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14. ABSTRACT Obesity has reached epidemic levels and yet the incidence continues to rise. The current study is seeking to examine the hypothesis that obesity may reflect dysfunctioning of the hypothalamic-pituitary-adrenal (HPA) axis in response to stressors. African American persons are at greatest risk, but reasons for this difference are unknown. We will study 127 men and women of Caucasian and African American ethnicity and examine their responses to physiologic stressors: exercise and ingestion of a meal. Methods: The HPA axis will be studied in some detail by using two stressor paradigms and two steroid regimens. We expect to be able to detect subtle differences in HPA axis reactivity in obese individuals that might contribute to morbidity and perhaps even make individuals resistant to therapeutic interventions. Results: We have enrolled 160 participants, with 123 completed. Data collection and analyses are proceeding on schedule. Two abstracts were presented in 2006, one presented in 2007, and one was submitted and accepted for presentation in Summer 2008. Conclusions: We are on schedule for all study milestones and look forward to being able to answer the important questions regarding the potential role of the HPA axis in obesity.					
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INTRODUCTION

Obesity has reached epidemic levels and yet the incidence continues to rise. The current study is seeking to examine the hypothesis that obesity may reflect a dysfunction of the hypothalamic-pituitary-adrenal (HPA) axis in response to stressors. African American persons are at greatest risk, but reasons for this difference are unknown. We are studying 127 men and women of Caucasian (CA) and African American (AA) ethnicity to examine their responses to physiologic stressors: exercise and ingestion of a meal.

BODY

Year Five:

1. *Recruit, screen, and test 20 Overweight/Obese and 8 Non-obese subjects*

Table 1 presents the participants recruited and tested over the past year.

Table 1. Breakdown of Normal/Overweight/Obese Participants by Ethnicity (Year 4)

Normal (NW) Overweight (OW) & Obese (OB)	CA-NW	AA-NW	CA-OW	AA-OW	CA-OB	AA-OB	TOTAL
Screened	1	35	2	16	12	33	99
Recruited/Enrolled	0	12	0	4	6	16	38
In Progress	0	3	0	0	0	1	4
Dropped	0	1	0	0	0	5	6
Completed	0	8	0	4	6	10	28

2. *Complete subject recruitment*

Since the start of the study, we have recruited 160 subjects. Of those, 123 subjects have completed the study and 4 are in progress; 33 have dropped out (Table 2). We have completed recruitment and will finish testing the remaining 4 subjects by the end of March. We have over recruited AA in every weight class as expected, but because the hormonal data suggest that some subjects did not take the prescribed treatments, we have increased our accrual in selected cells. Recruiting for the obese class in both CA and AA went extremely well this year compared to the past. With new staff and recruiting methods, we were able to recruit the last thirty subjects with a minimal drop out rate. Three subjects were excluded due to health issues and the other three subjects were unable to complete the study due to personal schedule changes.

For the past two years, we provided body composition services and health consultations as community service and as a recruitment tool at the Montgomery County Heart Health Symposium for African Americans in Germantown, MD in March 2006, June 2007, and March 2008.

Table 2. Years 1-5 Subject Recruitment and Progress

		Normal	Overweight	Obese	Total
AA	Enrolled	34	37	38	109
	In Progress	3	0	1	4
	Completed	22	28	25	75
	Dropped	9	9	12	30
	Needed	0	0	0	0
CA	Enrolled	18	16	17	51
	In Progress	0	0	0	0
	Completed	16	16	16	48
	Dropped	2	0	1	3
	Needed	0	0	0	0
	Total in Progress	4			
	Total Completed	123			
	Total Needed	0			

3. *Complete subject testing*

To date, 123 subjects have completed testing in the laboratory; 4 subjects are in progress and are scheduled to finish testing by the end of March.

4. *Continue evaluating, reducing, and analyzing data*

Multiple meetings have occurred among the PI, Co-investigators, with the Project Coordinator and other key staff on a monthly basis to discuss issues and examine data collected on all completed subjects. Preliminary hormone, psychological, and other physiological data have been analyzed while preparing for one abstract, two scientific presentations and two manuscripts for publication. We are ahead of schedule for this goal.

