Commun. Fac. Sci. Univ. Ank. Ser. C
V, 4. pp, 61 - 79 (1986)

THE EFFECTS OF THIOTEPA ON THE FINE STRUCTURE OF THE OVARIUM OF THE LAST INSTAR LARVAE OF AGROTIS IPSILON (HUFNAGEL) (LEPIDOPTERA: NOCTUIDAE)

ZEKİYE SULUDERE* and SEVİNÇ KAROL**

* *Department of Biology, Faculty of Art and Science, University of Gazi, Ankara, Turkey*
** *Department of Biology, Facutly of Science, University of Ankara, Turkey*

(Received in June 1986, Accepted in September 1986)

ABSTRACT:

Thiotepa was applied at two doses (0.01 % and 0.1 %) and for 3 or 10 minutes to the abdomens of *A. ipsilon* larvae.

The coating layers of ovaries are distinctly affected by 0.1 % Thiotepa for each period. The cells in the ovarioles show, also, degenerations after using 0.01 % and 0.1 % Thiotepa. 0.01 % Thiotepa application for three minutes has a less effect. The increase in concentration as well as the duration of application of Thiotepa increases also the degeneration more severely in the cells of ovarioles. A complete destruction is obtained after the application of 0.01 % Thiotepa for 10 minutes.

The apperances of these degenerations are shown with electronmicrographs in detail Thiotepa is a very effective chemosterialant for *A. ipsilon.*

INTRODUCTION

One of the harmful larvae of the insects in the fields is that of *Agrotis ipsilon.* In order to control this species several methods have been tested including insecticides, juvenoids, parasites, microorganisms, and some antifeedants. Aluminium sulphate and ammonium sulphate are the most effective salts which were used as deterrent agent against *A. ipsilon* in the laboratory experiments (DIMETRY and MANSOUR, 1975). Duter, Duter tetra, Brestan, Brestanol and Suzo are also some commercial substances which were used for inhibition of feeding of insects (MEGEED et al., 1974). Insecticides and juvenoids were used on some species of Noctuidae. The effective dose for *A. ipsilon* has been

ISSN 0256 - 7865, A. Üniv. Basımevi

found higher than that of the other noctuids (SEHNAL et al., 1976; STREIBERT and DITTRICH, 1977). Pathogenic microorganisms and insect parasites were also found to be pathogenic to *A. ipsilon* (IGNOFFO and GARCIA, 1979; JOHNSON and LEWIS, 1982; SCHOENBOHM and TURPIN, 1977).

Chemosterilants have been started to use for insect control. BORKOVEC (1962, 1966) and CAMPION (1972) reviewed these works. The effects of chemosterilants on insects were generally evaluated with the decrease of the number of the eggs laid and with the percentage of egg hatchability (CRYSTAL and LaCHANCE, 1963; COLLIER and DOWNEY, 1965; LaBRECQUE, 1972; ANWAR et al., 1974; OSMAN et al., 1975; STEFAN and STUEBEN, 1976). Metepa and Duter were examined as chemosterilants on *A. ipsilon* by SHAABAN et al., (1975).

The effects of chemosterilants differ according to the larval instars (SALAMA, 1976; SHARMA and THERIAULT, 1980) and to the structure, concentrations, methods, durations of the chemosterialants during the treatments (CHANG et al., 1964; COLLIER and DOWNEY, 1965; BORKOVEC et al., 1968; CHAUDHARY and KAPIL, 1976, 1977; TERRY et al., 1977; HAFEZ et al., 1978; GÜVEN, 1979).

The degeneration and the decrease of the volume of ovaries and testes caused by chemosterilants were shown on *Blatella germanica* (SMITTLE et al., 1966), on *Musca domestica* (MORGAN, 1967), on *Oncopeltus fasciatus* (ECONOMOPOULOS, 1971) and on *Trichoplucia ni* (HENNEBERRY et al., 1972). With the effect of chemosterilant, the vacuolisation of cytoplasm and nucleus, the aggregation of chromatin, the aberation of chromosome and the differentiation of cell content of different parts of reproductive organs occured (LANDA and REZABOVA, 1965; CLINE, 1968; LaCHANCE and LEVERICH, 1968; KLASSEN et al., 1969; SAXENA and ADITYA, 1969, 1974; WILSON and HAYS, 1969; ADDY, 1970; LANDA and MATOLIN, 1971; NATH and SHEIKHER, 1976).

Thiotepa, as an anticancerogen drug for the treatment of several cancerogenic tumors and Hodgkin disease, has also been shown that it could be used as a chemosterilant for insects (LANDA and REZABOVA, 1965; PARNELL and METTRICK, 1969; OSMAN et al., 1975; CHAUDHARY and KAPIL, 1976, 1977; TERRY et al., 1977; LADD et al., 1980).

On the control of *A. ipsilon*, a successful result has not been obtained yet. For this reason, it is necessary to try other methods on this insect.

There is no study with chemosterilant on the ovary of *A. ipsilon*. The aim of this work is to show the effect of Thiotepa on the structure of ovaries of the last instar larvae.

MATERIAL and METHODS

The methods of collection and rearing of the larvae and the method used for electron microscopy are the same as described in the previous paper (SULUDERE, 1986).

Thiotepa was prepared at 0.01 % and 0.1 % concentrations in distilled water. Thiotepa was dissolved till the liquid seemed clear. Thiotepa was prepared pior to use.

The abdomens of, at least, three specimens for each serie were dipped in 0.1 % Thiotepa or 0.01 % Thiotepa for 3 or 10 minutes. Distilled water was used for control experiments since it was the solvent of Thiotepa. After these treatments, the excess liquid on the abdomen was absorbed with filter paper, then the larvae were kept in normal conditions for 24 hours. Then the larvae were dissected and ovaries were prepared for the TEM observation.

RESULTS

The ovaries from the larvae which were dipped in 0.01 % or 0.1 % Thiotepa for 3 or 10 minutes and were dissected after 24 hours, have not shown any distinguishable differences with naked eyes. However, when they were studied histologically, the thin sections of these ovaries have shown some differences.

