

## **Insulin as an Effective Drug in Scorpion Venom Poisoning**

**A. Zare<sup>\*</sup>, K. Radha Krishna Murthy and M.B. Jafar**

*Department of Physiology, LTM Medical College, Sion, Bombay*

### **Summary**

*Acute myocarditis was induced in rabbits by subcutaneous injection of 4 mg/kg body weight of scorpion (*Mesobuthus tamulus concanensis*, Pocock) venom. Following venom injection the levels of blood sugar, free fatty acids and lactate dehydrogenase (LDH) significantly increased. Among LDH isoenzymes, LDH<sub>1</sub> and LDH<sub>5</sub> comparatively increased more than the other isoenzymes. Out of 5 rabbits which did not receive insulin treatment 4 died within 2 to five h. Insulin treatment in 8 rabbits, 90 minutes after venom injection, reversed blood sugar level, free fatty acids and LDH<sub>1</sub> isoenzyme within 20 hours of treatment and all the rabbits survived. It appears that administration of insulin can effectively reverse the metabolic changes, various manifestations of poisoning and prevent death due to scorpion venom toxicity.*

### **Introduction**

Death due to scorpion venom poisoning is common in developed and undeveloped countries(1). There is no effective, specific and definitive treatment for the metabolic, cardiovascular changes and many other manifestations which are brought about by the scorpion venom toxicity. Our earlier studies had shown the presence of acute hyperglycemia, increased free fatty acids level, rise in osmotic fragility of erythrocytes, depletion of glycogen content of liver and heart(2) with subsequent suppression of insulin release as well as enhanced insulin resistance (results observed but not presented in this paper). The use of insulin to reverse the

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*\*Present address: Department of biochemistry, Razi Inst., PO Box 11365-1558, Tehran, Iran*

electrocardiograph and metabolic changes, due to scorpion venom toxicity, has been proposed by our laboratory recently(3). Therefore, attempt has been made to see the efficacy of insulin in reversing the various metabolic changes and prevention of death in experimental animals poisoned with scorpion venom.

## **Materials and methods**

The lyophilised, crude scorpion venom (*Mesobuthus tamulus concanesis*, Pocock) was obtained from Haffkine Institute, Bombay. A dose of 4 mg/kg body weight venom was injected subcutaneously in 13 rabbits. The rabbits were randomly divided into 2 groups. Group I did not receive insulin treatment whereas Group II received 8 units of crystalline insulin subcutaneously, 90 minutes after venom injection. Blood was collected prior to venom injection, 90 minutes after venom injection and 20 hours after insulin treatment. The blood samples were utilised for determination of blood sugar(4), free fatty acids(5), amylase(6) and LDH(7). The LDH isoenzymes pattern was studied by polyacrylamide gel electrophoresis(8).

Results were analysed statistically using Student paired 't' test.

## **Results**

Signs and symptoms of envenomation including hypersalivation, lacrimation, frequent urination and defecation with diarrhoea appeared, in all the rabbits, within 25 to 30 minutes after venom injection. Following venom injection the levels of blood sugar free fatty acids and LDH activity increased significantly ( $P < 0.001$ ) (Table 1). Twenty h after insulin treatment these levels were found to be normal except for LDH level that remained high for some more time (Table 1).

The results of polyacrylamide gel electrophoresis of LDH isoenzymes pattern showed that following venom injection the rise in LDH level was mainly in LDH<sub>1</sub> and LDH<sub>5</sub> isoenzymes. Following insulin treatment the level of LDH<sub>1</sub> returned to normal whereas LDH<sub>5</sub> showed darker band (Fig. 1).

4 out of 5 rabbits, which did not receive insulin, died within 2 to 5 h whereas all the 8 rabbits that were treated with insulin survived.

## **Discussion**

In the present study insulin administration resulted in reversal of blood sugar, free fatty acids and LDH<sub>1</sub> isoenzyme to normal levels as well as prevention of death in envenomated animals. Eight units of crystalline insulin was used as a treatment dose. This was based on our previous studies of insulin therapy in scorpion venom poisoning(11). On the other hand, 1 to 2 units of insulin is usually suggested for reduction of 50 mg% of glucose in the blood.

Scorpion venom poisoning resulted in increased levels of blood sugar, free fatty acids, and various LDH, mainly LDH<sub>1</sub> and LDH<sub>5</sub>, isoenzymes. These results are in agreement with the results of our studies on envenomated dogs. No significant change was observed in the level of amylase. This was in contradiction with our previous report(10), which showed an increase in amylase level following the scorpion venom poisoning. However, this difference can be due to the route of venom administration as this was injected intravenously.

Results of our study showed the effectiveness of insulin in reversing the metabolic changes as well as preventing the death in poisoned animals. Four of the rabbits which did not receive insulin died and only 1 survived, whereas all 8 rabbits which received insulin survived. The surviving animals were observed for more than 8 weeks.

