ORGAN AND BODY WEIGHTS CHANGES IN FEMALE WISTAR RATS EXPOSED TO DIFFERENT STRESSORS

Nwogueze B. C.¹, Ojieh A. E.¹, Aloamaka C. P.¹, Igweh J. C.¹ and Onyesom I.²

¹Department of Human Physiology, Faculty of Basic Medical Sciences, Delta State University, Abraka, Nigeria.

²Department of Medical Biochemistry, Faculty of Basic Medical Sciences, Delta State University, Abraka, Nigeria.

*Corresponding author. E-mail: bukasono123@gmail.com.

Accepted 21st May, 2020

Understanding the changes in the absolute organ and body weights serves as the most sensitive indicator of assessing reproductive success and fertility concern. The study examined changes in the organ and body weights of female Wistar rats exposed to different stressors. 168 healthy rats weighing between 150-200 g, aged between 12-14weeks were used and distributed into groups of six rats each. Stress models; restraint, mirrored and intruder paradigm tests were used. The rats were euthanized via cervical dislocation; their ovary, uterus, kidney, liver and brain were carefully isolated and weighed. Data were analyzed for Mean ± SEM, and p-level <0.05 was considered statistically significant. Results obtained revealed that the weights of kidney, liver and brain organs of the rats exposed to restraint and intruder stressors were the most affected when compared with control. Findings from the study showed that body weights of the animals were significantly (p<0.05) reduced irrespective of the stressors applied. Exposure of the rats to different periods of 1, 3 and 5 h per day for 1 and 2 weeks caused apparent significant (p<0.05) reduction in body weights in the rats exposed to the different stressors when compared with the control. Similarly, body weights returned to almost the control levels at 3rd week especially in rats exposed to restraint and mirrored stressors while rats exposed to intruder stressor experienced continuous significant (p<0.05) decrease in their body weights. In conclusion, stress-induced effect on organ and body weights is variable and the efficacy in causing morphological changes is dependent on the stressor utilized.

Keywords: Stress, restraint, mirrored, intruder, weights and Wistar rats.

INTRODUCTION

Stress is a non-specific result of demand on the human body in response to stressful events within the environment designed to interfere with the set equilibrium. Stress, whether acute, episodic or chronic has been hypothesized to be a threat to metabolic and physiological responses, altering the homeostatic balance of living organisms (Dallman et al., 2003). Nayanatara et al. (2005) referred to this process as positive adaptive trend of stress; however, they are suggested series of behavioural, subjective, cognitive, psychological, cellular, immunological and neurochemical changes caused by stress. Exposure to stress alters the mechanism regulating hypothalamo-pituitary-

adrenal and the sympathetic adrenal medullary pathways, and this is dependent on the nature of the stress in question.

The severity of the stress model determines the nature of the stress and its manifestation differs significantly. In response to short term or long term effect of stress, a cascade of activity takes place in humans in the form of experience of stimulus to maintain homeostasis. Epel et al. (2001) opined that stress induction plays a significant role in altering caloric balance in food consumed, especially in emotional condition, as such, could have a synergy with hyperphagia, anorexia and reduced body weight. Such adjustment in humans, according to Turrens (2003), suggests excessive formation/imbalance

of oxidants in the presence of antioxidant protective mechanism.

Risk associated with stress has been identified by Amara and Aljunid (2014) to include cardiovascular dysfunctions, metabolic syndrome, traumatized conditions, possible psychological implications and changes in nutritional habits. At the cellular levels, stress exerts major inflammatory and oxidative response, thereby, affecting the physiologic status of the tissue. Evanthia et al. (2017) maintained that, such series of unfavorable metabolic differentiations is felt by the skeletal muscles, liver, pancreas and adipose tissue manifestation sub-clinical inflammatory dysfunction of endothelial and insulin response system and deregulation of mitochondria.

Stress induction is a clear evidence of divergent violation of cellular balance for determining organ or body weights in different settings in a given organism. Change in trends in body and organ weights following exposure to different natures of stress in experimental animals serves as a pointer of morphological and biological outcome. Yang et al. (2013) inferred that understanding the relationship between absolute organ and body weights is vital for improving weight managements. Various conditions have been documented to determine the onset of organ or/and body weight changes in animals such environment, age, strain, sex and experimental factors. Since stress induces changes at the cellular levels, it is most likely that stress may result in changes in the weights of organs and body weights; hence, this study examined the effects of stress of different stressors on tissues and body weights of female Wistar rats.

MATERIALS AND METHODS

Procurement and handling of animals

One hundred and sixty eight female Wistar rats weighing between 150-200 g, obtained from Emma Maria Laboratory, Abraka, Delta State, Nigeria, were used in the present study. The animals were randomly assigned to groups and housed in plastic cages with adequate ventilation based on the research design. All the female rats were allowed unrestricted

access to standard rat chaws and water and they were acclimatized for a period of 2 weeks. The normal 12 h light and dark cycle with temperature of 22-24°C and relative humidity of 70 -77% were maintained.