0.01 % Thiotepa for 3 minutes has not affected the sheet of ovary, the loose and the dense connective tissues, lamina, epithelial layer and tunica propria. But some of the cells in the ovarioles have started to degenerate. This degeneration has shown itself as a disintegration and vacuolisation in the karyoplasm (Fig. 1). There is no visible change in the nucleolus (Fig. 2). In some cells, there are some cytoplasmic

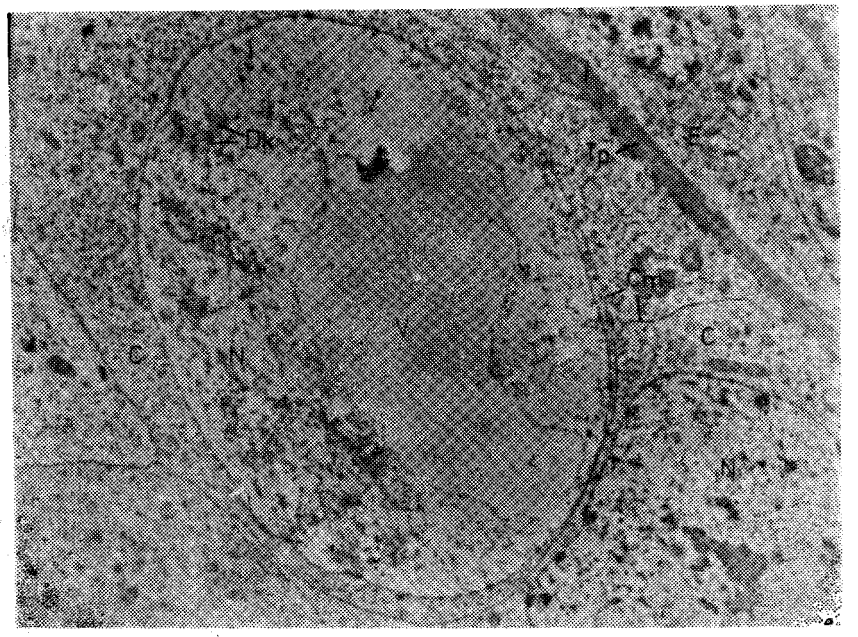

Figure 1. The vacuolisation and the disintegration of karyoplasm of the nucleus in the germ cell after the treatment with 0.01 % Thiotepa for 3 minutes. 5000X

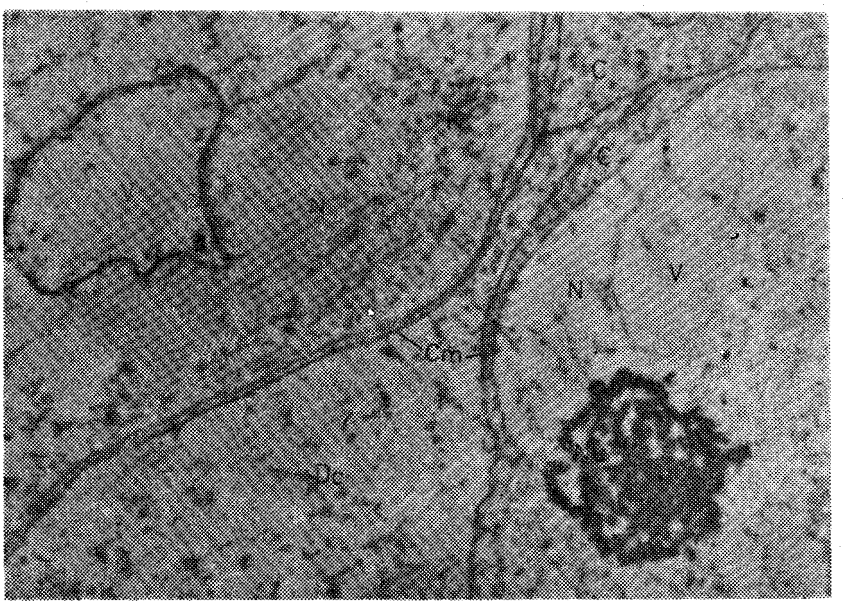

Figure 2. The vacuoles in the nuclei of germ cells and a nucleolus without any destruction after the treatment with 0.01 % Thiotepa for 3 minutes. 10 000 X

degeneration. Some vacuoles with incomplete membrane, some amorphous and membranous structures are typical apperances of these degenerations (Fig. 3). Sometimes, a big amorphous mass surrounded

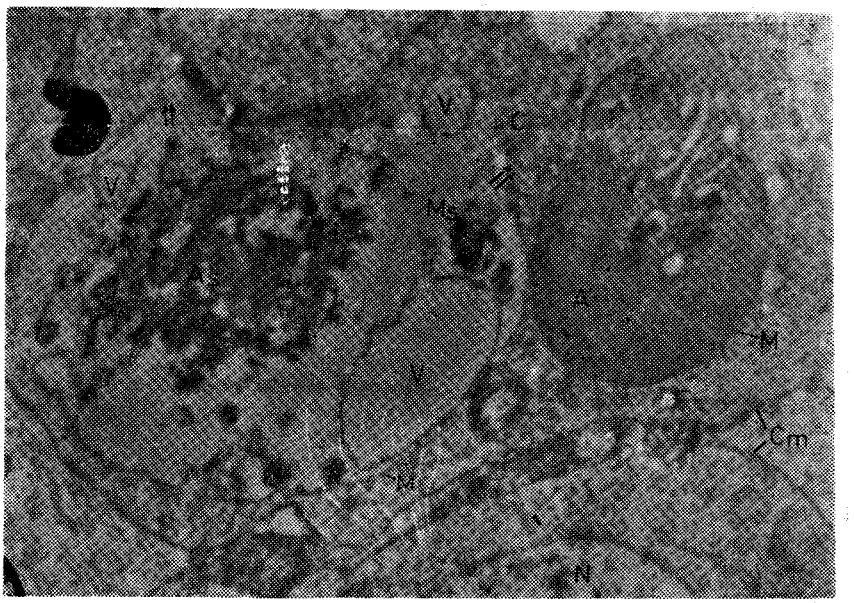

Figure 3. The degeneration of the cytoplasm of the germ cells and vacuoles with incomplete membrane (⇒) after the treatment with 0.01 % Thiotepa for 3 minutes. 7 000 X

with a membrane and several small vacuoles can be seen in the cytoplasm of the same cell. In the amorphous mass, small membranous structures the origin of which are not known, some unstructured places and less dense regions can be recognized.