5. *Continue with biochemical analyses.*

Radioimmunoassay (RIA) and ELISA analyses are in process for several of the different hormones of interest. Specifically, we have completed ACTH data on 115 subjects, Insulin data on 112 subjects, Cortisol (CORT) data on 115 subjects, DHEA data on 57 subjects, and DHEAS data on 103 subjects. Unfortunately, we have had similar problems with our Insulin and DHEA kits that we experienced last year. The manufacturer stopped production for several months, which caused a delay in analyzing samples for Insulin and DHEA. We plan to complete the biochemical analyses on ACTH, CORT, DHEAS, and Insulin by mid May and DHEA by August. We will begin testing CBG after completing our analyses of DHEA; this will take a few months to complete. This goal is on track.

6. *Begin statistical analyses on ethnicity/obesity and potential interactions.*

We have partial data from 127 subjects (123 have complete data and 4 have partial data). Table 3 presents basic demographic and anthropometric data for the groups by ethnicity and body mass index (BMI) category. From preliminary data analyses, age, weight, BMI, and maximal aerobic capacity were significantly different across weight groups. In contrast, fasting blood glucose was not significantly different between AA and CA. However, despite similar weights and BMI, maximal aerobic capacity (VO_{2max}) was significantly lower in AA than CA (Table 3). Overall, VO_{2max} was 42.6 ± 9.5 (ml/kg/min) for CA and 36.2 ± 10.0 (ml/kg/min) for AA. The values for AA are very low relative to normative data and indicate poor cardiovascular fitness.

Table 3. Characteristics and Physiological Measures by Ethnicity and Weight Group

	CA-NW (n=16)	CA-OW (n=16)	CA-OB (n=12)	AA-NW (n=20)	AA-OW (n=26)	AA-OB (n=21)
Age (yrs)	24.0 ± 3.7	29.9 ± 3.8	30.8 ± 5.8	28.3 ± 8.6	30.7 ± 8.4	34.9 ± 7.1
Weight (kg)	62.7 ± 9.5	85.4 ± 8.7	103.8 ± 11.0	64.8 ± 10.1	81.8 ± 12.1	94.8 ± 12.9
BMI (kg/m ²)	21.9 ± 1.5	27.1 ± 1.4	33.9 ± 2.5	22.3 ± 1.5	27.3 ± 1.5	32.6 ± 2.1
VO _{2max} (ml/kg/min)	50.4 ± 4.9	43.5 ± 8.4	33.8 ± 6.4	40.6 ± 9.5	39.6 ± 9.4	29.0 ± 6.8
Glucose (mmol/L)	5.1 ± 0.6	5.4 ± 0.7	5.4 ± 0.6	5.1 ± 0.67	5.1 ± 0.7	5.3 ± 0.7

Values are mean ± SD.

7. *Reduce and interpret data on HPA reactivity from the exercise and meal challenge tests as a function of ethnicity and obesity after all subjects have been tested.*

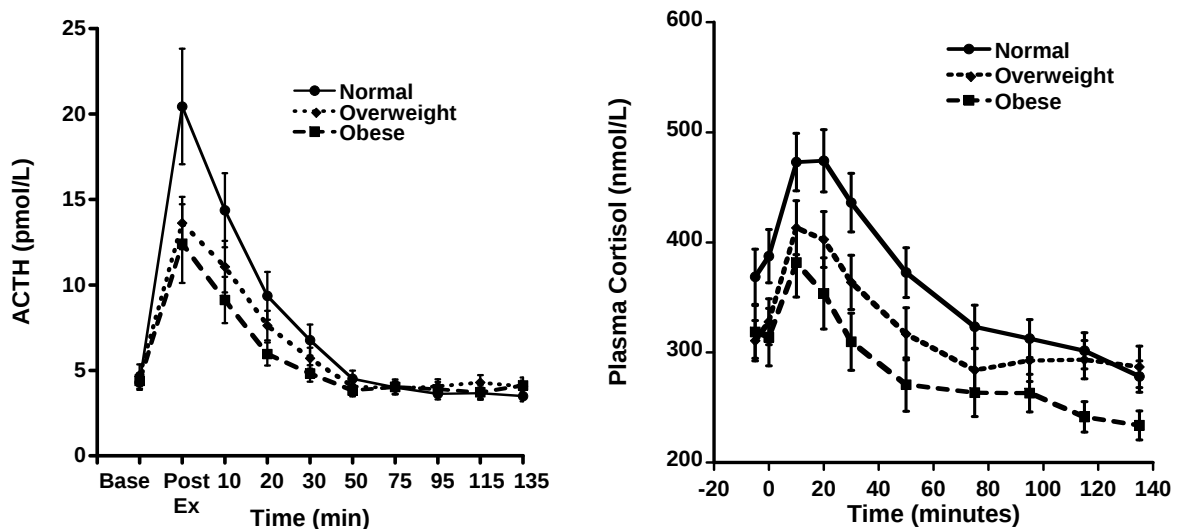
Areas under the curve (AUC) for the exercise challenge and areas under the curve for the meal challenge were calculated for ACTH (n = 44 CA, 60 AA) and CORT (n = 44 CA, 60 AA) responses. Preliminary analyses of the data showed higher ACTH and cortisol for CA in response to exercise as compared to AA (Table 4). With exercise and BMI class, only CA had a significantly higher response in cortisol levels. The HPA axis was not activated by the meal and did not differ by either ethnicity or weight category.

