DIFFERENCES BETWEEN MALE AND FEMALE STUDENTS IN CARDIOVASCULAR AND ENDOCRINE RESPONSES TO EXAMINATION STRESS

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Background: It is known that stress alters biological processes. The aim of the present study was to examine the effect of examination stress in young adult male and female students. **Methods:** Examination stress was studied in 28 young female and 21 young male volunteer students of Rafsanjan university of medical Sciences, 0.5 hour before Physiology examination (stress condition) at 10-12 a.m. and 45 days after examination (control condition) at the same time in the year 2003. **Results:** There were no differences in BMI of male and female groups at control and stress conditions. Subsequent analysis between two sexes showed that males had significantly higher systolic [SBP (124.7±4.01 mmHg)] and diastolic blood pressure [DBP (76.56±2.48 mmHg)], heart rate [HR (84.6±2.63)] increases in stress condition, in both sexes, but in males the increasing of HR is more than females, whereas females had higher repsiratory frequency increase in stress condition, compare to males. Moreover, there were no differences in SBP, DBP and HR responses to stress condition in different phases of the menstrual cycle. The increased amonut of the plasma cortisol in stress condition was significantly higher in males (485.3±37.9 in stress vs. 335.7±27.9 pg/ml in control) than females, stress also reduced females' ACTH in both phases of the menstrual cycle (13.3±0.8 in stress vs. 27.47±7.25 pg/ml in control), but in males stress increased ACTH (43.72±4.45 in stress vs. 49.29±3.25 pg/ml in control). In males, stress induced a significant decrease in plasma testosterone. Plasma progesterone in response to stress showed a significant decrease in the luteal phase. **Conclusion:** These data suggest that, the responses to physiology examination stress are different between two sexes.

Key words: Cardiovascular; Endocrine; Physiology Examination; Sex differences; Stress.

INTRODUCTION

Stress is the specific and nonspecific response of the body to any kind of physiologic pressure or unwanted forces due to environmental or peripheral effects. Stress acts in different axes including hypothalamus-pituitary–adrenal (HPA) axis, hypothalamus–pituitary–gonads (HPG) axis, and hyphothalamus–pituitary–thyroid (HPT) axis.[1,2] In addition, there are autonomic responses by sympathetic nerve system to the stressful condition i.e. the activation of sympathetic nerve system and the activation of the above axes specially HPA occur, simultaneously.[2,3] Many kinds of different stress need the responses of endocrine glands, for example responses to the stress of temperature, hypoglycemia, surgery, pain, occupations, or decreasing energy, are mediated by different endocrine glands. Individual diffrences in cardiovascular and neuroendocrine responses to stress are cardiovascular complication (hypertension and increasing heart beat), gastric ulcer, migrane headache, and asthma.[2]

Among neuroendocrine stress reactions the releasing of catecholamines, adrenaline and noradrenaline play a key role in human adjustment to environmental demands, for example, augmentation of the peripheral catecholamines level is accompanied by a series of changes in cardiovascular and metabolic function which facilitate adaptation to a wide range of stimulus conditions.[2]

Human studies suggest that cardiovascular responses to stress are sex- dependent and in females differ during the menstrual cycle.[4,5] Women usually have lower blood pressure and adrenaline in responses to stress than do men,[6] suggesting that determinants of women's stress responses may differe from men's responses. It has been shown that premenopausal and postmenopausal women differed in physiological responses to behavioral stressor.[7]

Most of the studies in different responses to stress carried out in middle age men and women, and there were not many reports that investigated the effects of stress on the young university students. The examination stress was accounted as an acute physiologic stress in some subjects. Examinations are anecdotally viewed as extremely stressful to Singapore schoolchildren.[8] Patients with acne may experience worsening of the disease during examinations.[9] A study in Netherland on PhD students showed that although the blood pressure did not changed significantly by the examination stress, but peripheral benzodiazepine receptor density, allopregnanolon, and cortisol concentration were significantly increased during examination.[10]

It is crucial to survey the determinants of young male's and female's stress induced by examination, therefore, the general aim of this study was: firstly, to measure the cardiovascular and neuroendocrine responses to examination stress in young students, secondly, to find whether the responses are sex dependent, and thirdly, is there any differences in the responses to stress in follicular and luteal phases of menstrual cycles.

