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DEPARTMENT OF KINESIOLOGY

BAROREFLEX FUNCTION DURING ORTHOSTATIC STRESS WITH  
ADMINISTRATION OF A SOMATOSTATIN ANALOG

DAVID MICHAEL LEONE

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Reviewed and approved\* by the following:

James A. Pawelczyk  
Associate Professor of Physiology and Kinesiology  
Thesis Supervisor

Stephen J. Piazza  
Associate Professor of Kinesiology  
Honors Advisor

Karl M. Newell  
Marie Underhill Noll Chair  
Professor of Kinesiology and Biobehavioral Health  
Kinesiology Department Head

\*Signatures on file in the Schreyer Honors College

## **Abstract**

Arterial baroreflexes provide short-term blood pressure regulation during orthostatic stresses, such as standing. In this study we evaluated the magnitude and frequency of baroreflex engagement during orthostatic stress before and after blood volume was redistributed by constricting the splanchnic (gut) vasculature selectively with octreotide acetate, a somatostatin analog. **Purpose:** To evaluate the baroreflex sensitivity (BRS), and effectiveness index (BEI) during orthostatic stress, with and without pharmacological protection from orthostatic intolerance. Two procedures were employed with healthy, young subjects ( $n = 52$ ; ages 18-37). In the first procedure, subjects were tilted upright to  $70^\circ$ , once with a placebo and once with octreotide. In the second procedure, subjects were tilted at intervals of  $15^\circ$ ,  $30^\circ$ , and  $50^\circ$ . BRS and BEI were both evaluated post-tilt using the spontaneous association between changes in R-R Intervals and changes in systolic blood pressure. BRS was measured by the slope of the relationship. BEI was determined as the percentage of ramps in systolic blood pressure that elicited a baroreflex-mediated change in R-R interval. **Results:** In all protocols there was a significant decrease in BRS with increased orthostatic stress ( $p < 0.0001$ ). Conversely, there was a significant increase in BEI with increased stress ( $p = 0.0018$ ). Orthostatic tolerance was significantly increased ( $p = 0.001$ ) with octreotide, but BRS and BEI were unaffected. **Conclusion:** Passive tilt reduces the sensitivity of the baroreflex, but increases the frequency of engagement. The increased baroreflex activation is necessary to maintain blood pressure during orthostatic stress. We found that octreotide significantly increases orthostatic tolerance, but has no effect on BRS or BEI. These findings suggest that this improvement to tolerance was independent of arterial baroreflexes.

## Table of Contents

Introduction.....	1
Methods.....	5
Results.....	11
Discussion.....	16
Conclusion .....	19
Acknowledgments.....	20
References.....	21
Appendix.....	23
A- Protocol 1: Informed Consent .....	23
B- Protocol 2: Informed Consent .....	33
C- Protocol 3: Informed Consent .....	43

Academic Vita

**List of Tables and Figures**

Table 1. Subject Demographics .....5

Figure 1. Baroreflex Sequences .....9

Table 2. Cardiovascular changes during HUT.....11

Table 3. Baroreflex changes during HUT .....12

Figure 2. Orthostatic Tolerance changes with Octreotide .....13

Figure 3. Panel Graph of changes during HUT .....15

Figure 4. Conceptual Stimulus-Response Curve .....17

## **Introduction**

An estimated 500,000 Americans suffer from some form of orthostatic intolerance, which is denoted by a decrease in blood pressure by 20/10 mmHg or more upon attaining upright posture (Robertson, 1999). Many of these individuals experience symptoms such as fatigue, blurred vision, palpitations, and syncope, and affects their day-to-day living (Streeten & Anderson, 1992). Orthostatic hypotension is caused by excessive decreases in stroke volume and cardiac output as a result of blood pooling in the legs upon standing (Minson *et al.*, 1999).

The syncope due to orthostatic stresses is unique because it involves a combination of systemic vasodilation and bradycardia (Julu *et al.*, 2003). This phenomenon was interpreted by Sir Thomas Lewis who coined the term ‘Vasovagal Syncope,’ as being caused largely by systemic vasodilation (Lewis, 1932).

Tilt table testing is often used to induce vasovagal syncope to simulate the stresses of standing and to discriminate between patients who show symptoms and those who do not (Benditt *et al.*, 1996). Head-up-tilt (HUT) at 70° produces a reliable response and is useful in assessing a person’s orthostatic tolerance (OT) (Benditt *et al.*, 2004). Subjects are passively tilted with no skeletal muscle engagement until they show presyncopal signs, or in some cases experience complete syncope. Because of the diminished hydrostatic gradient, tilt at lesser angles (15°, 30°, or 50°) are not usually adequate enough to provoke syncope, but can still illustrate hemodynamic changes (Benditt *et al.*, 1996).

Over seventy years after Lewis' work, this carotid sinus mechanism is recognized as an important contributing factor to orthostatic tolerance. Arterial baroreceptors, mainly located in the carotid sinus and aortic arch, sense changes in blood pressure each beat (Eckberg, 2008). These receptors activate inhibitory reflexes whose efferents innervate vasculature via the sympathetic nervous system and the heart through the vagus nerve. Decreases in arterial blood pressure lessen the neural inhibition generated by the parasympathetic nervous system. The result is an increase in heart rate, cardiac contractility, and systemic vascular resistance (La Rovere *et al.*, 2008).

Baroreflexes play a very important role as a person stands. As compliant veins fill, blood pools below the heart, which leads to a decrease in stroke volume from reductions in venous return (preload) and cardiac output (Rowell, 1986; Minson *et al.*, 1999). The high transmural pressure within peripheral capillaries causes interstitial edema and further drops in blood volume. A decrease in blood volume elicits additional decreases in stroke volume. Constriction of the arterial and venous vasculature, mediated by carotid and aortic baroreflexes, slows the rate and magnitude of venous pooling (Minson *et al.*, 1999; Benditt *et al.*, 2004).

Reducing blood pooling in compliant circulations may be an effective therapy for orthostatic intolerance. For example, somatostatin has been shown to both increase orthostatic tolerance, as well as stabilize low blood pressures of patients with postprandial hypotension (Hoeldtke *et al.*, 1986; Wong & Sheriff, 2007). This is most likely due to the movement of blood out of the splanchnic circulation, via selective vasoconstriction, and into the central circulation (Rowell, 1986; Jarvis *et al.*, 2010). Displacing blood from the splanchnic to the central circulation causes an increase in

preload which can in turn increase stroke volume and cardiac output. Theoretically, increased preload should increase pulse pressure, leading to more baroreflex engagement. However, this hypothesis has not been tested experimentally. Although producing a desired pharmacological effect, one major limitation of somatostatin is its short plasma half-life. To overcome this obstacle in longer experiments, an analog with a longer lasting half-life is used. A very popular long-acting somatostatin analog is octreotide acetate (Nubiola *et al.*, 1989).

The extent that somatostatin analogs might alter baroreflex function is not known. Several techniques are available to assess baroreflex function. These include altering carotid artery transmural pressure with neck suction and pressure, and the use of vasoactive drugs (Di Rienzo *et al.*, 1997). More recently, the spontaneous sequence technique has been utilized more widely in the literature (Julu *et al.*, 2003; Steptoe & Vogeles, 1990; Parlow *et al.*, 1995; Di Rienzo *et al.*, 2001; Laude *et al.*, 2004; Eckberg & Kuusela, 2005). The sequence technique is a non-invasive procedure that quantifies baroreflex sensitivity as the slope of the relation between changes in beat-to-beat measurements of cardiac depolarization (in the form of R-R interval) and systolic blood pressure (See Figure 1).

However, not every change in blood pressure elicits a baroreflex-mediated change in heart rate. In addition to the baroreflex sensitivity (BRS), another way to assess the function of the baroreflex is to quantify the percentage of changes in blood pressure associated with a baroreflex-mediated change in R-R interval. This value has been termed the baroreflex effectiveness index (BEI) and determines the frequency of baroreflex engagement (Di Rienzo *et al.*, 2001).

The overall purpose of the present study is to better understand the function of the baroreflex during orthostatic stress. To our knowledge, the effect of somatostatin and its analogs on BRS and BEI have not been assessed; thus a secondary purpose was to determine how baroreflex function changes with the administration of octreotide. In addition, it has been shown that there may be some relationship between a person's baseline baroreflex function and their overall orthostatic tolerance (Pitzalis *et al.*, 2003; Iacoviello *et al.*, 2008). Theoretically, a higher sensitivity to the baroreflex at baseline may mean that that person's baroreflex is working better to defend against changes in blood pressure. Because octreotide causes dramatic improvements in orthostatic tolerance, it is critical to understand whether baroreflex function will change, both in terms of BRS and BEI. Thus, the results of this study could provide insight to the importance of baroreflex function versus blood volume distribution in the maintenance of orthostatic tolerance.

## Methods

### *Subjects*

A total of 52 subjects (31 men, 21 women) were pooled from three different studies. All subjects signed written informed consent approved by The Pennsylvania State University Institutional Review Board. Descriptive statistics of the subjects are presented in Table 1. Each subject was pre-screened and was excluded if they exhibited any of the following:

- smoker
- body mass index  $< 18.5$  or  $\geq 30.0$  kg/m<sup>2</sup>
- waist-to hip ratio  $> 0.97$
- currently pregnant or breast feeding
- cardiovascular or peripheral vascular disease (including hyper- or hypotension)
- hormonal contraceptive use
- clinically diagnosed orthostatic intolerance, or
- allergies to any of the drugs or anesthetics utilized at any time during the experiments.

**Subject Demographics for all Studies**

	<b>Protocol 1</b>	<b>Protocol 2</b>	<b>Protocol 3</b>	<b>Average</b>
Age (years)	24 ± 6	22 ± 4	24 ± 4	23 ± 1
Height (cm)	173.11 ± 10.82	171.3 ± 8	169.3 ± 11.9	171.24 ± 1.91
Weight (kg)	75.6 ± 14.4	71.8 ± 12.2	69.1 ± 12.5	72.2 ± 3.2
BMI (kg m <sup>-2</sup> )	25.0 ± 3.1	24.4 ± 2.4	24 ± 3.3	24.5 ± 0.5
Body Surface Area (m <sup>2</sup> )	1.89 ± 0.23	1.83 ± 0.19	1.79 ± 0.22	1.84 ± 0.05
W:H Ratio	-	0.83 ± 0.04	0.77 ± 0.03	0.80 ± 0.04

**Table 1.** Subject descriptive data ( $n_{\text{tot}} = 52$ ; Protocol 1  $n = 21$ ; Protocol 2  $n = 17$ ; Protocol 3  $n = 14$ ); BMI = Body Mass Index; W:H Ratio = Waist to Hip Measurement Ratio. All data is reported as mean ± S.D.

Before each visit, subjects were instructed to fast for 8 hours, as well as refraining from alcohol and caffeine for 24 hours and 48 hours, respectively. All subjects were admitted to the General Clinical Research Center at the Pennsylvania State University for

a physical evaluation and family history prior to the study. Additionally, all female subjects were tested only during the early follicular phase (days 2-6) of the menstrual cycle and were required to undergo a pregnancy test during each testing day.

### *Experimental Design*

Subjects were included in 1 of 3 protocols:

***Protocol 1.*** On the first visit, subjects were evaluated with a resting 12-lead electrocardiogram and blood pressure measurements. They underwent a basic physical examination, as well as a routine blood chemistry profile. The next day consisted of a 70° head-up-tilt table test 30 minutes after administration of either a placebo or of 1.7 µg/kg octreotide acetate (mixed with 50 ml saline) infused over 15 minutes. On the next test day, the subject was again tilted with the opposite substance. All pharmacological interventions were double-blinded and randomized.

***Protocol 2.*** Like protocol 1, all subjects underwent a pre-screening physical with ECG, blood pressure measurements, and blood chemistry. Instead of undergoing a continuous 70° head-up-tilt, subjects were tilted at graded intervals of 15°, 30°, and 50° for six minutes each. Similarly to protocol 1, octreotide acetate or placebo was infused before the tilt in a double-blinded fashion. On the next test day, the opposite substance was infused before another graded tilt.

***Protocol 3.*** This experiment combined tilts from procedures 1 and 2. Along with similar pre-study procedures, subjects were tilted continuously for 45 minutes at 70° on one study day and in graded tilt at similar intervals as above, on the next. No octreotide acetate was administered during this procedure.

