KOREAN JOURNAL OF ZOOLOGY
Vol. 18, No. 3, 147—156 (1975)

Effect of Low Atmospheric Pressure on Serum Glutamic Oxaloacetic Transaminase and Lactic Dehydrogenase Activities of Rats

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低氣壓이 흰쥐의 血淸 Glutamic Oxaloacetic Transaminase 및 Lactic Dehydrogenase 活性에 미치는 影響

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(Received September 4, 1975)

摘 要

成熟한 Sprague-Dawley系 雄性 흰쥐를 對照群(760 mmHg)과 低氣壓群인 500 mmHg와 380 mmHg의 兩實驗群으로 나누어 15日間(1日當 1時間) 曝露시켜, 低氣壓이 血淸 glutamic oxaloacetic transaminase(GOT) 및 血淸 lactic dehydrogenase(LDH) 活性에 미치는 影響을 考察하였다.

血淸 GOT 및 LDH含量은 兩群 共히 對照群에 比하여 顯著한 變化를 招來하였으며, 특히 GOT는 初期에 減少되고, 後期에 恒定持續性을 나타내어 對照群의 값에 近接하는 傾向이 나타났으며, 한편 LDH의 含量은 初期에 增加하고 後期에는 若干의 減少傾向이 나타났다.

一般的으로 이러한 變化는 흰쥐가 低氣壓의 曝露에 一時的인 恒定持續性을 나타내는 것을 보여주며 酵素의 含量의 變化는 低氣壓의 强度와 順化期間에 따라 다르며, 一般的으로 380 mmHg 群은 500 mmHg 群에 比하여 顯著한 變化相을 가져오는 것으로 思料된다.

INTRODUCTION

It is a common knowledge that the mechanism by which the environment induces adaptive change is multi-integration by nervous, hormonal, behavioral, metabolic and enzymatic responses. Low atmospheric pressure is known to produce biochemical, electrophysical and morphological alterations in various tissues of intact animals.

Biochemical changes in various enzymes in the guinea pig(Tappan *et al.*, 1957), in the dog (Highman and Altland, 1960), and in the man (Reynafarje, 1962) have been induced when animals were experimentally exposed to low atmospheric pressure.

The present study was undertaken to look for adaptive changes in activity of serum glutamic oxaloacetic transaminase and lactic dehydrogenase of rats during acclimatization to low atmospheric pressure.

MATERIALS AND METHODS

Ninety laboratory-conditioned male rats of the Sprague Dawley strain weighing between 200—250 grams at the termination of the experimental period were used in this study. The animals were kept in cages maintained at room temperature of 25±1°C. They were fed *ad libitum* water and commercially prepared animal food and were continued on this regimen throughout the experimiental period. Conditions of caging, illumination and temperature were similar for all groups.

The animals were randomly grouped as the control maintained at normal atmospheric pressure (760 mmHg), and the experimental subjected to a series of one hour exposure a day to low atmospheric pressure: 500 and 380 mmHg up to the time of sacrifice; 1, 2, 3, 5, 9 and 15 days. An 18.5 liter desiccator connected to a vacuum pump by a rubber tubing was used as a low atmospheric chamber. Proper arrangement of inlet and outlet tubings and attached clamps permitted the test at desired pressure. The resulting change was indicated by a barometer. In the bottom of the desiccator, 1,000 m*l* of 1 N NaOH for the absorption of CO_2 expired by the animals was filled. While in the chamber, the animals were understrained to food and drinking water. Immediately after the rats were removed from the chamber and their weights were taken, blood samples of about 5 m*l* were obtained in Wintrobe tubes from the severed left saphenous vein of each specimen. After clotting, the sera were separated by centrifugation at 3,000 rpm for 30 minutes and were kept in a refrigerator until the determination of enzyme activity. Haemolysed sera were rejected.