The use of insulin as a treatment for scorpion poisoning has been recently proposed by our laboratory(9,11). We had observed that insulin can reverse electrocardiographic and metabolic changes brought about by scorpion venom. However, our previous studies were restricted to a limited time of observation. In the present study a few more parameters associated with insulin therapy were included.

In the present study insulin therapy resulted in reversal of blood sugar, free fatty acids and LDH<sub>1</sub> isoenzyme to normal level within 20 h of insulin administration.

LDH<sub>1</sub> is heart specific enzyme and LDH<sub>5</sub> is liver specific enzyme(12). The reversal of LDH<sub>1</sub> to normal levels indicate normal function of the cardiovascular system. This is in agreement with our earlier reports that showed the reversal of electro-cardiographic changes, due to scorpion venom poisoning, to normal following administration of insulin(11). However, LDH<sub>5</sub> did not return to normal values sooner than 20 h after

insulin therapy. Interestingly, the results of our serotherapy also showed that the levels of ALT and AST remained high in antivenom treated animals, whereas all other parameters returned to normal levels within 120 minutes (results observed but not presented in this paper). It can be assumed that liver enzymes require more time to return to normal values.

The success of insulin in reversing the metabolic changes, due to scorpion venom, is mainly due to the important role of insulin in carbohydrate, fat and protein metabolism. The most important role of insulin is the reduction of concentration of extracellular glucose due to increased translocation of glucose, primarily in muscle and fat tissues, with concurrent inhibition of glucose efflux from the liver(14). Insulin also enhances glucose oxidation, glycogen synthesis, potassium influx, aminoacid transport and protein synthesis(14). In addition, in man it has been shown to inhibit the release of aminoacids from the muscles(16). Associated with its facilitating the glucose entry into adipose cells, insulin restrains lipolysis and enhances lipogenesis. These effects are along with decreased circulating free fatty acids, decreased blood glucose and elevation of insulin concentration in the blood(14).

In mouse soleus muscle, there are around 80  $\text{Na}^+ \text{-K}^+$  pumps for every insulin receptor(15). Studies with different tissues have indicated that insulin induces the stimulation of  $\text{Na}^+ \text{-K}^+$  ATPase(17). Insulin was reported to inhibit the action of epinephrine. Exton and Park found that the minimum concentration of insulin that inhibits  $1 \times 10^{-6}$  M epinephrine was  $100 \mu\text{u/ml}$ (18). Therefore, all these functions of insulin may have

Table 1. Effect of insulin after 90 min of venom administration on bloodsugar, free fatty acids, lactate dehydrogenase and amylase  
(Mean  $\pm$  S.E.D.)

	<i>BEFORE VENOM (13)</i>	<i>90 MIN. AFTER VENOM (13)</i>	<i>20 H. AFTER INSULIN (13)</i>
<b>BLOOD SUGAR</b>		****	****
(mg/dl)	126.66	326.80 $\pm$ 38.5	117.00 $\pm$ 23.62
%CHANGES		+158	-8
<b>LACTATE DEHYDRGENASE</b>		**	NS
(10 $\mu$ mol)	28.00	45.00 $\pm$ 5.7	48.00 $\pm$ 6.3
%CHANGES		+61	+71
<b>AMYLASE</b>		NS	NS
(U/dl)	60.00	57.00 $\pm$ 3.6	58.00 $\pm$ 9.5
%CHANGES		-5	-3
<b>FREE FATTY ACIDS</b>		****	****
( $\mu$ Mol/l)	330.25	728.80 $\pm$ 42.00	469.50 $\pm$ 35.00
%CHANGES		+121	-18

\*-P<0.05; \*\*-P<0.02; \*\*\*-P<0.01; \*\*\*\*-P<0.001

NS - NOT SIGNIFICANT. NUMBER IN PARENTHESIS = NUMBER OF ANIMALS.

% CHANGES: BETWEEN BEFORE VENOM AND ALL OTHER OBSERVATIONS.

S.E.D.: STANDARD ERROR OF MEAN DIFFERENCE.

COMPARISON OF RESULTS FOR BIOSTATISTICAL ANALYSIS;

BEFORE VENOM WITH 90MIN. AFTER VENOM

90 MIN. AFTER VENOM WITH 20 HRS. AFTER INSULIN.

contributed in reversal of various biochemical changes and prevention of death in insulin treated scorpion venom poisoned animals.

It seems, insulin therapy can be used effectively to overcome scorpion venom poisoning. However, it should also be emphasized that it is essential to correct the acid-base balance gaseous exchange, fluids and maintenance of vital functions in all the scorpion sting victims besides insulin therapy.

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