Table 4. ACTH AUC and CORT AUC after Exercise Challenge

	ACTH (pM•135 min)	CORT (nM•135 min)
AA	519.7 ± 342.4	22,994 ± 8227*
CA	668.9 ± 508.0	29,082 ± 8938

Values are mean ± SD. *p = 0.001

Figure 1 presents the preliminary patterns in terms of the magnitude and timing of peak ACTH and cortisol responses to exercise as a function of BMI category. Peak ACTH and cortisol are lower in both overweight and obese participants.

Figure 1. Plasma ACTH and Cortisol over Time by BMI

Reduce and interpret data describing relationship between HPA axis resistance to feedback control and insulin resistance as a function of obesity and ethnicity after all subjects have been tested.

Fasting IR was calculated from serum insulin by using HOMA [(glucose (mmol/L) x insulin (μIU/mL))/22.5] for 66 AA and 44 CA (Figure 2). Preliminary analyses showed that individuals with a higher BMI have a greater IR. Differences were found among BMI groups, with the OB group being significantly different than the NW and OW group. There were no differences in ethnicity or age.

Preliminary analyses showed that both serum glucose and insulin responded differently by BMI category and Treatment. Treatment 2 (Dexamethasone) increased glucose and insulin levels in all weight groups (Figure 3 dashed lines).

