
35	61	53	51
36	63	49	81
37	42	46	54
39	78	55	101
40	24	32	22
41	72	53	82
42	54	36	52
43	46	45	56

**Appendix A-6: Raw Data for LF (ms<sup>2</sup>) during baseline, exercise and recovery****Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	309	163	260
2	171	79	253
3	142	310	321
4	187	259	179
5	734	331	1137
6	181	197	349
7	72	364	260
8	425	183	1219
9	385	-	446
10	131	33	170
11	687	462	415
12	163	292	414
13	1041	1271	1552
14	670	268	1119

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
15	610	395	624
16	269	875	836
17	131	130	177
18	166	164	148
19	491	2673	705
20	105	191	215
21	1703	1170	1619
22	167	80	187
23	450	721	1141
24	1049	911	568
25	695	984	1178
26	383	2472	942
27	3294	161	2946
38	615	2330	4675



**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	2114	170	7547
29	2050	322	2066
30	1332	336	1052
31	902	1215	2386
32	365	540	419
33	531	1479	1243
35	562	550	454
36	861	702	1772
37	637	1073	472
39	1313	1136	2268
40	217	174	164
41	918	1356	2115
42	1097	449	616
43	555	1011	759

**Appendix A-7: Raw Data for HF (ms<sup>2</sup>) during baseline, exercise and recovery****Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	315	89	462
2	50	32	56
3	72	98	98
4	81	147	91
5	1341	150	1136
6	150	205	185
7	26	86	87
8	249	37	1112
9	38	-	61
10	47	61	42
11	412	328	423
12	50	81	52
13	3812	5166	4720
14	864	532	805

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
15	446	249	585
16	127	149	110
17	328	138	128
18	33	61	33
19	7229	6727	8870
20	166	39	102
21	4913	1834	2423
22	43	27	61
23	322	131	1678
24	807	377	436
25	399	431	796
26	830	2037	2954
27	13318	168	9048
38	1808	2820	2682

**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	4705	112	4223
29	2712	301	3248
30	3639	1024	2480
31	2244	4573	3478
32	280	145	254
33	736	834	630
35	1262	1656	326
36	1133	940	2325
37	457	818	346
39	2324	514	6134
40	75	56	68
41	1399	790	2317
42	892	201	918
43	533	272	929

**Appendix A-8: Raw Data for LF/HF (ms<sup>2</sup>) during baseline, exercise and recovery****Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	98.3	183.2	56.4
2	345.6	247.4	455.8
3	195.5	315.5	326.5
4	231.9	176.6	197.7
5	54.7	220.3	100.1
6	120.2	96.4	188.8
7	278.3	424.3	299.4
8	170.4	501.4	109.7
9	1008.6	-	735.9
10	277.3	53.8	403.8
11	166.8	140.9	97.9
12	323.2	362.4	796.3
13	27.3	24.6	32.9
14	77.5	50.4	139.1

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
15	136.7	158.8	106.7
16	211.5	587.6	759.8
17	40	93.9	138.4
18	499.9	268.8	447.5
19	6.8	39.7	7.9
20	63.1	492.5	211.1
21	34.7	63.8	66.8
22	392.3	295.7	307.1
23	139.7	548.2	68
24	130.1	241.4	130.1
25	174.2	228.4	147.9
26	46.2	121.3	31.9
27	24.7	95.8	32.6
38	34	82.6	174.3

**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	44.9	151.9	178.7
29	75.6	107	63.6
30	36.6	32.9	42.4
31	40.2	26.6	68.6
32	130.6	371.9	165.2
33	72.2	177.4	197.2
35	44.6	33.2	139.1
36	76	74.7	76.2
37	139.4	131.2	136.4
39	56.5	221	37
40	289.8	307.4	239.3
41	65.6	171.6	91.3
42	123	223	67.1
43	104.2	372.2	81.7

