Zagazig J.Agric. Res., Vol .30 No.(3) 2003 943 - 956

HISTOPATHOLOGICAL STUDIES ON THE EFFECTS OF ANILOFOS (HERBICIDE) IN PREGNANT ALBINO RATS

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Received 15 / 2 / 2003 *Accepted 10 / 3 / 2003*

ABSTRACT: **In the present study anilofos which considered the most selective herbicide used till now for controlling the annual grasses, sedges and some broad- leaved weeds in transplanted and direct seeded rice, was chosen to study its histopathological efects in the white rats *Rattus norvegicus*.**

The doses (1/10 and 1/30 of LD_{50}) were orally given to pregnant female rats using a stomach tube. These doses were applied 5 times started from the 7^{th} to the 11^{th} day of gestation period.

Liver of rats treated with the dose of 1/10 of LD_{50} showed severe congestion of hepatic blood vessels and hepatic sinusoides, such congestion was moderate with the dose 1/30 of LD_{50}. Portal oedema, lymphocytic aggregation and hepatic degeneration of the hepatocytes were also detected in case of the higher dose. Kidneys of rat treated with both doses of anilofos showed congestion of the renal blood vessels, focal haemorrhage and perivascular beside interstitial lymphocytic infiltration.

Spleen of rats treated with a dose of 1/10 of LD_{50} showed severe hyperplasia in the lymphocytic elements of white pulp. The smallest dose revealed nearly the same lesions.

The cardiac muscle showed hyaline degeneration, vaculation and interstitial lymphocytic infiltration in case of the highest dose.

The heart of rats treated with the smallest one was normal. The lungs showed no pathological lesions, in all treated rats.

INTRODUCTION

The extensive agricultural usage of pesticides (insecticides, herbicides, fungicides, and rodenticides) carries potential health hazards to human and other desirable species of animals, directly by exposure to the toxic residues of pesticides in foods and indirectly through the pollution of surrounding environment. The literature showed that toxicity of pesticides may be acute or chronic and lead to, internal organs injuries. In addition to (teratogenic, mutagenic, and carcinogenic effects (Mohamed 1995; Fujitani *et al.*, 1997 and Farag 1998).

In studying the toxicological effects of pesticides, it must take in account, both the direct injuries effects on human and domestic animals and the indirect effects of the environment as well as their effects on food production which is essential to maintain a proper ecological balance.

Anilofos is a pre-emergence and early post-emergence selective herbicide used up now to control annual grasses, sedges and some broad - leaved weeds in transplanted and direct seeded rice. Very few toxicological studies have been carried out to evaluate the acute and chronic toxicity effects of anilofos on the laboratory animals.

The histo- pathological lesions caused by different pesticides had been reviewed by many investigators, (Abou El-Ross 1977; Kady *et al.*, 1987; Ram and Singh 1988; Mohamed 1995; Srivastava *et al.*, 1995; Fujitani *et al.*, 1997; Farag 1998 and Fujitani *et al.*, 2001).

The present study was designated to investigate some organs-histopathological changes, in pregnant albino rats orally treated for five successive days (from day 7 to day 11 of pregnancy) with sub lethal doses (1/30 or 1/10 of LD_{50}) of anilofos herbicide.

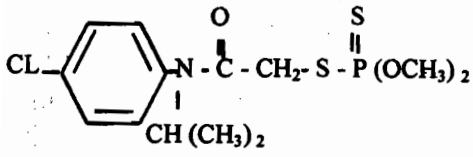
MATERIALS AND METHODS

1- Herbicide used:

Anilofos 30 EC, has trade names: Anilofos, Aniloguard, Nidan and Anilgard. Common name: anilofos.

Chemical name: S-4-Chloro-N-isopropyl carbaniloyl-methyl O,O-dimethyl phosphoro-dithioate and has a structural formula of

2- Animals used:



A total number of 30 mature female albino rats with an average initial body weight of 190 gm were used in the present study. The animals were distributed into three experimental groups (ten females for each group). The animals were kept under normal laboratory conditions in stainless cages provided with feeders and water supply ad libitum. The 1st and 2nd groups were treated by 1/10 and 1/30 of anilofos LD_{50}, respectively, while the 3rd was chosen as a cheek control.