Ethical clearance

Ethical approval was sought and obtained from the Ethical Committee of the Faculty of Basic Medical Sciences, Delta State University, Abraka. All the recommended regulations and experimental research guideline relating to animal handling and well-being were strictly adhered to.

Stress procedure

Three different stress models were adopted for this study as follows:

Restraint chamber test

Restraint stressor was used to induce physical stress in the rats by limiting their movement. The female Wistar rats were introduced into cylindrical plastic tubes measuring 5.7cm × 20cm adopting the methods of Ely et al. (1997). Both ends of the restrainer were covered with wire mesh to prevent the animals from escaping and allowing free flow of air, as well as back and forward movement.

Mirrored chamber test

To induce anxiety in the female Wistar rats following the methods of Paterson et al. (2010), the Mirrored chamber test was designed with wood, with mirrors placed internally on three walls measuring $42 \times 42 \times 42$ cm³ and the 4th wall was coloured opaque black in each of the box compartment.

Intruder paradigm test

The resident Paradigm intruder test apparatus was designed in the form of rat cage that consisted of six (6) enclosed rectangular partitions made of woods measuring $25 \times 13 \times 32$ cm and each of the walls was separated by a transparent wire mesh. The test apparatus was used to induce psychosocial stress in the female Wistar rats adopting the technique of McGregor et al. (2002). An aggressive male cat was used to induce emotional stress and elicit defensive reaction.

Experimental design

The animals were randomized into twenty

eight (28) experimental groups of six rats each. The rats were exposed to different stress models as described in Table 1.

Body and organ weights

The animals were stressed for a duration based on the protocol of the study. The weights of the

Table 1. Grouping of animals.

Category	Experimental protocol	Duration (Hours/Weeks)		
Α	Control (Non-stressed Wistar rats)	Hours	Hours	Hours
B ^{1,2,3}	Exposure of Wistar rats to Restraint Stressor for 1 week	1	2	3
$C^{1,2,3}$	Exposure of Wistar rats to Restraint Stressor for 2 weeks	1	2	3
$D^{1,2,3}$	Exposure of Wistar rats to Restraint Stressor for 3 weeks 1		2	3
E ^{1,2,3}	Exposure of Wistar rats to Mirrored Stressor for 1 week	1	2	3
F ^{1,2,3}	Exposure of Wistar rats to Mirrored Stressor for 2weeks	1	2	3
$G^{1,2,3}$	Exposure of Wistar rats to Mirrored Stressor for 3 weeks	1	2	3
$H^{1,2,3}$	Exposure of Wistar rats to Intruder Stressor for 1 week	1	2	3
I ^{1,2,3}	Exposure of Wistar rats to Intruder Stressor for 2 weeks	1	2	3
$J^{1,2,3}$	Exposure of Wistar rats to Intruder Stressor for 3 weeks	1	2	3

Indications: 1,2,3 are sub-groups of stressed rats at the rate of 1hours, 3hours and 5hours per day hr= hour; hrs=hours

experimental animals were determined using an electronic weighing balance (G&G 301, England) before and after stress induction. At the end of the experimental period for the different rats, the female Wistar rats were euthanized through cervical dislocation to prevent possible effect of anesthetic agents on samples collected. Subsequently, the kidney, liver, ovary, uterus, and brain organs of each of the rats were isolated and weighed; meanwhile all adhering adipose tissues in the isolated organs were carefully removed.

Statistical analysis

Data were statistically analyzed using IBM

SPSS tools (version 22). The quantitative data were presented as Mean±SEM; while comparison of mean difference across groups was done using one-way ANOVA followed by LSD post-hoc. Differences with p-value <0.05 were considered to have significant difference.

RESULTS

Changes in absolute body weights of Wistar rats after exposure to stress

The body weights of rats exposed to different stressor at 1, 3 and 5 h for 1, 2 and 3 weeks, respectively are illustrated in Tables 2, 3 and 4.

Table 2. Body Weight changes of rats exposed to different nature of stress for 1 week.

Darameter	Absolute boo	% Change in body		
Parameter	Initial	Final	weight	
Control	96.47±3.44	110.42±4.72	12.27	
Restraint_1 h	149.93±3.29 ^d	165.63±4.15 ^d	9.30	
Mirrored_1 h	148.70±9.86 ^d	151.95±9.24 ^d	2.22	
Intruder_1 h	122.8±10.21 ^{abd}	128.23±12.92 ^a	3.14	
Restraint_3 h	174.82±4.31 ^d	172.48±10.27 ^d	-2.88	
Mirrored_3 h	133.28±7.77 ^{ad}	136.05±9.13 ^a	1.44	
Intruder_3 h	140.60±17.18 ^{ad}	150.77±16.13 ^d	7.24	
Restraint_5 h	167.00±3.80 ^d	166.62±3.97 ^d	-0.28	
Mirrored_5 h	119.37±6.71 ^a	120.00±6.21 ^a	0.59	
Intruder_5 h	133.00±4.99 ^{ad}	126.77±4.87 ^a	-4.95	

Values obtained are expressed as mean ± S.E.M, (n=6), Superscripts designated with alphabets are described as follows: a = significant when compared to Restraint stressor, b = significant when compared to Mirrored stressor, c = significant when compared to Intruder stressor and, d = significant when compared to control group. P-value ≤0.05 level of significance

Table 3. Body Weight changes of rats exposed to different nature of stress for 2 weeks.

