

Histopathological change in the testis of the desert locust *Schistocerca gregaria* (Forsk.) induced by the IGR Consult and Lufox

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ABSTRACT

Histopathological changes in the testes were tested in normal adult males and those developed from treated one day old of the fifth nymphal instar of the desert locust with (LC₅₀) of Consult and Lufox.

Both testes are formed each of numerous testicular follicles containing germ cells in different stages of development. Certain aspects of spermatogenesis in the desert locust *S. gregaria* are described in this work using light microscop. Spermatogenesis commences by the division of spermatogonia giving spermatocytes which in turn give spermatids. The transformation process of spermatid to sperm involves several morphological reorganization of the cells.

The testicular follicles of those males developed from treated one day old of the 5th nymphal instar with Consult (LC₅₀) showed damage in zones of reduction and transformation and degeneration and necrosis appeared in many spermatids and spermatozoa. However those testicular follicles of males developed from treated 1 day old of the 5th nymphal instar with Lufox LC₅₀ showed severe degeneration and necrosis of the most spermatogenic stages.

Keywords: Locust, *Schistocerca gregaria*, testis, histopathology, IGR (Consult-Lufox).

INTRODUCTION

Plagues of desert locust, *Schistocerca gregaria* (Forsk.) have been recognized as a threat to agricultural production in Africa and Western Asia for thousands of years.

IGRs are diverse groups of chemical compounds that are highly active against immature stage of insects and have a good margin of safety to most non-target biota including invertebrates, fishes, birds and other wild life, they are also safe to man and domestic animals, they will play an important role in control programs in the future (Mulla, 1995).

The main types of insect growth regulators used commercially are juvenile hormone analogues and chitin synthesis inhibitors (Parrella and Murphy, 1998).

Histological studies of gonads may provide morphological evidence for functions of gonial, extragonial tissues and cells.

The aim of the present study is to examine the histopathological changes occurred in the testes of the male *S. gregaria* developed from treated 1 day old of the 5th nymphal instar with sublethal concentration (LC₅₀) of two compounds of insect growth regulators; Consult (chitin synthesis inhibitor) and Lufox (mixture of juvenile hormone mimic and chitin synthesis inhibitor).

MATERIAL AND METHODS

1-Origin of population:

The stock colony of *S. gregaria* was maintained for several years at the Luocst Research Division, Plant Protection Research Institute, Agricultural Research Center, Dokki, Giza. The insects were reared and handled under the following technique described by Abbassi *et al.* (2003). Leaves of leguminous plant, *Medicago sativa* were daily placed as feeding material. The cages were incubated in a constant room temperature ($32\pm 2^{\circ}\text{C}$) and (30-50RH).

2- Insect growth regulators used:

Two analogues of insect growth regulators (IGRs) were used:

2-1. Hexaflumuron (10% EC) (Chitin synthesis inhibitor):

Consult (Hexaflumuron), [N(((3,5-dichloro-4-(1,1,2,2-tetrafluoroethoxy) phenyl)-amino) carbonyl)-2,6- difluoroben-zamide].

2-2. Lufox: mixture of juvenile hormone mimic (fenoxycarb 7.5% EC), ethyl [2-(4-phenoxyphenoxy) ethyl] Carbamate and Chitin Synthesis inhibitor (lufenuron), (Axor 3% EC). N-[[[2,5-dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy) phenyl] amino] carbonyl] -2,6-difluorobenzamide.

Consult and Lufox have proved to be toxic to one day old of 5th nymphal instar *S. gregaria* (Bakr *et al.* 2009).

3- Experimental insects:

The First day of the 5th nymphal instar of *S. gregaria* were treated with the estimated LC_{50} of Consult and Lufox. The living individuals completed the development to be adults were studied as the treated generation.

The testes of the tenth day of males were dissected.

These testes were used in the light microscope histological study.

4- Light microscop studies:

The adult males were dissected out in Ringer's solution. The male reproductive organs were isolated from the freshly dissected insects.