The looseness is generally discerned in the layers of ovarian coat with the treatment of 0.01 % thiotepa for 10 minutes. The most severe pathological damage was occured inside of ovarioles. In addition to the normal cells, slightly deformed or completely degenerated cells have appeared (Fig. 4). There were some accumulated materials and vacuoles in the nuclei of slightly deformed cells. There were not any visible change in the fusome material and at the intercellular birdges. The plasma membranes of some cells have disappeared in various places. Their flattened nuclei were pushed aside and their cytoplasm had a disintegrated part and some accumulated materials (Fig. 4, 5).

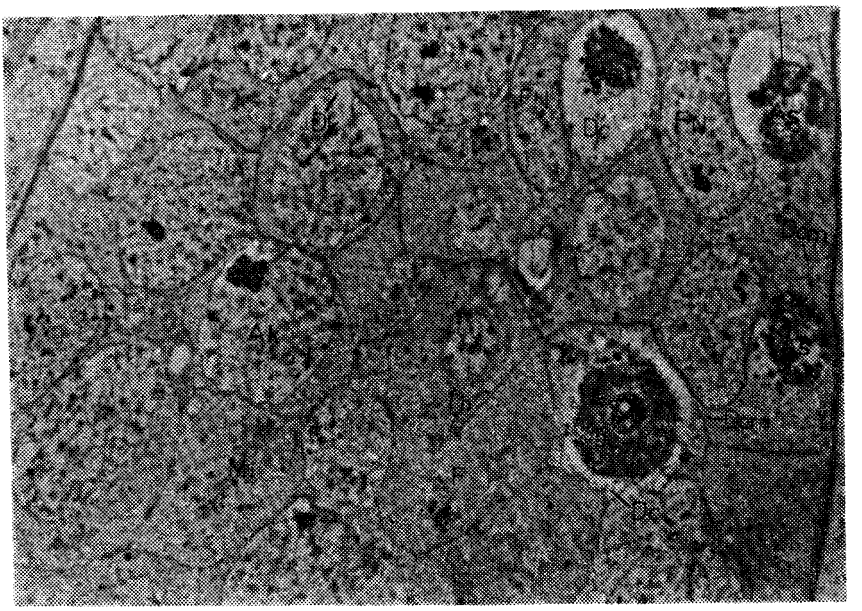

Figure 4. A longitudinal section of ovariole after the treatment with 0.01 % Thiotepa for 10 minutes. Slightly deformed or completely degenerated cells are seen. 2 000 X

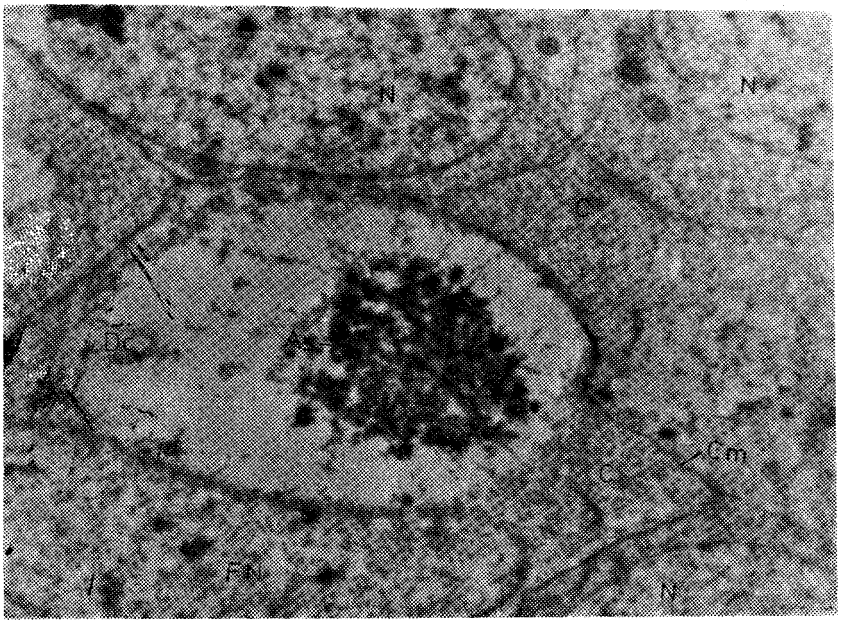

Figure 5. A flattened nucleus and a disintegrated cytoplasm of the germ cell after the treatment with 0.01 % Thiotepa for 10 minutes. 7000 X

Even an amorphous structure with a surrounded membrane and a dense center observed in the disintegrated cytoplasm of some cell (Fig. 6). The size of this amorphous structure may reach up to

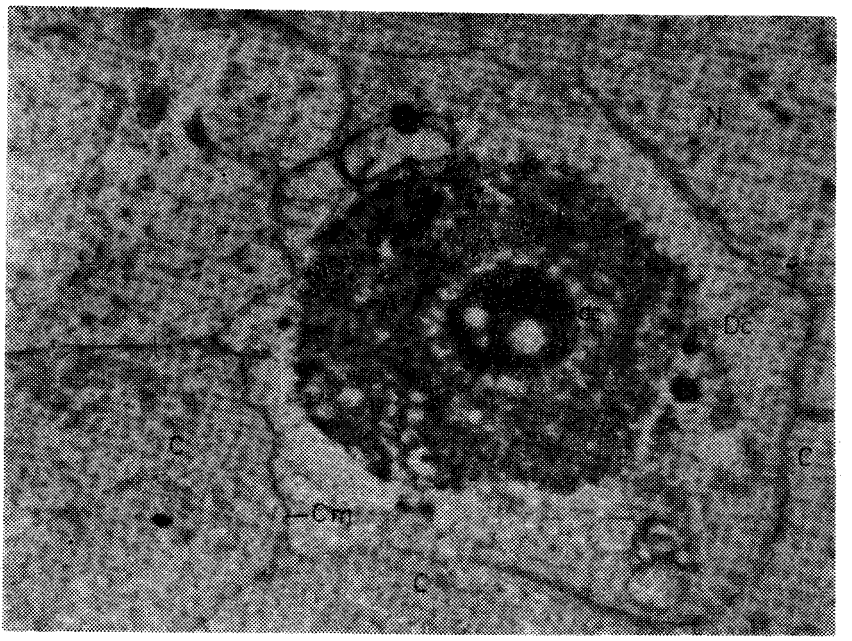

Figure 6. An amorphous structure within the degenerated cytoplasm of the germ cell after the treatment with 0.01 % Thiotepa for 10 minutes. 7 000 X

7 μ. In the cytoplasm of some cells, some small vacuoles and a large sized structure surrounded with a membrane were seen. There were not empty place in the cytoplasm of these cells (Fig. 7). A dense region and numerous membranous structures at various size were observed in this large sized structure. At this concentration of Thiotepa the karyoplasm of the pedicel cells have shown disintegration as well (Fig. 8).

