التأثير المرضى الوظيفى لسم ديور البلح
على الكلية والكبد

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أدى حقن سم ديور البلح فى الفئران الى اتساع وزيادة الدم الوارد

وكانت الاستحالات تتناسب من حيث درجتها وتوزيعها مع مقدار الجرعة ومدة تأثيرها . ولكن خروجا على هذه القاعدة كانت الجرعات المساوية لخمس وحدات من السم لا تتناسب من حيث مقدار الاستحالات الناتجة مع الكمية ومدة تأثيرها فى كل من الكبد والكلى .

وقد أعزى اتساع الاوعية الدموية وزيادة نفاذيتها الى وجود كل من الهيستامين والسيروتونين والكينينى وانزيم الهيالورونديز الموجودة فى تركيب السم . وقد أعزيت الاستحالات فى الخلايا البرنشيميه الى تأثير انزيم الفوسفوليبز .

وقد أظهرت دراسة كيمياء الانسجة الى وجود نقص فى نشاط التمثيل الغذائى البروتينى ، كذلك الى فقد وظيفة تخزين النشا فى الكبد . واقترح كتفسير لنقص سمية الجرعات الكبيرة من سم البلح على الكبد والكلى الى وجود مانع منافس لنشاط انزيم للفوسفوليبز .

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BIOLOGICAL STUDIES ON THE VENOM OF DATS WASP (VESPA ORIENTAIS)

III THE PATHOPHYSIOLOGICAL EFFECT ON THE LIVER AND KIDNEY

(With 6 Figures)

By

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SUMMARY

Injection of wasp venom in rats resulted in hyperaemia and increased vascular permeability in both liver and kidney. The parenchymatous cells of both organs manifested granular proteinous dystrophy and hydropic proteinous dystrophy up to focal lyses. The dystrophic changes were proportionated in distribution and severity according to the dose and time of application. An exception was with five stings dosage where the liver and kidney damage was neither proportionated with the magnititude of the dose nor with the prolonged time of application.

The vascular hyperaemia and permeability were related to the histamine, serotonin, kinins and hyalouronidase components of the venom. The parenchymal cell damage was related to the phospholipase A enzyme content of the venom. The histochemistry demonestrated proteinous metabolic hypoactivation and loss of the liver glycogenesis. A competive phospholipase inhibition was suggested for the decreased toxicity of higher doses.

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INTRODUCTION

Plenty of works have been done to investigate the natture and effects of wasp venom. Chemically, HABERMANN, and REIZ, 1965; reported that wasp venom centains histamine, serotonin, kinins, and the enzymes phospholipase A and phospholipase B and the hyalouronidase. HAMED et al. (1973) demonestrated that Egyption wasp venom contains 1.85 mg serotonin and free amino acids. Physiologically, MOHAMED et al. (1972) studied the effect of the venom on the anaethetised dogs. The venom resulted in a significant drop in blood pressure and a significant decrease in urine flow and a significant acceleration of respiration with no apparent change on the E. C. G. Phenergan blocked its action while allercur was only capable of blocking the lower doses of the venom. HAMED and MOHAMED (1975) studied the effect of the venom on the oxygen consumption by isolated tissue slices. Contradictive results were obtained. The venom produced a significant decrease in oxygen consumption by brain slices and significant increase in oxgen consumption by kidney tissue. The oxygen consumption by jujenal slices was increased only with two stings dose.

This contradictory physiological properties of the wasp venom specially with higher doses lead us to study structural changes of the tissue histopathologically and to correlate them with the functional changes of the tissue histochemically and physiologically in order to explain the actual way of tissue dammage by venom.

MATERIALS AND METHODS

Date wasps were collected from their nests during the date season from Assiut Governate. The entire venom apparatus was removed, cleaned and stored in deep freeze for use.

A definite number of the stings wetted with distilled water were ground in a small mortar. Additional amount of distilled water was added and the entire mixture was centrifuged. The supernatent fluid was collected and the volume was adjusted so that each ml contains the venom extract of 10 stings. Albino rats were divided into three groups (each of 9 animals). The venom prepared was injected intraperitoneally in the dose of one sting in the first group, two stings in the second group and five stings in the third group of rats. These rats from each group were decapitated 15,30 and 60 min. after venom injection. Specimens from the liver and kidney of each rat were fixed in neutral buffer formalin and carnoy fixatives. The materials were embeded in paraffin. Section of seven micron thickness were stained by: Hematoxyline and eosin, methyl green pyroinin, fulgen reaction, P. A. S. and Toludin blue. Control groups of nine rats injected with distilled water were used for comparison with the venom injected rats.