Dovementor	Absolute boo	0/ 0		
Parameter	Initial	Final	% Change in body weigh	
Control	96.47±3.44	110.42±4.72	12.27	
Restraint_1 h	154.60±16.20 ^d	152.12±6.11 ^d	-4.36	
Mirrored_1 h	140.43±7.46 ^d	147.35±4.92 ^d	4.96	
Intruder_1 h	117.08±6.36 ^a	119.97±5.46 ^a	1.69	
Restraint_3 h	155.32±16.48 ^d	160.22±16.19 ^d	3.33	
Mirrored_3 h	140.78±15.16 ^d	141.98±16.53 ^d	0.33	
Intruder_3 h	127.05±5.16 ^a	131.23±5.06	3.23	
Restraint_5 h	176.25±6.13 ^d	176.85±7.15 ^d	-0.14	
Mirrored_5 h	147.22±7.11 ^d	144.07±7.98 ^{ad}	-2.84	
Intruder_5 h	153.53±13.20 ^d	159.28±18.03 ^d	1.88	

Values obtained are expressed as mean \pm S.E.M, (n=6), Superscripts designated with alphabets are described as follows: a = significant when compared to Restraint stressor, b = significant when compared to Mirrored stressor, c = significant when compared to Intruder stressor and, d = significant when compared to control group. P-value \leq 0.05 level of significance.

Table 4. Body weight changes of rats exposed to different nature of stress for 3 weeks.

D	Absolute bod	0/ 04		
Parameter	Initial	Final	% Change in body weigh	
Control	96.47±3.44	110.42±4.72	12.27	
Restraint_1 h	130.98±10.97 ^d	147.33±15.47 ^d	9.15	
Mirrored_1 h	116.92±14.82	131.43±12.61	11.84	
Intruder_1 h	105.70±3.82	122.63±3.64	13.40	
Restraint_3 h	132.83±7.62 ^d	147.40±6.55 ^d	8.93	
Mirrored_3 h	137.90±10.93 ^{cd}	146.80±9.01 ^d	6.55	
Intruder_3 h	109.33±5.89	129.22±5.37	15.16	
Restraint_5 h	106.62±10.65	117.37±9.93	9.57	
Mirrored_5 h	118.95±5.62	142.40±4.99 ^d	16.46	
Intruder_5 h	126.83±10.56 ^d	127.52±11.31	-2.3	

Values obtained are expressed as mean \pm S.E.M, (n=6), Superscripts designated with alphabets are described as follows: a = significant when compared to Restraint stressor, b = significant when compared to Mirrored stressor, c = significant when compared to Intruder stressor and, d = significant when compared to control group. P-value \leq 0.05 level of significance

Table 2 shows the comparative effect of different stressors on body weights of the rats after 1 week of treatment. Result showed a percentage decrease in body weight in all experimental groups when compared with control. The observed percentage change was severe towards a negative loss in body weight following exposure to restraint stressor at 3 and 5 h respectively when compared with control. Similarly, rats exposed to intruder stressor at 5 h per day revealed severe loss in weights. Comparison of means with One-way ANOVA statistics revealed that there was significant (p<0.003) difference in the percentage change in means of body weight of rats exposed to the three nature of stressors at 1, 3 and 5 h for 1 week when compared to control weights. This implies stress of different that nature

significantly altered the body weights of rats irrespective of the stressor applied.

Table 3 represents the comparative effect of different stressors on body weights of the rats after two weeks. Result showed percentage change in the body weight in all experimental groups when compared with control. The observed percentage loss in body weights were severe in rats exposed to restraint stressor at 1 and 5 h durations for 2 weeks. Also, the body weights were significantly reduced towards the negative in rats exposed to Wistar rats at 5 h when compared with the control weights. One-way ANOVA statistics revealed that there was no significant (p>0.586) difference in the percentage change in means of body weight of rats exposed to the three nature of stressors at 1, 3 and 5 h for 2 weeks when compared to control weights. This

implies that stress of different nature mildly altered the body weights of the rats, although, not significantly.