The more concentrated Thiotepa caused the more damage in the cells. The treatment of 0.1 % Thiotepa for 3 minutes has started the destruction of ovarian coats. The disintegration and the vacuolisation of the cytoplasm of the cells of the loose connective tissue inc-

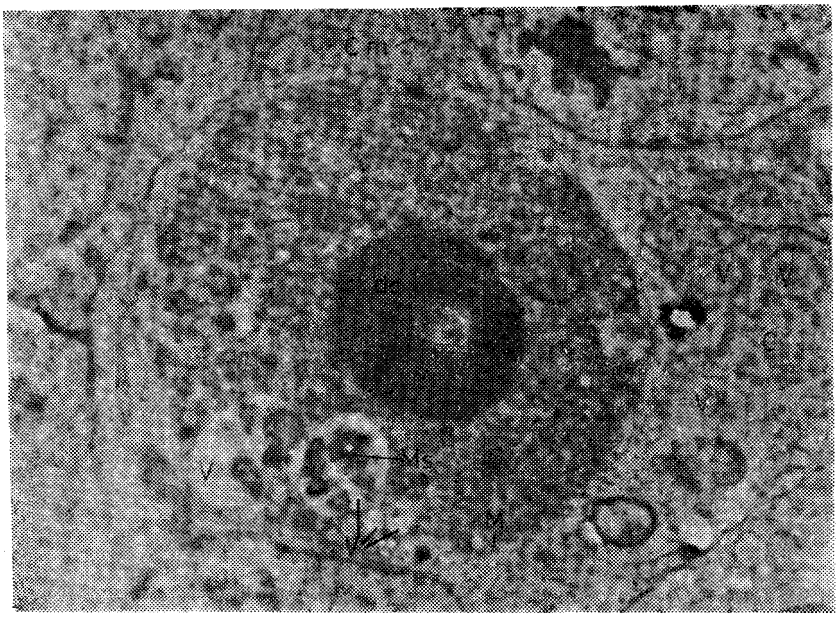

Figure 7. The membranous structures with dense center in the cytoplasm of germ cell after the treatment with 0.01 % Thiotepa for 10 minutes. 7 000 X

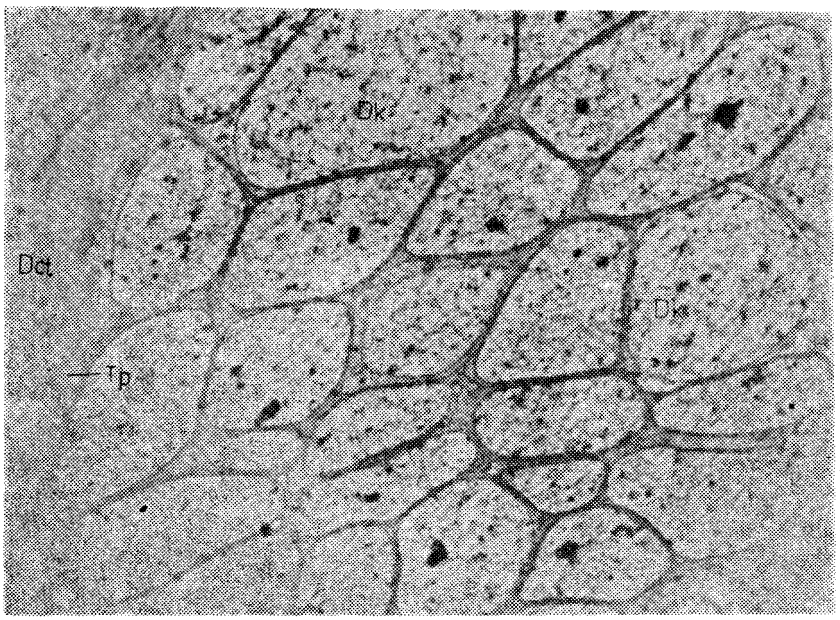

Figure 8. The disintegration of karyoplasm of the pedicel after the treatment with 0.01 % Thiotepa for 10 minutes. 3 000 X

reased and the intercellular spaces were widened (Fig. 9). All the cells of ovarioles seemed to be affected by Thiotepa (Fig. 10). The least

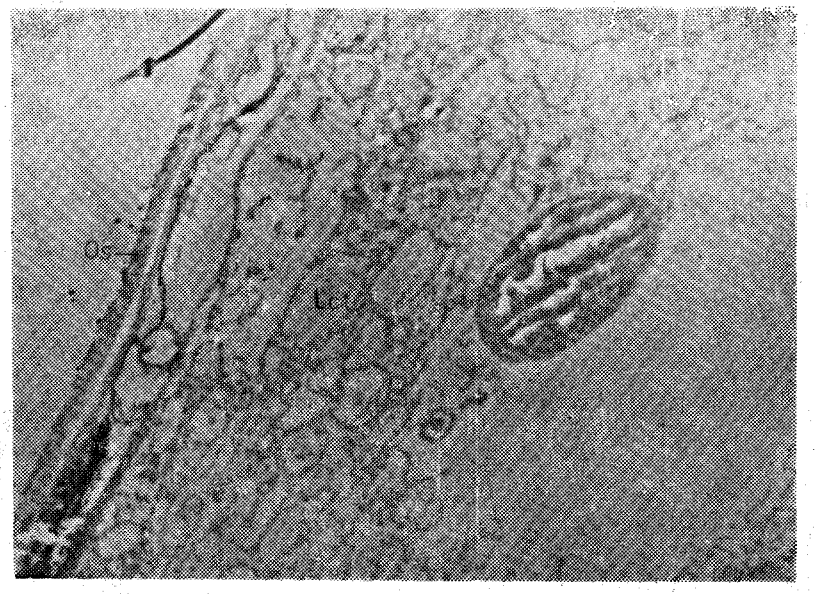

Figure 9. The disintegration and vacuolisation in the cytoplasm of the loose connective tissue cells after the treatment with 0.1 % Thiotepa for 3 minutes. 5 000 X