## *Measurements*

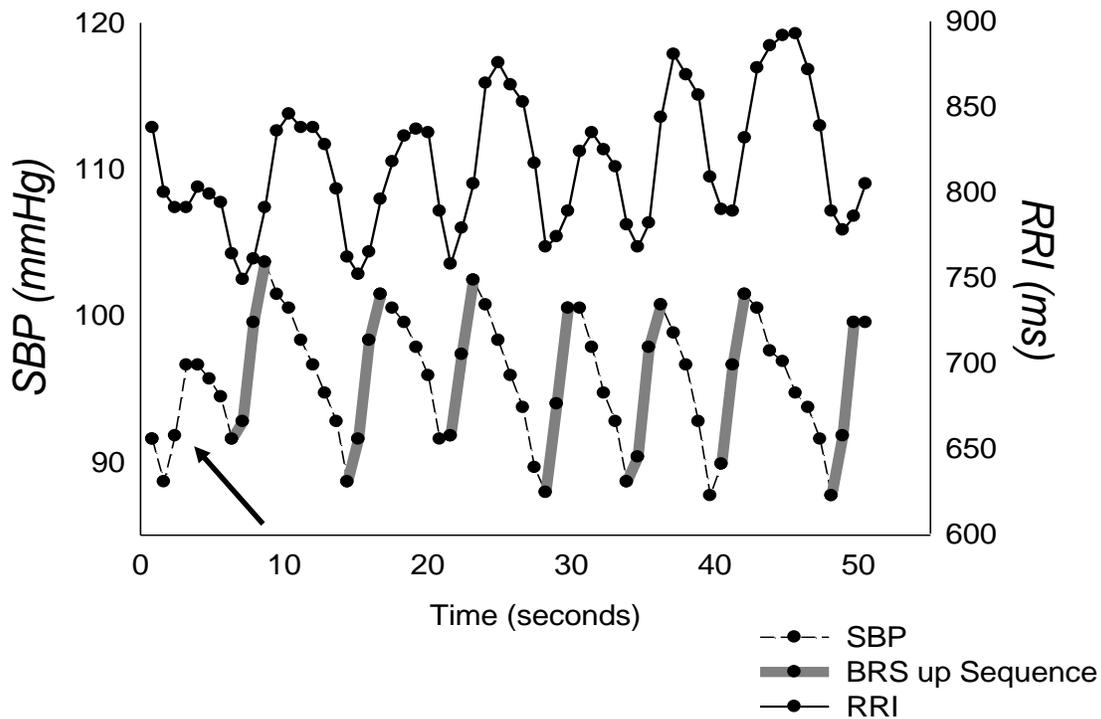
***Tilt Table Testing.*** For all tilt testing, subjects received a brief physical examination at the General Clinical Research Center prior to reporting to the lab. Upon arrival, subjects were instrumented by laboratory staff for electrocardiogram (3-lead ECG; Hewlett-Packard 78534A, Andover, MA, USA) and blood pressure (Colin 7000; Colin Medical Instruments, San Antonio, TX, USA). ECG and blood pressure were continuously sampled at 1,000 Hz and 100 Hz, respectively. Data were stored in beat-to-beat format using a customized data acquisition program. To ensure subjects were at hemodynamic equilibrium before test protocol, they were instructed to lie supine on the modified tilt table (model OT-9003, Omni Technologies, Valley City, ND, USA) for 70 minutes (instrumentation and equilibrium time). Subjects' arms were outstretched and supported at the level of the right atrium for blood pressure measurements.

**70° Head-Up-Tilt.** For all protocols, baseline measurements were taken for 10 minutes. After baseline, the subject was tilted up to 70° and was instructed to refrain from any movements to avoid skeletal muscle pump activity. Subjects remained at 70° for 45 minutes or until they voluntarily withdrew or showed signs of presyncope (decrease in blood pressure > 20/10 mmHg and/or rapid decline in heart rate > 25 beats/min). Other discomforts brought upon by the passive tilt such as nausea or light-headedness were criteria for termination. If a test was terminated due to presyncope, the subject was placed in the Trendelenburg position (-10°) until hemodynamics stabilized. Afterward, subjects were returned to supine and twenty minutes of recovery data were collected.

Graded Tilt. In protocols two and three, subjects were instrumented and prepared before the procedure in similar way as to the 70° HUT trial. After the 10 minute baseline recordings were taken, subjects were then tilted to 15°, 30°, and 50° for 6 minutes each. The test continued until all stages were complete, subject withdrawal, or evidence of presyncope. After tilt was completed, 20 minutes of recovery data were recorded.

**Baroreflex Function.** To assess the arterial BRS, systolic blood pressure (SBP) and R-R Interval (RRI) were evaluated on a beat-to-beat analysis using the sequence method (Eckberg & Kuusela, 2005; Steptoe & Vogele, 1990). Data were analyzed using the WinCPRS system (Absolute Aliens, Oy, Turku, Finland). The software algorithm scanned for a minimum 3-beat increase (or decrease) in blood pressure exceeding a criterion of at least 1 mmHg/beat. Sequences were only considered when there was a subsequent lengthening (or shortening) in RRI by at least 5 ms per beat that followed with 1 beat lag. The lag accounts for baroreflex latency and has been shown to be a better indicator of baroreflex function than a lag of 0 beats (Steptoe & Vogele, 1990). Sequences where there was a detectable increase in blood pressure but RRI stayed the same or shortened were not evaluated because these do not constitute physiological evidence of a baroreflex-mediated response (Parlow *et al.*, 1995).

Figure 1 portrays the association between SBP and RRI. In addition to the above criteria, in order to be counted as a valid baroreflex sequence, the regression between the SBP ramp and the RRI shortening/lengthening had to yield a linear correlation coefficient  $\geq 0.80$ . BRS, the slope between these two changes, was expressed in the units of ms/mmHg.



**Figure 1.** Baroreflex sequences. In order to be considered, SBP had to increase/decrease by at least 1 mmHg for 3 beats. RRI had to correspondingly increase or decrease for 5 ms per beat. Note the 1 beat lag between the SBP and the RRI value. BRS sequences are broken into up-slopes and down-slopes, and are averaged together to give a BRS value in  $\text{ms mmHg}^{-1}$ . Note the figure above only shows up-sequences for this example even though down-sequences exist. The arrow denotes a SBP ramp where blood pressure increases without the corresponding increase in RRI.

The baroreflex effectiveness index was determined using the WinCPRS software.

BEI was determined as the ratio of total number of valid BRS sequences to the total number of SBP ramps (Di Rienzo *et al.*, 2001). Expressed as a percentage, BEI measures how frequently rises or falls in blood pressure elicited a baroreflex response.

**Orthostatic Tolerance.** For protocols 1 and 3, orthostatic tolerance could be assessed.

Tolerance was calculated as the duration, in minutes, that a subject could be passively tilted at  $70^\circ$  without showing signs of presyncope. The maximum OT time was 45 minutes; the total length of the tilt.

***Data Summary and Statistical Analysis.*** Artifact-free continuous beat-to-beat data were analyzed in three minute segments during the end of each stage. An analysis of variance test (ANOVA) was performed using SAS 9.1 (Cary, NC, USA) to assess stage x condition changes. All data are presented as means  $\pm$  S.D. A p-value  $< 0.05$  was considered to be a significant change.

## Results

### *Cardiovascular Changes during Passive 70° HUT*

All cardiovascular changes are illustrated in Table 2. Baseline RRI for subjects who completed the 70° HUT under placebo conditions was  $983 \pm 108.6$  ms ( $61.8 \pm 6.9$  bpm). Systolic blood pressure was  $107.1 \pm 17.5$  mmHg, diastolic blood pressure (DBP) was  $58.5 \pm 9.1$  mmHg, and mean arterial pressure was  $77.1 \pm 9.4$  mmHg. No variables were influenced by octreotide, but RRI and SBP were significantly less during tilt than during supine at  $645.1 \pm 83.4$  ms and  $107.1 \pm 17.5$  mmHg, respectively ( $p < 0.0001$  and  $p = 0.0028$ ). Recovery data was similar to that of supine.

Cardiovascular Changes to Subjects During Head Up Tilt								
Stage	HR		SBP		DBP		MAP	
	Plac	Oct	Plac	Oct	Plac	Oct	Plac	Oct
<b>70° Upright Tilt</b>								
Supine	$61.8 \pm 6.9$	$63.2 \pm 7.1$	$116.5 \pm 15.0$	$119.5 \pm 14.0$	$58.5 \pm 9.1$	$61.7 \pm 9.7$	$77.1 \pm 9.4$	$81.4 \pm 10.0$
Tilt	$94.4 \pm 11.1^*$	$95.2 \pm 13.8^*$	$107.1 \pm 17.5^*$	$109.9 \pm 29.5^*$	$62.3 \pm 10.5$	$61.8 \pm 17.7$	$77.9 \pm 11.2$	$78.8 \pm 20.6$
Recovery	$64.5 \pm 8.6$	$67.0 \pm 6.4$	$118.6 \pm 15.3$	$122.8 \pm 15.8$	$61.0 \pm 9.1$	$62.9 \pm 9.6$	$81.2 \pm 10.9$	$84.2 \pm 9.9$
<b>Graded Tilt</b>								
Supine	$56.6 \pm 6.4$	$58.3 \pm 7.9$	$111.1 \pm 12.0$	$116.6 \pm 9.4$	$56.9 \pm 7.4$	$59.8 \pm 6.5$	$75.1 \pm 8.7$	$78.8 \pm 6.2$
15°	$58.6 \pm 7.6^*$	$59.1 \pm 7.6^*$	$108.8 \pm 11.5^*$	$111.1 \pm 10.5^*$	$57.6 \pm 6.5$	$58.1 \pm 6.4$	$75.2 \pm 7.1^*$	$76.5 \pm 6.3^*$
30°	$65.1 \pm 9.3^*$	$63.1 \pm 8.9^*$	$105.9 \pm 10.9^*$	$108.0 \pm 11.5^*$	$57.2 \pm 8.1$	$58.4 \pm 7.6$	$73.6 \pm 8.5^*$	$75.8 \pm 7.2^*$
50°	$72.4 \pm 8.6^*$	$68.2 \pm 10.9^*$	$104.5 \pm 9.8^*$	$106.8 \pm 11.0^*$	$57.4 \pm 8.3$	$57.4 \pm 6.2$	$73.9 \pm 7.8^*$	$74.9 \pm 6.1^*$
Recovery	$59.2 \pm 7.6$	$59.2 \pm 7.6$	$115.2 \pm 12.9$	$115.2 \pm 12.9$	$59.6 \pm 8.4$	$59.6 \pm 8.4$	$79.4 \pm 8.9$	$79.4 \pm 8.9$

**Table 2.** The data above is for the two tilt procedures conducted in the three protocols with control and administration of octreotide. All data is reported as mean  $\pm$  S.D. Plac = placebo, Oct = Octreotide, Heart Rate (HR) in beats  $\text{min}^{-1}$ ; Systolic Blood Pressure (SBP) in mmHg; Diastolic Blood Pressure (DBP) in mmHg; Mean Arterial Pressure (MAP) in mmHg. \* indicates significantly different from supine ( $p < 0.05$ ). Octreotide did not produce any significant differences.

*Baroreflex Changes during Passive 70° HUT*

The values for BRS and BEI can both be found in Table 3 for all angles of tilt. Average baroreflex sensitivity while supine was  $17.2 \pm 8.2$  ms/mmHg for all subjects in this protocol, and decreased to  $5.0 \pm 2.4$  ms/mmHg during for 70° HUT ( $p < 0.0001$ ). Baroreflex sensitivity upon return to supine for recovery was not different from baseline.