Table 4 illustrates the comparative effect of different stressors on body weights of the rats after three weeks. Results revealed that there was recovery of body weight in rats exposed to intruder stressor at 1 and 3 h respectively when compared with control. Comparison of means revealed the observed increase in the body weights of rats exposed to mirrored stress, while the progressive decrease in rats exposed to intruder stress for 5hours was statistically significant when compared across group. Although, one-way ANOVA statistics revealed that there was no significant (p>0.379) difference in the percentage change in means of body weight of rats exposed to the three nature of stressors at 1, 3 and 5 h for 2weeks when compared to control weights. This implies that adaption was observed in the body weights of rats exposed to stress of different

nature within 3 weeks.

Changes in relative organ weights of Wistar rats after exposure to stress

The changes in organ weights of rats exposed to different stressor at 1, 3 and 5 h for 1, 2 and 3 weeks are represented in Tables 5, 6 and 7, respectively. The weights of liver and brain decreased in rats exposed to restraint stress for 1 h within 1 week; subsequently, the kidney, liver and brain tissues of the rats decreased after 1 h exposure to Mirrored stress within 1week as shown in Table 5. A similar trend was observed in the tissues of kidney, liver and ovary tissues of rats exposed to 1 h Intruder stressor within 1 when compared to control Comparison of means using one-way ANOVA for the 1 h exposure to the three different stressors revealed that there was a significant difference (p<0.05) in the liver and uteri tissues. However, the kidney and brain tissues showed no significant difference (p>0.05).

Table 5. Organ weight of female Wistar rats exposed to stress for 1 week.

Parameter	Relative organ weight (g)				
Parameter	Kidney	Liver	Ovary	Uterus	Brain
Control	0.853±0.050	4.641±0.230	0.060±0.007	0.051±0.002	1.853±0.090
Restraint_1 h	0.442±0.090	2.930±0.100 ^{cd}	0.101±0.007 ^d	0.186±0.031 ^{cd}	1.014±0.040 ^d
Mirrored_1 h	0.526±0.020	3.108±0.110 ^{cd}	0.072±0.007 ^a	0.179±0.009 ^{cd}	1.041±0.040 ^d
Intruder_1 h	0.613±0.030	3.652±0.220 ^d	0.048±0.007 ^{ac}	0.060±0.008	1.160±0.100 ^d
Restraint_3 h	0.583±0.040 ^d	2.821±0.190 ^d	0.132±0.013 ^d	0.189±0.019 ^d	0.995 ± 0.070^{d}
Mirrored_3 h	0.568±0.019 ^d	3.312±0.070 ^{ad}	0.139±0.043 ^d	0.248±0.040 ^d	1.165±0.070 ^d
Intruder_3 h	0.563±0.020 ^d	3.433±0.070 ^{ad}	0.086±0.013	0.042±0.007	1.105±0.070 ^d
Restraint_5 h	0.579±0.010 ^d	3.020±0.160 ^d	0.109±0.012 ^d	0.221±0.017 ^d	1.003±0.040 ^d
Mirrored_5 h	0.565±0.020 ^d	3.129±0.070 ^d	0.071±0.007 ^a	0.145±0.060 ^d	1.227±0.060 ^{ad}
Intruder_5 h	0.576±0.030 ^d	3.473±0.130 ^d	0.060±0.015 ^a	0.052±0.006 ^a	1.206±0.040 ^{ad}

Values obtained are expressed as mean \pm S.E.M, (n=6), Superscripts designated with alphabets are described as follows: a = significant when compared to Restraint stressor, b = significant when compared to Mirrored stressor, c = significant when compared to Intruder stressor and, d = significant when compared to control group. p-value \leq 0.05 level of significance.

The organ weights of kidney, liver, and brain tissues of rats exposed to restraint and mirrored stressors for 3 h within 1week were significantly decreased. A marked decrease was equally observed in the kidney, liver, uterus and brain weights of rats exposed to Intruder paradigm stress within 1 week. Comparison of means with one-way analysis of variance shows a significant difference (p<0.05) in kidney, liver, uteri, and brain tissue

weights. However, the ovary tissue was not significant (p>0.05). The change in organ weights in rats exposed to different nature of stress for 5hours within 1week revealed that the kidney, liver and brain weights of the rats were markedly decreased when compared to control group. Meanwhile, comparison of mean using one-way ANOVA showed that there was a significant difference (p<0.05) in the relative weights of kidney, liver, ovary, uteri and brain tissues.

Table 6. Organ weight of female Wistar rats exposed to stress for 2 weeks.