MATERIAL AND METHODS

Examination stress was studied in 28 young females and 21 young males, 19-23 year old of Rafsanjan medical volunteer students, and there was no difference in age between males (21.5±1.3 years) and females (20.4±1 years). Mean value for weight and height for males, were 59.35±3.86 kg and 175±1.4 cm respectively, which significantly were greater than those were for females, 54.78±1.21 kg and 163.56±0.97 cm respectively. Students were asked to fill in a questionaire for the stressful examinations and specify which subject is more stressful. After reviewing the questionaires it was clarified that physiology examination is among the stressful subjects. Then for the control of stress condition and elimination the interfering factors, all of the individuals who had other stresses, except the examination i.e. psychological problems or took medicines were withdrawn from the study.

The effect of stress was investigated half an hour before final physiology examination in January 2003 (stress condition) and 45 days after examination (control condition, ordinary university work) in Feberuary at 10-12 a.m.

The body mass index (BMI), systolic and diastolic blood pressure (SBP and DBP respectively), heart rate (HR), respiratory freqeuncy, cortisol, estradiol, progesterone, testosterone and ACTH were measured at control and stress conditions. Blood pressure and heart rate were measured using sphygmomanometer and stethoscope. Blood samples were taken 30 minutes before examination and 45 days after examination at 10-12 a.m. The cortisol, ACTH, progesterone and testosterone of blood plasma were measured by radioimmunoassay (RIA). To determine the follicular or luteal phase, the females were asked how many days were passed from their last menstruationcyle and that was confirmed by the measurement of blood progesterone concentration.

All subjects had their informed written consent for participation in the study. The experimental design and the procedures followed were in accordance with the ethical standards laid down for human studies.

Data were analysed by analysis of variance (ANOVA) to compare the differences between groups, and to determine the differences between two groups the Tukey test were used. The data are presented as Mean±SEM and P<0.05 was considered as statistically significant.

RESULTS

The results of this study are summarized in tables 1 and 2 and Figures 1-5.

Table 1 summarises some demographic charactristics of the female and male students. Both weight and height of males are significantly more than females (P<0.001), but there was no significant difference in BMI between two sexes.

Table 2 shows the mean of systolic and diastolic blood pressure (SBP and DBP respectively), heart rate (HR) and respiratory frequency (RF) for male and female students in stress and control conditions. As data presents SBP is significantly higher in males than females (P<0.01). For males SBP is significantly greater in stress than control condition (P<0.05). DBP in stress condition increased in male compare to female significantly (P<0.01).

The HR for females in stress condition is significantly increased compare to control; this is the same for males. In control condition HR for females is significantly greater than males (P<0.001), therefore for males there is a higher increase of HR, than females in stress condition. RF for female was more than males in both control and stress conditions (P<0.05).

Table-1: Selected demographic charactristics of the female and male students in the physiology examination stress and control condition. Data reperesents mean±SEM.

Variable	**Conditions**			
	Control		**Stress**	
	Female	**Male**	**Female**	**Male**
Weight(kg)	54.78±1.21	***59.35±3.86	55.6±1.2	***66.68±1.8
Height (cm)	163.56±0.97	***175?1±1.4	164.32±0.97	***175.6±0.98
BMI(kg m^{-2})	20.4±0.4	21.52±0.46	20.56±0.45	21.84±0.57
Age(year)	21.4±1.5	22.5±1.75	21.7±1.4	22.9±1.65

***: Significant sex difference, P<0.001. n=28 for females and 21 for males.

Table-2: Mean and standard errors for systolic (SBP), diastolic blood pressure (DBP), Heart rate (HR), and respiratory frequency (RF) in female and male students under physiology examination stress and control conditions. Data repersents Mean±SEM

Variable	**Conditions**			
	Control		**Stress**	
	Female	**Male**	**Female**	**Male**
SBP (mmHg)	112.34±1.6	[b]116±2.7	110.8±2.25	[a] 124.76±4.01
DBP (mmHg)	67.96±1.27	74.2±1.9	68.22±2.03	[c] 76.56±2.48
HR (beat min^{-1})	77.09±1.04	70.2±1.64	[d] 84.9±1.25	84.6±2.63
RF (beat min^{-1})	18.48±0.28	16.88±0.21	[f] 20.81±0.52	17.12±0.28

a: Significant sex difference under stress condition, P<0.01, **b:** Significant difference in males under stress and control condition, P<0.05, **c:** Significant sex difference under stress condition, P<0.05, **d:** Significant difference in females under stress and control condition, P<0.001, **f:** Significant sex difference under stress condition, P<0.001, n=28 for females and 21 for males.

