Pharmacophore

ISSN-2229-5402

Journal home page: http://www.pharmacophorejournal.com

Pharmacophore
an International Journal
ISSN 2229-5402

EFFECTS OF CROCİN AND RESİSTANCE TRAİNİNG ON LİVER ENZYMES İN TOXİCATED MALE RATS BY NANDROLONE DECANOATE

Hadi LalAbadi[1*], Mohammad Ali Azarbayjani[2], Bahman Tarverdizade[3], Maziyar Amin[1]

1. *MA Sports Physiology, Bushehr Branch, Islamic Azad University, Bushehr, Iran*
2. *Professor of Sports Physiology, Central Tehran Branch, Islamic Azad University,Tehran, Iran*
3. *Assistant Professor of Sports Physiology, Bushehr Branch, Islamic Azad University, Bushehr, Iran*

ARTICLE INFO

Received:
03th Jun 2017
Accepted:
29th Nov 2017
Available online:
14th Dec 2017

Keywords: *Resistance training, Nandrolone decanoate, Crocin*

ABSTRACT

Introduction: The purpose of this study was to investigate the effects of crocin and resistance training on liver enzymes in toxicated male rats by Nandrolone decanoate.
Method & materials: Fifty six male Wistar rats (age: 9 weeks, weight: 190±10g) were randomly divided into eight groups: control group (Co, n=7), sham group (Sh), 10 mg/kg of Nandrolone decanoate group (Nd), resistance training group (RT), 12.5 mg/kg of crocin group (C1), 25 mg/kg of crocin group (C2), RT and 12.5 mg/kg of crocin group (RT-C1) , RT and 25 mg/kg of crocin group (RT-C2).All the groups were injected by Nandrolone decanoate (10mlg/kg of body) except (Co and Sh)groups. Resistance training was progressively and consisted of climbing the ladder and started with carrying a load of 50% of body weight. Twenty four hours after the last injection, the animals were sacrificed. Blood samples were obtained from left ventricle immediately and analyzed for liver enzymes (AST, ALT, ALP and GGT), albumin, total protein. Testicular and liver were brought out of their body for analyzing. Data were analyzed using two way analysis of variance.
Results: Data analysis revealed that the interaction effect of crocin and resistance training cause to significant changes on ALT, GGT, Albumin and Total protein ($P<0.05$). The main effect of crocin on albumin and total protein were significant ($P<0.05$). Also, there were not any significant changes on AST and ALP ($P>0.05$).
Conclusion: In general, resistance training and crocin consumption result in eliminate or improve of complication of Nandrolone decanoate in liver organ.

To Cite This Article: Hadi LalAbadi, Mohammad Ali Azarbayjani, Bahman Tarverdizade, Maziyar Amin , (2017), "Effects of Crocin and Resistance Training on Liver Enzymes in Toxicated Male Rats by Nandrolone Decanoate", ***Pharmacophore, 8(6S),*** *e-1173149.*

Introduction

Anabolic-androgenic steroids (AAS) are compound derivatives of the male testosterone hormone [1]. Nandrolone decanoate (ND) has been indicated to improve physical and muscular performance [2], and resistance exercise training enhances strength [3]. The long term use of steroids causes heart attack, liver toxicity, suppression of endocrine nervous glands [4], change in sexual tendencies and test is atrophy [5]. The liver is the largest organ of the human body and by help of difference enzymes has important roles in regulation of hormones activity and metabolism during resting, training and recovery. The main role of liver is detoxification of steroid hormones which long term consumption of them by people causes to accumulate theses steroids and result in liver toxicity [6].The best way for clinical assessing of liver is via

Corresponding Author: Hadi LalAbadi, MA Sports PhysiologyBushehr Branch, Islamic Azad University, Bushehr, Iran. E-mail: Hadi.lal64@gmail.com