3- Preparation of internal organs for histopathological examina-tions:

The internal organs (liver, kidneys, heart, spleen and lungs) of the sacrified dams were grossty examined, weighed and specimens of them were taken and fixed in 10% neutral buffered formation. Such specimens were processed by the routine methods and serial sections were cutted (5 micrometers thickness) by rotary microtome. Sections were stained with haematoxylin and eosin (Carleton *et al.*,1980).

RESULTS AND DISCUSSION

1-The histo- pathological findings in internal organs of rats treated with anilofos:

The internal organs of pregnant albino rats orally treated for 5 successive days with 1/10 or 1/30 of LD_{50} of anilofos herbicide revealed the following histo-pathological findings:

a- Liver:

Liver of rats treated with the dose 1/10 of LD_{50} showed severe congestion of the hepatic blood vessels and hepatic sinusoids (Figure 1). Portal edema, lymphocytic aggregation and hydropic degeneration of the hepatocytes were also detected (Figure 2). Liver of rats treated With the dose 1/30 of LD_{50} revealed moderate congestion of hepatic blood vessels, lymphocytic infilteration and vacular degeneration of the hepatocyts (Figure 3 and 4).

b- Kidneys:

Kidneys of rats treated with the dose level 1/10 of LD_{50} of anilofos showed congestion of the renal blood vessels, focal haemorrhage perivascular and interstitial lymphocytic infilteration (Figure 5). The renal tubules revealed degenerative changes, intratubular hyaline casts (Figure 6) and moderate to severe dilatation of the renal tubules (Figure 7). Kidneys of rats treated with the dose 1/30 of LD_{50} suffered mild congestion of the renal blood vessels, perivascular lymphocytic infilteration, intratubular hyaline casts and degenerative changes of the renal tubules (Figure 8). In some cases cystic dilatation of some renal tubules was also detected (Figure 9).

c- Spleen:

Spleen of rats treated with the dose 1/10 of LD_{50} showed severe hyperplasia in the lymphocytic elements of the white pulp (Figure 10). In some cases the spleen showed mild to moderate hyperplasia of the lymphocytic elements of the white pulp, together with congestion and hemosiderosis (Figure 11). The spleen of rats treated with the dose 1/30 of LD_{50} revealed nearly the same lesions (Figure 12).

d- Heart:

The cardiac muscle of rats treated with the dose 1/10 of LD_{50} showed hyaline degeneration, vaculation and interstitial lymphocytic infilteration (Figure 13). The heart of rats treated with the dose 1/30 of LD_{50} was normal.

e- Lungs:

It showed no pathological lesions, either in rats orally treated with the dose level 1/10 or 1/30 of LD_{50} of the anilofos.

Although there was some similarity in the lesions affected the internal organs of rats treated with either the dose level 1/10 or 1/30 of LD_{50} of anilofos, the lesions that appeared in the organs of rats treated with 1/10 of LD_{50} were more severe, prominent and widespread than those found in the rats treated with the dose 1/30 of LD_{50} of anilofos.

Figure (1): Cross section in liver of pregnant albino rats orally treated for 5 successive days with one dose 1/10 of LD_{50} of anilofos herbicide, showing severe congestion of the hepatic blood vessels (A) *(H&E* x 300*)*.

Figure (2): Cross section in liver of female rats orally treated for 5 successive days with one dose of 1/10 of LD_{50} of anilofos, showing hydropic degeneration of the lymphocytes *(H&E* x 300).