Figure 2. Insulin Resistance by HOMA by BMI and Ethnicity

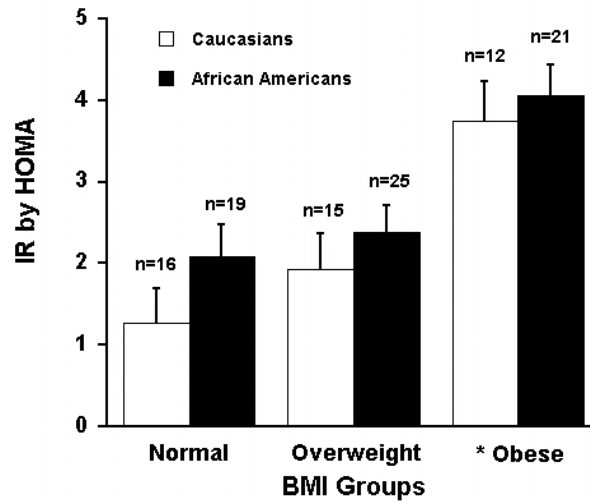
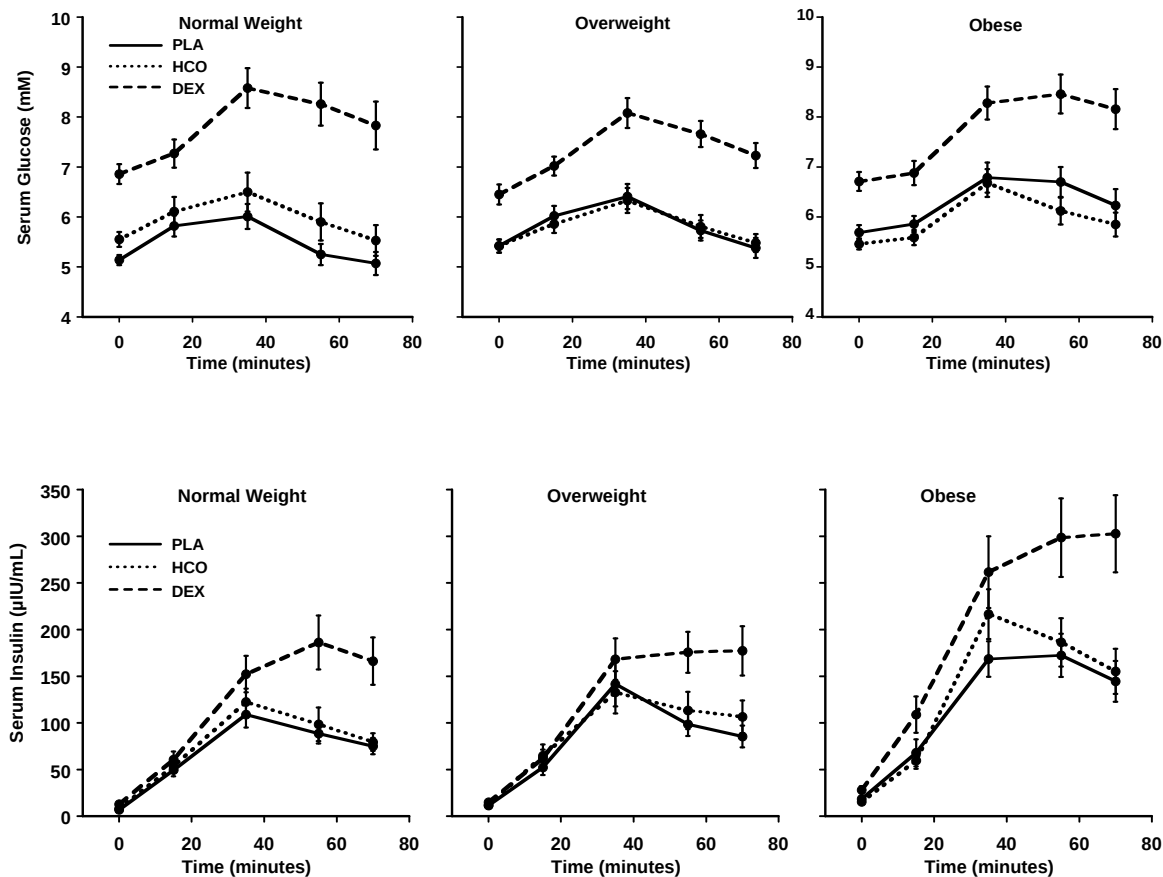


Figure 3. Serum Insulin and Glucose by BMI and Treatment after a Meal



8. *Reduce and interpret data describing relation between exercise-associated increases in insulin and glucocorticoid sensitivity as a function of ethnicity.*

Areas under the curve (AUC) for insulin were calculated over the course of the meal challenge. Preliminary analyses of the data showed significant differences between AA and CA across all three treatment groups. AA produced more insulin than CA for every treatment condition (Table 5). Individuals were grouped by BMI (normal weight/NW: $18 \leq 25$, over weight/OW: $25 \leq 30$, and obese/OB: $30 \leq 38$). Significant differences across BMI groups were also noted for all treatments, with the OB group releasing significantly greater amounts of insulin than NW and OW. When analyzed by ethnicity, significant differences in BMI groups were found only among CA with Treatments 1 and 3 while there was a significant difference for AA in Treatment 2.

Table 5. Insulin AUC ($\mu\text{IU/mL}/70\text{min}$) by ethnicity and BMI

Group	n	TRT 1	TRT 2	TRT 3
AA *	58	9,687 $\pm$ 837	14,212 $\pm$ 1203	8,929 $\pm$ 664
CA	42	5,820 $\pm$ 570	8,100 $\pm$ 760	5,060 $\pm$ 471
AA NW	18	7,378 $\pm$ 988	11,730 $\pm$ 1880	7,118 $\pm$ 908
AA OW	20	9,999 $\pm$ 1684	12,464 $\pm$ 1597	8,883 $\pm$ 1229
AA OB	20	11,451 $\pm$ 1432	18,319 $\pm$ 2437	10,696 $\pm$ 1181
CA NW	16	5,377 $\pm$ 753	7,508 $\pm$ 657	4,750 $\pm$ 481
CA OW	16	4,199 $\pm$ 559	6,716 $\pm$ 1157	4,102 $\pm$ 654
CA OB	10	* 9,124 $\pm$ 1471	11,261 $\pm$ 2197	7,090 $\pm$ 1360