measuring the liver enzymes activity (AST[1], ALT[2], ALP[3]and GGT[4]), because if the liver cells get injured, these transaminases would increase in blood and would finally damage the liver[7]. Silva and Beneto reported that testosterone undecanoate (TU) treatment along with moderate physical training increased liver damage.Nowdays, inside the chemical drug treatments, try to use physical activity and herbs in treatment of liver cells and testis cells damages. One of these herbalsisCrocus sativus L, which is commonly known as saffron[8]. The effective components of saffron arecrocin, crocetin and safranal. Crocin is a carotenoid and is responsible for the red color of saffron[9]. Crocinhas different pharmacological effects such asprotective effects againstcardiovasculardiseases, nervous system protection, liver protection and inhibition of tumor cells proliferation. However, among the different protective mechanisms, the antioxidant activity of crocin is responsible for the pharmacological effects. With due attention to hypolipidemic andantioxidant activity effects of crocin, probably it can protect the liver fromSteatosis and oxidative stress [10]. Also, compound of saffron stigma acts as a stimulant to produce LH, FSH and testosterone hormones, proliferatethe possibility of seminiferous tubules epithelial cells and increase of leydig cells activity , and thus it causes to significant increase inSpermatocytes and Spermatogenesis levels [11].With due attention to protective and impressive effects of physical activity and crocin on liver and prevalence of consumption of Nandrolone decanoate between athletics and adults, this study is investigate the effects of crocin and resistance training and interaction of them on liver enzymes in toxicated male rats by Nandrolone decanoate.

Methods

1. Subjects

Fifty six male Wistar rats (age: 9 weeks, weight: 190±10g) were randomly divided into eight groups. Control group were fed standard rodent chow (Co, n=7), sham group were injected by 0.2 CC of saline(SH, n=7), 10 mg/kg of Nandrolone decanoate (ND,=7), resistance training (RT, n=7), 12.5 mg/kg of crocin (C1, n=7), 25 mg/kg of crocin (C2, n=7), RT and 12.5 mg/kg of crocin (RT-C1, n=7) , RT and 25 mg/kg of crocin (RT-C2, n=7). Animals were kept in a cage under a 12/12 h light/dark cycle at 22 ± 2°C, humidity of 50-60% had free access to water and food. Animals were acclimatized and familiarized to the laboratory conditions and exercised for the duration of fourteen days before the beginning of the protocol.

2.Experimental design

After obtaining the approval of the Institutional Review Board of our medical school, all experiments were carried out in accordance with the Guidelines of the Animal Care and use ethics committee of Baqiyatallah University of medical sciences. Training exercise protocol was progressive resistance training form including 3 sessions per week for 8 weeks with three sets of five repetitions. The rest among sets and repetition were 90 s and 60 s, respectively. The resistance training consisted of climbing the ladder (1 meter with 26 escalators inclined at 85 degrees) carrying a load suspended from the tail. The initial load in the first week was 50% of their body weight and increased10%of their bodyweight gradually each week so that at 8th week arrived to 120% of their body weight [12]. The rats were familiarized with the climbing the ladder two weeks ago. All injections were done between 12 AM to2 PM. The supplementation groups received crocin daily. The control group presented in training place but did not have any activities, consumption of any crocin or ND. All the groups received intramuscular injections, every week, in the quadriceps muscles using insulin syringe containing 10 mg / kg of ND except control and sham groups. The weight of animals were measured in initial and the end of protocol.

3.Crocin supplement

Crocin was powdered and then was packed in 1.5gr packs. Crocin was the product of Sigma company made in Germany. They were kept at +4°C.Both doses of crocin diluted in 0.2 CC of saline and was intraperitoneally injected (IP) daily at 2 PM.

4.Blood sampling and analysis

Twenty four hours after the last injection, the animals were anaesthetized by ether (Merk, Germany) and sacrificed. Blood samples were obtained from left ventricle immediately. Plasma was separated by centrifugation (3000g, 15min at 20°C) immediately after blood sampling and was frozen and stored at −20°C for subsequent analyses. The livers and testises were removed. The samples were analyzed for liver enzymes (AST, ALT , ALP and GGT), total protein and albumin . Serum AST, ALT , ALP,GGT, total protein and albumin were measured by radioimmunoassay (RIA) using kits (Monobind, USA).

5. Statistical analysis

All statistical analyses were performed using the software statistical package SPSS version 23.0 (SPSS, Inc., Chicago, IL, USA). Data were expressed as mean ± standard deviation. All data were normalized by a Kolmogorov-Smirnov test. Levene test was used to assess the homogeneity of variances for a calculable variable. Data changes before and after training protocol were analyzed using the two way analysis of variance (ANOVA), if any significant differences had been observed Bonferroni post hoc test had to be used to determine this differences. Statistical significance was set at $P<0.05$.