Serum glutamic oxaloacetic transaminase (SGOT) and serum lactic dehydrogenase (SLDH) activities were determined according to the procedures outlined by Rietman and Frankel (1957) and Cabaud and Wroblewski (1958), respectively, utilizing reagents supplied by the Sigma Chemical Company, Missouri, USA. Spectrophotometric determinations were made with a Coleman model 295 E spectrophotometer at 505 *n*m for SGOT and 550 *n*m for SLDH. SGOT activity level was expressed in units per milliliter and SLDH in units.

Statistical evaluation of the data was accomplished by calculating the mean activity and the standard error of the mean for each group. Comparision of means was made by the "t" test. A probability level of 0.05 and 0.01 were used as the criteria of statistical significance.

RESULTS

The influence on serum glutamic oxaloacetic transaminase and lactic dehydrogenase levels of rats of low atmospheric pressure was followed up to to 15 days. All the rats became restless during decompression. At the desired simulated atmospheric pressure the rats seemed to be asleep. They were conscious, however, as each rat would lift its head or stand occasionally. As soon as recompression to sea level(760 mmHg) was attained, most of the rats were standing and alert. All rats survived without mortality.

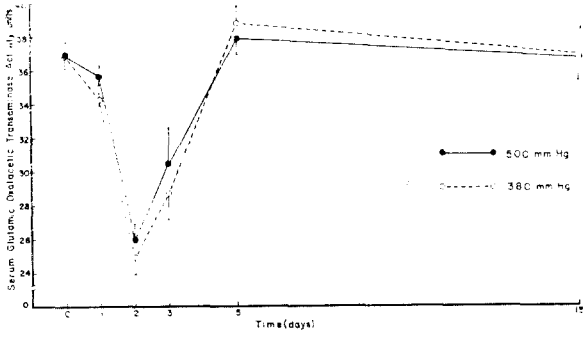

Fig. 1. Fffect of environmental low atmospheric pressures (500 and 380mmHg) on glutamic oxaloacetic transaminase activity in serum of rats at various time intervals during acclimation. Each point represents the average value for the number of rats given in Table 1 and the vertical bars indicate the standard error of the mean.

Table 1 shows mean values of SGOT in sera of control rats exposed to 500 mmHg and rats exposed to 380 mmHg, and standard error. All data were expressed in units/m*l* and are summarized graphically in Fig. 1.

The average value of the control was 36.93 units/m*l* with a standard error of ±0.76. The SGOT mean values of the experimental rats on the first and second days after exposure to 500 mmHg for one hour a day were 35.68±0.59 and 26.00 ±0.98 respectively, for those treated at 380 mmHg were 34.30±0.32 and 24.99±0.88 respectively. These data indicate a decrease of SGOT levels on the above mentioned days. However, at 3 days both decompression groups manifested a remarkable rise in SGOT and continued to increase to a maximum level (37.88± 0.94 for 500 mmHg and 38.75±1.03 at 380 mmHg) on the fifth day, although these increases were not statistically significant. Thereafter, at 15 days after exposure, SGOT values for both groups returned very close to the control.

A comparision of the results obtained from rats exposed to 500mmHg with that the same order of change has occurred in enzyme levels.

It is evident from Table 1 and Fig. 1 that the differences in SGOT levels between the control and both experimental groups are strikingly significant. In the statistical test of significance for SGOT, changes at 2 and 3 days were markedly significant ($P<0.01$) for 380mmHg exposed rats.

Fig. 1 clearly shows that the serum enzyme reached steady state from 5 to 15 days. It can be inferred that the rats became acclimated to low atmospheric

Table 1. Glutamic oxaloacetic transaminase activity in serum of rats acclimated to low atmospheric pressures of 500 and 380 mmHg.

atmospheric pressure	Time (days) of acclimation	No. of rats	Serum GOT activity level (units/ml)	
			Mean	Standard error
Control (760mmHg)	0	8	36.93	±0.76
500 mmHg	1	7	35.68	±0.59
	2	7	26.00	±0.98**
	3	7	30.54	±2.11*
	5	7	37.88	±0.94
	15	6	36.73	±1.38
380 mmHg	1	7	34.30	±0.32
	2	7	24.99	±0.88**
	3	7	28.70	±1.64**
	5	7	38.75	±1.03
	15	7	36.85	±1.93