Values are mean $\pm$ SE. * $p < 0.05$

Areas under the curve (AUC) for glucose were calculated over the course of the meal challenge. The ratios of the AUC for insulin to glucose were calculated and significant differences were found across all treatments by ethnicity (Figure 4). Despite similar plasma glucose levels for AA and CA, AA released a larger amount of insulin in response to the same meal challenge (Figure 5). The code for the treatments was broken due to the questioning of several subjects taking treatment 2; data for these participants were unusually high relative to the rest of the subjects – a value less than 5 $\mu\text{g/dl}$ or 138 nM is expected after taking 4 mg of DEX.

Figure 4. Ratio of AUC for Insulin to Glucose across Treatments

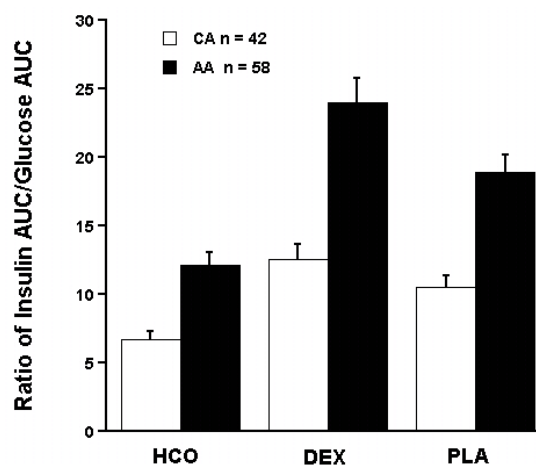
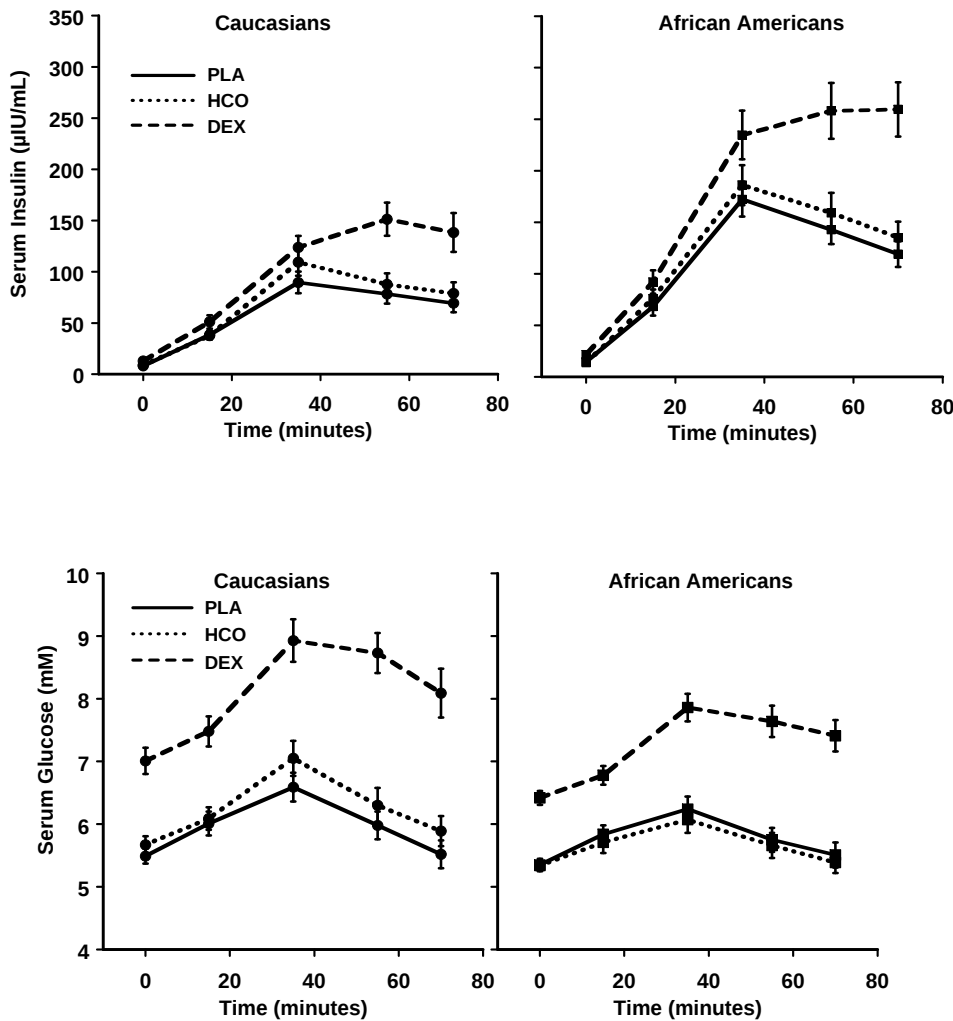