1. Aspartate aminotransferase
2. Alanine aminotransferase
3. Alkaline Phosphatase
4. Gamma Glutamyl transferase

Results

All obtained data from the variables, based on mean and standard deviation (Mean±SD) are presented in tables 1, 2 and 3.The results of this study showed that there was significant difference among the body weight of groups ($F_{7,46}$=4.099, P=0.001). The highest and the lowest Body weight (67.46% and 35.58% , respectively) among the groups were in (Co)group and C1 group, respectively (table 1).

The main effect of crocin on albumin ($F_{2,34}$=19.730, P=0.000) and total protein ($F_{2,34}$=19.741, P=0.005) were significant (table2).

The interaction effect of crocin and resistance training significantly decreased ALT ($F_{2,34}$=3.298, P=0.049) (fig 1), albumin ($F_{2,34}$=12.590, P=0.000)(fig 4)and total protein ($F_{2,34}$=6.091, P=0.005)(fig 3) levels in all the groups but this reduction in RT-C1 group was more (table 2). The interaction effect of crocin and resistance training significantly increase GGT levels ($F_{2,34}$=0.174, P=0.039) in all the groups but this increase in RT-C1 group was more (fig.2 and table 2). Also, the interaction effect of crocin and resistance training did not cause any significant changes in ALPand AST in all the groups (P>0.05).

Discussion

The purpose of this study was to investigate the effects of crocin and resistance training on liver enzymes in toxiciated male rats by Nandrolone decanoate. Based on the protocol, all the groups except the control and sham groups were confronted with high amount of ND (10 mg / kg of Nandrolone decanoate) every week. Body weight of all the groups increased. The highest and the lowest Body weight (increase of 67.46% and 35.58% respectively) among groups were in control group and C1 group, respectively. The results demonstrated that ALT levels were at least amount in resistance training group and this reduction along with crocin supplement especially dose of 12.5 mg/kg of crocin was significant and more clear. Interaction effect of resistance training and crocin decreased AST and ALP activity but were not significant but it is important clinically aspect and should not be unseen. Although, exercise that itself is one of the damaging factors on liver, it had made it possible that with reducing the complication of ND and the mediation of crocin, eliminate the harmful effects of using ND in high amount. It is interesting that training groups had the lowest albumin levels and the groups which had not use crocin had the highest albumin levels. This result showed that probably the groups which ND complications of them had been seen lower or been reduced by resistance training and crocin , had lower osmosis stress. Another match in results was repetition of similar findings about total protein. This means, there was less total protein in resistance group and crocin supplement groupsbut it was higher in Nandrolone group and the groups without crocin. Crocin and Safranal singly have strong immediate influences on and against high toxic drug and can keep the amounts of albumin and total protein in normal range [13]. Perhaps, as it was mentioned the presence and enzymes activity were decreased by one day recovery of physical activity or training adaptation. However, significant differences among interaction groups with other groups were seen. Also , changes in body weights of groups confirmed other findings. This result even in patients that used blood dialysis treatment along with combination of strength training and ND were seen [2]. Although, there were significant statistical differences among body weight of groups before present study, the most change weight occurs in groups that consumed dose of 12.5 ml/kg crocin. Because of utilization of similar food for all the groups to control of pesky variable, probably having been doing resistance training had cause to enhance energy consumption and lower increase body weight in interaction groups. Likely,the use of high dose of crocin because of impression on catabolism specially lipids induce to lesser body weight changes [14]. This finding can be indicative of improvement of liver function.The less liver weight in interaction effect of resistance training and crocin supplement groups (dose of 25mg/kg) could be due to fatty liver catabolism [15]. Also, it could be because of the changes in ions and blood flow or decrease of body water that result in Shrinkage of the liver [16]. Also, ND caused to increase in AST, ALT activity and the ratio of them in albino mice [17].Physical activity in accordance with duration and intensity could be from different mechanisms specially ischemic reperfusion and invasion of free radicals in to liver, cause kinds of damage in this organ [18,19]. This report suggested that even oxygen and blood flow were decreased after high intensity physical activity [18]. Although, these findings were not repeat about the liver enzymes changes after one bout aerobic training in untrained persons [20]. The various adaptation to a regular training such as enzymatic and non enzymatic antioxidants defense reinforcement and anti- inflammatory andantioxidant properties of crocin decrease the effects of liver damage or improve them [21,22]. Pay attention to it that anabolic steroids due to steroid structure do not need to membrane receptors and easily result in severe oxidative stress in all the body cells. In usual, additional steroids for treatment of patient, excrete via the liver and kidney [8], but exercise trainings cause the consumption of them. Because steroids have anabolic and androgenic impressions. In treatment of patient, its androgenic property was cleared but exercise trainings induces using both of steroids effects [23]. In general, there were not any certain changes in numberof spermatogonia, primary spermatocytes, spermatids and sertoli cells in experimental groups to control but leydig cells increased dramatically in experimental groups that had received crocin. Also, decrease of leydig cells in Nandrolone decanoategroup (ND group) was seen. Our results showed that the C1 group (dose of 12.5 mg/kg of crocin) had the greatest testicle weight and C2 group (dose of 25 mg/kg of crocin) had the lowest testicle weight among groups.