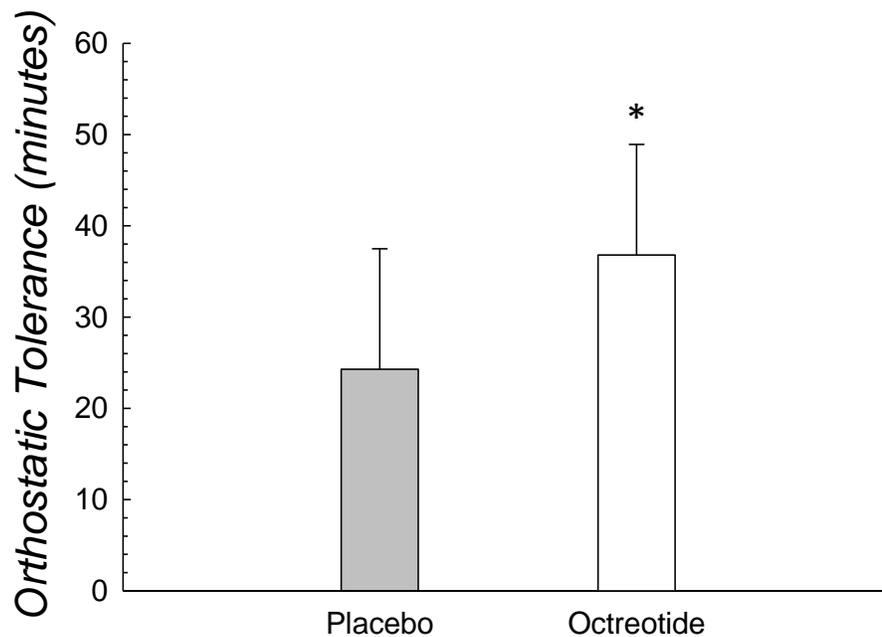
The baseline baroreflex effectiveness index for the control trial was  $34.1 \pm 21\%$ . This value increased significantly with the addition of orthostatic stress during tilt to  $47.6 \pm 16.3\%$  ( $p = 0.0018$ ). Like all the other variables, BEI showed no difference between baseline and recovery values. BRS and BEI, during octreotide trials did differ from the corresponding control (placebo) conditions.

<b>Baroreflex Changes to Subjects During Head Up Tilt</b>				
<b>Stage</b>	<b>BRS</b>		<b>BEI</b>	
	Placebo	Octreotide	Placebo	Octreotide
<b>70° Upright Tilt</b>				
Supine	$17.2 \pm 8.2$	$16.8 \pm 8.4$	$0.341 \pm 0.210$	$0.296 \pm 0.133$
Tilt	$5.0 \pm 2.4^*$	$4.5 \pm 1.8^*$	$0.476 \pm 0.163^*$	$0.418 \pm 0.209^*$
Recovery	$18.9 \pm 12.6$	$14.4 \pm 7.6$	$0.412 \pm 0.202$	$0.390 \pm 0.173$
<b>Graded Tilt</b>				
Supine	$27.0 \pm 18.7$	$25.2 \pm 16.0$	$0.264 \pm 0.179$	$0.304 \pm 0.203$
15°	$21.8 \pm 15.3^*$	$21.1 \pm 12.5^*$	$0.346 \pm 0.217^*$	$0.367 \pm 0.183^*$
30°	$14.7 \pm 11.6^*$	$16.3 \pm 10.0^*$	$0.427 \pm 0.208^*$	$0.442 \pm 0.154^*$
50°	$9.2 \pm 5.4^*$	$13.0 \pm 7.8^*$	$0.489 \pm 0.193^*$	$0.475 \pm 0.182^*$
Recovery	$22.8 \pm 14.5$	$22.8 \pm 14.5$	$0.309 \pm 0.159$	$0.309 \pm 0.159$

**Table 3.** The data above is for the two tilt procedures conducted in the three protocols with control and administration of octreotide. All data is reported as mean  $\pm$  S.D. Baroreflex Sensitivity (BRS) in  $\text{ms mmHg}^{-1}$ ; Baroreflex Effectiveness index (BEI) is the ratio of BRS sequences to SBP ramps. \* indicates significantly different from supine ( $p < 0.05$ ). Octreotide did not produce any significant differences.

### *Changes to Orthostatic Tolerance with Octreotide*

Tilt tolerance time with the administration of a placebo was averaged at  $24.28 \pm 13.19$  minutes for all subjects. The intervention with octreotide acetate extended this overall average tilt time to  $36.80 \pm 12.12$  minutes ( $p=0.001$ ). Orthostatic tolerance was the only variable that was statistically different with octreotide.



**Figure 2.** Average duration subjects could be tilted before displaying signs of presyncope or achieving the maximum tilt time (45 minutes). Octreotide significantly increased the duration in which subjects could be tilted. \* indicates a statistical difference from placebo ( $p=0.001$ ).

### *Cardiovascular Changes during Graded HUT*

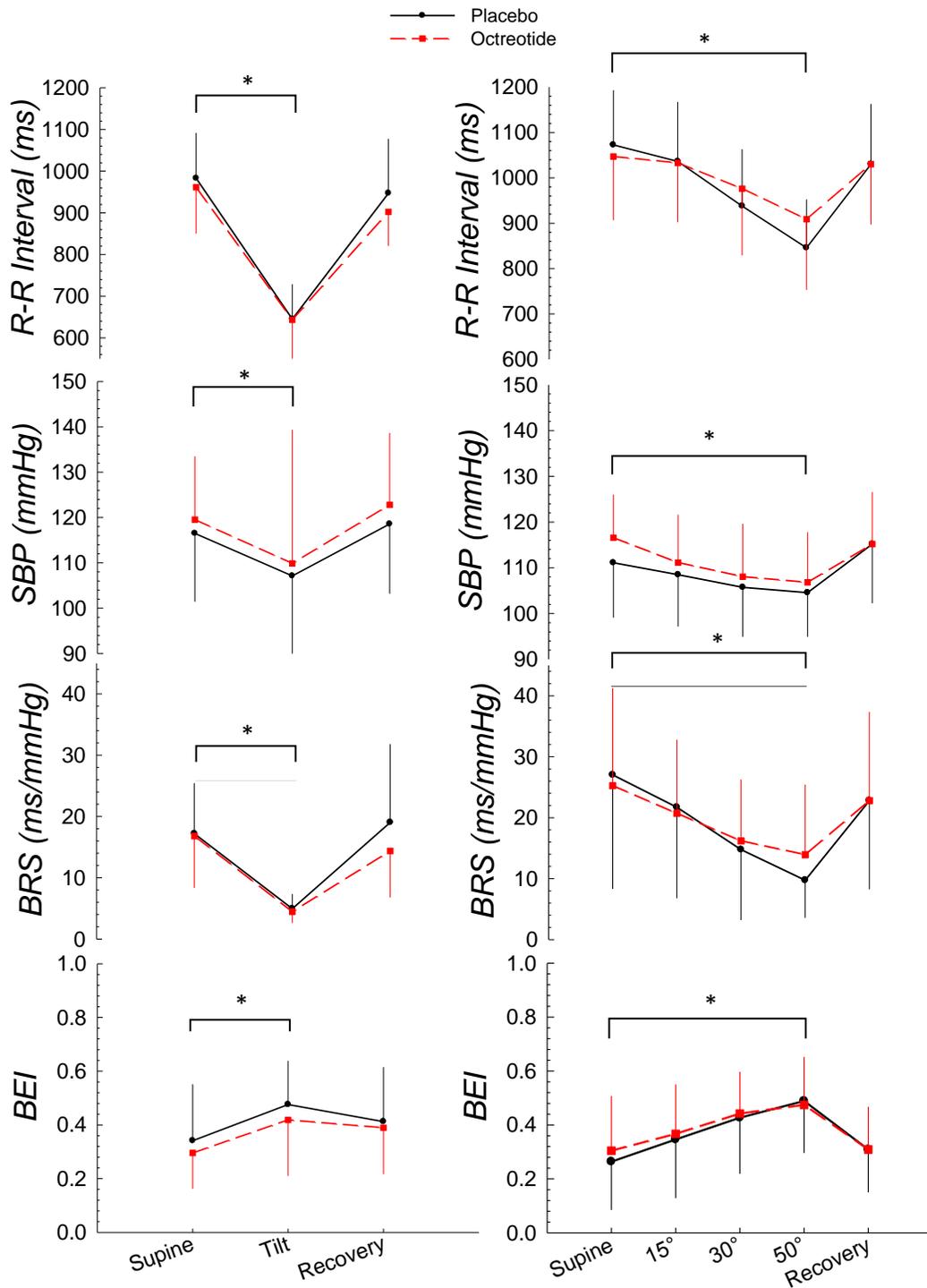
All cardiovascular variables were affected similarly during graded and  $70^\circ$  HUT procedures. The supine value for RRI with placebo was  $1072.7 \pm 120.9$  ms ( $56.6 \pm 6.4$  bpm). This steadily decreased until it reached a nadir of  $840.4 \pm 103.2$  ms ( $72.4 \pm 8.6$  bpm) at  $50^\circ$  ( $p<0.0001$ ). SBP was  $111.1 \pm 12.0$  mmHg at rest, and reached its lowest

value of  $104.5 \pm 9.8$  mmHg ( $p < 0.0001$ ) also at  $50^\circ$ . DBP was  $56.7 \pm 7.4$  mmHg but did not change during tilt. MAP decreased from  $75.1 \pm 8.7$  mmHg to  $73.9 \pm 7.8$  mmHg ( $p < 0.0001$ ). Similar to  $70^\circ$  tilt, octreotide had no effect on any variable.

#### *Baroreflex Changes during Graded HUT*

Baroreflex sensitivity was highest in the supine position with a value of  $27.0 \pm 18.7$  ms/mmHg, decreasing to  $9.2 \pm 5.4$  ms/mmHg at  $50^\circ$  tilt ( $p < 0.0001$ ). There was no difference between baroreflex sensitivity during baseline (supine) and recovery positions.

During graded tilt, BEI increased with escalating orthostatic stress. From supine values of  $26.4 \pm 17.9\%$ , it increased to a maximum value of  $48.9 \pm 19.3$  ( $p < 0.0001$ ) at  $50^\circ$  tilt. There was no statistical difference between recovery and baseline values. Octreotide had no affect on BRS and BEI during graded tilt.



**Figure 3.** Above left: this panel shows cardiovascular and baroreflex changes during 70° HUT. RRI, SBP, and BRS all decreased during tilt, but did not statistically change in recovery. BEI increased during tilt, but also had no difference in recovery. Above right: this panel shows similar variables during graded tilt. RRI, SBP, and BRS continued to decrease during each stage. Similar to 70° tilt, BEI increased during each stage of graded HUT. All four variables also showed no change in recovery compared to baseline. \* indicates a statistical difference from baseline ( $p < 0.05$ ). Octreotide yielded no change for any variable.

## **Discussion**

This study was conducted to observe the effects of octreotide acetate on baroreflex function during orthostatic stress. Octreotide is extremely effective in decreasing a person's susceptibility to orthostatic stresses. This is consistent with the literature and appears to be a reliable and repeatable effect of octreotide (Wong & Sheriff, 2007). The movement of blood out of the splanchnic circulation via selective vasoconstriction caused by this drug seems to be adequate in aiding in the defense of cranial perfusion.

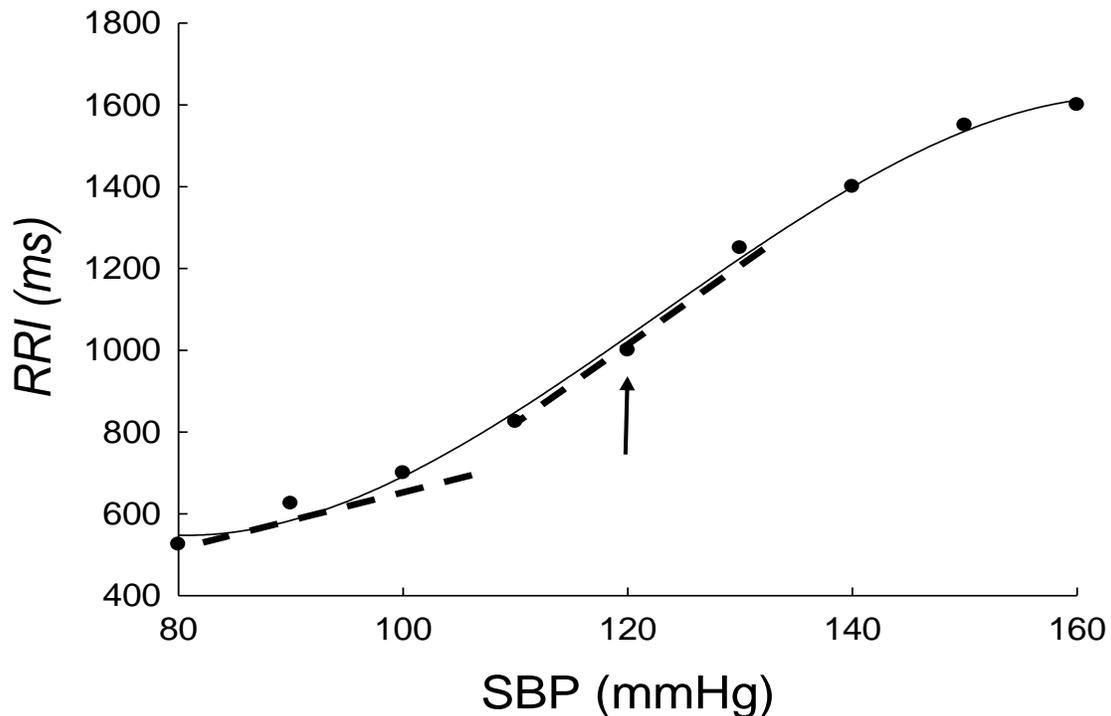
Because baroreceptors are very important in regulating mean arterial pressure we hypothesized baroreflex function would change with the administration of octreotide. However, we failed to identify any difference.