RESULTS

The pathophysiological effect on the liver:

15 min. aplication of one sting of the poison lead to a limited damage of the liver cell inform of granular proteinous dystrophy. The damage was clear and limited in the cells around the portal triad; center of the vascular lobule or the periphary of the anatomic lobule; as they are the first cells supplied by the blood borne toxins; Vasodilatation was clear in the central and portal veins with mucoid oedema of their wall, while the sinusoidal hyperaemia was slight. There was also dilatation of the periportal lymphatic space of male.

The desoxyribonucleic acid content of the nuclei was lowered, on account of the decrease in the number of dark

nuclei of the relatively highly active liver cells, in the zone of dystrophy and moderate staining of the other nuclei.

The swollen dystrophied cells lost the pyrininophilia totally or it was in a few granules in a ring around the nuclei.

The PAS reaction demonstrated that few lobules contained small amount of glycogen granules in the periphary of the cell cytoplasm. The rest of the lobules were free.

Application of one sting for 30 min. resulted in more severe cell damage manifested in form of diffuse granular proteinous dystrophy and foci of cell lysis near by or related to portal triad (Fig. 1). Also there were few cell foci of necrobiosis. The degree of hyperaemia was more or less the same. The lymphatic dilation increased to involve the pericellular spaces of Disse.

The desoxyribonucleic acid content was lowered to half its normal content on account of the decrease of the dark nuclei diffusely and focally and the necrobiosis and lysis of the nuclei. Pyrininophilia was decreased to half its normal centent. The liver was totally free of glycogen.

60 min. application led to a severe liver cell damage as more or less diffuse early hydropic proteinous dystrophy with foci of cell lysis.

The decrease in the desoxyribonucleic and pyrininophilia corresponded to the degree of cell dammage.

Only sporodic cells in some lobules and the peripheral zone of the others contianed some PAS positive granules while the rest of parenchyma was free.

The cell dammage was corresponding to the increase of the dosage. 2 stings application for 15 min. caused diffuse

granular proteinous dystrophy with more cells showing necrobiosis. 30 min. application of 2 stings initiated diffuse hydropic proteinous dystrophy of some lobules while the rest of the lobules suffered granular proteinous dystrophy. By 60 Min. application, the hydropic proteinous dystrophy was advanced,severe and diffuse (Fig. 2).

The decrease of desoxyribonucleic acid and pyrininophilia was correspondant to the severity of damage.

The glycogen content with 15 and 30 min. application was in form of few PAS positive granules present in the cytoplasm of sporadic liver cells (Fig. 3). Within 60 min. application the liver was totally free of glycogen.

The severity of liver damage was neither proportionated with the magnititude of 5 stings dosage, nor with the prolonged time of application.

5 stings application for 15 min. gave diffuse granular proteinous dystrophy but of slighter degree. The number of necrobiotic cells were fewer. The condition was more or less the same with 30 min. application. In 60 min. application only zones of light swelling of liver cells appeared in addition. (Fig. 4).

The desoxyribonucleic acid and pyrininophilia was correspondant to the amount of damage.

The glycogen was present in cells around the central veins of few lobules.

The pathophysiological effect on the kidney:

With one sting of the poison introduced for 15 min. only individual nephrons demonstrated granular proteinous dystrophy or mild hydropic dystrophy which was epecially pronounced

in the proximal convoluted tubules. The distal convoluted tubules demonstrated drivatives of haemoglobin pigment in form of fine granules in the cytoplasm and lumen.

In 30 min. application nearly all the nephrons manifested cloudly swelling (Fig. 5). 60 min. application resulted in severe degree of hydropic dystrophy of some nephrons on the back ground of diffuse cloudy swelling of the parenchyma.

2 stings application for 16 min. resulted in a magnitude of dammage equal to that of one sting spplication for 60 min. The histopathological picture was the same beside abundant haemoglobin corpusles and casts.

With 2 stings application for 30 min. the number of nephrons suffering hydropic degeneration became more while in 60 min. application the hydropic degeneration was diffuse. (Fig. 6).