The present study, however, has some limitations. One of the limitations of our study relies on the absence of amount of variables such as liver enzymes before initiate of protocol in order to comparison of data to precise determination of variations. It is recommended that future studies measure some of these enzymes before protocol.

Conclusions

Totally the result of present study has showed that interaction effect of resistance training and both dose of crocin resulted in decrease of the complication of ND in liver. Probably, antioxidants properties of crocin is one of the important factor in improvement of liver organ.

Acknowledgment

The authors wish to thank volunteers for their enthusiastic participation in this study.

Table legends

Table 1.Mean and standard deviation (Mean±SD) of body weight

Table 2.Mean and standard deviation (Mean±SD) of liver enzymes, albumin, total protein

Figure legends

Fig.1 The mean of ALP in different groups

Fig.2 The mean of GGT in different groups

Fig.3 The mean of total protein in different groups

Fig.4 The mean of aibumin in different groups

Fig.5 The mean of AST in different groups

Fig.6 The mean of ALT in different groups

Table.1

group	Weight (gr)		Percentage of increase
	Pre test	Post test	
C	180.00±3.83	301.43±19.03	67.46
SH	184.43±1.90	288.71±13.11	56.54
ND	190.43±8.20	283.14±33.38	47.68
RT	196.00±8.06	281.28±21.48	43.51
C1	200.83±5.00	285.67±15.65	42.24
C2	199.14±7.36	270.00±30.17	35.58
RT+C1	197.33±6.22	277.00±20.27	40.37
RT+C2	199.71±8.03	272.71±23.63	36.55

C: Control, SH: sham, ND: 10 mg/kg of Nandrolone Decanoate, RT: Resistance Training, C1: 12.5 mg/kg of crocin, C2: 25 mg/kg of crocin, RT+C1: Resistance training and 12.5 mg/kg of crocin, RT+C2: Resistance training and 25 mg/kg of crocin

Table.2

group	Liver weight (g)	AST(u/L)	ALT(u/L)	ALP(Iu/L)	GGT(Iu/L)	Total protein(g/dl)	Albumin
C	12.22±1.41	299.14±45.60	98.00±25.88	949.57±423.84	1.43±0.53	9.86±0.43	**5.34±0.36**
SH	11.11±1.83	262.86±80.78	110.14±21/00	914.71±105.10	1.86±0.86	10.51±0.48	**5.63±0.22**
ND	10.26±1.11	293.28±82.17	94.43±14.03	968.14±253.26	2.43±0.78	10.36±0.46	**5.48±0.21**
RT	9.74±1.77	294.00±50.43	102.28±12.26	898.57226.89	1.86±0.69	9.03±1.32	**4.81±0.46**
C1	9.91±1.50	292.00±67.30	107.50±11.73	882.17±187.78	2/00±0.63	8.22±0.20	**4.52±0.15**
C2	9.64±1.06	202.57±51.83	88.57±20.70	801.14±276/07	2.14±0.90	8.13±0.39	**4.54±0.13**
RT+C1	9.13±1.03	207.67±52.25	78.50±12.04	665.67±228.10	3/00±0.89	8.37±0.22	**4.70±0.14**
RT+C2	9.90±0.85	198.28±55.20	91.00±33.70	669.43±166.78	2.71±0/75	8.43±0.63	**4.71±0.24**