Doromotor	Relative organ weight (g)				
Parameter	Kidney	Liver	Ovary	Uterus	Brain
Control	0.853±0.05	4.641±0.24	0.060±0.007	0.051±0.002	1.853±0.09
Restraint_1 h	0.586±0.03 ^{bd}	3.099±0.24 ^{bd}	0.082±0.023	0.068±0.013	1.514±0.29
Mirrored_1 h	0.720±0.05 ^{ad}	3.886±0.20 ^d	0.070±0.006	0.030±0.004 ^a	1.361±0.09 ^d
Intruder_1 h	0.462±0.03 ^{acd}	2.939±0.29 ^{bd}	0.073±0.007	0.038±0.005 ^a	1.050±0.01 ^d
Restraint_3 h	0.607±0.04 ^d	3.516±0.36 ^d	0.061±0.015	0.050±0.003	1.135±0.12 ^d
Mirrored_3 h	0.515±0.09 ^d	3.384±0.12 ^d	0.068±0.010	0.053±0.009	1.258±0.12 ^d
Intruder_3 h	0.573±0.02 ^d	3.088±0.11 ^d	0.066±0.011	0.031±0.001 ^{bd}	1.247±0.06 ^d
Restraint_5 h	0.549 ± 0.02^{d}	2.798±0.09 ^d	0.069±0.011	0.051±0.005 ^b	0.951±0.07 ^d
Mirrored_5 h	0.577±0.02 ^d	3.203±0.19 ^d	0.075±0.013	0.035±0.004 ^d	1.139±0.04 ^d
Intruder_5 h	0.554±0.01 ^d	3.045±0.12 ^d	0.049±0.007	0.023±0.003 ^{abd}	1.046±0.08 ^d

Values obtained are expressed as mean \pm S.E.M, (n=6), Superscripts designated with alphabets are described as follows: a = significant when compared to Restraint stressor, b = significant when compared to Mirrored stressor, c = significant when compared to Intruder stressor and, d = significant when compared to control group. p-value \leq 0.05 level of significance

Table 7. Organ weight of female Wistar rats exposed to stress for 3 weeks.

Donomoton	Relative organ weight (g)					
Parameter	Kidney	Liver	Ovary	Uterus	Brain	
Control	0.853±0.05	2.967±0.23	0.060±0.007	0.051±0.002	1.853±0.09	
Restraint_1 h	0.594 ± 0.07^{d}	3.140±0.41 ^d	0.036±0.007	0.036 ± 0.004^{bd}	1.09±0.11 ^d	
Mirrored_1 h	0.677 ± 0.05^{d}	3.141±0.14 ^d	0.076±0.013 ^a	0.060±0.004	1.292±0.11 ^d	
Intruder_1 h	0.573±0.01 ^d	3.047±0.08 ^d	0.059±0.008	0.043±0.004 ^b	1.219±0.03 ^d	
Restraint_3 h	0.581 ± 0.04^{d}	3.381±0.14 ^d	0.031 ± 0.003^{bd}	0.048±0.006	1.087±0.08 ^d	
Mirrored_3 h	0.662 ± 0.02^{d}	3.550±0.53 ^d	0.09 ± 0.006^{d}	0.059±0.007	1.250±0.07 ^d	
Intruder_3 h	0.708±0.02 ^{ad}	4.207±0.07 ^{ad}	0.062±0.006 ^{ab}	0.042±0.003 ^b	1.457±0.09 ^{ad}	
Restraint_5 h	0.808±0.02	4.249±0.05	0.064±0.015	0.063 ± 00.8^{c}	1.540±0.14 ^d	
Mirrored_5 h	0.746±0.02	3.994±0.25	0.066±0.009	0.063±0.004 ^c	1.377±0.06 ^d	
Intruder_5 h	0.773±0.02	2.967±0.13	0.073±0.011	0.048±0.003	1.621±0.09	

Values obtained are expressed as mean \pm S.E.M, (n=6), Superscripts designated with alphabets are described as follows: a = significant when compared to Restraint stressor, b = significant when compared to Mirrored stressor, c = significant when compared to Intruder stressor and, d = significant when compared to control group. p-value \leq 0.05 level of significance.

The organ weight of kidney, liver and brain tissues exposed to restraint stress for 1 h within 2 weeks revealed significant decrease, whereas there was reduction in the organ weights of kidney, liver, uterus and brain of rats exposed to mirrored stress for 1hour as well as intruder stress respectively within 2 weeks when compared to control group (Table 6). Comparison of means revealed that there was significant interaction (p<0.05) between the organ weights of kidney, liver, uteri and brain tissues, however, there was no significant difference (p>0.05) in the ovary tissues across the different stress models utilized.

The organ weight of rats exposed to restraint and intruder stress showed significant decrease in the kidney, liver uterus and brain tissues. Meanwhile, the rats exposed to mirrored stress with similar stress duration of 3hour revealed a reduction in the kidney, liver and brain organ weights when compared to control group. Comparison of means showed that there was significant interaction (p<0.05) between the organ weights of kidney, liver, and brain tissues. However, this was not same for ovary and uteri organ weights which showed no significant difference (p>0.05). The organ weights of kidney, liver, ovary, uterus and brain were significantly affected by intruder stress in experimental rats when compared to control group. Comparison of means across all the stressors revealed that there were significant interactions (p<0.05) in the organ weights of kidney, liver, uteri and brain tissues except for the ovary weight.

