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THE EFFECT OF A HYPOTHALAMIC LESION ON BLOOD CHOLESTROL IN THE LIZARD, *UROMASTIX HARDWICKII*

MAHMOOD AHMAD, ALMAS HUMERA, RUQAIYA HASAN, HABIB FATIMA, MUHAMMAD ASLAM SIDDIQUI, AISHA JAVAID, TAZEEN NAIM AND *MANSOOR AHMAD

Department of Physiology, University of Karachi, Karachi-75270, Pakistan
**Department of Anatomy, Hamdard College of Medicine & Dentistry, Karachi, Pakistan*
***Department of Physiology, New York Medical College, Valhalla, N.Y., U.S.A.*

ABSTRACT

The study evaluates TBC (Total Blood Cholesterol) during complete starvation following a hypothalamic lesion. For this purpose 240 lizards were divided into three groups. Animals of groups A & B were kept in captivity. Group A represented only starved animals. While group B represented starved lesioned Group. Group C consisted of 10 fresh animals purchased every day to represent a fed group. The cholesterol estimation of A, B and C groups was done from day 1 to day 7 by Sachet's method. Results showed presence of hypocholestrolemia in group A animals during starvation. The decline being great from day 5 to 7. In response to the hypothalamic lesion in group B animals cholesterol increased on day 1, steeply declined on day 2 and then gradually increased till day 7.

INTRODUCTION

The reptile Uromastix hardwickii is a typical desert adapted animal found in dry and sandy regions of IndoPak subcontinent. The whole body of the animals is covered with scales. The lizard enjoys green grasses, grass seeds, fleshy green leaves, flowers and fruits of shrubs. The lizard however has never been seen drinking water (Lyddeker 1910). It under goes hibernation to meet the frost of the winter. Ahmed and Taqawi (1978) have reported that the tissues of this reptile are more fatty and that this extra fat is used as a source of energy and metabolic water during hibernation. They have also suggested that the lizard acquires low metabolic rate and reduces its energy expenditure during hibernation. Captivity and starvation therefore has no considerable adverse effect on this animal. Lizards are cold blooded animals i.e. their body temperature falls in line with that of the surroundings. Temperature therefore, exerts notable influence on their metabolism (Ahmad *et al.*, 1980).

MATERIALS AND METHOD

In this investigation one hundred and sixty lizards, roughly similar *in* size and weight were selected from a large stock. They were then divided into two equal groups A and B.

Animals of group A were kept as starved control, while those of group B served as starved lesioned test group. These animals were kept in captivity for starvation at 29 ± 1°C during the whole study period.

In addition to the above two groups, ten fresh animals obtained each day for 8 days were used as a fed group C. In all 240 animals were used in this study.

Production of the Lesion:

For the sake of producing lesion in the posterior region of the median eminence of hypothalamus, a 220 volt Brawn Melsungcn Electrical Stimulator, type 2240 was employed with 12 volts at 0.5 Amp (Ahmad *et al.*, 1980; Bogdanove and Schoen, 1959).

Collection of Blood:

At the end of each day, 10 animals of starved group A, were quickly decapitated one by one and blood from jugular vein was collected. In a similar way, blood of starved lesioned animals was obtained and cholesterol determinations of each group were done from day 0 to day 7.

Group C animals which were obtained from fields each day were also killed and the estimation of their samples served the analysis of a fed group.

Sackett's method (1925) was adopted for the estimation of total cholesterol in blood (TBC). Alcohol-ether-mixed was prepared by mixing in 3: 1 volumes respectively. A mixture of acetic anhydride-sulphuric acid was also prepared by adding 1 ml of concentrated H_2SO_4 with 20ml of acetic anhydride.

The solutions were made freshly. The stock standard solution of cholesterol was made by dissolving 200mg of pure cholesterol in small quantity of chloroform to be made up finally to 100ml with the further addition of this reagent. The stock solution was kept well stoppered. 1ml of this solution was diluted to 25 ml with chloroform, so that 5 ml of this diluted solution contained 0.4 mg of cholesterol.

PROCEDURE

0.2 ml of blood, added to 10-12 ml of the alcohol-ether mixture in a centrifuge tube, was tightly stoppered before being shaken vigorously for about a minute. After standing horizontally for about 30 minutes, the evenly distributed precipitation was centrifuged for a few minutes to get firm deposit. The supernatant was decanted completely into a suitable test tube and dried through evaporation in a water bath. The residue was then dissolved in 5 ml chloroform.

Moreover, 5 ml of the standard cholesterol solution containing 0.4 mg cholesterol was introduced simultaneously into another tube. To each of these tubes wad added 2 ml of acetic anhydride and H_2SO_4 mixture. After standing in the dark at 25°C for 15 minutes, the samples were read without any delay in the Pye Unicam SP 500 Service 2 Ultraviolet Spectrophotometer at 640 mu.

RESULTS

A consideration of table 1 indicates that the serum cholesterol value remained at 371 ± 14.0 mg/100 ml of blood in fed group C through out the study period. In starved group A it was 371 ± 12 on day 0 and 371 ± 11.4 on 1st day. It dropped to 271 ± 5.7on the 2nd day and further dropped to 257 ± 5.7 on the 3rd day. It was 243 ± 2.3 on the 4th day and 200 ± 0.0 on the 5th day. It decreased to 101 ± 4.1 on the 6th day and 78 ± 2.4 mg/100 ml of serum on the 7th day.