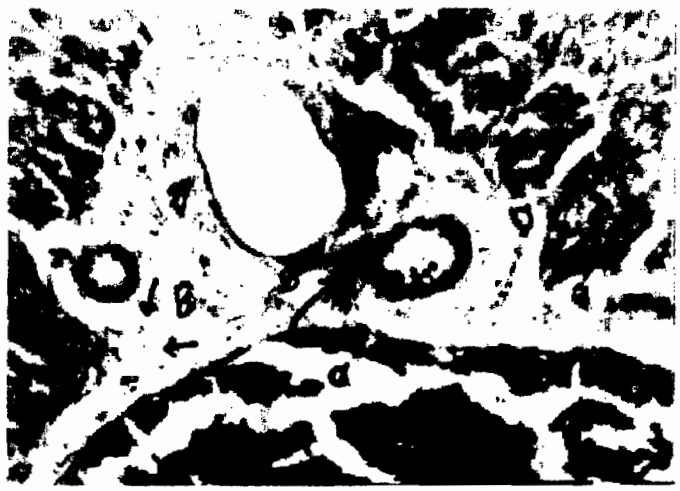


Figure (3): Cross section in liver of female rats orally treated with the dose level 1/30 of LD_{50}, showing moderate congestion of hepatic blood vessels (A) and portal oedema (B) *(H&E* x 300*)*.

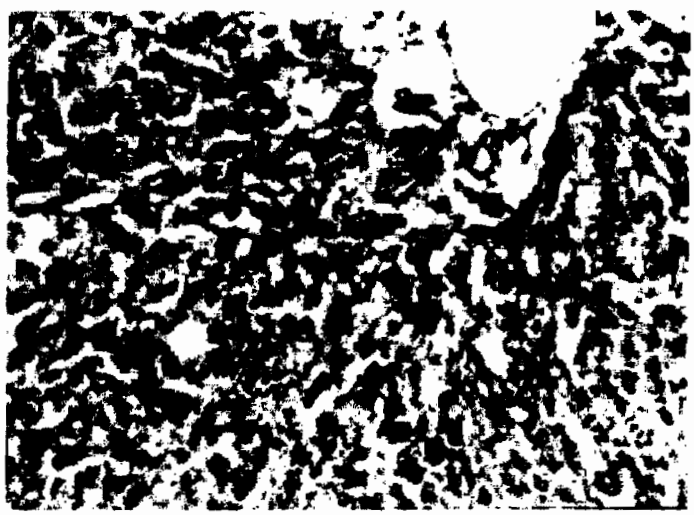


Figure (4): Cross section in liver of female rats orally treated with the dose level 1/30 of LD_{50} of anilofos, showing vacular degeneration of the hepatocytes *(H&E* x 300*)*.

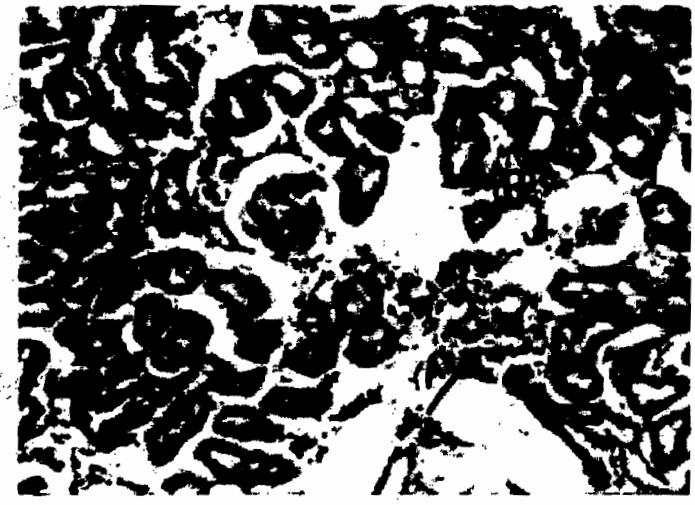


Figure (5): Cross section in kidney of female rats orally treated with the dose level 1/10 of LD_{50} of anilofos, showing perivascular (A) and interstitial (B) lymphocytic infiltration (H & E x 300).

Figure (6): Cross section in kidney of female rats orally treated with the dose level 1/10 of LD_{50} of anilofos, showing degenerative changes in the renal tubules (A) and intratubular hyaline casts (B) (H&E x 300).