The organ-weights of rats exposed to Restraint and Intruder stressors for 1hour within 3weeks

revealed a continuous decrease in the organ tissues of kidney, liver, ovary, uterus and brain. Meanwhile, the rats exposed to mirrored stress showed significant decrease only in kidney, liver, and brain tissues when compared to control group. Comparison of means of different nature of stress model for organ weights showed significant difference (p<0.05) in the organ weights of kidney, liver, uteri and brain tissues except for the ovary tissue. The organ weights of kidney, liver, ovary, uterus and brain exposed to restraint and intruder for 3hours were significantly stressors decreased as was the case with rats exposed to 1hour stress within 3weeks. However, the rats exposed to mirrored stress for 3hours within 3weeks revealed reduction in the organ weights of kidney, liver, and brain weights when compared to control group. Comparison of means across the different stress model showed significant interactions (p<0.05) between the organ weights of kidney, liver, ovary, and brain tissues except for the uteri. The organ weights of kidney, liver and brain exposed to restraint and mirrored stressors for 5hours within 3weeks were markedly decreased respectively. Meanwhile, the organ weights of rats exposed to intruder stress for 5hours within 3weeks revealed a gross decrease in the weights of kidney, liver, uterus and brain except for the uterus when compared with the control group. Comparison of means with One-way ANOVA revealed that there was no significant difference (p>0.05) between the organ weights of kidney, liver, ovary and uterus tissues respectively except for the brain weight.

DISCUSSION

The relationship between organ and body weight changes when subjected to stress was examined in this study. Stress may be beneficial or harmful and this can be ascertained by its outcome on the body. It is beneficial if it is able to improve on the survival capacity of the body; on the other hand, it is harmful if it leads to degradation or a pathological state in the body (Habib *et al.*, 2017). Restraint, mirrored and intruder paradigm stressors are important stress models used to measure the response of an organism in

stressful situation. The physical restraint is considered as physical stressor (Chagra et al. 2013). Also, Glavin et al. (1994) believe that the restraint stress model is the most commonly employed for studying depression and anorexia, social dominance and subordination in animals as it mimics potent physical and psychological stress. Mirrored chamber is a putative test designed to induce anxiety-like behaviour and precipitate anxiogenic response experimental animals depending on the time spent. The intruder paradigm is often a stress model associated with emotional and social defeat as well as emergent of depressive-like behaviour due to high levels of aggressiveness in the rodent (Carnevali and Sgoifo, 2014).

In the present study, Wistar rats were exposed to restraint stressor, mirrored chamber stressor and resident intruder stressor and it was observed that the responses of the rats to the different stressors varied with respect to alteration in the organ and body weights, suggesting that responses of rats to exposure to stressors depends, at least to some degree, on the extent of stress induced in the respective rats. Results obtained from this study revealed that body weight changes (%) for initial and final weights here significantly decreased during exposure to different stress models of 1, 3 and 5 h for 1 and 2 weeks respectively. In support of the observations made in the experiments concerning the effect of stress on body weight changes in rats are the reports by Berton et al. (1998) who found that exposure of rats to intruders decreased body weight, and food intake, thereby increasing anxiety-like behaviour and hyper locomotion in Lewis rats; while Razzoli et al. (2009) reported body weight loss and anxiety and depressive behaviours in Wistar rats. Similarly, the study by Anil et al. (2014) revealed that exposure of mice to mirrored chamber test imposed locomotion activity, body weight loss, and caused anxiety following oxidative stress.

The results revealed that there was decrease in the body weights of rats exposed to intruder stressor at 5h for 3weeks. This probably explains why Keeney et al. (2006) posited that when compared to other types of stressors, the resident intruder paradigm model elicits the most robust physiological response due to their aggressive dominant behaviour. Such loss in body weights as observed supports the assertion of Bhatnagar et al. (2006) who found that chronic stress has been related to alterations in body weight and physiology response in different organs of experimental animals. As such, following exposure to this stressor, rats have been shown to show decrease in their food intake and body weights as reported by Wang et al. (2012) which is a clear indication of the potent effect of the stressor.

The study revealed that there was a gradual recovery in the body weights of the experimental animals exposed to restraint and mirrored stressor for 3weeks except for the animals exposed to the intruder paradigm stressor. According to Herman et al. (2013) based on time, type and severity of applied stimulus, stress exerts actions on the body, thereby, altering its homeostasis. Exposure of an organism to threats within the environment in the form of stressors initiates a cascade of events through the action of HPA-axis to restore homeostasis. These observed changes in this study according to Nagaraja and Jeganathan (1999) could be due to adaptation or habituation of the continued stress. It is likely that stress modifies the pathways that normally sense and respond to a reduction in weight thereby opposing the mechanism which promotes weight recovery (Harris et al. 2006).