Figure 5. Serum Insulin ($\mu\text{IU/mL}$) and Serum Glucose (mM) over Meal Time



9. *Examine data as a function of gender after 20 men and 20 women have been tested.*

Expected gender differences were found in the preliminary data analyses between men and women. Men have a greater maximal aerobic capacity ($\text{VO}_{2\text{max}}$) (Men: 43.2 ± 8.9 vs. Women 34.1 ± 9.7 ml/kg/min), waist circumference (Men: 90.5 ± 13.9 vs. Women: 85.0 ± 13.9 cm), and a lower percent body fat (Men: 26.1 ± 6.0 vs. Women: 36.8 ± 7.4 %) than women. A significant gender difference was also found with fasting glucose, but not insulin resistance, as measured by HOMA IR.

Gender differences were also noted for select psychological variables on the Stress Profile and the Beck Depression Inventory (BDI) questionnaire.

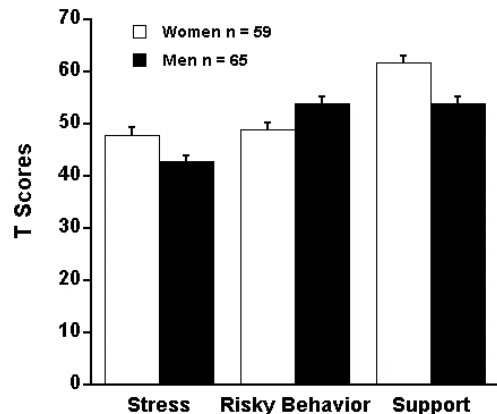


Figure 6. Differences in Stress Profile Subscales

Although women had significantly higher scores for self-reported stress, they also reported having greater social support network (ns) than men (Figure 6). Men reported participating in risky behaviors more frequently than women. Men also had significantly lower scores on the BDI (Men: 2.7 ± 0.4 vs. Women: 5.0 ± 0.6) as compared to women. The trend for greater coping strategies among women through social support may offset the negative feelings of stress and depression in women.

10. *Prepare final report on results.*

This has not been prepared since an extension has been granted for the study until 31 March 2008.