**Appendix A-9: Raw Data for nLF (nu) during baseline, exercise and recovery****Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	0.482	0.585	0.35
2	0.716	0.661	0.761
3	0.6	0.737	0.715
4	0.585	0.612	0.6
5	0.211	0.647	0.424
6	0.412	0.461	0.584
7	0.599	0.725	0.641
8	0.599	0.806	0.517
9	0.886	-	0.856
10	0.702	0.316	0.782
11	0.607	0.525	0.463
12	0.757	0.777	0.876
13	0.168	0.16	0.189
14	0.422	0.32	0.563

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
15	0.539	0.58	0.474
16	0.647	0.852	0.839
17	0.238	0.253	0.5
18	0.789	0.718	0.783
19	0.041	0.218	0.061
20	0.37	0.801	0.652
21	0.243	0.368	0.336
22	0.741	0.733	0.634
23	0.534	0.765	0.356
24	0.527	0.675	0.493
25	0.477	0.647	0.54
26	0.199	0.491	0.208
27	0.181	0.476	0.197
38	0.24	0.429	0.623

**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	0.298	0.557	0.626
29	0.405	0.35	0.38
30	0.215	0.232	0.257
31	0.181	0.178	0.345
32	0.537	0.782	0.517
33	0.35	0.587	0.636
35	0.287	0.233	0.553
36	0.407	0.389	0.416
37	0.537	0.558	0.522
39	0.338	0.67	0.259
40	0.722	0.724	0.676
41	0.381	0.599	0.466
42	0.539	0.654	0.395
43	0.481	0.751	0.437

**Appendix A-10: Raw Data for nHF (nu) during baseline, exercise and recovery****Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	0.49	0.319	0.62
2	0.207	0.267	0.167
3	0.307	0.234	0.219
4	0.252	0.347	0.304
5	0.386	0.294	0.423
6	0.343	0.479	0.309
7	0.215	0.171	0.214
8	0.352	0.161	0.471
9	0.088	-	0.116
10	0.253	0.588	0.194
11	0.364	0.372	0.473
12	0.234	0.214	0.11
13	0.616	0.65	0.575
14	0.544	0.634	0.405

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
15	0.394	0.365	0.444
16	0.306	0.145	0.11
17	0.595	0.27	0.362
18	0.158	0.267	0.175
19	0.604	0.549	0.762
20	0.587	0.163	0.309
21	0.701	0.577	0.503
22	0.189	0.248	0.206
23	0.382	0.139	0.524
24	0.405	0.279	0.379
25	0.274	0.283	0.365
26	0.431	0.404	0.653
27	0.732	0.496	0.604
38	0.705	0.519	0.357



**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	0.664	0.366	0.35
29	0.535	0.327	0.598
30	0.586	0.706	0.607
31	0.451	0.669	0.504
32	0.411	0.21	0.313
33	0.485	0.331	0.323
35	0.645	0.702	0.398
36	0.535	0.521	0.546
37	0.385	0.425	0.383
39	0.599	0.303	0.702
40	0.249	0.236	0.282
41	0.58	0.349	0.51
42	0.438	0.293	0.589
43	0.462	0.202	0.534

**Appendix A-11:** Raw Data for Respiration (breaths/min) during baseline, exercise and recovery

**Normal**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	18	27	21
2	26	24	25
3	23	26	23
4	23	27	25
5	29	35	29
6	26	29	27
7	27	28	26
8	28	33	24
9	25	***	25
10	19	23	21
11	20	22	21
12	36	35	30
13	28	32	26
14	32	29	23

**Overweight**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
1	29	43	28
2	29	31	26
3	24	24	24
4	28	26	27
5	25	27	24
6	23	26	23
7	18	20	20
8	30	39	28
9	23	20	22
10	22	33	26
11	25	29	25
12	24	25	21
13	19	33	21
14	23	20	19