The different internal organs of animal perform varying functions and this is so because apart from structural differences, the chemical (enzymatic and non-enzymatic) constituents of the different organs vary, thereby giving rise to different chemical reactions in the cells of the different organs. It is therefore not surprising that the internal organs from non-pregnant female Wistar rats examined responded differently to exposure to different nature of stressors at 1, 3 and 5hours for 1 and 2weeks respectively. For instance, results from this study revealed that stressful situations induced organ weight loss especially that of kidney, liver and brain organs irrespective of the stressors used. Findings from this study were similar with previous studies. Liu et al., (2014) noted that restraint stress reduces the organ weights, though, that of the uterus and ovary as well as or promotes

reduction in body weights.

In the present study, while the weights of the ovary and uterus were not significantly altered by exposure of the Wistar rats to restraint, mirrored and intruder stressors at 1, 3 and 5hours, the weights of the kidney, liver and brain were significantly (p<0.05) decreased. Also, the weights of the liver were significantly (p<0.05) increased when exposed for 3 weeks. Bali et al. (2014) maintain that such decrease in organ weights following stress-induced depression could be attributed to differential changes in miRNA expression in a brain-specific manner. This is why Cao et al. (2013) identified the brain region as hippocampus in rats responsible for stress-signaling pathways. In contrast to our study, exposure to stress model using forced swimming test increased various organ weights of liver, kidney and brain as reported by Najanatara et al. (2005); although, the stressor applied was different from that used in this study.

The marked decrease in organ and body weights of female Wistar rats exposed to restraint, mirrored and intruder paradigm stressors at 1, 3 and 5 h for 1 and 2 weeks as observed in our study may be due to the reduction in food intake and stress activation of HPA axis which is subsequently, maintained by increase in the level of energy expenditure and elevated body temperature as suggested by Bhatnagar et al. (2006). Based on available data from this study, it is not possible to provide precise mechanisms for variable responses of internal following exposure of rats to a particular stressor. However, Korner et al. (2001) asserted that the central regulation of organ-body weights food intake, growth and hormone secretion occurs in the hypothalamus via regulation of energy homeostasis, specifically functional balance orexigenic (neuropeptide Y) between anorexigenic (proppionclano) hypothalamic well as amphetamine-regulated peptides; as transcript.

Conclusion

This study has revealed that the organ weights of kidney, liver and brain of the female Wistar rats exposed to the different stressors were significantly affected. The body weights of the animals were equally significantly affected irrespective of the stressor utilized. Exposure of the rats to restraint and mirrored stressors up to the 3rd week caused restoration of body weights while the intruder stressor caused continuous decrease in body Therefore, stress-induced effect on organ and body weights is variable on the nature of stressor and the observed alterations is dependent on the intensity of the stressor applied; thus, this can play significant role in the determination of reproductive success in females.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENTS

The authors are grateful to Emma Maria Diagnostic and Research Laboratory for their technical support and assistance with the experiments and valuable guidance.

REFERENCES

- Amara, A.H., and Aljunid, S.M. (2014). Non-communicable diseases among urban refugees and asylum-seekers in developing countries: A neglected healthcare need. *Global Health*, 10: 24.
- Anil, K., Gurleen, K. and Puneet, R. (2014).

 Buspirone along with melatonin alternates oxidative damage and anxiety-like behaviour in a mouse model of immunization stress: *Chinese Journal of Natural Medicine*, 12(8): 582-589.
- Bahi, A., Chandrasekar, V. and Dreyer, J.L. (2014). Selective lentiviral-mediated suppression of microRNA/24a in the hippocampus evokes anti-depressants-like effects in rats. *Psychoneuroendocrinology*, 46: 78-87.
- Berton, O., Agucore, S., Sarrieau, A., Mormede, P. and Chaouloff, F. (1998). Differential effects on social stress in central serotonergic activity

- and emotional reactivity in Lewis and spontaneously hypertensive rats. *Neuroscience*, 82:147-159.
- Bhatnagar, S., Vining, C., Iyer, V. and Kinni, V. (2006). Changes in hypothalamic-pituitary adrenal function, body temperature, body weight and food intake with repeated social stress exposure of rats. *Journal of Neuroendocrinology*, 18:13-24.
- Cao, M., Chen, D., Zhang, C. and Wu, Z. (2013). Screening of specific microRNA in hippocampus of depression model rats and intervention effect of Chaihu Shugan San Zhongguo, *Zhongguo Zhong Yao Za Zhi*, 38(10): 1585-1589.
- Carnevali, L. and Sgoifo, A. (2014). Vagal modulation of resting heart in rats: The role of stress, psychological factors, and physical exercise. *Frontiers in Psychology*, 5:118.
- Chagra, S.L., Zavala, J.K., Hall, M.V. and Gosselink, K.L. (2013). Acute and repeated restraint differentially activate orexigenic pathway in rats hypothalamus. *Regulatory Peptides*, 167:70-78.
- Dallman, M.F., la Fleur, S.E., Pecoraro, N.C., Gomez, F., Houshyar, H. and Akana, S.F. (2004). Minireview: glucocorticoidsfood intake, abdominal obesity, and wealthy nations in 2004. *Endocrinology*, 145:2633-2638.
- Ely, D.R., Dapper, V., Marasca, J., Correa, J.B., Gamaro, G.D., Xavier, M.H., Michalowski, M.B., Catelli, D., Rosat, R., Ferreira, M.B.C., Dalmaz, C., 1997. Effect of restraint stress on feeding behavior of rats. *Physiology & Behavior*. 61, 395-398.
- Epel, E., Lapidus, R., McEwen, B. and Brownell, K. (2001). Stress may add bite to appetite in women: a laboratory study of stress-induced cortisol and eating behaviour. *Psychoneuroendocrinology*, 26:37-49.
- Evanthia, D.R., Olya, P., Eleni, A.K. and Georgia, K. (2017). Nutrition as a mediator of oxidative stress in metabolic and reproductive disorders in women.