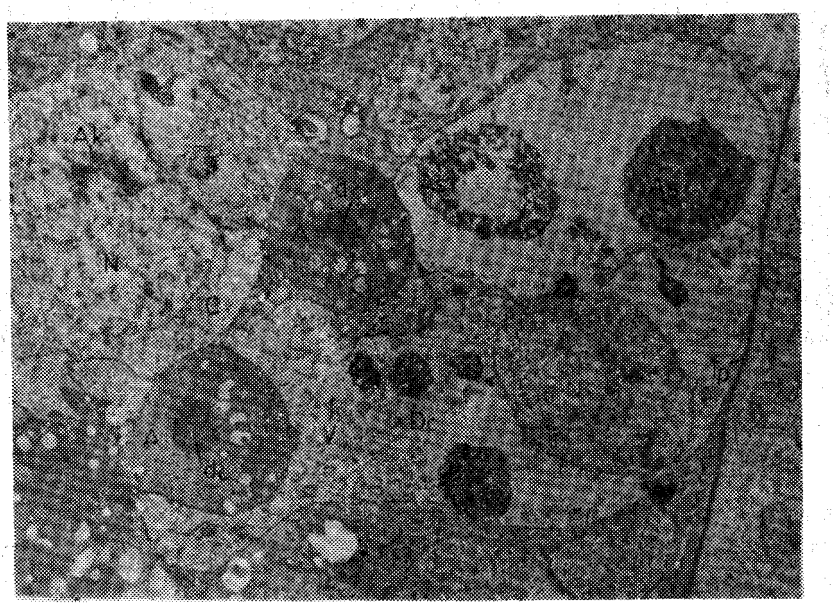

Figure 10. The degeneration of the cells in the ovariole after the treatment with 0.1 % Thiotepa for 3 minutes. Some amorphous structures with dense center are seen. 3 000 X

affected cells also displayed disintegration and accumulation to a certain extent in various places. Cytoplasmic organelles were displaced by small vacuoles. Mitochondria have distributed all over the cell instead of getting acumulated in untreated cells. But sometimes a few mitochondria have observed together (Fig. 11). Small vacuoles and small or large amorphous structures surrounded with a membrane have seen in the cytoplasm of some cells. These amorphous structures had a dense center and some vesicles. Some accumulated and granulated materials were observed in other cells (See Fig. 10, 11).

When the solution of 0.1 % Thiotepa was applied for 10 minutes, the destruction of the ovarian structure has increased. In addition to the enlargement of the intercellular spaces in the loose connective

Figure 11. The degeneration of the cells in the ovariole after the treatment with 0.1 % Thiotepa for 3 minutes. Amorphous structures with a surrounded membrane and some vacuoles are seen. Some mitochondria are observed together. 3 000 X

tissue (Fig. 12), the dense connective tissue which were among the ovarioles showed a distinct looseness as well (Fig. 13). The cells surrounding tracheae and tracheoles are separated from their attached parts.

Figure 12. The loose connective tissue of ovarium after the treatment with 0.1 % Thiotepa for 10 minutes. The enlargement of intercellular spaces are seen. 5 000 X

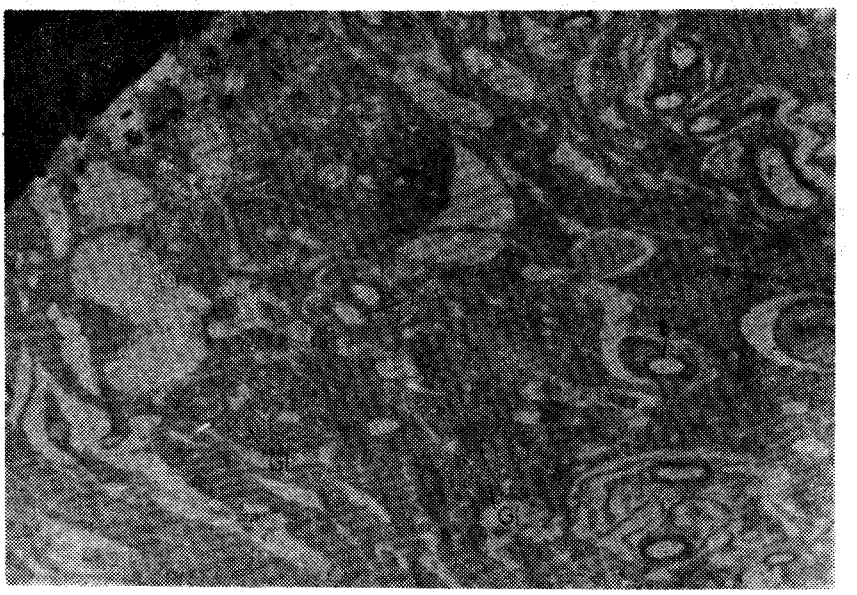

Figure 13. The looseness of dense connective tissue after the treatment with 0.1 % Thiotepa for 10 minutes. 7 000 X

The cells of ovarioles displayed a complete degeneration. Some structures which are not known their origins have arised (Fig. 14).

Figure 14. Some structures with unknown origin in the ovariole after the treatment with 0.1 % Thiotepa for 10 minutes. 10 000 X

The cytoplasm around the amorphous structure is completely disintegrated in some cells. The amorphous structure is round and homogenous (Fig. 15).

DISCUSSION

When Thiotepa is applied to the *A. ipsilon* larvae, any malformation could not be seen with naked eyes in the ovaries. Twenty-four hours is not a long period to see any shrinkage which would be produced after the Thiotepa treatments. ECONOMOPOULOS (1971) observed the shrinkage of ovaries after five days following Tretamin application in *O. fasciatus*. In *A. ipsilon*, since the size of ovaries is very small, 1–1.5 mm, such an effect is not possible to be observed with naked eye.