The salient results have been summarized in figures 1-5. Figure-1 shows plasma ACTH concentration in females (luteal and follicular phases) and males under control and stress conditions.

Figure-2 illustrates females' cortisol mean concentration in control and stress condition in both phases and the comparison of their serum cortisol with males. Figure-3 represents males' serum testosterone mean concentration in control and stress condition. Figure-4 shows the mean concentration of estradiol in females' follicullar and luteal phase, while the mean concentration of progesterone in females at both phases is shown in figure 5.

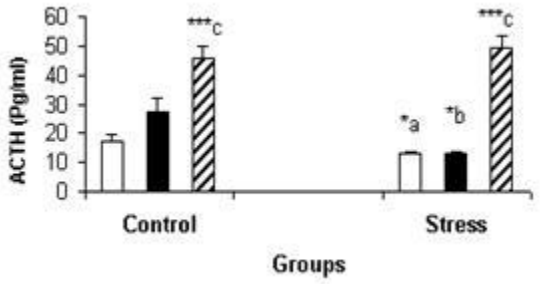

Figure-1: Mean ± SEM of ACTH plasma levels during stress and control conditions in males (▒), follicular (□) and luteal (■) phases. a: significant difference in follicular phase under stress compare to control. b: significant difference in luteal phase under stress compare to control. c: significant sex difference between males and females, in the stress and control conditions.*: P<0.05, ***: P<0.001.

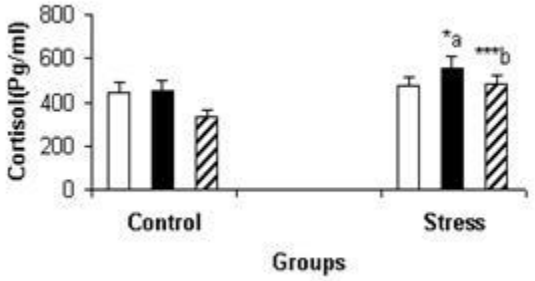

Figure-2: Comparison of the serum cortisol concentration in males (▒), and females [follicular (□) and luteal (■) phases] in the stress and control conditions. a: significant difference in luteal phase under stress and control conditions. b: significant difference for males in control and stress conditions. *: P<0.05, ***: P<0.001.

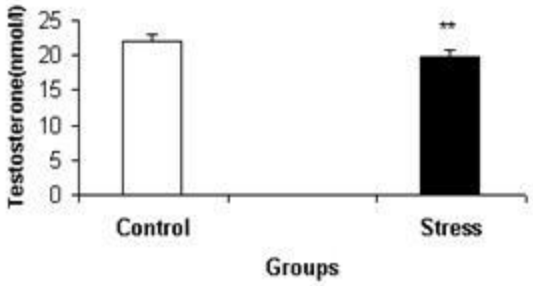

Figure-3: Comparison of serum testosterone in males under stress (■), and control condition (□). **:Significant difference between control and stress, P<0.01.

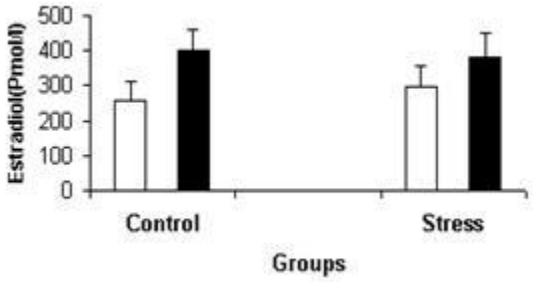

Figure-4: Mean ± SEM of serum estradiol of females' follicular (□), and luteal (■) phases in control and stress situation.

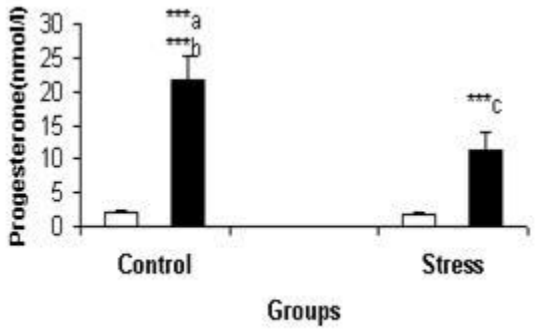

Figure-5: Serum progesterone comparison in females' follicular (□) and luteal (■) phases in control and stress conditions. a: significant difference between stress and control situations in luteal phase. b: significant difference between follicular and luteal phases in control condition. c: significant difference between follicular and luteal phases in stress condition. ***: P<0.001.