**Obese**

<b>Subject</b>	<b>Baseline</b>	<b>Exercise</b>	<b>Recovery</b>
28	22	30	21
29	19	28	17
30	32	41	31
31	24	30	24
32	31	32	27
33	26	27	26
35	23	23	24
36	28	29	26
37	26	26	22
39	24	20	18
40	22	26	23
41	21	28	23
42	24	28	21
43	22	27	19

**Appendix B-1: Means for normal, overweight and obese children - demographics**

<b>Variable</b>	<b>Group</b>	<b>N</b>	<b>Mean</b>
<b>HT</b>	Normal	14	103 ± 1
	Overweight	14	106 ± 1
	Obese	14	111 ± 3
<b>WT</b>	Normal	14	17 ± 0
	Overweight	14	20 ± 1
	Obese	14	24 ± 1
<b>BMI</b>	Normal	14	16 ± 0
	Overweight	14	17 ± 0
	Obese	14	19 ± 0
<b>SAP</b>	Normal	14	88 ± 3
	Overweight	14	87 ± 3
	Obese	14	92 ± 2
<b>DAP</b>	Normal	14	59 ± 1
	Overweight	14	59 ± 1
	Obese	14	59 ± 1
<b>MAP</b>	Normal	14	69 ± 2
	Overweight	14	68 ± 2
	Obese	14	70 ± 2
<b>MVC</b>	Normal	14	2 ± 0
	Overweight	14	2 ± 0
	Obese	14	2 ± 0

HT, height (cm); WT, weight (kg); BMI, body mass index (kg/m<sup>2</sup>);  
 SAP, systolic arterial blood pressure (mmHg); DAP, diastolic  
 arterial blood pressure (mmHg); MAP, mean arterial blood  
 pressure (mmHg); MVC, maximal voluntary contraction (p.s.i.).

**Appendix B-2: Means for normal, overweight and obese children – HRV variables**

<b>Variable</b>	<b>Group</b>	<b>N</b>	<b>Baseline</b>		<b>Exercise</b>		<b>Recovery</b>	
<b>HR</b>	Normal	13	102	± 3	109	± 3	102	± 3
	Overweight	14	98	± 3	104	± 3	98	± 3
	Obese	14	93	± 2	102	± 2	93	± 2
<b>RRI</b>	Normal	13	599	± 17	559	± 17	597	± 16
	Overweight	14	630	± 22	588	± 21	629	± 20
	Obese	14	661	± 17	603	± 14	656	± 18
<b>RRISD</b>	Normal	13	41	± 8	39	± 7	46	± 7
	Overweight	14	64	± 13	59	± 10	70	± 11
	Obese	14	65	± 7	53	± 5	72	± 7
<b>LF</b>	Normal	13	378	± 85	324	± 85	588	± 133
	Overweight	14	723	± 229	947	± 244	1140	± 336
	Obese	14	961	± 154	751	± 120	1667	± 497
<b>HF</b>	Normal	13	575	± 290	539	± 387	713	± 352
	Overweight	14	2198	± 1025	1085	± 496	2136	± 820
	Obese	14	1599	± 364	874	± 309	1977	± 484
<b>LF_HF</b>	Normal	13	182	± 29	215	± 42	246	± 59
	Overweight	14	138	± 39	237	± 49	188	± 54
	Obese	14	93	± 18	172	± 31	113	± 17
<b>nLF</b>	Normal	13	0.5	± 0.1	0.6	± 0.1	0.6	± 0.1
	Overweight	14	0.4	± 0.1	0.6	± 0.1	0.5	± 0.1
	Obese	14	0.4	± 0.0	0.5	± 0.1	0.5	± 0.0
<b>nHF</b>	Normal	13	0.4	± 0.0	0.4	± 0.1	0.3	± 0.0
	Overweight	14	0.5	± 0.1	0.3	± 0.0	0.4	± 0.1
	Obese	14	0.5	± 0.0	0.4	± 0.1	0.5	± 0.0
<b>Resp</b>	Normal	13	26	± 1	28	± 1	25	± 1
	Overweight	14	25	± 1	28	± 2	24	± 1
	Obese	14	24	± 1	28	± 2	23	± 1