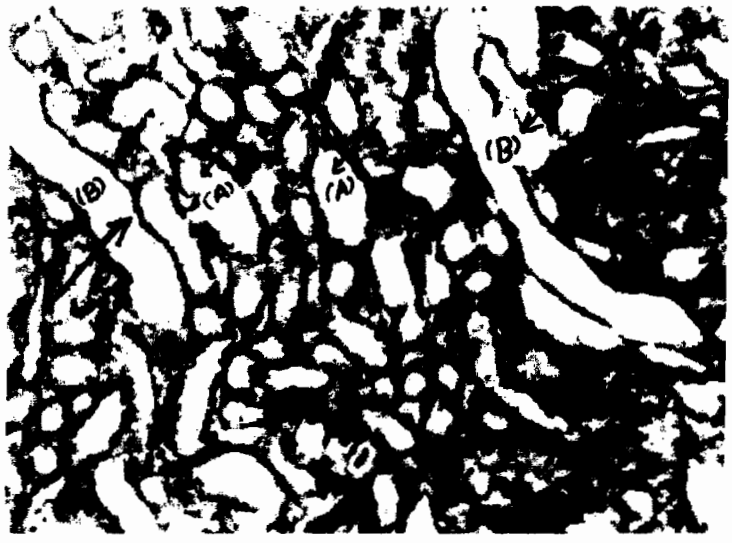


Figure (7): Cross section in kidney of female rats orally treated with the dose level 1/10 of LD_{50} of anilofos, showing moderate (A) to severe (B) dilatation of the renal tubules *(H&E* x 300*)*.

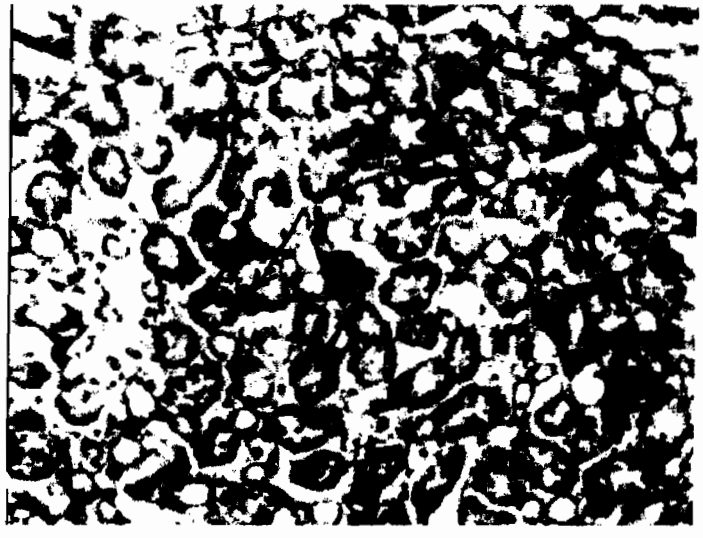


Figure (8): Cross section in kidney of female rats orally treated with the dose level 1/30 of LD_{50} of anilofos, showing degenerative changes in the renal tubules *(H& E* x 300*)*.

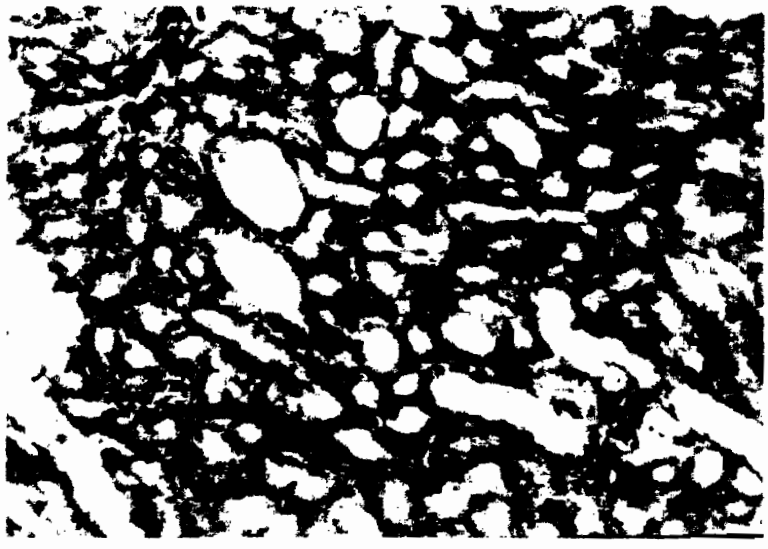


Figure (9): Cross section in kidney of female rats orally treated with the dose level 1/30 of LD_{50} of anifolos, showing moderate cystic dilatation of some renal tubules *(H&E* x 300*)*.