DISCUSSION

The results of this study showed that: there were no differences in BMI of male and female groups at control and stress conditions. Subsequent analysis of sex diferences showed that males had higher systolic and diastolic blood pressure

and increase in heart rate in stress condition, compare to those of females. Whereas, females had higher respiratory frequency in stress condition compare to that's of males. Moreover, there were no differences in SBP, DBP and HR responses to stress condition, during the menstrual cycle. Our finding in cardiovascular responses is in agreement with the studies of Matthews, that reported sex differences in SBP and DBP responses during stress in middle age individuals[6] and Tersman that found increasing in SBP[11], in this study, the cardiovascular responses to physical and mental stresses in both menstrual cycle phases has been examined in psychiatric female students. Zeller et al. have recently shown that during medical licensing examination, DBP was significantly icreased but the SBP did not change significantly.[12] The present report is shown that, the sex differences also exsist in young adult individuals, but is in oppose to the result of Bijlani.[13] It is supposed that the differences between our finding and Bijlani are due to the situation of experiments, because, the Bijlani experiments were done one week before examination, but our experiment was on the day of examination.

Heart rate increasing in this study is in agreement with the results of Stoney[5] and Matthews[6] that determined the cardiovascular responses to physical and psychological stresses, if so Collins[14] and Tersman[11] that reported heart rate in females is more than males during stress condition but is opposed to the report of Zeller et al.[12] It has been shown that subchronic physiological stress in human increased alpha2-adrenergic receptor densithy, which is related to stress-induced anxiety.[15] Examination stress cause a decrease in the parasympathetic influences on the heart rate.[16] Examination stress also changes the activity of sympathetic and parasympatheticnervous system.[17] Our finding in respiratory frequency (RF) suggests that increasing in RF along with increasing the activity of vagus nerve instead of accompanying with decrease of sympathic activity, it is also possible that females may be more sensitive to stimulation of vagus nerve than males. In the other part of this study it was shown that cardiovascular responses are independent from different phases of menstrual cysle both in control and stress conditions. This result is consistent to the Tersman study that has been reported there is no phase dependent difference in SBP and DBP or heart rate under mental stress.[11]

Our results showed that the plasma ACTH in male students is higher than females in both control and stress conditions. Also the level of cortisol in males increased under stress, whereas in females it was not changed. These results are in agreement with the results of other investigators.[4,11,19] The results of this study are also in areement with the finding of Martinek[18] who studied the effect of routine written examination on salivary cortisol and Johansson that has shown the hormonal changes in male and female medical students in response to examination stress.[20] Furthermore Komesaroff showed that only under stress condition the amount of cortisol in luteal phase is greater than follicular phase, whereas in control condition there is no significant difference, that is parallel with our finding,[21] our results is also simillar to Tersman finding that has measured the contisol concentration in different phases.[11]

In other part of study we measured the changes of sex hormones in response to examination stress. The results showed that stress induced a significant decrease in male serum testosterone compare to control, this result is consistent to previous study that reported stress inhibits testosterone secretion in males.[1,13] The possible mechanism of stress inhibitory effect on testosterone secretion is due to the effect of stress on the hypothalamus–pituitary–gonad (HPG) axis, because, gonadotropins secretion are correlated to the secretion of CRH, ?-endorphins, ACTH, and glucocorticoids. In females, serum progesterone response to stress reperesented a significant decrease in luteal phase. This suggests that the changes in female's sex hormone are a result of stress inhibitory effect on HPG axis in women. Variation in the concentration of progestrone under stress could be the results of alteration in opioids secretion, decrease in ACTH secretion under the stress is an indication of the inhibitory role of estrogen due to decreased progestrone during stress that affect ACTH secretion.[21]

Based on the above mentioned results we could conclude that there are differences in physiological responses to examination stress between males and females, so that males showed a greater increase in systolic and diastolic blood pressure and also cortisol, in response to stress. Therefore we suggest that when the examination is stressful, different effects of stress in male and female students should be considered, even in female students it is supposed that stress affect in luteal phase differently from follicular phase.

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