Five stings application for 15 min. only resulted in slight damage. Few nephrons suffered cloudy swelling, the rest of parenchyma was more less normal.

5 stings application for 30 min. intiated diffuse cloudy swelling with hydropic degeneration of few nephrons.

60 min. application of 5 stings caused only mild degree of diffuse proteinous dystrophy.

Normal control kidneys and the kidneys under different doses and variable duration were free of glycogen. Also no pyrininophilia was observed. The ammount of desoxyribonucleic acid decreased and was proportional to the amount of damage.

DISCUSSION

Hyperaemia of the blood vessel in both liver and kidney was evident with the application of the smallest dose for the

shorter time (15 sec). Increased capillary permeability was manifested by oedema of the perisinusoidal space of Disse and the space of Male. The wasp venom contains mainly active principales that alter the vascular permeability of the tissue. HAMED et al. (1973) found that the wasp venom contains 38.5 mg histamine and 1.85 mg serotonin for each gram of the poison. JAQUES and SCHACHTER, (1954) discovered kinins in wasp venom. Histamin is the main factor controling permeability physiologically and in an inflammation. Serotonin the main intiator also seconderly increases permeability. WALTER and ESRAEL (1965) stated that kinins are cabable of causing vasodilation and increase the capillary permeability. They suggested that they are responsible for the important prolonged second phase of increased vascular permeability in an inflammation. In addition to histamine, serotonin and kinins present in wasp venom, JAQUES, 1955 established the presence of hyalouronidase enzyme with its characteristic effect on the vascular and connective tissue permeability. The presence of hyalouronidase explains the occurrence of mucoid oedema in the wall of the central and portal veins.

The action of wasp venom on the parenchymatous cells of the liver and kidney was dystrophic changes which varies in distribution and severity according to the dose and the time of application. The damage varied from regional granular protein dystophy to diffuse and diffuse hydropic dystrophy. In the pathogenesis of the granular proteinous dystrophy and hydropic distrophy, hypoxia lead to dissociation of lipoprotion complex of the metochondria and of the endoplasmic reticulum. As it passes to hydropic changes there is increase accumulation of fluid in the cytoplasm which is partely due to osmosis and mainly due to large increase in the cell wall permeability. DAVYDOVSKY, (1971). This alteration explained

only the back ground of the biological action of the phospholipase A content of the wasp venom. HABERMANN and REIZ,(1965) found that phospholipase constitute 12% of the dry venom mass. Phospholipase A splits one fatty acid from phosphatidyl compounds thus leaving monoachylophosphatid as a lysocompound. These lyso-compound causes universal membrane damage.

It causes increased permeability of the muscle cells (HEYDENREICH, 1957), perfused organs (KELIAWAY and TRETHEWIE, 1940) and brain (GAUTRELET and CORTAGGIANT, 1939). Phospholipase inactivates the respiratory enzymes either directly or through the liberation of lysolecithins and its action on the mitochondria where lipids are intergal parts (HABERMANN and ZPURER, 1971). Phospholipase directly inhibits oxidative phospherylation (HABERMANN, 1954). Thus phospholipase A is the compenent responsible for the intiation of granular proteinous and hydropic dystrophy of the liver and kidney cells.

The presence of haemoglobin pigments in the kidney tubules was due to the haemolytic effect of the venom. This can be also related to the action of phospholipase A (HAVERMANN , 1954). Phospholipase A acts on plasma lecithines and liberates lysoicithins which will increase the permeability of erythrocytic membrane leading to swelling and haemolysis.

The severity of the liver and kidney damage was neither proportionated with the magnitude of 5 stings dosage nor with prolonged time of application for this dose. Similar results were obtained by HAMED and MOHAMED (1975) where the venom increased the oxygen consumption of jujenal slices only when the dose of 2 stings is applied. Five stings have no influence. They explained this contradictory results by the complex composition and the different components present in this venom. The histopathological results demonestrated that

the damaging effect on the liver and kidney cells was mainly due to phospholipase A content of the venom. It is suggested that there is a competive enzyme inhibition with the higher doses either from the other venom components or from products of their action. Further work is suggested for isolation and identification of this component. Steroisomers may be used for competive inhibition of the enzyme as antidote.