Baroreflex sensitivity decreased with added orthostatic stress; this was consistent regardless of intervention with octreotide. Conversely, the percent of activation, or the baroreflex effectiveness index, rose under both conditions with tilt.

The decrease in BRS is primarily caused by the shortening of RRI and the decrease in SBP during tilt. The increase in BEI suggests that as orthostatic stress increases, the baroreflex is activated more often in order to defend the changes in blood pressure brought on by blood volume displacement.

In regards to the change in baroreflex function, we believe that the lowering of the slope is indicative of its movement on the stimulus-response curve, and not resetting of the operating point. The hypothesis is that the BRS slope moves closer to the less sensitive threshold (low SBP, low RRI) from where it normally lies at the operating point

within the steeper (i.e. the more sensitive) relationship. In order to test this hypothesis, further studies would need to be conducted. Options for the designs of these studies would involve constructing stimulus-response curves using vasoactive drugs or neck chamber suction/pressure. These techniques allow for more extreme SBP changes (or analogous transmural pressure changes in the neck chamber experiment), and can properly differentiate between the operating (as evaluated in our study), threshold, and saturation regions. Creating stimulus-response curves during rest and tilt will allow one to determine if the BRS response represents same function with the operating point reset to a lower value (as outlined in Figure 4.), or to an entirely different stimulus-response curve (i.e. baroreflex “resetting”)



**Figure 4.** A conceptual stimulus-response curve of the baroreflex. The dashed line indicates the baroreflex sensitivity at a given blood pressure, and is the slope of the tangent line. This would be representative of the baroreflex moving to the less sensitive region of the curve. The arrow denotes the operating point. If the baroreflex were to reset during tilt, the curve would shift to the left, but the slope of the tangent line would not change.

Even though there is an augmentation in the baroreflex engagement during changes to posture, octreotide does little to change baroreflex function. This infers that the increase in orthostatic tolerance is brought on by another, discrete mechanism.

El-Sayed and Hainsworth demonstrated that increasing plasma volume via salt supplementation is the cause for increased orthostatic tolerance in patients with unexplained syncope (El-Sayed & Hainsworth, 1996). It is conceivable that octreotide mimics the increase in available plasma volume by increasing central blood volume. However, there is an inconsistency between the El-Sayed & Hainsworth data and ours in reference to BRS. With an increase in blood volume, they found that resting BRS actually decreased; whereas in our study, the value for BRS did not change (El-Sayed & Hainsworth, 1996).

### *Limitations*

We advise caution because baroreflex sensitivity is measured differently in many labs. The EuroBaVar study has shown that many different laboratories have different criteria to define baroreflex sensitivity (Laude *et al.*, 2004). This can make comparisons of BRS values found by different groups very difficult to interpret.

## **Conclusion**

Overall, we conclude from this study that octreotide acetate significantly increases orthostatic tolerance, but has little or no effect to baroreflex function. During tilt, sensitivity decreases with a reduction in both RRI and SBP. Conversely, BEI increases due to increased number of SBP ramps that elicit a response by the baroreflex. More research needs to be directed at the mechanism that causes this decrease in sensitivity. Comparison of stimulus-response curves at rest and during tilt can give insight into whether the baroreflex function actually changes during orthostatic stress, or merely changes location upon the same function's curve.

Our results imply that prudence needs to be used when using baseline baroreflex values to predict orthostatic tolerance. Our analysis shows no effect on either BRS or BEI, even though octreotide has a noteworthy augmentation to OT. That is, by pharmacologically increasing a person's tolerance to tilting, baroreflex function (in terms of sensitivity and percent activation) did not change. This implies that there may not be a primary relationship between the variables, and that baroreflex sensitivity should not be a sole predictor of a person's orthostatic tolerance.

## **Acknowledgements**

We would like to thank the subjects who volunteered for these studies for their cooperation. We express appreciation to the staff of the General Clinical Research Center for their aid in patient screening and preparation. We thank the graduate students and research assistants, particularly Sara Jarvis and Kyle Preston, who devoted their time and effort into making this project possible. We would also like to extend our gratitude to Tom Kuusela and Absolute Aliens for working with us to implement BEI analysis. Finally, we would like to thank NASA, the NIH, the Noll Endowment, and Phi Beta Kappa for their financial support.

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**Appendix**

A- Protocol 1 Informed Consent

**INFORMED CONSENT FORM FOR CLINICAL  
RESEARCH STUDY**

**The Pennsylvania State University**

**Protocol 2**

ORP USE ONLY: IRB#02M0187 Doc.#2

The Pennsylvania State University

Office for Research Protections

**Title of Project:** Improving Orthostatic Tolerance in Women: Control of Splanchnic and Cutaneous Vascular Capacitance

**Principal Investigator:** James A. Pawelczyk, Ph.D.

**Other Investigators:** W. Larry Kenney, Ph.D.

Urs Leuenberger, M.D.

Nancy Williams, Sc.D.

**Graduate Students:** John Florian

Sara Jarvis

**Research Assistant:** Sandra Smithmyer

This is to certify that I, \_\_\_\_\_, have been given the following information with respect to my participation as a volunteer in a program of investigation under the supervision of Dr. James A. Pawelczyk.

**1. Purpose of the study:**

The purpose of this investigation is to test your ability to control blood pressure with and without an injection of a drug called octreotide, which causes blood flow in your splanchnic (gut) region to decrease and may improve your body's ability to control its blood pressure. These measurements will be done by tilting subjects at different angles on a tilt table. A total of 16 male and 16 female volunteers will be included in this investigation. To complete this study you will be asked to visit the Noll Physiological Research Center and/or the General Clinical Research Center (GCRC), on five separate occasions.

**2. Procedures to be followed:**

General information: A total of 5 days are required to complete this study. These include:

- Pre-study Screening Days:
  - Blood draw and lab tour (45 minutes)
  - Physical and exercise test (2 hours)
- Preparation Day: Splanchnic (gut) extraction before and after octreotide injection (2 hours)
- Study Day 1: Tilt test with octreotide injection (4 hours)
- Study Day 2: Tilt test without octreotide injection (4 hours)

Pre-study Screening Day: During your first visit we will give you a tour of Noll laboratory. You will complete a detailed medical history and a GCRC clinician will draw a small amount of blood from your arm (less than a tablespoon or 15 milliliters) with a needle in order to perform routine blood checks for anemia, kidney function, and liver function. On a second day, about 3 days later, you will receive a physical examination by a designated GCRC clinician. We will record the electrical activity of your heart (electrocardiogram). In addition, you will undergo a graded exercise test (GXT). This test is detailed on the separate GXT informed consent sheet.

Menstrual History (female participants): You will complete a menstrual history questionnaire to document your gynecological age, prior history of menstrual cycle lengths, and menstrual cycle characteristics. The study days will take place on the day of your menstrual cycle when estrogen levels are lowest (approximately 3-5 days after your period begins).

Pregnancy Test (female participants): An over the counter urine pregnancy test will be performed before each experimental day. If the test result is positive you will be excused from participating in the study.

Preparation Day- Splanchnic (gut) removal of dye: The ability of your liver to remove the dye used later in the study will be measured before and after an injection of octreotide. Following the application of a numbing cream, a catheter (small tube) will be inserted

into each hand or forearm. After 30 minutes of rest, this dye, indocyanine green (ICG), will be injected into one of the tubes inserted into a vein. Small samples of your blood will be taken from the second catheter every three minutes for one-half hour. The total amount of blood removed will be about 2 tablespoons (30 mls). This will be repeated after you are given an injection of octreotide. Right before and then following the octreotide injection, we will take small amounts of blood (about 1 teaspoon total) to measure your blood sugar levels. This is done to make sure that your blood sugar doesn't become too low following the injection of octreotide.

ICG is a safe non-toxic substance that is easily cleared by the liver and can be safely injected with little risk when subjects are properly screened. **People who are sensitive or allergic to penicillin, iodides, shellfish, or sulfa drugs should not participate because they have a greater chance of being allergic to the dye.** Therefore, you will be asked if you have had any reactions to these types of medicines in the past, and if you have, you will not be allowed to participate in the project. This is a necessary requirement to reduce the slight possibility of an allergic reaction to ICG.

Study Day 1 and 2: Following the application of a numbing cream, a catheter (small tube) will be inserted into each hand or forearm. You will then be placed on a tilt table and again given an injection of either octreotide or saline. After twenty minutes, you will be slowly raised to a tilt angle of 70° (almost standing). While you are lying flat and during the tilting, we will measure:

- heart rate from sticky patches placed on your chest,
- blood pressure from your arm and a sensor on your wrist,
- blood flow from cuffs attached to your wrist and upper arm,
- your heart's pumping ability by breathing in and out of a bag containing a small amount of a gas called acetylene,
- blood sugar levels by drawing small amounts (about 1 teaspoon total) of blood from your arm,
- Fluid volume changes in your abdomen,
- splanchnic (gut) blood flow by similar infusion of ICG described above.

You will remain tilted for no more than 45 minutes. During that time you should stay as relaxed and motionless as possible. If your blood pressure begins to fall or you begin to feel like you may pass out, we will return you to a flat position and you will feel better within minutes. After the tilt test, you will be lowered to a flat position.

After you are finished with day 1 of the study, you will be asked to return to the lab one more time to repeat the tilt test during the same phase of your menstrual cycle if you are a female. If you received an injection of octreotide before the first tilt test, you will receive an injection of saline before the second tilt test and vice versa.

On rare occurrences we may need to infuse more ICG during the study. This would add an additional 30-40 minutes to the length of the study.

If we discover that you are pregnant at any time during your involvement in the study, you will be notified immediately and eliminated from the study.

Researchers of either sex will be available to administer tests if you so desire.

A more detailed description of these days, and the procedures to be conducted, is provided below. All procedures will take place at the Noll Physiological Research Center or the GCRC. For two days before each day of the study you should be sure to drink normal amounts of fluid (8 glasses water, juice, or sports drinks per day). You should avoid using any type of stimulant (including cold medications and chocolate), drinking caffeine (coffee, tea, cola), and drinking alcohol after 9 PM the night before any procedure.

**Detailed description of the procedures to be used:**

Please read the detailed description of each procedure below and initial each one to indicate that you have read and understand them.

\_\_\_ Heart Rate. Electrodes (sticky patches) will be applied to the skin of your chest to measure your heart's electrical activity. Other than skin irritation from the sticky electrodes, there is no known risk involved with this procedure.

\_\_\_ Blood Pressure. A wrist brace and sensor will be placed over your wrist and a cuff will be placed on your upper arm. The sensor will push down against your skin and may leave a mark when it is removed. The mark will go away within a few minutes after the sensor is removed. There is no known risk involved with this procedure.

\_\_\_ Blood Flow. Periodically we will measure blood flow to your forearm. Velcro blood pressure cuffs will be wrapped around your upper arm and around your wrist, and thin rubber tubes filled with mercury will be wrapped around the middle of your arm. The wrist cuff will be inflated to a pressure high enough to stop blood flow to your hand. This causes no problems for the 2-5 minute period the cuff will be inflated. While this cuff is inflated, the other cuff will be inflated to a much lower pressure for 10-15 seconds, about 3 times a minute. While this cuff inflates you may feel that your arm is swelling. This sensation will cease when the cuffs are released.

\_\_\_ Abdominal fluid measurement (bioimpedance). We will place sticky tape around your chest, waist, legs, and lower neck. A cable will be connected to the tape around your legs and around your neck. A very small current of electricity will

flow through the tape and through your body. You will not be able to detect the current as it passes through your body.