Bouin's fluid was used to fix the male reproductive organs. The latter were then dehydrated in a series of ethanol, then cleared in xylene and embedded in paraffin wax. Serial sections, 5-7 μm were stained with haematoxylin and eosin, then cleared mounted in DPX.

RESULTS AND DISCUSSION

1) Testis of normal male:

The male gonads consist of two testes fused to a single oval body. Each testes is formed of tubular (testicular follicles), each follicle is enclosed by a layer of epithelial cells. It contains few apical cells followed series of successive zones in which the germ cells are in different stages of development. containing spermatogonia, spermatocytes spermatids and sperms, respectively.

2) Structure of testicular follicle of normal male:

The testis is enclosed in transparent non pigmented membrane, the peritoneal membrons. The testis follicle are composed of aggregation of large sacs. The adjacent follicles are separated by epithelial septa Fig. (1).

Germinal cells:

The testicular follicle is filled with germinal cells in different stages of development. These include spermatogonia, spermatocytes, spermatids, and

spermatozoa. The sperm cysts are formed by the grouping of germ cells of the some stage.

1- Spermatogonia

These are spherical cells have dark stained round nuclei Fig. (1).

2- Spermatocytes

Spermatocytes are generally larger than spermatogonia but with less intensity stainable nuclei Fig. (1).

3- Spermatids

Young spermatids are relatively small in size with intensively stainable round nuclei which are much smaller than those of the spermatocytes Fig. (1).

4- Sperm

The sperm differentiated from the presperm by the great elongation of the tail and further thinning of the nucleus, sperm bundle disperses in the **interstitial** tissue within one bundle, sperms are aligned, roughly parallel to one another and oriented in the same direction Fig. (1).

3) Histopathological studies:

The histopathological examination of the testicular follicle of adult male *S. gregaria* developed from treated 1- day old of the 5th nymphal instar with Consult at LC₅₀ showed damage and vacuoles in zones of reduction and transformation. So the degeneration and necrosis appeared in many spermatids and spermatozoa. Fig (2).

Histopathological abnormalities were appeared in the testis of adult male *S. gregaria* developed from treated 1 day old of the 5th nymphal instar with Lufox LC₅₀. Degeneration and necrosis of testicular germ cells, spermatogonia, spermatocytes, spermatids and spermatozoa was clearly obvious Fig. (3).

As in most other orthoptera, the testicular follicles in *S. gregaria* are composed of aggregations of sessile sacs. The various zones of development described in the testicular tubules of other insects (Snodgrass, 1935; Wigglesworth, 1972 and Chapman, 1973) are clearly distinguishable in male *S. gregaria*

The present study showed histopathological abnormalities in testes of ten day old adult males. Degeneration and necrosis in spermatogenic stages and inhibition in the formation of sperm bundles were observed.

These results agree with that on *L. migratoria* and grasshopper *P. pictus* after treatment with Apholat (Saxen & Aditya, 1969 and Sheikher & Mittal, 1986); on *L. migratoria* treated with Hempa (Nath *et al.*, 1976), on *E. insulana* treated with pyriproxyfen (Hussein *et al.*, 1993) and on 4th and 5th nymphs of *Heteracris litoralis* (Orthoptera) when treated with Azadirachtin (Ghazawi *et al.*, 2007).

Finally, it is worthy to know that protein presented in all viable cells is essential to the process of cell division control of many chemical reactions in the metabolism of cells. Application of different IGR's (Consult and Lufox) caused a disturbance in the chemical reactions of cell metabolism, which in turn resulted an inhibitory action on the biological characters that appears as morphological and histological malformation in all different treated locust tissues.