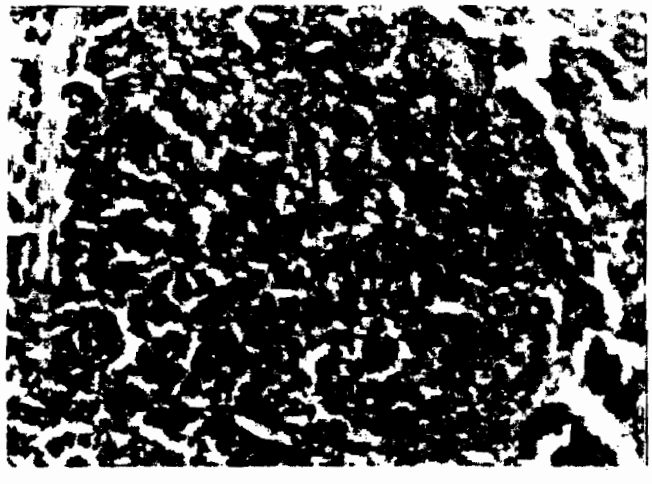


Figure (10): Cross section in spleen of female rats orally treated with the dose level 1/10 of LD_{50} of anilofos, showing severe hyperplasia in the lymphocytic elements of the white pulp, (H&E x 300).

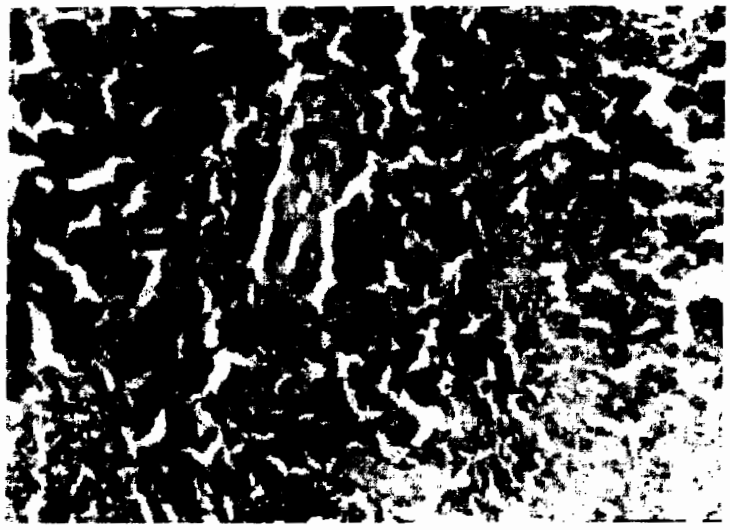


Figure (11): Cross section in spleen of female rats orally treated with the dose level 1/10 of LD_{50} of anilofos, showing mild to moderate hyperplasia of the lymphocytic elements of the white pulp, (A) together with congestion (B) and hemosiderosis (C) (H&E x 300).

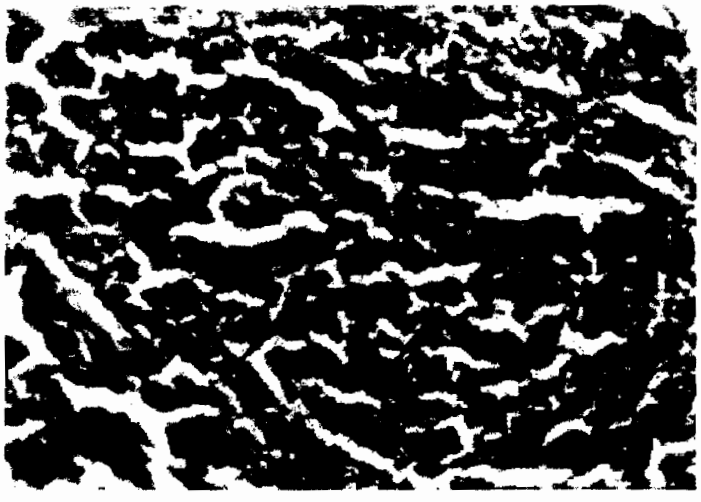


Figure (12): Cross section in spleen of female rats orally treated with the dose level 1/30 of LD_{50} of anilofos, showing moderate hyperplasia in the lymphocytes of the white pulp (H&E x 300).