\_\_\_\_\_ Cardiac Output (acetylene rebreathing). We will measure your heart's pumping rate by analyzing the air you rebreath in and out of a bag using fairly deep breaths for 15-20 seconds. The bag will contain a small concentration of two gases, acetylene and helium. The concentrations of these gasses are so small that there is no risk of them catching fire. Some people report a slightly "tangy" taste from the rebreathing gas. Although you may become light-headed for a few seconds during rebreathing or develop a slight headache from repeated rebreathing, there are no other known risks to performing this procedure. The gas disappears from your lungs and blood in less than 5 minutes.

\_\_\_\_\_ Splanchnic (gut) blood flow. The greenish dye called ICG will be pumped into a tube inserted into a vein in your forearm throughout the test. A little more than 1 tablespoon (15 mls) of ICG will be injected just prior to starting the pump. Small samples of your blood will then be taken from a second catheter inserted into a vein near your hand every 5 minutes during tilting. ICG is a safe non-toxic substance that is easily cleared by the liver and can be safely injected with little risk in subjects who show no allergic reactions. An allergic reaction to ICG, though very rare, could be life-threatening.

\_\_\_\_\_ Head-up Tilt. You will lie on a table for 20 minutes and then you will be tilted at an angle of 70° on a tilt table for up to 45 minutes. During that time we ask that you relax and stay as motionless as possible. We will stop tilting if your blood pressure becomes too low, you experience an abnormal heart beat, or you feel light-headed or dizzy.

**3. Discomforts and risks:**

**All procedures carry risk. Risk has two aspects: severity and frequency. Severe risk might threaten the loss of life or limb, while a mild risk might be discomfort. The frequency of a risk is the chance that a problem will occur. In this section we have summarized the risks associated with the procedures used in this experiment. The risks in this experiment have different severity and frequency, and some could be life threatening. Please feel free to ask about the severity or frequency of these risks at any time. To help you decide whether or not you are willing to accept the risks associated with this experiment, the table below provides some commonly mentioned risks and the estimated chance they will occur to you:**

Contracting meningitis while living in a dorm	1 in 20,000
Being struck by lightning in your lifetime	1 in 10,000

Contracting AIDS if you avoid "high risk" activity	1 in 3,000
Dying of liver disease if you drank one beer per day	1 in 1,000
Developing breast cancer by age 25	1 in 1,000
Contracting a disease caused by radon in your home	1 in 440
Being killed in a car accident in your lifetime	1 in 60
Contracting cancer at some point during your life	
	Men 1 in 2
	Women 1 in 3

General: If during the screening procedures we should detect any abnormality, we will inform you as soon as possible so that you can contact your personal doctor for treatment. Some people find medical procedures to be scary, and feel faint as a result (vasovagal response). To keep you comfortable and informed, we encourage you to share your concerns with us at any time.

Heart rate, blood flow, and blood pressure: The only known risk associated with measuring heart rate, blood flow, and blood pressure is the possibility that your skin will be irritated by the adhesive, cuffs, or tape. The blood pressure sensor will push down against your skin and may leave a mark when it is removed. The mark will go away within a few minutes after the sensor is removed.

Abdominal fluid measurement (bioimpedance): There are no risks involved with this procedure. The sticky tape may cause your skin to become irritated when we remove the tape.

Rebreathing cardiac output: Because you breathe slightly deeper than normal, there is a chance you will feel light headed for a few seconds during rebreathing or develop a slight headache after repeated measurements. There are no other known risks to this procedure.

Indocyanine Green Dye (ICG): ICG is a safe non-toxic substance that is easily cleared by the liver and can be safely injected with little risk when subjects are properly screened. **The only risk involved is when a person is sensitive or allergic to penicillin, iodides, or sulfa drugs.** Therefore, you will be asked if you have had any reactions to these types of medicines in the past, and if you have, you will not be allowed to participate in the project. This is a necessary requirement to insure that the slight possibility of an adverse response to ICG in a subject sensitive to the drugs listed above is alleviated. For those people that deny an allergy, there is a 1 in 250 risk of a mild to moderate reaction. Symptoms include lightheadedness, nausea, hives and itching. Rarely, (1 in 2000) a serious allergic reaction which affects the entire body (anaphylaxis) may develop. An anaphylactic reaction can be life-threatening. You are free to discontinue these tests at any time.

Octreotide: Octreotide is very similar to a naturally occurring hormone called somatostatin. It is generally well tolerated, though about 1 in 10 people experience diarrhea or an upset stomach. Because this drug makes blood vessels narrow (constrict), your blood pressure will probably rise and your heart rate slow down during the injection. This might be accompanied by a flushed face or a feeling of heaviness in your chest. This is a natural result of a slow heart rate. Less often people become light-headed or tired (1 in 20). If your heart rate or blood pressure changes too much, or at your request, we will end the octreotide infusion. Any unusual sensations should last only a minute or two after the infusion ends.

Topical Anesthetic Cream: Numbing cream will not be used in those who have sensitivity to lidocaine. Eye contact should be avoided. When used, all sensations within the treated area are blocked. For this reason, unintentional trauma to the treated area, such as scratching, rubbing or exposure to hot or cold temperatures should be avoided until complete sensation has returned. During or immediately after application, mild swelling, skin redness or abnormal sensation may develop at the site of treatment. In clinical studies, no serious reactions resulted from the use of the cream. Allergic reactions can occur and can be managed by usual allergic means. Whole body adverse reactions following appropriate use are unlikely due to the small dose absorbed. If effects do occur, they are similar in nature to those seen with other local anesthetic agents and may include lightheadedness, nervousness, apprehension, dizziness, drowsiness, twitching, and vomiting. Reactions may be brief or not at all.

Blood sampling/venous catheter: The risks of a blood sample include bruising and/or discomfort from the needle, venous inflammation from the catheter, infection (less than 1 in 10,000), or the chance that you will become lightheaded. Should you feel this way we will stop the experiment, and you will be given fluids to drink (water or juice). We ask that you remain in the laboratory until we have checked your blood pressure and we are sure that you feel OK.

Head-up tilt: The risk of head-up tilt is that you become lightheaded, nauseous, or pass out. Should you feel this way or we suspect that see that your blood pressure is starting to fall, we will stop the procedure and you should begin to feel better within minutes. In one case (less than 1 in 10,000) a person developed an unusually slow heart rhythm that returned to normal with a drug (atropine) injected in their arm vein.

Treadmill: It is possible for you to stumble or fall on a treadmill leading to cuts, scrapes, dislocations, broken bones, head injury, abnormal cardiac rhythms, or even death. However, this risk is minimal. The risk of heart attack, although minor (1 in 15,000) does exist. You will be taught the safe use of the treadmill and watched closely during exercise. All changes in speed will be made gradually, and you will be assisted in mounting and dismounting.

4. a. **Benefits to the subject:**

None

b. **Potential benefits to society:**

This information will be used to help people or patients who have difficulty maintaining their blood pressure. An example is patients who have recently undergone dialysis, some elderly people after eating a meal, and some otherwise healthy people who experience problems with lightheadedness. These problems are much more common in women.

5. **Alternative procedures which could be utilized:**

Recording heart rate, blood pressure, and blood flow is routine. Measuring your splanchnic (gut) blood flow can be estimated using sound waves (ultrasound), but this method is not as accurate as the green dye.

6. **Time duration of the procedures and study:**

This study will require two visits of two hours and two visits of four hours duration to the Noll Laboratory and the General Clinical Research Center (located next to Noll Laboratory).

7. **Statement of confidentiality:**

You have the right to privacy. All information that is obtained in connection with this study will remain confidential within the limits of State Law. Information gained from this study will be released only to the investigators, and if appropriate, to your physician and the sponsors of the study with your approval. Any information provided to the sponsors of the study will not include your identity. The following may review and copy records related to this research; The

Office of Human Research Protections in the U.S. Dept. of Health and Human Services; The U.S. Food and Drug Administration (FDA) when applicable; the Penn State University Biomedical Institutional Review Board (IRB); The Penn State University Office for Research Protections. The results of this study may be published in scientific journals without identifying you by name.

**8. Right to ask questions:**

If you have any questions about the research or about your rights as a subject, we want you to ask us. If you have questions later, or if you wish to report a research-related injury, please contact Dr. Pawelczyk at (814) 865-3453 (W) or (814) 861-1379 (H). Questions regarding this statement or your rights as a subject of this research should be directed to the Office for Research Protections in 201 Kern Graduate Building, University Park, PA (814-865-1775). Please initial the statement below to indicate your understanding of this right.

\_\_\_ I have been given an opportunity to ask any questions I may have, and all such questions or inquiries have been answered to my satisfaction.

**9. Compensation:**

There will be no charge for any tests required for the study. You will receive \$200 for participation in this investigation to compensate your travel and loss of time. If you choose to withdraw early from the investigation, this amount will be prorated accordingly. If you are an employee of Penn State University, the compensation you receive for participation will be treated as taxable income and therefore taxes will be taken from the total amount. If you are not employed by Penn State University, total payments within one calendar year that exceed \$600 will require the University to annually report these payments to the IRS. This may require you to claim the compensation that you receive for participation in this study as taxable income.

**10. Voluntary participation:**

Participation in this research study is entirely voluntary. Refusal to participate will involve no penalty or loss of benefits to which you are otherwise entitled. If you decide to participate, you are free to withdraw your consent and discontinue participation at any time without affecting your status (as a patient, student, employee, etc.), or the medical care that you will receive. Also, you may decline to answer any specific questions asked that you might be asked. Under certain circumstances the study may be discontinued by the sponsor or the investigator.

**11. Event of Injury:**

Medical care is available in the event of injury resulting from research but that neither financial compensation nor free medical treatment is provided. You are not waiving any

rights that you may have against the University for injury resulting from negligence of the University or the investigators.

This is to certify that I consent to and give permission for my participation as a volunteer in this program of investigation. I understand that I will receive a signed copy of this consent form. I have read this form, and understand its contents.

---

Volunteer

Date

I, the undersigned, have defined and explained the studies involved to the above volunteer.

---

Investigator

Date

B- Protocol 2 Informed Consent

**INFORMED CONSENT FORM FOR CLINICAL  
RESEARCH STUDY**

**The Pennsylvania State University**

**Protocol 1**

ORP USE ONLY: IRB# 24393 Doc. #1

The Pennsylvania State University

Office for Research Protections

**Title of Project:** Determination of the Human Volume Indifferent Point

**Principal Investigator:** Sara S. Jarvis

227 Noll Laboratory

(814) 865-0476

ssj120@psu.edu

**Other Investigators:** James A. Pawelczyk, PhD

107 Noll Laboratory

(814) 865-3453

jap18@psu.edu

**Graduate Students:** John P. Florian

Ellen E. Spiller

**Research Assistants:**

This is to certify that I, \_\_\_\_\_, have been given the following information with respect to my participation as a volunteer in a program of investigation under the supervision of Dr. James A. Pawelczyk.

**1. Purpose of the study:**

The purpose of this investigation is to locate your "volume indifferent point" or VIP, the point where volume in your veins does not change when you stand up. We suspect this point is closer to the feet in women than men, which causes women to pass out more often than men when they stand up. To measure the VIP we will place electrodes (sticky patches on your skin) then tilt you at different angles on a tilt table. We will repeat this measurement after we give you a drug called octreotide. Octreotide causes blood flow in your splanchnic (gut) region to decrease and should move your VIP closer to your head. A total of 32 volunteers will be included in this investigation (16 men and 16 women). To complete this study you will be asked to visit Noll Laboratory and/or the General Clinical Research Center (GCRC), on four separate occasions.

**2. Procedures to be followed:**

General information: A total of 4 days are required to complete this study. For female participants, study days 1 and 2 need to be scheduled around your menstrual cycle. Depending on your availability, this study could take up to three months to complete. These days include:

- Day 1 (Screening Day 1): Blood draw and electrocardiogram (1 tablespoon) (30 minutes)
- Day 2 (Screening Day 2): Physical exam and exercise test (2 hours)
- Day 3 (Study Day 1): VIP determination (3.5 hours)
- Day 4 (Study Day 2): VIP determination (3.5 hours)
- Either on Day 3 or Day 4: VIP determination following octreotide infusion (additional 1.5 hours)

Women of childbearing-age will submit a urine sample at the beginning of each visit for a pregnancy test. If you are pregnant, you will not be able to participate in the project.

A more detailed description of these days, and the procedures to be conducted, is provided below. For two days before each day of the study you should be sure to drink normal amounts of fluid (up to 8 glasses water, juice, or sports drinks per day). You should avoid using any type of stimulant (including cold medications and chocolate), drinking caffeine (coffee, tea, cola), and drinking alcohol after 9 PM the night before any procedure. You will not be allowed to eat or drink anything, except for water, after 9 PM the night before. You should not participate in strenuous physical activity for 12 hrs before the experiment.