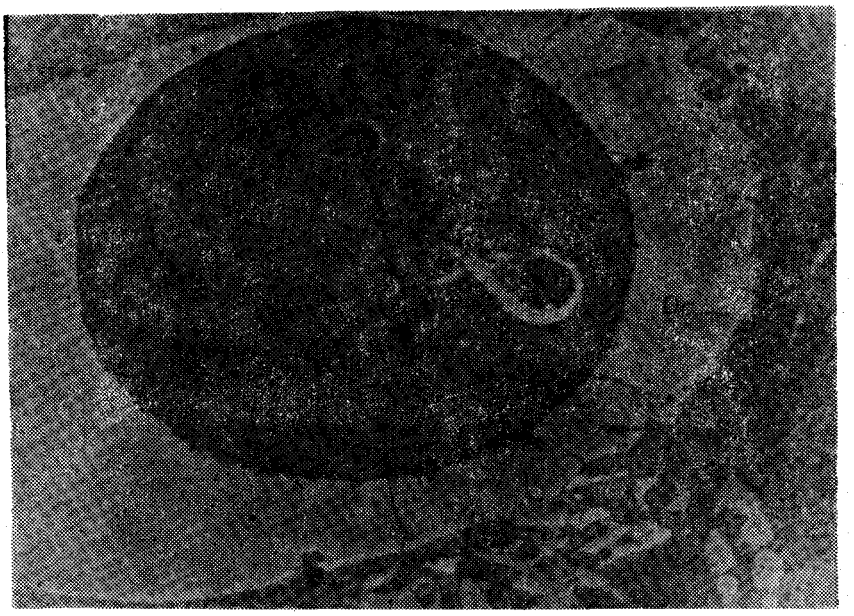

Figure 15. An amorphous structure in the cell of ovariole after the treatment with 0.1 % Thiotepa for 10 minutes. 10 000 X

The effect of Thiotepa on the cells of *A. ipsilon* ovarioles are clearly seen by electron microscope. After the treatment of 0.01 % Thiotepa for 3 minutes, there is no effect on the ovarian coat. If the same dose is used for 10 minutes, it will seen a general looseness in the ovarian coat. The ovarian coat shows a discernable destruction when 0.1 % Thiotepa is applied for 3 or 10 minutes. In the cytoplasm of the cells of the loose and dense connective tissue, the increase of vacuolisation and the enlargement of intercellular spaces show the effects of Thiotepa. SAXENA and ADITYA (1969) pointed out that Apholate did not make any difference on the connective tissue of testes of *Poecilocerus pictus* (Orth.). There is not any explanation about the effects of chemosterilants on the connective tissue of insect ovaries.

The cells in the ovarioles of *A. ipsilon* are affected by Thiotepa more or less, according to the concentration and to the period of application. The degeneration of the cells increases as long as the doses and the treatment period of Thiotepa increase. The degeneration of the cells shows itself as disintegration, accumulation, vacuolisation and formation of some amorphous or membranous structures surrounded with a membrane in cytoplasm or in nuclei.

The effects of some chemosterilants on the ovarian tissue were studied especially in Diptera. In *Musca domestica*, LANDA and REZABOVA (1965) reported that Ribozidin, Aminopterin, 6-Azauracil and Thiotepa caused destruction of nuclei of follicular epithelial cells and vacuolisation in the cytoplasm and production of some small pycnotic nuclei in the nurse cells and formation of big basophilic amorphous masses in the egg chambers. In *M. domestica*, KISSAM et al. (1967) showed that there was a yolk degeneration in the developing oocyte and in the follicular cells, but there was no effect on the fully developed oocyte by the effects of Tretamin, Hydroxyurea and Methylmethane sulphonat. On the other hand, they pointed out that the increase in the treatment duration also increased the effects of chemosterilants. In *M. domestica*, WILSON and HAYS (1969) stated that the yolk degeneration in the oocyte occured after feeding the matures with p.p-bis (1-aziridinyl)-N-Methylphosphinic amide and p,p-bis (1-aziridinyl)-N-(3-Methoxypropyl) phosphinothioic amide. MORGAN (1967) also observed that vacuolisation, chromatin accumulation and destruction of oocyte and nurse cell were seen when the females were fed on a nutrient containing 1–2 % Hempa. Similar effects were seen by the authors in *A. ipsilon* in this work. MORGAN (1967), also, showed that the egg in the first chamber in each ovariole developed, but there was not any development in the following chambers. In the ovary of *Cochliomyia hominivorax* (Dipt.), CRYSTAL and LaCHANCE (1963) and LaCHANCE and LEVERICH (1968) pointed out that the most effective result was obtained during the endomitosis of nurse cells after the treatments of Thiotepa, Tretamin, Methyl tretamin, and a derivative of Benzoquinon.

In *Thermobia domestica* (Thysanura), LANDA and MATOLIN (1971) stated that Metepa caused the degeneration of young oocyte and degenerative pycnosis in the nuclei of profollicular cells. In *Locusta migratoria* (Orth.), NATH and SHEIKHER (1976) explained that a complete degeneration such as the abberation of chromatin, pycnosis of nuclei, formation of empty spaces in the cytoplasm were occured by the effect of Hempa. In *Trichopliusia ni* (Lep.), HENNEBERRY et al. (1972) showed that the egg development was inhibited by the treatment of Tepa and Metepa, but the oocyte was not affected at the late developmental stage.

It is obvious that the effects of chemosterilants are seen especially during the oocyte development, and in addition to the developing

oocytes, the follicular epithelial cells and nurse cells are also affected severely. Generally, cell degeneration, as it was observed in *A. ipsilon*, is seen as the vacuolisation of nuclei and cytoplasm, disintegration or accumulation of chromatin in the germ cells.

Among the damages seen after Thiotepa application, the amorphous masses with a dense center and membranous structures have not been encountered except that a study of LANDA and REZABOVA (1965). The big basophilic masses in the cytoplasm that LANDA and REZABOVA mentioned in the mature *M. domestica* after the Thiotepa application, are similar structures to those of *A. ipsilon*. Similar structures were stated by GRIMSTONE et al. (1967) in the insect blood cell capsules and were called as cytolysomes. GRIMSTONE et al. explained that the large and small structures with a dense center were found in great number in the cell. These structures show acid phosphatase acrtivity and appear under the pathological conditions. NOVIKOFF (1960) observed these structures in the cells of pathological liver and kidney that were undergone to cytolisis and since they were larger than normal lysosomes, he called them cytolysomes. NAPOLITANO (1963) found similar structures in the active brown adipose tissue in mice and claimed that these were not the signs of degeneration. These structures were not seen by the author in normal brown adipose tissue, but were seen in that of chilled animal under experimental conditions. PITT (1975) explained that cytolysomes were used as synonyms with autophagic vacuoles and secondary lysosomes. It is quite obvious that these structures are the results of pathological situations and are formed in the cells of ovarioles ofter the Thiotepa treatment in *A. ipsilon*.