KEY RESEARCH ACCOMPLISHMENTS

- No significant differences were found between AA and CA in the measurements of weight (AA: $\pm$ vs. CA: $\pm$ kg), BMI (AA: 27.6 ± 4.6 vs. CA: 26.9 ± 4.9 kg/m²), waist circumference (AA: 87.7 ± 12.0 vs. CA: 86.4 ± 14.4 cm), and fasting plasma glucose (AA: 5.1 ± 0.7 vs. CA: 5.3 ± 0.7 mmol/L).
- Maximal aerobic capacity (VO_{2max}) was significantly lower in AA than CA (36.5 ± 10.1 and 43.6 ± 9.1 ml/kg/min for AA and CA, respectively) [$F_{(1,91)} = 14.0$, $p < 0.001$].
- Insulin Resistance (IR) by HOMA (μ IU/mL x mmol/L) was significantly higher in Obese (3.9 ± 0.29) compared to Normal (1.7 ± 0.28) and Overweight (2.2 ± 0.26)
- Fasting glucose and insulin by HOMA IR differed significantly by BMI category [$F_{(2,79)} = 16.1$, $p < 0.001$].
- In AA: IR was positively correlated with BMI ($r = 0.39$) and hip-waist ratio ($r = .26$) and negatively correlated to VO_{2max} ($r = -0.32$).
- In CA: IR was positively correlated with BMI ($r = 0.70$), hip-waist ratio ($r = 0.42$), and body fat percent ($r = .49$); and negatively correlated with VO_{2max} ($r = -0.59$).
- Morning basal ACTH concentrations were comparable in CA and AA, but morning cortisol concentrations were significantly lower in AA than CA [$F_{(1,22)} = 16.38$, $p < 0.001$].
- Peak ACTH response to exercise differed significantly by ethnicity: AA: 18.9 ± 14.2 vs. CA: 28.9 ± 22.6 ; likewise, AA had significantly blunted cortisol responses [$F_{(1,22)} = 12.8$, $p = 0.001$].
- Glucose levels after a liquid meal did not differ between AA and CA, but insulin responses were significantly higher in AA [$F_{(1,75)} = 15.3$, $p < 0.001$; AUC - $F_{(1,75)} = 9.86$, $p = 0.002$].
- Women reported significantly higher scores for stress (Women: 47.8 ± 1.4 vs. Men: 42.9 ± 1.1) than men [$F_{(1,88)} = 8.771$, $p = 0.004$].
- Men had significantly higher scores for participating in risky behaviors (Men: 54.20 ± 1.0 vs. Women: 48.08 ± 1.2) than women [$F_{(1,88)} = 15.989$, $p = 0.004$].
- Men also had significantly lower scores on the Beck Depression Inventory (Men: 2.8 ± 0.4 vs. Women: 4.9 ± 0.5) as compared to women [$F_{(1,85)} = 4.435$, $p = 0.004$].

REPORTABLE OUTCOMES 2006-2008

ABSTRACTS

1. Abraham PA, McGill C, Davis J, Zeno SA, C, Deuster PA and Poth M. Physiological Correlates of the Insulin Response to a Test Meal in African American and Caucasian Women. Accepted for presentation at the Endocrine Society 2008.
2. Kim SJ, Oates C, Fendrick N, Zeno S, Faraday M, Sbrocco T, Poth M, and Deuster P. Physiological and Psychological Correlates of Insulin Resistance in African Americans and Caucasians. Presented at the Society for Behavioral Medicine 2006.
3. Fendrick N, Stephens Q, Oates C, Kim S, Zeno S, Faraday M, Poth M, and Deuster P. Physiologic and Behavioral Correlates of Obesity in African Americans and Caucasians. Presented at the 2006 DoD Military Health Research Forum (MHRF).
4. Kim SJ, Poth M, Deuster P, Fendrick N, and Zeno S. Differential Hormone Responses to a Liquid Meal and Exercise in African Americans and Caucasians. Presented at The Endocrine Society 2007.
5. Stephens Q, Deuster P, and Poth M. Applicability of VO₂ max Criteria in African American and Caucasian Individuals. Submitted to the American College of Sports Medicine.

MANUSCRIPTS

1. Kim SJ, Oates C, Poth M and Deuster PA. Psychological and physiological correlates of insulin resistance at fasting and in response to a meal in African Americans and Caucasians. Submitted to Annals of Behavioral Medicine.

PRESENTATIONS

1. Kim SJ, Sbrocco T, and Deuster PA. Ethnicity and Insulin Resistance: Role of Chronic Stress, Exercise, and Eating Behaviors. Presenting at the Society of Behavioral Medicine Annual Meeting, San Diego, CA in March 2008.
2. Kim SJ, Deuster PA, Fendrick NA, Zeno SA and Poth M. Differential Hormone Responses to Food and Exercise in African Americans and Caucasians. Poster presented at The Endocrine Society's 89th Annual Meeting in Toronto, Canada in June 2007.
3. Stephens Q. Featured Guest Speaker – Montgomery County Cable Community Comments Television Program (Topic: Cardiovascular Disease and Exercise) in May 2006.
4. Kim S. HPA Axis Responsivity and Ethnicity. Oral presentation for American Psychological Association meeting in New Orleans, LA in August 2006
5. Deuster P. "Stress Reactivity: Models, Markers, Meaning, Muddlement" at the Office of Naval Research (ONR) Stress Physiology Program Review Meeting in San Diego 25 – 27 July 2007.
6. Deuster PA. "Physiologic and Ethnic Effects on Stress Reactivity" for the Department of Medical and Clinical Psychology on 11 February 2008.
7. Deuster PA. "Exercise as a neuroendocrine stimulus: Exercise responses in health and disease" for the Foundation for Advanced Education in the Sciences Course: Neural Immune Interactions in Health and Disease. April 2007.