*P<0.05
**P<0.01

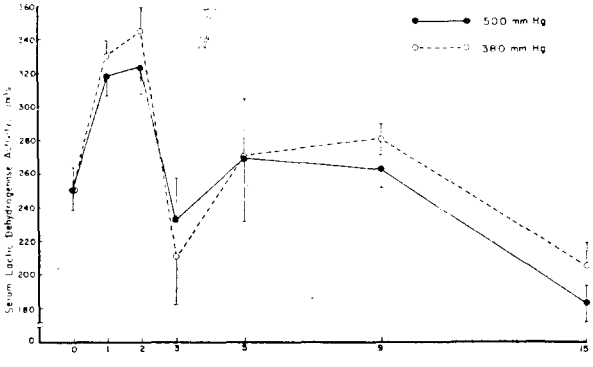

Fig. 2. Effect of environmental low atmospheric pressures (500 and 380 mmHg) on lactic dehydrogenase activity in serum of rats at various time intervals during acclimation. Each point represents the average value for the number of rats given in Table 2 and the vertical bars indicate the standard error of the mean.

pressure during this period.

Table 2 and Fig. 2 summarize data on SLDH levels of control and low atmosphere-exposed rats. Average value for control was 251.19±13.51 units. In the experimental group, large increases in SLDH were observed on the first and second days, 317.78±12.17 and 322.50 ±16.49 respectively for those exposed to 500 mmHg, and 330.28±9.33 and 344.58±15.14 respectively for those at 380mmHg. A fall in this enzyme level was noted in both experimental groups on the third day. However, a slight increase was seen after 5 days, although it was not significant. A further rise in mean values was observed on the 9th day, after which a significant drop of this enzyme level was registered at the termination of this experiment for both decompression groups.

The general pattern of fluctuations was the same in the 380 mmHg and 500 mm Hg treated rats though the variation was greater in the former than in the latter. These fluctuations are considered as to indicate the change in enzyme concen-

tration according to the intensity of the stress.

Table 2. Lactic dehydrogenase activity in serum of rats acclimated to low atmospheric pressures of 500 and 380mmHg.

atmospheric pressure	Time (days) of acclimation	No. of rats	Serum LDH activity level (units)	
			Mean	Standard error
Control (760mmHg)	0	8	251.19	±13.51
500mmHg	1	7	317.78	±12.17**
	2	7	322.50	±16.46**
	3	7	231.66	±26.21
	5	6	269.17	±49.56
	9	7	261.67	±11.67
	15	6	183.33	±9.37*
380mmHg	1	7	330.28	± 9.33**
	2	7	344.58	±15.14**
	3	7	210.00	±28.07
	5	7	270.33	±33.74
	9	6	280.00	± 9.40
	15	7	205.00	±13.70*

*$P<0.05$
**$P<0.01$

From Table 2 and Fig. 2 it can be deduced that the most striking change in enzyme level was an increase in SLDH which was set at its highest point at 2 days for both experimental groups. Furthermore, SLDH was decreased to its lowest at 15 days.

The analysis of variance by the "t" test revealed that the different levels of SLDH enzyme in both groups were significantly elevated on the first and second days ($P<0.01$) and markedly fell at 15 days ($P<0.05$).

The irregularity of these enzyme levels throughout the course of the experimental period indicated that no steady-state was arrived by the animals during exposure to low atmospheric pressure. Nevertheless, this anomaly in SLDH values might be indication of organisms' struggle for acclimation.

The present results for SGOT and SLDH values during acclimation to decompression treatments manifested similar responses to the experimental groups.