Researchers of either sex will be available to administer tests if you so desire.

Screening: During your first visit we will give you a tour of Noll laboratory. You will complete a detailed medical history and a GCRC clinician will draw a small amount of blood from your arm (less than a tablespoon or 15 milliliters) with a needle in order to perform routine blood checks for anemia, kidney function, and liver function. We will record the electrical activity of your heart (electrocardiogram). On a second day, about 3 days later, you will receive a physical examination by a designated GCRC clinician. In addition, you will undergo a graded exercise test (GXT).

VIP determination: On the day of the study, you will be checked into the GCRC. A nurse will insert one catheter (a thin plastic tube) into a vein in your arm to draw blood during the study. If you choose, a numbing cream will be applied to your arm 15 minutes before the catheter is inserted to lessen discomfort. After the catheter is inserted, you will be taken to the lab for testing. During the experiment we will measure:

- heart rate from sticky patches placed on your chest,
- blood pressure from your arm and a sensor on your wrist,
- blood flow in your forearm using blood pressure cuffs,
- your heart's pumping ability by breathing in and out of a bag containing a small amount of a gas called acetylene,
- hormone levels, blood sugar levels and hematocrit by drawing small amounts (about 3 tablespoons total) of blood from your arm,
- fluid shifts in your abdomen and legs using sticky patches that will be placed along your chest, abdomen, legs, and on your hand and foot.

These signals will be recorded as you lie flat on your back and while you are tilted for 12 minutes each at 15°, 30°, and 50° on a motorized tilt table. For comparison, lying on your back is 0 degrees, and standing up is 90 degrees. Once all measurements are taken at the different tilt angles, you will again lie flat on your back. We will repeat these measurements after giving you an injection of octreotide on one of the visits. You will return on another day for a repeat of the VIP determination.

**Detailed description of the procedures to be used:**

Please read the detailed description of each procedure below and initial each one to indicate that you have read and understand them.

\_\_\_\_\_ Heart Rate. Electrodes (sticky patches) will be applied to the skin of your chest to measure your heart's electrical activity. Other than skin irritation from the sticky electrodes, there is no known risk involved with this procedure.

\_\_\_\_\_ Blood Pressure. A wrist brace and sensor will be placed over your wrist and a cuff will be placed on your upper arm. The sensor will push down against your skin and may leave a mark when it is removed. The mark will go away within a few minutes after the sensor is removed. There is no known risk involved with this procedure.

\_\_\_\_\_ Venous Catheter (antecubital). When you arrive to the GCRC a numbing lotion will be applied to your forearm. Once the anesthetic has taken effect the nurse will insert a soft plastic tube (catheter) into your arm vein. The catheter will remain in place during the experiment so that blood may be drawn without having to insert a needle each time. The catheter will be removed by a nurse before you are discharged from the GCRC.

\_\_\_\_\_ Blood draw. Skilled GCRC staff will remove blood from the catheter in your arm. The staff uses standard safety measures and sterile techniques that are used in hospitals. A small amount of blood (less than 1/2 teaspoon each time) will be withdrawn 9 times during the VIP only determination study day and 22 times during the octreotide study day. This amount is much less than when you donate blood.

\_\_\_\_\_ Blood Flow. Periodically we will measure blood flow to your forearm. Velcro blood pressure cuffs will be wrapped around your upper arm and around your wrist, and thin rubber tubes filled with mercury will be wrapped around the middle of your arm. The wrist cuff will be inflated to a pressure high enough to stop blood flow to your hand. This causes no problems for the 2-5 minute period the cuff will be inflated. While this cuff is inflated, the other cuff will be inflated to a much lower pressure for 10-15 seconds, about 3 times a minute. While this cuff inflates you may feel that your arm is swelling. This sensation will cease when the cuffs are released.

\_\_\_\_\_ Abdominal and leg fluid measurement (bioelectrical impedance). We will place electrodes (sticky patches) on your chest, abdomen, legs, hand and foot. A cable will be connected to each patch. A very small current of electricity will flow through the electrode and through your body. You will not be able to detect the current as it passes through your body. For comparison, this measurement is similar to body fat scales available in gyms or for home use.

\_\_\_\_\_ Cardiac Output (acetylene rebreathing). We will measure your heart's pumping rate by analyzing the air you rebreathe in and out of a bag using fairly deep breaths for 15-20

seconds. The bag will contain a small concentration of two gases, acetylene and helium. The concentrations of these gasses are so small that there is no risk of them catching fire. Some people report a slightly "tangy" taste from the rebreathing gas. Although you may become light-headed for a few seconds during rebreathing or develop a slight headache from repeated rebreathing, there are no other known risks to performing this procedure. The gas disappears from your lungs and blood in less than 5 minutes.

\_\_\_\_ Octreotide. On either study day 1 or 2, 50 milliliters (a little more than 3 Tablespoons) of an octreotide/saline mix will be slowly infused through a tube (catheter) inserted into a vein in your forearm. Octreotide is generally well tolerated.

\_\_\_\_ Head-up Tilt. You will lie on a table and then you will be tilted at an angle of 15°, 30°, and 50° on a tilt table for 12 min at each angle. During that time we ask that you relax and stay as motionless as possible. We will stop tilting if your blood pressure becomes too low, you experience an abnormal heart beat, or you feel light-headed or dizzy.

\_\_\_\_ Graded Exercise Test (GXT). The GXT tests your fitness level and cardiovascular system. Your blood pressure and heart rate will be measured. During the test, you will wear a nose clip and breathe into a tube to measure the oxygen and carbon dioxide you breathe out. You will help the researcher adjust the harness that holds the tube so that you are comfortable. During the test, you will rate how hard you are working by using a numbered scale matched to short phrases (rating of perceived exertion or RPE scale). At first, you will warm up by walking at a comfortable pace on the treadmill for about 4 minutes. Then you will begin to run at a comfortable pace. The grade of the treadmill will increase a little every 2 minutes. The exercise will become harder. The test will be most accurate if you do your best to exercise for as long as you can. However, you can stop at any time. The test is 10-20 minutes long.

### 3. Discomforts and risks:

**All procedures carry risk. Risk has two aspects: severity and frequency. Severe risk might threaten the loss of life or limb, while a mild risk might be discomfort. The frequency of a risk is the chance that a problem will occur. In this section we have summarized the risks associated with the procedures used in this experiment. The risks in this experiment have different severity and frequency, and some could be life threatening. Please feel free to ask about the severity or frequency of these risks at any time. To help you decide whether or not you are willing to accept the risks associated with this experiment, the table below provides some commonly mentioned risks and the estimated chance they will occur to you:**

Contracting meningitis while you are at a large university	1 in 20,000
Being struck by lightning in your lifetime	1 in 10,000
Contracting AIDS if you avoid "high risk" activity	1 in 3,000
Dying of liver disease if you drank one beer per day	1 in 1,000
Developing breast cancer by age 25	1 in 1,000
Contracting a disease caused by radon in your home	1 in 440
Being killed in a car accident in your lifetime	1 in 60
Contracting cancer at some point during your life	
Men	1 in 2
Women	1 in 3

Please read the detailed description of each procedure below and initial each one to indicate that you have read and understand them.

\_\_\_ General: If during the screening procedures we should detect any abnormality, we will inform you as soon as possible so that you can contact your personal doctor for treatment. Some people find medical procedures to be scary, and feel faint as a result (vasovagal response). To keep you comfortable and informed, we encourage you to share your concerns with us at any time.

\_\_\_ Heart rate, blood pressure, and blood flow: The only known risk associated with measuring heart rate and blood pressure is the possibility that your skin will be irritated by the adhesive, cuffs, or tape. The blood pressure sensor will push down against your skin and may leave a mark when it is removed. The mark will go away within a few minutes after the sensor is removed.

\_\_\_ Blood sampling/venous catheter: The risks of a blood sample include bruising and/or discomfort from the needle, venous inflammation from the catheter, infection (less than 1 in 10,000), or the chance that you will become lightheaded. Should you feel this way we will stop the experiment, and you will be given fluids to drink (water or juice). We ask that you remain in the laboratory until we have checked your blood pressure and we are sure that you feel OK.

\_\_\_ Topical Anesthetic Cream: Numbing cream will not be used in those who have sensitivity to lidocaine. Eye contact should be avoided. When used, all sensations within the treated area are blocked. For this reason, unintentional trauma to the treated area, such as scratching,

rubbing or exposure to hot or cold temperatures should be avoided until complete sensation has returned. During or immediately after application, mild swelling, skin redness or abnormal sensation may develop at the site of treatment. In clinical studies, no serious reactions resulted from the use of the cream. Allergic reactions can occur and will be managed. Whole body adverse reactions following appropriate use are unlikely due to the small dose absorbed. If effects do occur, they are similar in nature to those seen with other local anesthetic agents and may include lightheadedness, nervousness, apprehension, dizziness, drowsiness, twitching, and vomiting. Reactions may be brief or not at all.

\_\_\_ Abdominal and leg fluid measurement (bioelectrical impedance): There are no risks involved with this procedure. The sticky electrodes may cause your skin to become irritated when we remove them.

\_\_\_ Rebreathing cardiac output: Because you breathe slightly deeper than normal, there is a chance you will feel light headed for a few seconds during rebreathing or develop a slight headache after repeated measurements. There are no other known risks to this procedure.

\_\_\_ Octreotide: Octreotide is very similar to a naturally occurring hormone called somatostatin. It is generally well tolerated, though about 1 in 10 people experience diarrhea or an upset stomach. Because this drug makes blood vessels narrow (constrict), your blood pressure will probably rise and your heart rate slow down during the injection, but this should not last more than a minute after stopping the infusion. This might be accompanied by a flushed face or a feeling of heaviness in your chest. This is a natural result of a slow heart rate. Less often people become light-headed or tired (1 in 20). If your heart rate or blood pressure changes too much, or at your request, we will end the octreotide infusion. Any unusual sensations should only last a minute or two after the infusion ends.

\_\_\_ Head-up tilt: The risk of head-up tilt is that you become lightheaded, nauseous, or pass out. Should you feel this way or we suspect that your blood pressure is starting to fall, we will stop the procedure and you should begin to feel better within minutes. In one case (less than 1 in 10,000) a person developed an unusually slow heart rhythm that returned to normal with a drug (atropine) injected in their arm vein.

\_\_\_ Treadmill: It is possible for you to stumble or fall on a treadmill leading to cuts, scrapes, dislocations, broken bones, head injury, abnormal cardiac rhythms, or even death. You will be taught the safe use of the treadmill and watched closely during exercise. All changes in speed will be made gradually, and you will be assisted in mounting and dismounting.

4. a. **Benefits to the subject:**

None

b. **Potential benefits to society:**

This information will be used to help people or patients who have difficulty maintaining their blood pressure. An example is patients who have recently undergone dialysis, some elderly people after eating a meal, and some otherwise healthy people who experience problems with lightheadedness. These problems are much more common in women.

5. **Alternative procedures which could be utilized:**

Recording heart rate, blood pressure, and blood flow is routine. Bioelectrical impedance analysis is the most non-invasive way to obtain these data.

6. **Time duration of the procedures and study:**

This study will require four visits (a total of about 10 hours) to Noll Laboratory and the General Clinical Research Center (a wing of Noll Laboratory). The screening visits will last about 2 hours and each study day will last about 4 hours each.

7. **Statement of confidentiality:**

You have the right to privacy. All information that is obtained in connection with this study will remain confidential within the limits of State Law. Information gained from this study will be released only to the investigators, and if appropriate, to your physician and the sponsors of the study with your approval. Any information provided to the sponsors of the study will not include your identity. The following may review and copy records related to this research The Office of Human Research Protections in the U.S. Dept. of Health and Human Services; The U.S. Food and Drug Administration (FDA); The Penn State University Biomedical Institutional Review Board (IRB); The Penn State University Office for Research Protections. Any information provided to the sponsors of

the study will not include your identity. The results of this study may be published in scientific journals without identifying you by name.

**8. Right to ask questions:**

If you have any questions about the research or about your rights as a subject, we want you to ask us. If you have questions later, or if you wish to report a research-related injury, please contact Sara Jarvis at (814) 865-0476 (W) or (814) 441-2113 (H) or Dr. Pawelczyk at (814) 865-3453 (W) or (814) 861-1379 (H). Questions regarding this statement or your rights as a subject of this research should be directed to the Office for Research Protections in 201 Kern Graduate Building, University Park, PA (814-865-1775). Please initial the statement below to indicate your understanding of this right.