The desoxyribonuclic acid content of the nuclei and the ribonucleic acid content (pyrininophilia) were decreased correspondingly to the degree of cell damage in both liver and kidney. Wasp venom with its content phospholipase A not only caused structural changes in the parenchymatous cell but also proteinous metabolic hypoactiveation which was completely arrested in the zone of focal liver cells lysis. Abscence of glycogen in the liver indicated the loss of important liver function glycogenesis. The liable energy depo was exhausted in such toxication.

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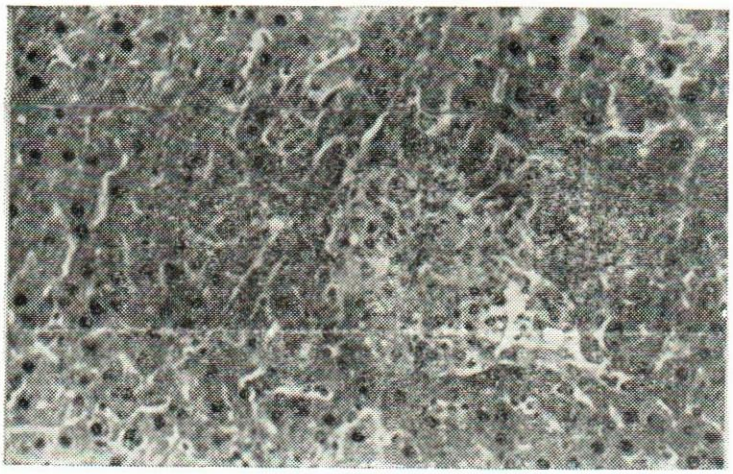

Figure [1]

One stings application for 30 second. Diffuse granular proteinous dystrophy and foci of cell lysis. H, E. [10 x 12.5].

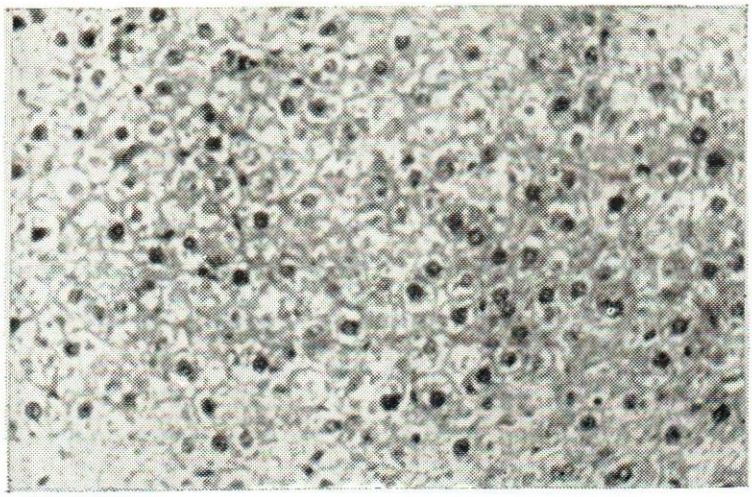

Figure [2]

Two stings application for 60 second. Diffuse and advanced hydropic dystrophy. H, E. [10 x 12.5].

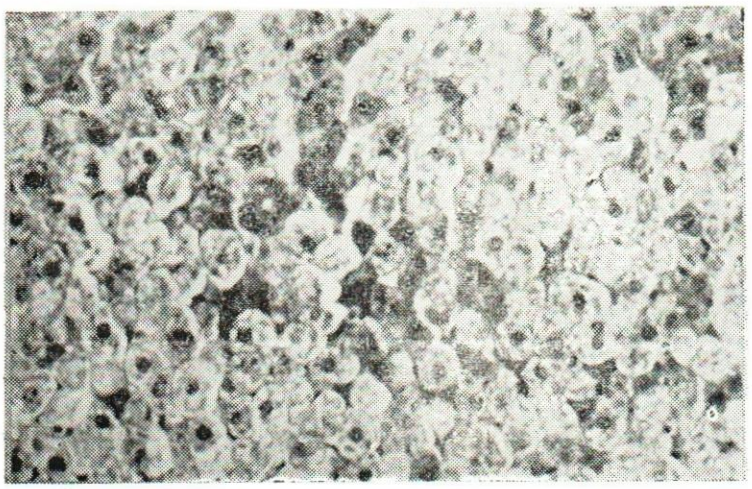

Figure [3]

Two stings application for 60 second. Few PAS + granules present in the cytoplasm of sporadic liver cells.