As a result, it is rather correct to say that Thiotepa causes a severe degeneration according to the concentration and the treatment period of Thiotepa in the ovary of *A. ipsilon* larvae. It is certain that this degeneration would inhibit the egg production of insect.

ABBREVATIONS

A : Amorphous structure

Ak : Accumulation of karyoplasm

As : Accumulated substance

C : Cytoplasm

Cm	: Cell membrane	L	: Lipid
dc	: Dense center	Lct	: Loose connective tissue
Dc	: Disintegration of cytoplasm	M	: Membrane
Dct	: Dense connective tissue	Mi	: Mitochondria
Dk	: Disintegration of karyoplasm	Ms	: Membraneous structure
Dcm	: Disapperance of cell membrane	N	: Nucleus
E	: Epithelial layer	Nu	: Nucleolus
F	: Fusome	Os	: Ovarian sheet
FN	: Flattened nucleus	t	: Tracheole
Gc	: Germ cell	T	: Trachea
Gl	: Glycogen	Tp	: Tunica propria
Ib	: Intercellular bridge	V	: Vacuole
K	: Karyoplasm		

ACKNOWLEDGEMENT

We would like to express our thanks to Dr. T. Güven for his kind cooperation and to physicist Mr. Y. Üstün for his kind assistance in the electron microscopy.

REFERENCES

ADDY, N.D., 1970. Some Cytological Changes in The Fat Body and Testes of Male Spruce Budworm *(Choristoneura fumiferana* Clem.) Pupae After Treatment With Tepa. *Can. J. Zool.*, 49: 831–836

ANWAR, M., D.A. WOLFENBARGER, E. CANTU, S.H. ROBINSON, 1974.Tobacoc Budworm: Chemosterilization by Fumigation With Thiotepa. *J. Econ. Ent.*, 67: 322–324

BORKOVEC, A.B., 1962. Sexual Sterilization of Insects by Chemicals. *Science*, 137: 1034–1037

BORKOVEC, A.B., 1966. Advances in Pest Control Research VII. Insect Chemosterilants. *John Wiley and Sons Inc., New York.*

BORKOVEC, A.B., S. NAGASAWA, H. SHINOHARA, 1968. Sterilization of The Azuki Bean Weevil, *Callosobruchus chinensis*, by Metepa and Hempa. *J. Econ. Ent.*, 61: 695–698

CAMPION, D.G., 1972. Insect Chemosterilants a Reviw. *Bull. Ent. Res.*, 61: 577–635

CHANG, S.C., P.H. TERRY, A.B. BORKOVEC, 1964. Insect Chemosterilants With Low Toxicity For Mammals. Science, 144: 57–58

CHAUDHARY, J.P. and R.P. KAPIL, 1976. Sterilizing *Trogoderma granarium* Ev. With Some Alkylating and Miscellaneous compounds. *Z. Angew. Ent.*, 81: 148–156

CHAUDHARY, J.P. and R.P. KAPIL, 1977. Sterilizing *Trogoderma granarium* Everts With Volatile Alkylating Compounds. *Z. Angew. Ent.*, 84: 184–191

CLINE, R.E., 1968. Evaluation of Chemosterilant Damage to The Testes of The Housefly, *Musca domestica*, by Microscopic Observation and by Measurement of the Uptake of C^{14} Compounds. *J. Insect Physiol*, 14: 945–953

COLLIER, C.W. and J.E. DOWNEY, 1965. Laboratory Evaluation of Certain Chemosterilants Against The Gypsy Moth. *J. Econ. Ent.*, 58: 649–651

CRYSTAL, M.M. and LaCHANCE, 1963. The Modification of Reproduction in Insects Treated With Alkylating Agents. I. Inhibition of Ovarian Growth and Egg Production. *Biol. Bull.*, 125: 270–280

DIMETRY, N.Z. and M.H. MANSOUR, 1971. Effect of some Inorganic Salts As A Deterrent Agent Against The Greasy Cutworm *Agrotis ipsilon* Hfn. and The Spiny Bollworm *Earias insulana* Boisd. *Z. Ang. Ent.*, 69: 315–317

ECONOMOPOULOS, A.P., 1971. Effects of Tretamine on The 4th-and 5th-instar *Oncopeltus fasciatus* Nymphs: Development of Eggs from Tretamine-Treated Females. Sterility Principle For Insect Control or Eradication, *IAEA, Vienna*, pp 259–270

GRIMSTONE, A.V., S. ROTHERAM, G. SALT, 1967. Capsule Formation by Insect Blood Cells. *J. Cell Science*, 2: 281–292

GÜVEN, T., 1979. Ergin *Locusta migratoria migratorioides* (R.F.) (Orthoptera: Acrididae) Testisinin Folikül Duvarı, Kist Hücreleri, Spermatogonyum ve Spermatositlerinin İnce Yapısı ve Bu İnce Yapıya Kemosterilant Hempa (Hexamethylphoshorictriamide) nın Etkisi *Unpublished Ph. D. Thesis*

HAFEZ, M., A.E. ABDUL-NASR, H.S. SALAMA, 1978. Studies On The Effect of Two Chemosterilants on *Aedes caspius* Pallas (Dipt., Culicidae). *Z. Ang. Ent.*, 86: 105–110

HENNEBERRY, T.J., M.W. STIMMAN, S. HARREL, 1972. Effects of Tepa and Metepa on The Reproductive Tissues of Cabbage Looper Moths. *J. Econ. Ent.*, 65: 93–97

IGNOFFO, C.M. and C. GARCIA, 1979. Susceptibility of Larvae of The Black Cutworm *(Agrotis ipsilon)* to Species of Entomopathogenic bacteria, Fungi, Protozoa and Viruses. *J. Econ. Ent.*, 72: 767–769

JOHNSON, B.T. and L.C. LEWIS, 1982. Pathogenicity of Two Nuclear Polyhedrosis Viruses in The Black Cutworm, *Agrotis ipsilon* (Lepidoptera: Noctuidae). *Can. Ent.*, 114: 311–316

KISSAM, J.B., J.A. WILSON, S.B. HAYS, 1967. Selective Effects of Certain Anti-Fertility Compounds on The Housefly As Shown by Reciprocal Crosses and Histological Sectioning. *J. Econ. Ent.*, 60: 1130–1135