Fig.1

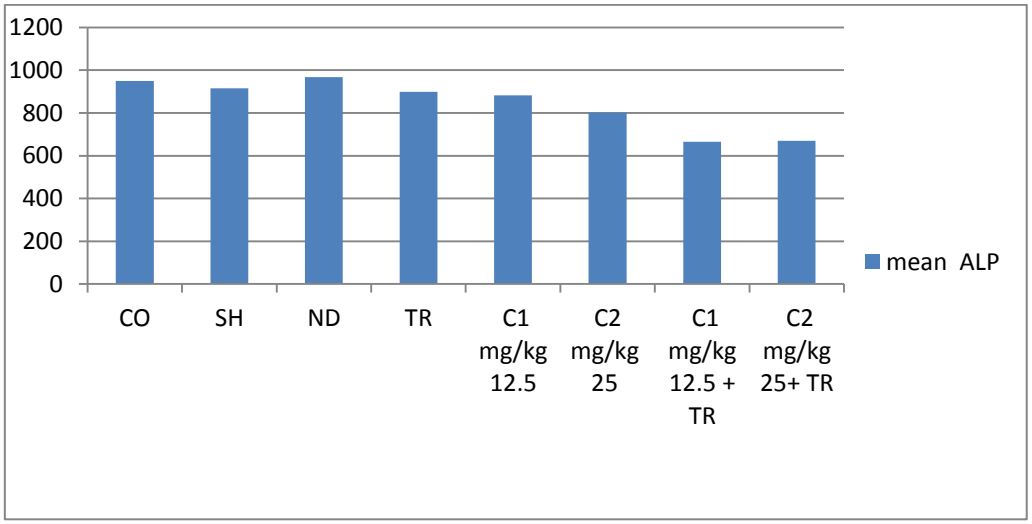


Fig.2

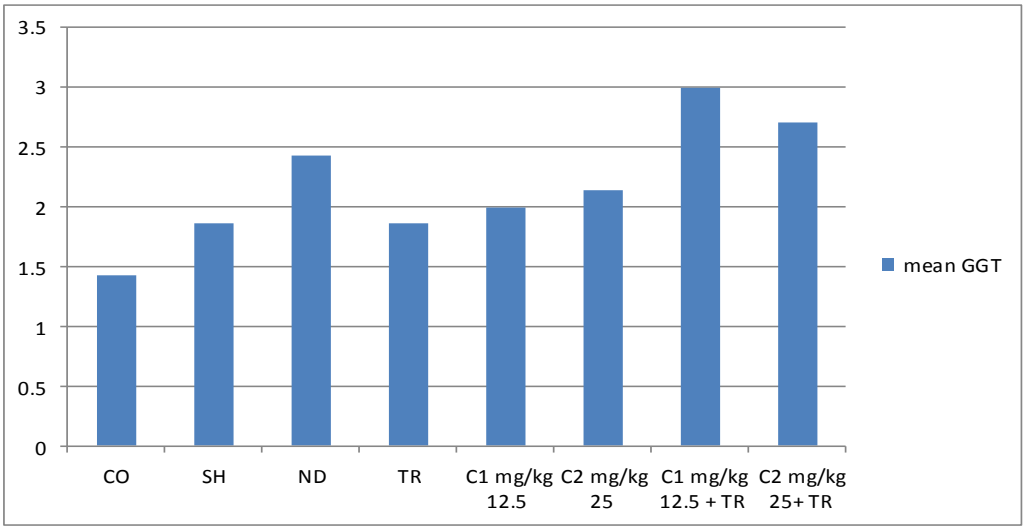