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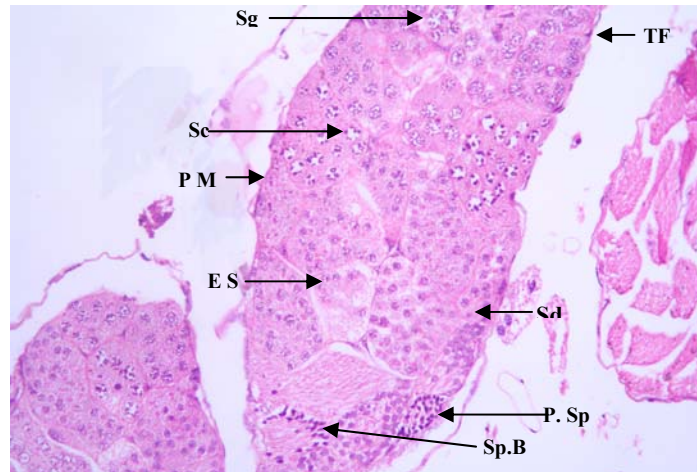


Fig. 1: light micrograph of longitudinal section of testis of normal male desert locust , *Schistocerca gregaria* shown typical testicular follicles (TF) with successive zones of spermatogonia (Sg), spermatocytes (Sc), spermatids (Sd) and presperm (P.Sp), sperm bundle (Sp.B) , peritoneal membrane (PM) and epithelial septa (ES). (H&E. x400)



Fig. 2 : light micrograph of longitudinal section of testis of desert locust , *Schistocerca gregaria* developed from treated one day old of the 5th nymphal instar with LC50 of Consult shown degeneration and necrosis in the spermatogonia(Sg) , spermatocytes (Sc) spermatids (Sd) and sperm bundles (Sp.B) (H&E, X400)

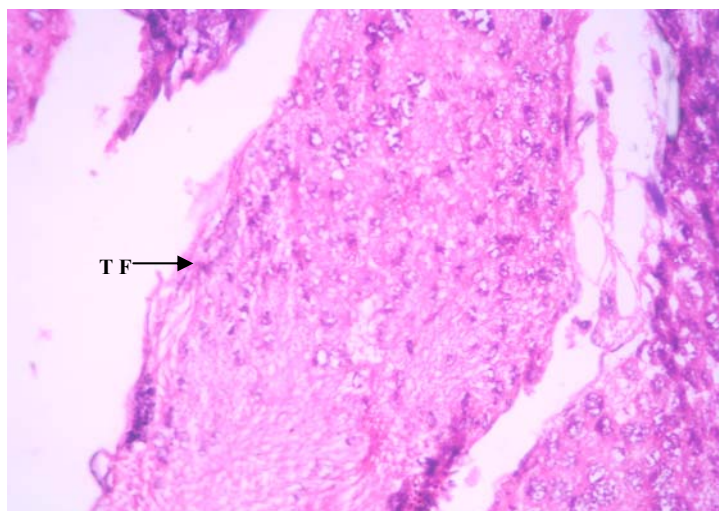


Fig. 3: light micrograph of longitudinal section of testis of desert locust , *Schistocerca gregaria* developed from treated one day old of the 5th nymphal instar with LC50 of Lufox shown severe degeneration and necrosis in the most germ cells of testicular follicle (TF), spermatogonia (Sg), spermatocytes (Sc), spermatid (Sd) and sperm (Sp). (H&E, X400)

ARABIC SUMMARY

التغيرات النسيجية فى خصية الجراد الصحراوى شيستوسيركا جريجاريا الناتجة عن منظمات النمو الحشرية الكونصلت واللوفكس

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تم دراسة التغيرات النسيجية فى خصية الطور اليافع للجراد الصحراوى بعد عشر أيام من انسلاخها من العمر الخامس المعامل أول يوم بالكونصلت واللوفكس. وأوضح الفحص النسيجى للحويصلات الخصوية المعاملة بالكونصلت خلل فى الانقسامات الخلوية فى منطقة النضج والاختزال مما أدى إلى حدوث تحلل وأماتة لمعظم طلائع المنى والحيوانات المنوية مما ينتج عنه عقم الذكور غالباً. كما أظهر الفحص النسيجى خلل فى كل مراحل الانقسامات الخلوية فى الحويصلة الخصوية المعاملة باللوفكس مما أحدث تحلل وأماتة شديدة لمعظم الخلايا المنوية وكذلك الطلائع المنوية والحيوانات المنوية مما يؤدى الى ظهور ذكور عقيمة.