KLASSEN, W., T.H. CHANG, P.E. EIDE, 1969. Effects of Apholate on Chromosomes of Germ cells in The Grasshopper Testes. *Can. J. Genet. Cytol.*, 11: 829–833

LaBRECQUE, G.C., 1972. Induction of Sterility in Adult House Flies and Stable Flies by Chemosterilization of Pupae. *J. Econ. Ent.*, 65: 751–753

LaCHANCE, L.E. and A.P. LEVERICH 1968. Cytology of Oogenesis in Chemosterilized Screw Worm Flies, *Cochliomyia hominivorax*, As Related to Endomitosis in Nurse Cells. *Ann. Entomol. Soc. Ame.*, 61: 1188–1197

LADD, T.L. Jr., A.B. BORKOVEC, C.R. BURIFF, M.G. KLEIN, 1980. Japanese Beetle: Chemosterilization by Fumigation. *J. Econ. Ent.*, 73: 12–14

LANDA, V. and REZABOVA, 1965. The Effect of Chemosterilants on The Development of Reproductive Organs in Insects. *Proc. XII th Int. Congr. Ent., London*, pp. 516–517

LANDA, V. and S. MATOLIN, 1971. Effects of Chemosterilants on Reproductive Organs and Embryogenesis in Insects. Sterility Principle For Insect Control or Eradication, *IAEA, Vienna*, pp. 173–182

MEGEED, M.I.A., Z.H. ZIDAN, R.A. KHALID, 1974. Response of The Greasy Cutworm, *Agrotis ypsilon* Rott. to Certain Antifeedants. *Z. Ang. Ent.*, 76: 106–110

MORGAN, P.B.. 1967. Effects of Hempa on The Ovarian Development of House Flies *(Musca domestica L.). Ann. Entomol. Soc. Ame.*, 60: 812–818

NAPOLITANO, L., 1963. Cytolysomes in Metabolically Active Cells. *J. Cell Biol.*, 18: 478–481

NATH, W.P.K. and C. SHEIKHER, 1976. Effects of Hempa on The Gonads of *Locusta migratoria* (L.) (Orth.: Acrididae). *Bull. Ent. Res.*, 66: 313–315

NOVIKOFF, A.B., 1960. Biochemical and Staining Reaction of Cytoplasmic Constituents, in Developing Cell Systems and Their Control. *Ronald Press. Co., New York*

OSMAN, M.F., M. HAFEZ, M.S. OMAR, 1975.Sterilization of *Musca domestica vicina* and *M. sorbens* by Adult Feeding of Thiotepa. *J. Econ. Ent.*, 68: 145–146

PARNELL, J.R. and D.F. METTRICK, 1969. The Action of Thiotepa on The Fertility of The Confused Flour Beetle. *J. Econ. Ent.*, 62: 585–588

PITT, D., 1975. Lysosomes and Cell Function. *Longman Inc., New York*

SALAMA, H.S., 1976. Sterilization and Spermatogenesis of The Gypsy Moth *Pothetria dispar* L. As Affected by Thiotepa. *Z. Ang. Ent.*, 81: 292–304

SAXENA, S.C. and V. ADITYA, 1969. Histopathology and Histochemistry of The Insects Treated With Chemosterilants I. On The Observation in Active Testes of *Poecilocerus pictus* (Orth.: Acrididae) Treated With Apholate. *Cytologia*, 34: 399–404

SAXENA, S.C. and V. ADITYA, 1974. Histopathology and Histochemistry of The Insects Treated With Chemosterilants V. Nucleic Acids, Phospholipids and Phosphatases in The Female Reproductive Organs of Chemosterilised *Poecilocerus pictus*. *Cytologia*, 39: 573–580

SCHOENBOHM, R.B. and F.T. TURPIN, 1977. Effect of Parasitism by *Meteorus leviventris* On Corn Foliage Consumption and Corn Seedling Cutting by The Black Cutworm. *J. Econ. Ent.*, 70: 457–459

SEHNAL, F., M.M. METWALLY, I. GELBIC, 1976. Reaction of Immature Stages of Noctuid Moths to juvenoids. *Z. Ang. Ent.*, 81: 85–102

SHAABAN, A.M., H.I. YOUSSEF, A.A. KAMEL, M.R. ABULGHAR, 1975. Effect of Certain Chemosterilants On The Larvae of The Greasy Cutworm, *Agrotis ipsilon*, Rott. *Z. Ang. Ent.*, 78: 386–392

SHARMA, M.L. and L.M. THERIAULT, 1980. Effect of The Chemosterilant Thiotepa on The Pea Aphid, *Acyrthosiphon pisum* (Homoptera: Aphididae). *Can. Ent.*, 112: 103–105

SMITTLE, B.J., J.B. SCHMITT, G.S. BURDEN, 1966. Effect of Tepa on The Reproductive Organs and Embryogeny of The German Cockroach. *J. Econ. Ent.*, 59: 1419–1423

STEFAN, A.W. and M. STUEBEN, 1976. Towards The Elimination of The Reproductive Capacity of Parthenogenetic Females of *Aphis fabae* (Homoptera: Aphididae) After Contact Treatment With Chemosterilants. *Z. Ang. Ent.*, 80: 56–69

STREIBERT, H.B. and V. DITTRICH, 1977. Toxicological Response of Insect Eggs and Larvae to A Saturated Atmosphere of Chlordimeform. *J. Econ. Ent.*, 70: 57–59

SULUDERE, Z., 1987. A Study on The Fine Structure of The Ovarium of The Last Instar Larvae of *Agrotis ipsilon* (Hufnagel) (Lepidoptera: Noctuidae). *Communs. Fac. Sci. Univ. Ankara, CB,*

TERRY, P.H., D.G. McHAFFEY and A.B. BORKOVEC, 1977. Uptake and Residues of Chemosterilants in Boll Weevils Treated by Fumigation or Dipping. *J. Econ. Ent.*, 70: 427–430

WILSON, J.A. and S.B. HAYS, 1969. Histological Changes in The Gonad and Reproductive Behaviour of House Flies Following Treatment With Chemosterilants p,p-bis (l-aziridinyl) –N– Methylphosphinic amide and p,p-bis (l-aziridinyl) –N– (3– Methoxypropyl) – Phosphinothioic amide. *J. Econ. Ent.*, 62: 690–692