HR, heart rate (bpm); RRI, RR intervals (ms); RRISD, RR interval standard deviation (ms); LF, low frequency (ms<sup>2</sup>); HF, high frequency (ms<sup>2</sup>); LF/HF, low to high frequency ratio (ms<sup>2</sup>); nLF, normalized low frequency (nu); nHF, normalized high frequency (nu); Resp, Respiration breaths per minute. Values reported as mean ± standard error.

### Appendix C-1: Repeated Measures ANOVA

#### HR

Within Subjects Effect	Mauchly's W	Approx. Chi-Square	df	Sig.	Greenhouse - Geisser
Condition	0.454	29.199	2	0.000	0.647

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Condition	Greenhouse-Geisser 1465.373	1.294	1132.571	58.861	0.000
Condition * Group	Greenhouse-Geisser 12.06	2.588	4.661	0.242	0.839

#### RRI

Within Subjects Effect	Mauchly's W	Approx. Chi-Square	df	Sig.	Greenhouse - Geisser
Condition	0.532	23.363	2	0.000	0.681

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Condition	Greenhouse-Geisser 56188.650	1.362	41247.322	57.910	0.000
Condition * Group	Greenhouse-Geisser 1701.686	2.724	624.592	0.877	0.451

## RRISD

Within Subjects Effect	Mauchly's W	Approx. Chi-Square	df	Sig.	Greenhouse - Geisser
Condition	0.600	18.872	2	0.000	0.715

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Condition	Greenhouse-Geisser	3018.054	1.429	2111.943	6.583	0.007
Condition *	Greenhouse-Geisser	647.545	2.858	226.566	0.706	0.546

## LF

Within Subjects Effect	Mauchly's W	Approx. Chi-Square	df	Sig.	Greenhouse - Geisser
Condition	0.773	9.509	2	0.009	0.815

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Condition	Huynh-Feldt	5555901.377	1.783	3115909.418	4.225	0.022
Condition *	Huynh-Feldt	2475830.308	3.566	694258.452	0.941	0.438

## HF

Within Subjects Effect	Mauchly's W	Approx. Chi-Square	df	Sig.	Greenhouse - Geisser
Condition	0.533	23.265	2	0.000	0.682

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Condition	Greenhouse-Geisser	1.385E+07	1.364	1.016E+07	4.073	0.037
Condition * Group	Greenhouse-Geisser	5535858.337	2.727	2029937.26	0.814	0.482

## LF/HF

Within Subjects Effect	Mauchly's W	Approx. Chi-Square	df	Sig.	Greenhouse - Geisser
Condition	0.885	4.502	2	0.105	0.897

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Condition	Sphericity Assumed	103625.585	2	51812.793	4.677	0.012
Condition * Group	Sphericity Assumed	36360.964	4	9090.241	0.821	0.516



## nLF

<b>Within Subjects Effect</b>	<b>Mauchly's W</b>	<b>Approx. Chi-Square</b>	<b>df</b>	<b>Sig.</b>	<b>Greenhouse - Geisser</b>
<b>Condition</b>	0.842	6.368	2	0.041	0.863

<b>Source</b>		<b>Type III Sum of Squares</b>	<b>df</b>	<b>Mean Square</b>	<b>F</b>	<b>Sig.</b>
<b>Condition</b>	<b>Huynh-Feldt</b>	0.219	1.897	0.115	7.945	0.001
<b>Condition *</b> <b>Group</b>	<b>Huynh-Feldt</b>	0.060	3.794	0.016	1.097	0.363

## nHF

<b>Within Subjects Effect</b>	<b>Mauchly's W</b>	<b>Approx. Chi-Square</b>	<b>df</b>	<b>Sig.</b>	<b>Greenhouse - Geisser</b>
<b>Condition</b>	0.888	4.387	2	0.112	0.899