Fig.3

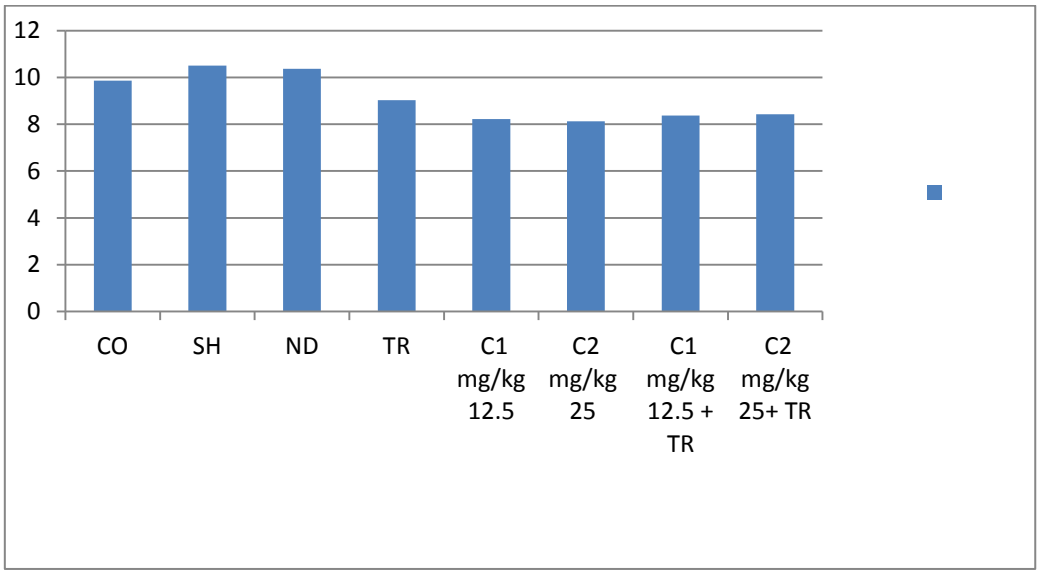


Fig.4

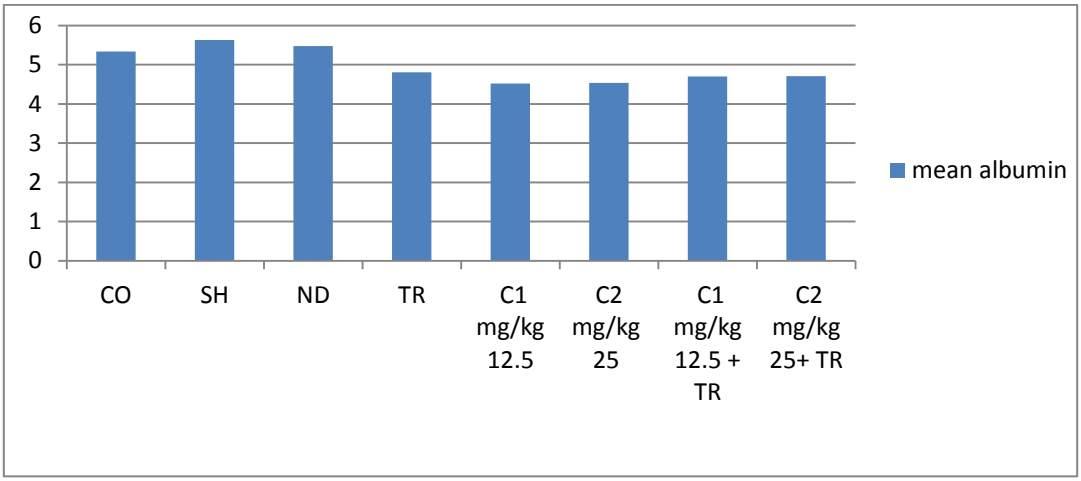