European Journal of Endocrinology, 176:79-99.

- Glavin, G.B., Pere, W.P., Sandbak, T., Bakke, H.K. and Murison, R. (1994).

 Restraint Stress in Biomedical Research: An Update. *Neuroscience & Biobehavioral Reviews* 18:223-249.
- Habib, Y., Yunes, P., Hadriyat, S., Thomas, P.J. and Amirhossein, S. (2017). The impact of stress on body function: A review. *EXCLI Journal*, 16: 1057-1072.
- Harris, R.B., Palmondon, J., Lesh, S., Flatt, W.P., and Richard, D. (2006). Chronic disruption of body weight but not stress peptides or receptors in rats exposed to repeated restraint stress. *Hormones and Behavior*. 49:615-625.
- Herman, T.P., Figueiredo, H., Mueller, N.K., Ulrichlai, Y.M. Ostrander, M.M. Choi, D.C. and Cullinan, W.E. (2013). Central mechanism of stress integration hierarchical circuitry controlling hypothalamo-puituitary-adrenocortical responsiveness. *Frontiers in Neuroendocrinology*. 24:151-180.
- Keeney, A., Jessop, D.S., Harbuz, M.S., Marsden, C.A., Hogg, S. and Blackburn-Munro, R.E. (2006).Differential effects of acute and chronic social defeat stress on hypothalamic pituitary adrenal axis function and hippocampal serotonin release in mice. Journal of Neuroendocrinology, 18:330-338.
- Korner, J., Sarontaus, E., Chua, S.C., Leibel, R.L. and Wardlaw, S.L. (2001). Leptin regulation of Agrp and Npy MRNA in the rats hypothalamus. *Journal of Neuroendocrinology*. 13: 959-966.
- Liu, G.H., Dong, Y.L., Wang, Z.X., Cao, J. and Chen, Y.X. (2014). Restraint stress alters immune parameters and induces oxidative stress in the mouse uterus during embryo implantation stress. *The International Journal of Stress*, 17; 494-503.
- McGregor IS, Schrama L, Ambermoon P, Dielenberg RA. (2002). Not all 'predator odours' are equal: cat odour but not 2,4,5 trimethylthiazoline (TMT;

- fox odour) elicits specific defensive behaviours in rats. *Behavioural Brain Research*, 129:1–16.
- Nagaraja, H. and Jeganathan, P. (1999). Forced swimming stress-induced changes in the physiological and biochemical parameters in Albino Wistar rats. *Indian Journal of Physiology and Pharmacology*, 43(1): 53-59.
- Nayanatara, A.K., Nagaraja, H.S. and Anupama, B.K. (2005). The effect of repeated swimming stress on organ weights and lipid peroxidation in rats. *Thai Journal of Physiological Sciences*, 18:3-9.
- Paterson, N.E., Iwunze, M., Davis, S.F., and Malekiani, S.A., & Hanania, T. (2010). Comparison of the predictive validity of the Mirrored Chamber and Elevated Plus Maze Test in Mice. *Journal of Neuroscience Methods*, 188 (1): 62-70.
- Razzoli, M., Carboni, L. and Arban, R. (2009).

 Alterations of behaviour and endocrinological activity induced by 3 brief defeats in rats: relevance of human psychopathology.
 - *Psychoneuroendocrinology*, 34: 1405-1416.
- **Turrens, J.F (2003)**. Mitochondrial formation of reactive oxygen species; *The Journal of Physiology*, 552(2): 335–344.
- Wang, S.X., Chen, J.X., Yue, G.X., Bai, M.H., Kou, M.J. and Jinzy (2012). Xiaoyaosan decoction regulates changes in neuropeptide Y and leptin receptor in rats arcuate nucleus after chronic immunization stress. Evidence-Based Complementary and Alternative Medicine, 3(8): 127-128.
- Yang, P., Yunen, L. and Xiaodong, X. (2013). Change trend of organ weights. Background data on Sprague Dawley Rats at different Ages: *Journal of Toxicologic Pathology.*, 26:29-34.