DISCUSSION

If a system at equilibrium is subjected to any stress, a change which reduces the stress will occur; or if a system is in stable equilibrium and one of the conditions changes, then the homeostasis will shift in such a way as to tend to restore the original conditions (Leake, 1964). The data presented in tables and

figures in the present paper demonstrate that the exposure to low atmospheric pressures, 500mmHg and 380mmHg, altered the levels of serum glutamic oxaloacetic transaminase (SGOT) and serum lactic dehydrogenase (SLDH) enzymes in the sera of rats. In most cases, the magnitude of change was greater in rats treated at 380mmHg than 500mmHg. Such data agree with Luft's (1964) statement that departure from sea level pressure to elevation into the higher regions of the atmosphere gives rise to a variety of adaptive changes, or manifests the same adaptive change with resulting levels that differ, the extent of which depends upon the degree of the prevailing stress.

On the first 2 days of the experimental period, decreases in SGOT were observed in both low atmosphere-exposed rats, the fallb eing statistically significant on the second day. This result is not conformity to that of Highman and Altland (1960) who noted an immediate rise in serum transaminase, alkaline phosphatase and lactic dehydrogenase after 4 hours exposure of dogs to a simulated altitude of 32,000 ft. Lemley-Stone *et al.* (1955) found an average in serum transaminase of 35% in dogs subjected to coronary artery ligature. According to Merrill *et al.* (1957), however, a 27% decrease in GOT content of anoxic myocardial tissues was seen after exposure of rats to acute anoxia. This is in good agreement with the results obtained on the first two days of the present investigation. These conflicting results may be due to the variety of experimental conditions used by the workers. The differences indicate that alterations do occur at some locus in the system, and that the same components of complex systems are not influenced equally by the adaptation process as well as type of environmental stress. A relative decrease in SGOT as shown in this study may therefore be important during the immediate stress of the adaptive period.

As is evident from Table 1, however, on the third day both low pressure-treated groups the SGOT values increased significantly. The cause of this delay is not known. Probably there was local inactivation of this enzyme and slow reactivation after absorption and elimination by the circulation of inactivators as was suggested by Merrill *et al.* (1957). This rise in SGOT level coming close to the initial level is in fair agreement with Highman and Altland's (1960) observation that the values of serum enzymes usually returned within 3 or 4 days to inital levels following the exposure. From this time on SGOT gradually maintained a steady-state until it came near the control value at the terminaion of the experimental period.

It was first speculated that exposure to hypoxia aids in the release of catecholamines from the sympahetic adrenal system and that large doses of this chemical substance produce marked elevations in serum transaminase, alkaline phosphatase and lactic dehydrogenase (Highman and Altland, 1960).

It is postulated that hypoxia alters serum enzyme levels by (a) damaging cells

which is evidenced by the development of pathologic changes in heart, skeletal muscle and other organs after repeated exposures to high altitude(Altland and Parker, 1955; Merrillet *et al.*, 1957); (b) tissue destruction where these enzymes are released into the blood stream by the disrupted tissues (Cantarow and Schepartz, 1967; Nam and Chang, 1970; Harper, 1971); (c) elevated level of enzymes in the blood serum of animals to physiological stress is a general increase in cellular permeability (Hawrylewicz and William, 1961; Nam and Chang, 1970); (d) liberation of enzmes into the blood stream from malignant cells (White *et al.*, 1968); (e) loss or inactivation of cellular tissue enzymes induced by hypoxia.

From the results obtained in this experiment it has been confirmed that exposure to various degrees of low atmospheric pressure resulted in an increase of serum lactic dehydrogenase(SLDH) in sera of rats immediately after exposure to the stress. Such sudden rise in SLDH level was observed in dogs by Barbashova (1964), and Highman and Altland (1960). One may speculate that such proximate elevation following exposure may be due to the liberation of enzyme into the blood stream from damaged cells. It has been suggested that LDH enzyme is loosely held to the mitochondrial membrane and readily removed in the process of cellular damage (Brody and Engel, 1964).