The 0-day cholesterol value in starved-lesioned group was similar (371 ± 1.4) to the values of the former two groups. It increased to 485 ± 12.2 mg/100 ml of blood on the 1st day, decreased to 157 ± 1.8 on the 2nd day and 107 ± 12.0 on the 3rd day. But it showed a rise again on the following day. It was 221 ± 0.8 on 4th day and 209 ± 1.6 mg/100 ml of blood on the 5th day post lesion. It dropped to 139 ± 13.3 on the 6th day to show a final rise of 285 ± 11.8 mg/100 ml blood on the 7th day.

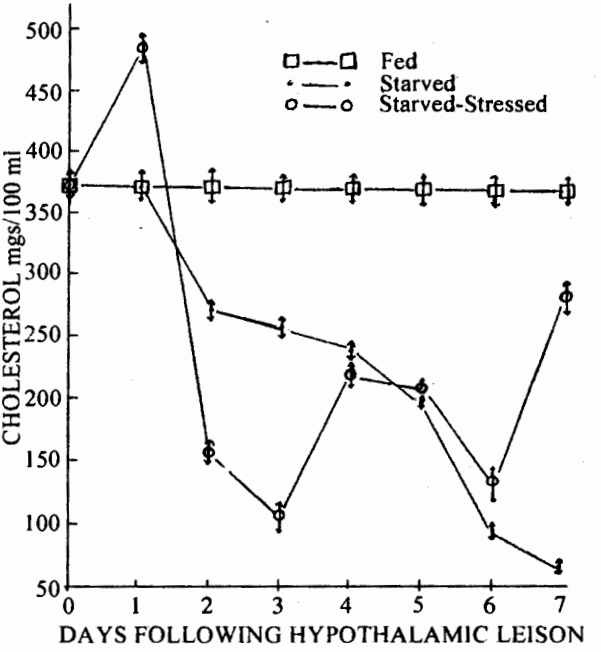
DISCUSSION

Available evidence indicates that the normal total blood cholesterol in Uromastix hardwickii lies between a wide range of 350 to 400 mg/100 ml. The range of variation in the freshly caught lizards in our study however is narrower and the serum level does not appear to be much altered *in* the fed animals.

Hypocholestrolemia is found most characteristically in starvation and the fall roughly parallels with the basal metabolic rate. There is a general agreement that cholesterol synthesis is reduced during

Table 1: Cholesterol estimation in *U. hardsickii* following ahypothaiamic lesions

Groups	Animals per Group	Day Temp. °C	0	1	2	3	4	5	6	7
Fed Group C	80	29 ± 1	371 ± 12.0 (10)	371 ± 12.0 (10)	371 ± 14.0 (10)	371 ± 12.4 (10)	371 ± 11.0 (10)	371 ± 12.5 (10)	371 ± 13.0 (10)	371 ± 11.4 (10)
Starved A	80	29 ± 1	371 ± 12.0 (10)	371 ± 11.0 (10)	371 ± 5.7 (10)	257 ± 5.7 (10)	243 ± 2.3 (10)	200 ± 0.9 (10)	101 ± 4.1 (10)	73 ± 2.4 (10)
Starved B lisioned	80	29 ± 1	371 ± 11.4 (10)	485 ± 12.2 (10)	15 ± 7.8 (10)	107 ± 12.0 (10)	221 ± 0.8 (10)	209 ± 1.6 (10)	139 ± 13.3 (10)	285 ± 1.8 (10)



starvation because of a marked decrease in the activity of B Hydroxy B methyl glutaiyle-CoA (HMG-CoA) reductase in this fasting animals. A fall of 100 mg/100 ml is seen as an initial loss on 2nd day of starvation (Tab. 1)

Compared with the fall on the 2nd day, cholesterol level was more consistent during the 3rd to the 5th day in starved group when a total loss of 75 mg/100 ml was observed. 125 mg/100 ml were lost from day 5 to day 7. This indicates that the activity of HMG-CoA reductase was less reduced during 2nd to the 5th day than on the 2nd day and more so on the 5th to the 6th day in starved group.

It is also a well established fact that thyroxine stimulates the hepatic mechanisms involved in the elimination of cholesterol from the circulation and inhibits as well the synthesis of cholesterol (Money *et al.*, 1960) resulting into decline of plasma cholesterol.

As an immediate reaction to the hypothalamic lesion, the blood cholesterol showed an initial sharp rise of more than 100 mg/ml above the normal values on the 1st day. HMG-DoA reductase is inhibited in liver by this increased cholesterol (Siperstein 1960) thereby inhibiting cholesterol synthesis in liver. Thus, a sharp fall of more than 325 mg/100 ml occurred on the 2nd day and a fall of 50 mg/100 ml on the 3rd day. However, there was a rise of 100 mg/100 ml on the 4th and of 150 mg / 100 ml on the 7th day in starved lesioned group.

The fall in the plasma cholesterol on the 2nd, 3rd and the 6th day was due to:

(a) the decline in resting TSH and insulin secretion (Ganong 1967).
(b) adaptive response to stress and secretion of glucocorticoids (Dempsey, 1974).
(c) decreased HMG-CoA reductase activity in liver, and
(d) raised cholesterol inhibiting cholestertol synthesis.

CONCLUSION

It can be concluded from this study that a normal hypocholestremine response to starvation is governed by hypothalamus because its damage resulted into a highly fluctuating TBC (total blood cholesterol) level.

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