Figure (13): Cross section in the cardiac muscle of female rats orally treated with the dose level 1/10 of LD_{50} of anilofos, showing hyaline degeneration, (A) vaculation (B) and interstitial lymphocytic infiltration (C) *(H&E* x 1200*)*.

The afore- mentioned histopathological findings were nearly similar to those reported by many investigators (Abou El-Roos, 1979; Mohamed, 1995; Fujitani *et al*, 1997; Farag, 1998 and Fujitani *et al*, 2001) who used other organophosphorus or carbamate pesticides.

Abou El-Roos (1979) found that rats treated with sublethal doses of Tamaron (Methamidophos), including 2.5, 7.5, 10, and 30 mg/kgb.wt. showed enlargement in the liver and spleen. Lymphocytic infiltration was seen in the lungs and liver. Lymphocytic hyperplasia and hemosiderosis were also observed in the malpigian corpuscles of the spleen. Mohamed (1995) mentioned that rats orally treated with doses 1/20 of LD_{50} of Selecron (Profenfos) insecticide for different times and different intervals revealed haemorrhage, haepatocytes necrosis, lympho-cytic infiltration and periportal inflammatory cells in the liver. The kidneys showed interstitial

inflammatory cells, cortical haemorrhage and glomerular and tubular fibrosis. The heart revealed congestion of the coronary and intermuscular blood vessels.

Fujitani *et al.* (1997) reported that rats given in their diet for 13 weeks the chlorpropham herbicide at rate of 7500, 15000, or 30000 ppm showed histopathological lesions in the internal organs, summarized in congestion and hemosiderosis of the red pulp of the spleen. The liver exhibited enlargement of hepatocytes. The kidneys of rats treated with 15000 and 30000 ppm of chlorpropham showed hemosiderosis and atrophy in renal proximal tubular cells.

Farag (1998) found that rats given in the drinking water for 90 days carbosulfan at concentrations of 43 and 86 ppm revealed histopathological lesions in the internal organs included, hepatic haemorrhage, hyperplasia in the bile duct epithelium and infiltration of lymphocytes. The kidneys showed cloudy swelling, hydropic degeneration of the renal tubules and necrosis of individual cells.

Fujitani *et al.* (2001) reported that male F344 rats were given 3% chlorpropham in the diet and at 2, 4, 6, 8 or 13 weeks of adminstration five rats in each group were killed for clinical and microscopic examination. Marked splenomegaly and hepatomegaly were observed in the treated rats at 2-13 weeks of adminstration. Microscopic examination revealed congestion, hemosidrein deposits, extramedullary hemopoiesis and lymphoid atrophy in spleen and hyperplasia of the hemopoietic cells in bone marrow of treated rats at 2-13 weeks and fibrosis in splenic capsule at 4-13 weeks.

Finally, it could be concluded that anilofos induced direct toxic effects on the internal organs of pregnant female rats and care showed be taken to avoid the hazards of its misuse in both human and animals also to avoid its environmental pollution effects.

REFERENCES

Abou El-Roos, A.A.M. (1979). Experimental histopathological studies of Tamaron "organophosphorous compound' toxicosis on rats. M.Sc. thesis, Fac. Of Agric., Zagazig Univ., Egypt.

Carleton,R.A.; E.B. Drury and E.A. Wallington (1980). Histochemical techniques for normal and pathological tissue and identification of parasites. Fifth edition, Oxford

University Press, New York and Toronto.

Farag, A.A.G. (1998): Toxicological studies of some pesticides on white albino rats. M.Sc. thesis, Fac. of Agric., Zagazig Univ., Egypt.