<b>Source</b>		<b>Type III Sum of Squares</b>	<b>df</b>	<b>Mean Square</b>	<b>F</b>	<b>Sig.</b>
<b>Condition</b>	<b>Sphericity Assumed</b>	0.103	2	0.052	4.036	0.022
<b>Condition *</b> <b>Group</b>	<b>Sphericity Assumed</b>	0.078	4	0.019	1.515	0.206

**Respiration**

<b>Within Subjects Effect</b>	<b>Mauchly's W</b>	<b>Approx. Chi-Square</b>	<b>df</b>	<b>Sig.</b>	<b>Greenhouse - Geisser</b>
<b>Condition</b>	0.665	15.080	2	0.001	0.749

<b>Source</b>		<b>Type III Sum of Squares</b>	<b>df</b>	<b>Mean Square</b>	<b>F</b>	<b>Sig.</b>
<b>Condition</b>	<b>Greenhouse- Geisser</b>	365.599	1.498	243.987	24.503	0.000
<b>Condition * Group</b>	<b>Greenhouse- Geisser</b>	10.942	2.997	3.651	0.367	0.777

**Appendix C-2: Post Hoc analysis for demographics****PAIRED T-TESTS**

		95% Confidence Interval				
	<b>Mean</b>	<b>Lower</b>	<b>Upper</b>	<b>t</b>	<b>Df</b>	<b>Sig. (2-tailed)</b>
<b>HT</b>						
N vs. OV	-3.109	-7.139	0.921	-1.667	13	0.119
N vs. OB	-7.261	-13.145	-1.377	-2.666	13	0.019
OV vs. OB	-4.151	-9.569	1.267	-1.655	13	0.122
<b>WT</b>						
N vs. OV	-2.863	-4.336	-1.389	-4.198	13	0.001
N vs. OB	-7.151	-10.346	-3.956	-4.836	13	0.000
OV vs. OB	-4.289	-7.216	-1.362	-3.166	13	0.007
<b>BMI</b>						
N vs. OV	-1.609	-2.182	-1.036	-6.066	13	0.000
N vs. OB	-3.646	-4.717	-2.575	-7.353	13	0.000
OV vs. OB	-2.036	-3.028	-1.045	-4.439	13	0.001
<b>SAP</b>						
N vs. OV	1.650	-8.503	11.803	0.351	13	0.731
N vs. OB	-3.740	-11.484	4.003	-1.043	13	0.316
OV vs. OB	-5.390	-15.236	4.456	-1.183	13	0.258
<b>DAP</b>						
N vs. OV	0.371	-4.465	5.208	0.166	13	0.871
N vs. OB	0.271	-4.234	4.777	0.130	13	0.898
OV vs. OB	-0.100	-5.039	4.839	-0.044	13	0.966
<b>MAP</b>						
N vs. OV	0.798	-4.555	6.151	0.322	13	0.753
N vs. OB	-1.066	-5.982	3.851	-0.468	13	0.647
OV vs. OB	-1.863	-7.529	3.792	-0.712	13	0.489
<b>MVC</b>						
N vs. OV	-0.214	-0.534	0.105	-1.450	13	0.171
N vs. OB	-0.450	-0.909	0.009	-2.116	13	0.054
OV vs. OB	-0.236	-0.577	0.105	-1.494	13	0.159

HT, height (cm); WT, weight (kg); BMI, body mass index (kg/m<sup>2</sup>); SAP, systolic arterial blood pressure (mmHg); DAP, diastolic arterial blood pressure (mmHg); MAP, mean arterial blood pressure (mmHg); MVC, maximal voluntary contraction (p.s.i.); N, normal weight; OV, overweight; OB (obese).