Tissue destruction in line with cellular damage had been associated with augmentation in SLDH level since degrees of hypoxia which alter the functional integrity of the capillary membrane will also damage cells since average oxygen tension in capillaries exceeds that of the tissues (Korner, 1959). In addition to this, liberation of preformed enzymes from the damaged tissues of heart, liver, skeletal muscles and other organs have been documented.

Goodale and Hackel (1949) accounted for lactate level increases due to anoxia as an immediate source of energy. Korner (1959) concluded that such rise in lactate levels after exposure to low atmospheric pressure may be attributed to enhanced liberation of lactate as a result of increased activity of the sympathetic-adrenal system.

A factor attributable to SLDH activity elevation is the increased glycolysis and increased rate of glycogen breakdown(Barbashova, 1964; Lemley and Meneely, 1952; Tappan *et al.*, 1957). Many of the organism's physiological and chemical functions as well as anatomical characters are modified in such a way that active tissue cells are able to receive and utilize properly the oxygen which is transported in the circulating blood. At any degree of oxygen lack the maximum response occurs during the first few minutes following exposure (Rahn and Otis, 1947). Prolonged living in low oxygen reduces the energy source thus, a variety of adaptive responses in organisms, one of which is the anaerobic breakdown of substance to provide the energy to cells necessary for the maintenace of normal metabolic activities for the preservation of life, occurs. Increase in the activity

of enzymes which catalyze anaerobic oxidizing processes and anaerobic transformation of carbohydrates, particularly glycogen, by enzymes such as phosporylase, lipase and LDH is of primary importance. The strengthening of glycolysis as a method of cellular adaptation to changing environmental conditions is a widespread biological reaction. Needless to say, the increase in the effectiveness of anaerobic processes with acclimatization to hypoxia is interesting from the point of view of the evolution of functions (Barbashova, 1964).

Tappan *et al.* (1957) suggested that as a result of altitude adaptation, increase in enzyme level is due to increase in cell number. This proposition may not well apply to the findings of this study since increased LDH in some sera was recorded right after exposure to the stress giving no ample time for cell reproduction to occur.

Reynafarje (1962) reported no difference in muscle LDH activity between high altitude and sea level human residents suggesting that the glycolytic enzymes are not significantly involved in the adaptive processes to high altitude. This conflicts with the findings of the previous investigations already discussed as well as the present experiment. It may, therefore, be deduced that the process of acclimatization to a new environment must not necessarily be similar in all species. The more evolved the organism the more delicate the process may be.

In conclusion, therefore, one may speculate that basically the essential effect of low atmospheric pressure is the stimulation of the activity of enzyme systems in the cell. In the final analysis this change provides for an increase in utilization of oxygen and consequently for a sufficiently high level of oxidizing processes during conditions of anoxemia. True adaptation of an organism to life in a changed environment is achieved mainly through metabolic and physiochemical reorganization of the tissue or cellular level by means of tissue or cellular adaptation. Animals moved from one low atmospheric pressure to the other show changes in enzyme concentrations indicative of acclimation.

SUMMARY

In order to determine the influence of low atmospheric pressure on serum glutamic oxaloacetic transaminase (SGOT) and serum lactic dehydrogenase (SLDH) activities of rats, blood samples were collected from laboratory-conditioned male rats of the Sprague-Dawley strain which were randomly grouped into control and the experimental subjected to a series of one hour-exposure a day to low atmospheric pressures of 500 and 380mmHg up to the the time of 15 days.

Results obtained indicated that decompression caused marked alterations in SGOT and SLDH levels when compared to that of the control. The trend of increases or decreases in these enzyme levels were similar in both 500 and 380mmHg exposed rats although the changes were greater in the latter group. Thus,

generally all the experimental rats showed temporary steady state to low atmospheric pressure. Changes in enzymatic contents depended on the intensity and extent of the environmental stress under study. The lower the atmospheric pressure the greater is the effect on these serum enzyme levels.

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