Fig.5

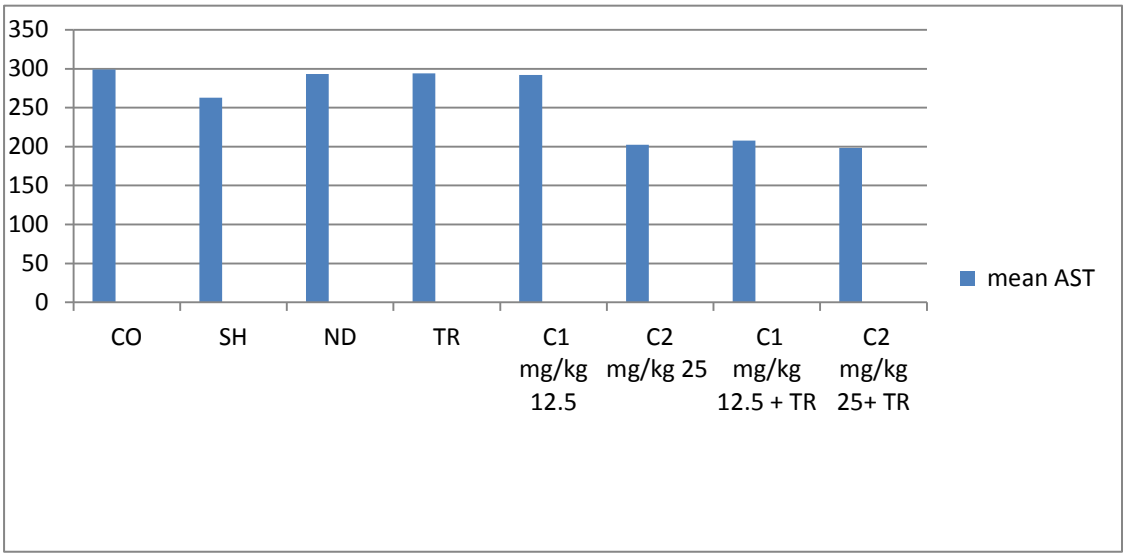
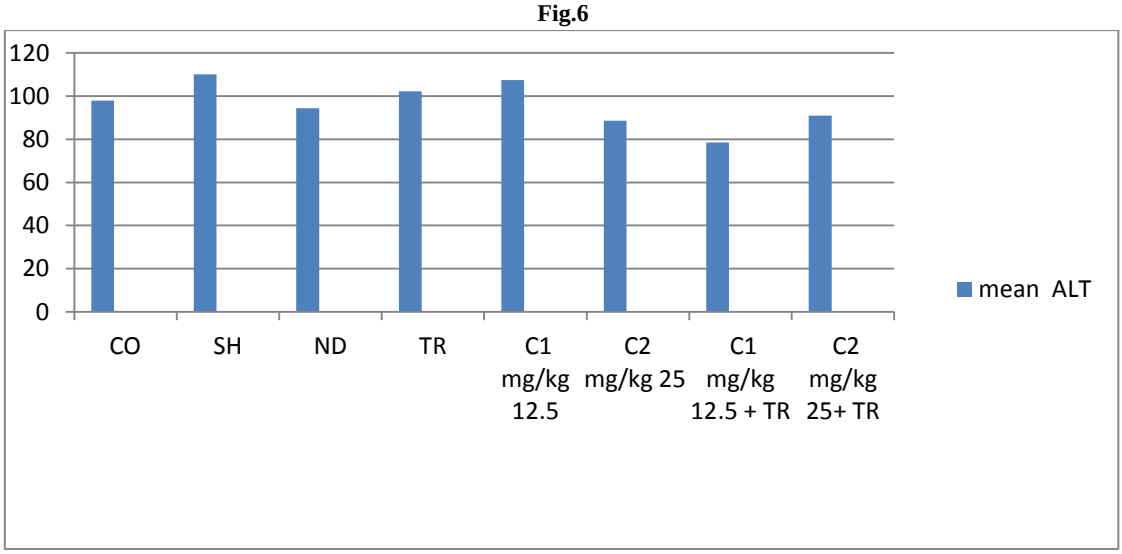