**Appendix C-3: Post Hoc analyses for HRV variables****PAIRED T-TESTS**

		95% Confidence Interval				
	<b>Mean</b>	<b>Lower</b>	<b>Upper</b>	<b>T</b>	<b>df</b>	<b>Sig. (2-tailed)</b>
<b>HR</b>						
B vs. E	-7.529	-1.154	0.402	-0.977	40	0.000
B vs. R	-0.376	-1.154	0.402	-0.977	41	0.334
<b>RRI</b>						
B vs. E	46.829	35.578	58.081	8.412	40	0.000
B vs. R	2.452	-2.864	7.769	0.932	41	0.357
<b>RRISD</b>						
B vs. E	6.488	-1.797	14.773	1.5834	40	0.121
B vs. R	-5.801	-10.071	-1.548	-2.753	41	0.009
<b>LF</b>						
B vs. E	12.512	-275.386	300.410	0.088	40	0.930
B vs. R	-440.67	-775.705	-105.629	-2.656	41	0.011
<b>HF</b>						
B vs. E	638.854	-94.472	1372.179	1.761	40	0.086
B vs. R	-148.690	-514.623	217.242	-0.821	41	0.417
<b>LF/HF</b>						
B vs. E	-71.161	-116.633	-25.689	-3.163	40	0.003
B vs. R	-36.798	-78.403	4.807	-1.786	41	0.081
<b>nLF</b>						
B vs. E	-0.105	-0.155	-0.054	-4.186	40	0.000
B vs. R	-0.055	-0.098	-0.012	-2.597	41	0.013
<b>nHF</b>						
B vs. E	0.073	0.022	0.123	2.907	40	0.006
B vs. R	0.027	-0.015	0.069	1.294	41	0.203
<b>Resp</b>						
B vs. E	-3.057	-4.495	-1.619	-4.297	40	0.000
B vs. R	1.000	0.211	1.789	2.558	41	0.014

HR, heart rate (bpm); RRI, RR intervals (ms); RRISD, RR interval standard deviation (ms); LF, low frequency (ms<sup>2</sup>); HF, high frequency (ms<sup>2</sup>); LF/HF, low to high frequency ratio (ms<sup>2</sup>); nLF, normalized low frequency (nu); nHF, normalized high frequency (nu); Resp, Respiration breaths per minute; N (normal weight); OV (overweight); OB (obese).

**Appendix C-4: Statistics for BMI****NONPARAMETRIC CORRELATIONS**

Spearman's rho

	<b>n</b>	<b>r</b>	<b>Sig.</b>
<b>HR</b>	42	-0.257	0.100
<b>RRI</b>	42	0.264	0.091
<b>RRISD</b>	42	0.375	0.014
<b>LF</b>	42	0.427	0.005
<b>HF</b>	42	0.432	0.004
<b>LF/HF</b>	42	-0.322	0.038
<b>nLF</b>	42	-0.280	0.072
<b>nHF</b>	42	0.406	0.008
<b>Resp</b>	42	-0.140	0.376
<b>SAP</b>	42	0.118	0.455
<b>DAP</b>	42	0.070	0.661
<b>MAP</b>	42	0.068	0.668
<b>HT</b>	42	0.367	0.017
<b>WT</b>	42	0.753	0.000
<b>MVC</b>	42	0.242	0.123

HR, heart rate (bpm); RRI, RR intervals (ms); RRISD, RR interval standard deviation (ms); LF, low frequency (ms<sup>2</sup>); HF, high frequency (ms<sup>2</sup>); LF/HF, low to high frequency ratio (ms<sup>2</sup>); nLF, normalized low frequency (nu); nHF, normalized high frequency (nu); Resp, Respiration breaths per minute; SAP, systolic arterial blood pressure (mmHg); DAP, diastolic arterial blood pressure (mmHg); MAP, mean arterial blood pressure (mmHg); HT, height (cm); WT, weight (kg); BMI, body mass index (kg/m<sup>2</sup>); MVC, maximal voluntary contraction (p.s.i.).