\_\_\_ I have been given an opportunity to ask any questions I may have, and all such questions or inquiries have been answered to my satisfaction.

**9. Compensation:**

There will be no charge for any tests required for the study. You will receive \$50 after completing the first study day and \$75 after completing the second study day (for a total of \$125) for participation in this investigation to compensate your travel and loss of time. If you choose to withdraw early from the investigation, this amount will be prorated accordingly. Total payments within one calendar year that exceed \$600 will require the University to annually report these payments to the IRS. This may require you to claim the compensation that you receive for participation in this study as taxable income.

**10. Voluntary participation:**

Participation in this research study is entirely voluntary. Refusal to participate will involve no penalty or loss of benefits to which you are otherwise entitled. If you decide to participate, you are free to withdraw your consent and discontinue participation at any time without affecting your status (as a patient, student, employee, etc.), or the medical care that you will receive. You may decline to answer specific questions. However, your acceptance into the study may be contingent upon answering these questions. Under certain circumstances the study may be discontinued by the sponsor or the investigator.

**11. Event of Injury:**

Medical care is available in the event of injury resulting from research but neither financial compensation nor free medical treatment is provided. You are not waiving any rights that you may have against the University for injury resulting from negligence of the University or the investigators.

**12. Abnormal Test Results:**

In the event that abnormal test results are obtained, you will be made aware of the results within 2 weeks and recommended to contact your private medical provider for follow-up.

*This is to certify that I am 18 years of age or older and I consent to and give permission for my participation as a volunteer in this program of investigation. I understand that I will receive a signed and dated copy of this consent form. I have read this form, and understand its contents.*

---

Volunteer                      Date

I, the undersigned, have defined and explained the studies involved to the above volunteer.

---

Investigator                      Date

C- Protocol 3 Informed Consent

**INFORMED CONSENT FORM FOR CLINICAL  
RESEARCH STUDY**

**The Pennsylvania State University**

**Protocol 2**

ORP USE ONLY: IRB#28670 Doc.#1

The Pennsylvania State University

Office for Research Protections

**Title of Project:** Experimental Manipulation of the Volume Indifferent Point with Lower  
Body Negative Pressure

**Principal Investigator:** Sara S. Jarvis

227 Noll Laboratory

(814) 865-0476

ssj120@psu.edu

**Other Investigators:** James A. Pawelczyk, PhD

107 Noll Laboratory

(814) 865-3453

jap18@psu.edu

**Research Assistants:** Sandy Smithmyer

201 Noll Laboratory

(814) 863-3182

**Undergraduate Assistants:** Dave Leone

Kylie Weaver

This is to certify that I, \_\_\_\_\_, have been given the following information with respect to my participation as a volunteer in a program of investigation under the supervision of Dr. James A. Pawelczyk.

**1. Purpose of the study:**

The purpose of this investigation is to locate your "volume indifferent point" or VIP, the point where volume in your veins does not change when you stand up. We suspect this point is closer to the feet in people susceptible to passing out when they stand up. To measure the VIP we will place electrodes (sticky patches on your skin) then tilt you at different angles on a tilt table. We will repeat this measurement during lower body negative pressure. Lower body negative pressure causes more blood to shift toward your feet and should move your VIP closer to your feet. We will also test your ability to control blood pressure while you are tilted on the tilt table. A total of 16 volunteers will be included in this investigation (8 men and 8 women). To complete this study you will be asked to visit Noll Laboratory and/or the General Clinical Research Center (GCRC), on five to six separate occasions.

**2. Procedures to be followed:**

General information: A total of 5 to 6 days are required to complete this study. For female participants, the blood volume determination visit and Study Days 1, 2, and 3 need to be scheduled during a certain time of your menstrual cycle (days 2-7). Depending on your availability, this study could take up to four months to complete. These days include:

- Day 1 (Screening Day 1): Blood draw (1 tablespoon), blood pressure (sitting and standing), height and weight, waist-to-hip ratio measurements, resting electrocardiogram, medical/family history (1 hour)
- Day 2 (Screening Day 2): Physical exam and lab familiarization (1 hr)
- Day 3 (Blood volume determination): Blood volume determination (1 ½ hours; this visit may be added to another visit or may be completed separately)
- Day 4 (Study Day 1, 2, or 3): VIP determination and VIP determination with lower body negative pressure (3 ½ hours)
- Day 5 (Study Day 1, 2, or 3): Tilt table tolerance test (2 ¾ hours)
- Day 6 (Study Day 1, 2, or 3): Tilt table tolerance test with lower body negative pressure (2 ¾ hours)

Women of childbearing-age will submit a urine sample at the beginning of day 2, 3, 4, 5, and 6 for a pregnancy test. If you are pregnant, you will not be able to participate in the project.

A more detailed description of these days, and the procedures to be conducted, is provided below. For two days before each day of the study you should be sure to drink normal amounts of fluid (up to 8 glasses water, juice, or sports drinks per day). You should avoid using any type of stimulant (including cold medications and chocolate), drinking caffeine (coffee, tea, cola), and drinking alcohol after 9 PM the night before any procedure. You will not be allowed to eat or drink anything, except for water, after 9 PM the night before. You should not participate in strenuous physical activity for 12 hrs before the experiments.

Screening: You will complete a detailed medical history and a GCRC nurse will draw a small amount of blood from your arm (less than a tablespoon or 15 milliliters) with a needle in order to perform routine blood checks for anemia, kidney function, and liver function. We will take measurements of height, weight, and your waist-to-hip ratio. We will record the electrical activity of your heart (electrocardiogram) and obtain blood pressure measurements during sitting and standing.

Physical exam and lab familiarization: About 3 days after your initial screening visit you will receive a physical examination by a designated GCRC clinician. After your physical exam you will attend a familiarization session in the lab (227 Noll Lab).

Blood volume determination: On the day of the study you will be checked into the GCRC. For female subjects this visit will need to be timed around your menstrual cycle. This visit may be added to another visit or can be completed separately. A nurse will insert one catheter (a thin plastic tube) into a vein in each arm to draw blood. If you choose, a numbing cream will be applied to your arms 20 minutes before the catheters are inserted to lessen discomfort. After the catheters are inserted, a small sample of blood will be drawn and then you will receive an injection of a dye, which will be followed by blood sampling every 3 min for 30 min. During this visit we will measure:

- heart rate from sticky patches placed on your chest,
- blood pressure from your arm and a sensor on your wrist,
- hematocrit by drawing a small amount (less than 1 teaspoon) of blood from your arm,
- disappearance of the dye from your blood by drawing small amounts of blood from your arm (about 3 tablespoons total)

VIP determination: On the day of the study you will be checked into the GCRC. A nurse will insert one catheter (a thin plastic tube) into a vein in your arm to draw blood during the study. If you choose, a numbing cream will be applied to your arm 20 minutes before the catheter is inserted to lessen discomfort. After the catheter is inserted, you will be taken to the lab for testing. During the experiment we will measure:

- heart rate from sticky patches placed on your chest,
- blood pressure from your arm and a sensor on your wrist,
- your heart's pumping ability by breathing in and out of a bag containing a small amount of a gas called acetylene,
- hematocrit by drawing small amounts (about 1 tablespoon total) of blood from your arm,
- fluid shifts in your abdomen and legs using sticky patches that will be placed along your chest, abdomen, legs, and on your hand and foot.

These signals will be recorded as you lie flat on your back and while you are tilted for 6 minutes each at 15°, 30°, and 50° on a motorized tilt table. For comparison, lying on your back is 0 degrees, and standing up is 90 degrees. Once all measurements are taken at the different tilt angles, you will again lie flat on your back. We will repeat these measurements with lower body negative pressure.

Tilt table tolerance: On the day of the study you will be checked into the GCRC. A nurse will insert one catheter (a thin plastic tube) into a vein in your arm or hand. If you choose, a numbing cream will be applied 20 minutes before the catheter is inserted to lessen discomfort. After the catheter is inserted, you will be taken to the lab for testing. You will be placed on a tilt table and will be slowly raised to a tilt angle of 70° (almost standing). While you are lying flat and during the tilting, we will measure:

- heart rate from sticky patches placed on your chest,
- blood pressure from your arm and a sensor on your wrist,
- your heart's pumping ability by breathing in and out of a bag containing a small amount of a gas called acetylene,

You will remain tilted for no more than 45 minutes. During that time you should stay as relaxed and motionless as possible. If your blood pressure begins to fall or you begin to feel like you may pass out, we will return you to a flat position and you will feel better within minutes. After the tilt test, you will be lowered to a flat position.

Tilt tolerance with LBNP: On the day of the study you will be checked into the GCRC. A nurse will insert one catheter (a thin plastic tube) into a vein in your arm or hand. If you choose, a numbing cream will be applied 20 minutes before the catheter is inserted to lessen discomfort. After the catheter is inserted, you will be taken to the lab for testing. You will be placed on a tilt table and will be slowly raised to a tilt angle of 70° (almost standing) while undergoing lower body negative pressure. While you are lying flat and during the tilting, we will measure:

- heart rate from sticky patches placed on your chest,
- blood pressure from your arm and a sensor on your wrist,
- your heart's pumping ability by breathing in and out of a bag containing a small amount of a gas called acetylene,

You will remain tilted for no more than 45 minutes. During that time you should stay as relaxed and motionless as possible. If your blood pressure begins to fall or you begin to feel like you may pass out, we will return you to a flat position and stop the lower body negative pressure and you will feel better within minutes. After the tilt test, you will be lowered to a flat position.

**Detailed description of the procedures to be used:**

Please read the detailed description of each procedure below and initial each one to indicate that you have read and understand them.

\_\_\_\_\_ Heart Rate. Electrodes (sticky patches) will be applied to the skin of your chest to measure your heart's electrical activity. Other than skin irritation from the sticky electrodes, there is no known risk involved with this procedure.

\_\_\_\_\_ Blood Pressure. A wrist brace and sensor will be placed over your wrist and a cuff will be placed on your upper arm. The sensor will push down against your skin and may leave a mark when it is removed. The mark will go away within a few minutes after the sensor is removed. There is no known risk involved with this procedure.

\_\_\_\_\_ Venous Catheter. When you arrive to the GCRC a numbing lotion will be applied to your arm or hand. Once the anesthetic has taken effect the nurse will insert a soft plastic tube (catheter) into your vein. The catheter will remain in place during the experiment so that blood may be drawn without having to insert a needle each time. The catheter will be removed by a nurse before you are discharged from the GCRC.

\_\_\_\_\_ Blood draw. Skilled GCRC staff will remove blood from the catheter in your arm. The staff uses standard safety measures and sterile techniques that are used in hospitals. Small amounts of blood will be drawn during screening (about 1 tablespoon), on the blood volume determination day (about 3 tablespoons), and on the VIP determination day (about 1 tablespoon). The total amount is much less than when you donate blood.

\_\_\_\_\_ Indocyanine green (ICG). To determine your total blood volume we will look at the

ability of your liver to remove a dye from your body. After 30 minutes of rest, this dye, indocyanine green (ICG), will be injected into a tube inserted into a vein in your forearm. Small samples of your blood will be taken from a second catheter inserted into a vein near your hand every three minutes for one-half hour. The total amount of blood removed will be about 3 tablespoons (45 mls). ICG is a safe non-toxic substance that is easily cleared by the liver and can be safely injected with little risk when subjects are properly screened. **People who are sensitive or allergic to penicillin, iodides, shellfish, or sulfa drugs should not participate because they have a greater chance of being allergic to the dye.** Therefore, you will be asked if you have had any reactions to these types of medicines in the past, and if you have, you will not be allowed to participate in the project. This is a necessary requirement to reduce the slight possibility of an allergic reaction to ICG.

\_\_\_\_\_ Abdominal and leg fluid measurement (bioelectrical impedance). We will place electrodes (sticky patches) on your chest, abdomen, legs, hand and foot. A cable will be connected to each patch. A very small current of electricity will flow through the electrode and through your body. You will not be able to detect the current as it passes through your body. For comparison, this measurement is similar to body fat scales available in gyms or for home use.