Fig.6



References

1. Joo, Y. I. A. Sone.. "Effects of endurance exercise on three – dimensional trabecular bone microarchitecture in young growing rats". Bone,2003, 33:PP: 485-493.
2. Johansen K, Mulligan K, Schambelan M: Anabolic effects of nandrolone decanoate in patients receiving dialysis: A randomized controlled trial. JAMA, 1998, 281: 1275–1281.
3. Headley S, Germain M, Mailloux P, Mulhern J, Ashworth B, Burris J, Brewer B, Nindl B, Coughlin M, Welles R, Jones M: Resistance training improves strength and functional measures in patients with end-stage renal disease. Am J Kidney Dis,2002, 40: 355–364.
4. Kanayama G, Barry S, Hudson JI, Pope HG. Body image and attitude toward male role in Anabolic AndrogenicSteroids use. Am J Psychiatry. 2006; 163(4):697–703.
5. Padersen W, Wichstrom L, Blekesaune M. Violent behaviors, violent victimization, and doping agent: Anormal population study of adolescents. J Interpers Violence. 2001; 16(8): 808 -32.
6. Singh Anita, Bhat Tej K and Sharma Om P,Clinical Biochemistry of Hepatotoxicity, J Clinic Toxicol, 2011,S:4.
7. chung HY, Radak Z, Goto S. Systemic adaptation to oxidative challenge induced by regular exercise. Free radic Biol Med. 2007;44 (2):153-9.
8. Silva, M. T. Bento. Maria do carmo de carvalho e martins, Francisco Leonardo Torres Leall, Talvany Luiz Barros, Ingrid Lara do Nascimento Ferreira de Carvalho, Hugo aparecido carvalho filho,Fernanda regina de Castro almeida Effects of administering testosterone undecanoate in rats subjected to physical Brazilian. Brazilian journal of Phar maceutical scienes,2010,vol.46,n. 1.
9. Mohamadpour AH, Ayati Z, Parizadeh MR, Rajbai O, Hosseinzadeh H. Safety Evaluation of Crocin (a constituent of saffron) Tablets in Healthy Volunteers. Iran J Basic Med Sci. 2013;16:39–46.
10. Sheng, L., Qian, Z., Zheng, S., and Xi L, Mechanism of hypolipidemic effect of
crocin in rats: crocin inhibits pancreatic lipase. European Journal of Pharmacology ,2006, 543:
116-22.
11. Modaresi, M., Messripour, M., Nazem, H. and Asadi, M., Effect of Saffron Extract on PituitarytestisAxis in Mice. International Journal of HealthScience,2008.
12. De Paiva Foletto, M.; Mareze-Costa, C.; Ferrari, F.; Franzói- de- Moraes, S. & Mara Segatelli, T. Effect of the nandrolone decanoate on the efficiency of spermatogenesis of sedentary rats and rats subjected to physical training. Acta Scientiarum Health,2010,Sci., 32(1):17-22.
13. Hariri AT1, Moallem SA, Mahmoudi M, Memar B, Hosseinzadeh H. Sub-acute effects of diazinon on biochemical indices and specific biomarkers in rats: protective effects of crocin and safranal. Food Chem Toxicol, 2010 Oct;48(10):2803-8. doi: 10.1016/j.fct.2010.07.010.
14. Xu GL1, Yu SQ, Gong ZN, Zhang SQ. Study of the effect ofcrocin on rat experimental hyperlipemia and the underlyingmechanisms]. Zhongguo Zhong Yao Za Zhi, 2005 Mar;30(5):369-72.
15. Hallsworth Kate , Fattakhova Gulnar, G Hollingsworth Kieren,Thoma Christian, Moore Sarah, Taylor Roy, P Day Christopher, I TrenellMichael, Resistance exercise reduces liver fat and its mediators in non-alcoholic fatty liver disease independent of weight loss,Gut,2011,Sep; 60(9): 1278–1283.
16. Latour, M. G., A. Brault, et al. "Effects of acute physical exercise on hepatocyte volume and function in rat." American Journal of Physiology-Regulatory, Integrative and Comparative Physiology,1999, 276(5): R1258-R1264.
17. Chowdhury Parmita and Dr. Mahanta Rita,2014, Hepatic transaminase activity in nandrolone decanoate treated Albino mice, International Journal of Scientific and Research Publications, Volume 4, Issue 5.
18. Fojt E, Ekelund LG, Hultman E. Enzyme activities in hepatic venous blood under strenuous physical exercise. Pflugers Arch. 1976 Feb 24;361(3):287-96.
19. Praphatsorna Panu, Thong-Ngama Duangporn, Onanong Kulaputanaa, Naruemon Klaikeaw Onanong, Effects of intense exercise on biochemical and histological changes in rat liver and pancreas ،Asian Biomedicine Vol. 4 No. 4 August ,2010; 619-625.
20. Ajami Nezhad M, Saberi kakhki A, Jahromi M " The Effects of a Single Bout of Aerobic Exercise at Different Intensities on Markers of Liver Function and Blood Hemoglobin in Healthy untrained Male" Quarterly of the Horizon of Medical Sciences Vol. 19, No. 4, Win 2014 Pages: 184- 191.
21. Altinoz E1, Ozmen T, Oner Z, Elbe H, Erdemli ME, Bag HG. Effect of crocin on oxidative stress in recovery from single bout of swimming exercise in rats. Gen Physiol Biophys. 2015 May 22.
22. Bakhtiary Z1, Shahrooz R1, Ahmadi A1, Zarei L2. Evaluation ofantioxidant effects of crocin on sperm quality in cyclophosphamidetreated adult mice. Vet Res Forum. 2014 Summer;5(3):213 8.
23. Sakr SA1, Zowail ME2, Marzouk AM1.(2014). Effect of saffron (Crocus sativus L.) on sodium valporate induced cytogenetic and testicular alterations in albino rats. Anat Cell Biol. 2014 Sep;47(3):171-9. doi: 10.5115/acb.2014.47.3.171.