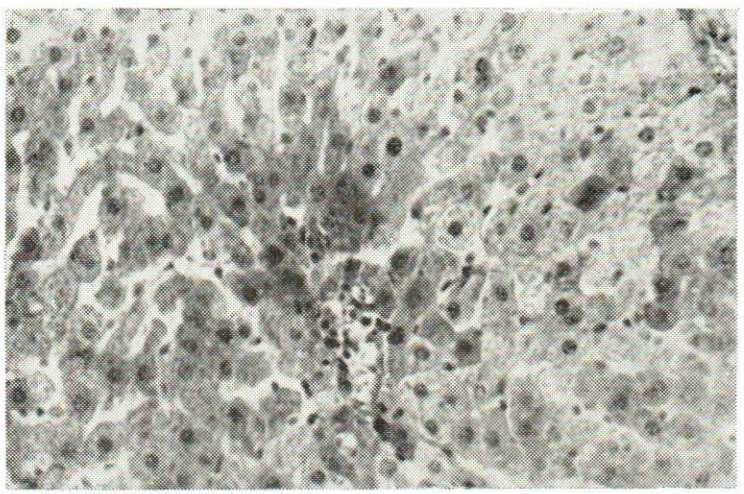

Figure [4]

Five stings application for 60 second. Zones of light swelling of the liver cells.
H, E. [10 x 12.5].

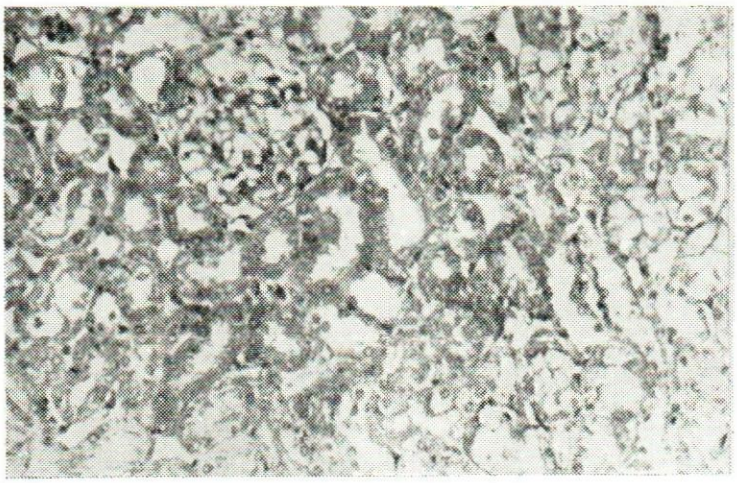

Figure [5]
One sting application for 30 second. Diffuse granular proteinous dystrophy. H, E. [0 x 12.5].

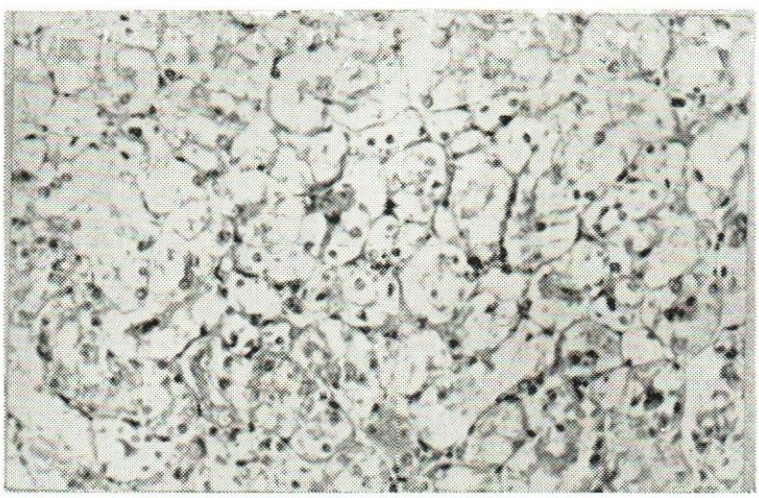

Figure [6]
Two stings application for 60 second. Diffuse hydropic dystrophy. H, E. [10 x 12.5].