\_\_\_\_\_ Cardiac Output (acetylene rebreathing). We will measure your heart's pumping rate by analyzing the air you rebreath in and out of a bag using fairly deep breaths for 15-20 seconds. The bag will contain a small concentration of two gases, acetylene and helium. The concentrations of these gases are so small that there is no risk of them catching fire. Some people report a slightly "tangy" taste from the rebreathing gas. Although you may become light-headed for a few seconds during rebreathing or develop a slight headache from repeated rebreathing, there are no other known risks to performing this procedure. The gas disappears from your lungs and blood in less than 5 minutes.

\_\_\_\_\_ Lower Body Negative Pressure. You will lie on your back during this procedure. From the waist down you will be sealed in a box-like chamber. Mild suction will be applied that will cause blood to shift into your legs similar to when you stand. We will stop the suction at your request or when we see that your blood pressure is starting to fall. Occasionally, light-headedness occurs and, rarely, a patient may faint. If this should happen, we will immediately stop the procedure, and you should begin to feel better in less than a minute.

\_\_\_\_\_ Head-up Tilt. You will lie on a table and then you will be tilted to an angle of 15°, 30°, and 50° on a tilt table for 6 min at each angle. During that time we ask that you relax and stay as motionless as possible. We will stop tilting if your blood pressure becomes too low, you experience an abnormal heart beat, or you feel light-headed or dizzy. The tilt series will be repeated with lower body negative pressure.

\_\_\_\_\_ Tilt Table Tolerance Test (with and without lower body negative pressure). You will lie on a table and then you will be tilted to an angle of 70° for up to 45 min. During that time we ask that you relax and stay as motionless as possible. We will stop tilting (and the lower body negative pressure) if your blood pressure becomes too low, you experience an abnormal heart beat, you feel light-headed or dizzy, or you ask to stop.

### 3. Discomforts and risks:

**All procedures carry risk. Risk has two aspects: severity and frequency. Severe risk might threaten the loss of life or limb, while a mild risk might be discomfort. The frequency of a risk is the chance that a problem will occur. In this section we have summarized the risks associated with the procedures used in this experiment. The risks in this experiment have different severity and frequency, and some could be life threatening. Please feel free to ask about the severity or frequency of these risks at any time. To help you decide whether or not you are willing to accept the risks associated with this experiment, the table below provides some commonly mentioned risks and the estimated chance they will occur to you:**

Contracting meningitis while you are at a large university <sup>2</sup>	1 in 33,000
Dying from being struck by lightning in your lifetime <sup>3</sup>	1 in 30,000
Dying of liver disease if you drank one beer per day <sup>3</sup>	1 in 1,000
Contracting a disease caused by radon in your home <sup>3</sup>	1 in 440
Being killed in a car accident in your lifetime <sup>3</sup>	1 in 60
Contracting cancer at some point during your life <sup>1</sup>	
	Men 1 in 2
	Women 1 in 3

References:

<sup>1</sup> American Cancer Society. Statistics for 2008. [www.cancer.org](http://www.cancer.org).

<sup>2</sup> Harrison LH, Dwyer DM, Maples CT, Billman L. (1999). Risk of Meningococcal Infection in College Students. *JAMA* 281:1906-1910, 1999.

<sup>3</sup> Walsh J. *True Odds: How Risk Affects Your Everyday Life*. 1996, p 16.

Please read the detailed description of each procedure below and initial each one to indicate that you have read and understand them.

\_\_\_\_\_ General: If during the screening procedures we should detect any abnormality, we will inform you as soon as possible so that you can contact your personal doctor for treatment. Some people find medical procedures to be scary, and feel faint as a result (vasovagal response). To keep you comfortable and informed, we encourage you to share your concerns with us at any time.

\_\_\_\_\_ Heart rate, blood pressure, and blood flow: The only known risk associated with measuring heart rate and blood pressure is the possibility that your skin will be irritated by the adhesive, cuffs, or tape. The blood pressure sensor will push down against your skin and may leave a mark when it is removed. The mark will go away within a few minutes after the sensor is removed.

\_\_\_\_\_ Blood sampling/venous catheter: The risks of a blood sample include bruising and/or discomfort from the needle, venous inflammation from the catheter, infection (less than 1 in 10,000), or the chance that you will become lightheaded. Should you feel this way we will stop the experiment, and you will be given fluids to drink (water or juice). We ask that you remain in the laboratory until we have checked your blood pressure and we are sure that you feel OK.

\_\_\_\_ Topical Anesthetic Cream: Numbing cream will not be used in those who have sensitivity to lidocaine. Eye contact should be avoided. When used, all sensations within the treated area are blocked. For this reason, unintentional trauma to the treated area, such as scratching, rubbing or exposure to hot or cold temperatures should be avoided until complete sensation has returned. During or immediately after application, mild swelling, skin redness or abnormal sensation may develop at the site of treatment. In clinical studies, no serious reactions resulted from the use of the cream. Allergic reactions can occur and will be managed. Whole body adverse reactions following appropriate use are unlikely due to the small dose absorbed. If effects do occur, they are similar in nature to those seen with other local anesthetic agents and may include lightheadedness, nervousness, apprehension, dizziness, drowsiness, twitching, and vomiting. Reactions may be brief or not at all.

\_\_\_\_ Abdominal and leg fluid measurement (bioelectrical impedance): There are no risks involved with this procedure. The sticky electrodes may cause your skin to become irritated when we remove them.

\_\_\_\_ Rebreathing cardiac output: Because you breathe slightly deeper than normal, there is a chance you will feel light headed for a few seconds during rebreathing or develop a slight headache after repeated measurements. There are no other known risks to this procedure.

\_\_\_\_ Indocyanine Green Dye (ICG): ICG is a safe non-toxic substance that is easily cleared by the liver and can be safely injected with little risk when subjects are properly screened. **The only known risk involved is when a person is sensitive or allergic to penicillin, iodides, or sulfa drugs.** Therefore, you will be asked if you have had any reactions to these types of medicines in the past, and if you have, you will not be allowed to participate in the project. This is a necessary requirement to insure that the slight possibility of an adverse response to ICG in a subject sensitive to the drugs listed above is alleviated. For those people that have no known allergy, there is a 1 in 250 risk of a mild to moderate reaction. Symptoms include lightheadedness, nausea, hives and itching. Rarely, (1 in 2000) a serious allergic reaction which affects the entire body (anaphylaxis) may develop. An anaphylactic reaction can be life-threatening. You are free to discontinue these tests at any time.

\_\_\_\_ Lower body negative pressure: The risk of lower body negative pressure is that you become lightheaded, nauseous, or pass out. Should you feel this way or we see that your blood pressure

is starting to fall, we will stop the procedure and you should begin to feel better within seconds. If we continued the suction you would develop an abnormally slow heart rhythm (bradyarrhythmia). This stops when suction is turned off, but in one case (less than 1 in 10,000) a person was injected with a drug (atropine) in their arm vein to restore their heart rhythm to normal. Because we observe your blood pressure closely, it is not likely (about 1 in 20) that you will develop a slow heart rhythm.

\_\_\_\_ Head-up tilt: The risk of head-up tilt is that you become lightheaded, nauseous, or pass out. Should you feel this way or we suspect that your blood pressure is starting to fall, we will stop the procedure and you should begin to feel better within minutes. In one case (less than 1 in 10,000) a person developed an unusually slow heart rhythm that returned to normal with a drug (atropine) injected in their arm vein. Because we observe your blood pressure closely, it is not likely (about 1 in 20) that you will develop a slow heart rhythm.

4. a. **Benefits to the subject:**

None

b. **Potential benefits to society:**

This information will be used to help people or patients who have difficulty maintaining their blood pressure. An example is patients who have recently undergone dialysis, some elderly people after eating a meal, and some otherwise healthy people who experience problems with lightheadedness. These problems are much more common in women.

5. **Alternative procedures which could be utilized:**

Recording heart rate, blood pressure, and blood flow is routine. Bioelectrical impedance analysis is the most non-invasive way to obtain these data.

6. **Time duration of the procedures and study:**

This study will require five to six visits (a total of about 13 hours) to Noll Laboratory and the General Clinical Research Center (a wing of Noll Laboratory). The screening visit will last about 1 hour, the physical exam and familiarization visit about 1 hour, the blood volume determination visit about 1 ½ hrs, the VIP determination visit about 3 ½ hrs, and the tilt table tolerance visits up to 2 ¾ hours each.

7. **Statement of confidentiality:**

Your participation in this research is confidential. The data will be stored and secured in Noll Laboratory in a locked file. Electronic files will only identify you by a subject identification number, not by name. In the event of any publication or presentation resulting from this research, no personally identifiable information will be disclosed. Penn State's Office for Research Protections, the Biomedical Institutional Review Board, the U.S. Food and Drug Administration (FDA), the Office for Human Research Protections in the Department of Health and Human Services, and the National Aeronautics and Space Administration may review records related to this research study.

8. **Right to ask questions:**

Please contact Sara Jarvis at (814) 865-0476 (W) or (814) 441-2113 (H) or Dr. Pawelczyk at (814) 865-3453 (W) or (814) 861-1379 (H) with questions, complaints or concerns about the research. You can also call this number if you feel this study has harmed you. Questions about your rights as a research participant may be directed to Penn State University's Office for "Research Protections at (814) 865-1775.

Please initial the statement below to indicate your understanding of this right.

\_\_\_ I have been given an opportunity to ask any questions I may have, and all such questions or inquiries have been answered to my satisfaction.

9. **Compensation:**

There will be no charge for any tests required for the study. You will receive:

- \$25 after completing the blood volume determination visit,
  - \$35 after completing the VIP determination visit,
  - \$30 after completing the first tilt table tolerance visit, and
  - \$35 after completing the second tilt table tolerance visit
- (for a total of \$125) for participation in this investigation to compensate your travel and loss of time. If you choose to withdraw early from the investigation, this amount will be prorated accordingly. Total payments within one calendar year that exceed \$600 will require the University to annually report these payments to the IRS. This may require you to claim the compensation that you receive for participation in this study as taxable income.

**10. Voluntary participation:**

Participation in this research study is entirely voluntary. Refusal to participate will involve no penalty or loss of benefits to which you are otherwise entitled. If you decide to participate, you are free to withdraw your consent and discontinue participation at any time without affecting your status (as a patient, student, employee, etc.), or the medical care that you will receive. You may decline to answer specific questions. However, your acceptance into the study may be contingent upon answering these questions. You may not be accepted into the study if you participate in other research studies, depending on what those studies involve. Under certain circumstances the study may be discontinued by the sponsor or the investigator.

**11. Event of injury:**

Medical care is available in the event of injury resulting from research but neither financial compensation nor free medical treatment is provided. You are not waiving any rights that you may have against the University for injury resulting from negligence of the University or the investigators.

**12. Abnormal test results:**

In the event that abnormal test results are obtained, you will be notified within 3 business days so that you can contact your private medical provider for follow up.

*This is to certify that I am 18 years of age or older and I consent to and give permission for my participation as a volunteer in this program of investigation. I understand that I will receive a signed copy of this consent form. I have read this form, and understand its contents.*

\_\_\_\_\_

Volunteer

Date

I, the undersigned, have defined and explained the studies involved to the above volunteer.

\_\_\_\_\_

Investigator

Date

## ACADEMIC VITA OF DAVID MICHAEL LEONE

David M. Leone  
17 Overlook Avenue  
Randolph, New Jersey 07869  
dml5064@psu.edu  
201-919-1759

### Education:

Bachelor of Science Degree in Kinesiology, Penn State University, Spring 2010  
Honors in Kinesiology  
Thesis Title: Baroreflex Function during Orthostatic Stress with Administration of a Somatostatin Analog  
Thesis Supervisor: James A. Pawelczyk

### Related Experience:

Volunteer at Mount Nittany Medical Center: Cardiac Catheterization Laboratory  
Supervisor: Alice Clark  
2009 to 2010

### Work Experience:

Penn State Fitness Centers: Trainer/ Assistant Supervisor  
Supervisor: Stacey Krupski  
2007 to 2010

### Awards and Honors:

Eagle Scout (2006)  
Schreyer Honors College  
Dean's List every semester  
Inducted into the Health and Human Development Honors Society (2007)  
Inducted into the Phi Kappa Phi Honors Society (2008)  
Noll Endowment Research Grant recipient (2008)  
Phi Beta Kappa Research Grant recipient (2009)  
Undergraduate Research Exhibition: Honorable Mention for the Information Literacy Award (2010)

### Community Service:

Pennsylvania State University IFC/Panhellenic Dance MaraTHON: Rules and Regulations Committee