GRANTS ACCEPTED

1. P20 NIH USH Health Disparities Research Center

CONCLUSIONS

Metabolic syndrome is a cluster of cardiovascular risk factors, including abdominal obesity, dyslipidemia, hypertension, insulin resistance, proinflammatory state, and prothrombotic state.

Those with metabolic syndrome are at risk for other diseases, such as coronary heart disease, stroke, and Type 2 diabetes. Recent investigations have demonstrated that ethnic and gender-based differences contribute to the prevalence, associated factors, and status of this disease.

Several socioeconomic variables are significantly associated with metabolic syndrome. Low socioeconomic position (SEP) is associated with a high prevalence of metabolic syndrome in AA and CA and the association is stronger in women than men. Furthermore, low education, low poverty income ratio, and socioeconomic position were specifically related to the metabolic syndrome in women. Despite the gender differences, education continues to be a variable associated with the components of metabolic syndrome. Improving access to higher education and targeting efforts at intervention and prevention towards low-SEP populations could reduce the risk for metabolic syndrome.

Ethnicity is a significant factor in the prevalence of individual and clustered components of the metabolic syndrome. Specifically, the HTN component of metabolic syndrome is significantly more common among AA with (73.1%) and without (47.5%) diabetes in comparison to their CA (58.6% and 32.4%) and Mexican American (50.8% and 23.4%) counterparts (with and without diabetes), respectively. Additionally, AA have a higher prevalence of elevated serum C-reactive protein and high plasma fibrinogen than CA. In diabetics, abdominal obesity is more prevalent among CA (80.6 %) than Mexican Americans (67.8 %).

Insulin resistance is more common in AA than CA. However, a single bout of exercise has been shown to improve insulin sensitivity in sedentary overweight/obese African American women in response to a 75 min brisk treadmill walk. This response was also previously observed in sedentary CA individuals. Since it has been established that insulin resistant AA and CA exhibit several differences with regard to the prevalence of metabolic syndrome and associated factors, it is important to examine the responsiveness to interventions by ethnicity.

In the abstract to be presented in June 2007, we have intriguing results of differential hormonal responses to daily activities. We found that anthropometric measures were independent of the insulin response to a meal and that the insulin response was higher in AA. The hormonal profiles of AA differ from those in CA and appear to be similar to those seen in more obese individuals: lower morning cortisol levels and higher insulin concentrations in response to a meal. This exaggerated insulin response in healthy AA may be an early indication of pre-insulin resistance, a risk factor for cardiovascular disease and Type 2 diabetes mellitus. The relations among these hormonal differences, life stress and ethnicity deserve further exploration.

Our goals for the final year of the study include completion of testing participants, analyzing data, continuing and completing biochemical and statistical analyses, and examining data for HPA reactivity, resistance to feedback control and insulin resistance, and exercise-associated increases in insulin and glucocorticoid sensitivity. We have successfully met our previous goals and are ahead of schedule in many areas. We have enrolled over 160 participants to date and have completed testing of 123 participants. We also submitted seven abstracts and gave five poster presentations based on our physiological, biochemical, and psychological testing. In addition, we have prepared two manuscripts discussing the criteria of maximal aerobic testing in AA and the correlates of insulin resistance between AA and CA. Over the next year we will continue to examine differences between CA and AA in terms of potential underlying causes of metabolic syndrome and how different physiologic stressors activate the HPA axis. Importantly, we will relate and explore metabolic processes intrinsic to obesity and associated CHD risk factors as a function of ethnicity. We expect to add to the body of knowledge that

surrounds metabolic syndrome, because of our comparative design with factors of ethnicity, weight, and gender. We will also continue to examine the data, prepare manuscripts, and complete the final report on the results.

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APPENDICES

None.