Fujitani, T.; Y. Tada; A.T. Noguchi and M. Yoneyana (1997). Hemotoxicity of chlorpropham (CIPC) in F344 rats. Journal of Toxicology. 123; 111-124.

Fujitani, T.; Y. Tada; A.T. Noguchi and M. Yoneyana (2001). Effects of chlorpropham (CIPC) on the hemopoietic system of rats. Food Chem. Toxicol., 39(3); 253-259.

Kady, M.M.; S.E. Negm and A.A. Said (1987): Early proliferative lesion induced in some rat organs by two organophosphorous insecticides. J. Agric. Sci., Mansoura University 12 (2): 335-340.

Mohamed, Y.M. (1995). Toxicological studies on albino rats. M.Sc. Thesis, Faculty of Agriculture, Zagazig University.

Ram, R.N. and S.K. Singh (1988). Carbofuran induced histo-pathological and biochemical changes in liver of the teleost fish, *Channa punctatus*. (Bloch) Ecotoxicology and environmental safety. 16:3,194-201.

Srivastava, M.K. and K.B. Raizada (1995). Development toxicity of substituted phenyl urea herbicide isoproturon in rats. Vet. Hum. Toxicol., 37 (3): 220 – 223.

دراسات هستوباثولوجية على إناث الفئران المعملية البيضاء نتيجة معاملتها بمبيد الحشائش انيلوفوس

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يعتبر انيلوفوس واحد من أكثر مبيدات الحشائش الاختيارية الشائع استخدامها لمكافحة الحشائش الحولية ونباتاتها وبعض الحشائش العريضة الأوراق فى حقول الأرز المشتول أو المنزرع بداراً.

ولدراسة مخاطر سمية هذا المركب للإنسان وحيواناته الاليفه تم اختبار جرعتين (١/٣٠، ١/١٠ من قيمة الجرعة النصفية القاتلة).

وتم فيها دراسة بعض التأثيرات الهستوباثوليوجية الناتجة عن المعاملة بهاتين الجرعتين لمدة خمسة أيام متتالية من اليوم السابع حتى اليوم الحادى عشر من الحمل لإناث الفئران المعملية البيضاء باستخدام أنبوبة اللى المعدى. علماً بأنه لم تكن لهذه الجرعات أى تأثيرات ظاهرية على سلوك الفئران المعاملة أو على النسبة المئوية للموت.

ويمكن تلخيص النتائج المتحصل عليها فى التأثير على بعض الأعضاء الداخلية (الكبد- الكلى- الطحال- القلب- الرئتان) نتيجة المعاملة بهاتين الجرعتين على النحو التالى:

الكبد: تأثر نسيج الكبد فى الإناث المعاملة بالجرعة (١/١٠ من قيمة LD_{50}) بشكل أكبر منه فى الجرعة الأقل (١/٣٠ من قيمة LD_{50}) فى شكل احتقان شديد وواضح فى أنسجة الوعاء الدموى الكبدى وبالتجاويف الكبدية بجانب تجمع الخلايا الليمفاوية وتدهور خلايا الكبد مع أوديما بابية بينما ظهرت هذه الأعراض بشكل أقل حدة فى الجرعة الأقل.

الكلى: تأثرت الكلى تبعاً للجرعة المعامل بها وقد اتضح ذلك فى صورة احتقان الأوعية الدموية بالكلى مع نزف وارتشاح ليمفاوى لخلايا هذه الأوعية.

الطحال: تأثر الطحال تأثراً واضحاً فى صورة تضخم شديد فى الأنسجة الليمفاوية.

القلب: تأثرت عضلة القلب نتيجة المعاملة بالجرعة (١/١٠ من قيمة LD_{50}) وظهر ذلك بشكل تنكس هيالينى (زجاجى) وارتشاح ليمفاوى مصحوب بوجود فراغات ما بين الأنسجة بينما لم يتأثر نسيج عضلة القلب بالجرعة الأقل.

الرئتان: لم يكن لكلتا الجرعتين تأثير على أنسجة الرئتين.