PATHOLOGICAL CHANGES OF ACUTE TOXICITY INDUCED BY ORAL ADMINISTRATION OF MALATHION IN PIGEONS

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ABSTRACT

The objective of the present study was to investigate the pathological changes of the acute toxicity of Malathion as organophosphorus insecticide in wild pigeons (Rock dove). Liver enzyme Alanine Transaminase (ALT) in the serum of treated pigeons was measured. The maximum tolerated dose of malathion was 3.525mg / kg B.W. However, when the maximum tolerated dose of the malathion was given to group **A**, two out of eight pigeons died within two hours after treatment .The insecticide caused clinical signs appeared within two hours before death, they included salivation, lacrimation , gasping, frequent defecation, drooping of wings, tremors, convulsion and recumbancy. Also the histopathological changes of the acute toxicity of present study included vacculation of nerve fibers in the spinal cord and sciatic nerve, meningitis associated with lymphocytic infiltration in the brain, myocardial fibrosis, aggregation of lymphocytes and hepatic septal fibrosis. Cortical fibrosis with inflammatory cells, regenerating renal cortical tubules and dilated cortical tubules were also seen. Compared to control values the exposure to insecticide caused increase of the ALT level in the serum for treated pigeons, and this increment was significant. In conclusion Malathion insecticide administered orally at maximum tolerated dose induced clinical signs of poisoning, pathological changes in different organs of pigeons and increased the ALT value.

INTRODUCTION

Malathion is an organophosphours insecticide and has a wide range of use in agriculture, veterinary medicine and public health. However, the unregulated use and its aerial application over large agricultural and urban area has caused sever environmental pollution and potential health hazards (1).

Malathion is extensively used in commercial poultry industries in middle east, including Iraq to control external parasites (ticks, lice, mites etc…) of poultry birds (2).

Exposure to insecticide, include malathion may cause a wide variety of clinical signs depending upon the nature and concentration of chemical, the duration of exposure , the species, age, nutritional and health status of animal (3)

The signal most important mechanism of the toxic action of these insecticide in animals is inhibition of acetylcholine esterase at the never terminals, and this causes acetylcholine accumulation that sub sequently causes a series of muscarinic, nicotinic and central nervous system effects (4).

(5) have reported the Arial spraying of malathion reduced the hatchability of embrynoated chicken eggs. (6) Reported that Malathion dipping causes marked alteration in different enzymic profile and gross parameter related to carbohydrate protein and lipid metabolism in poultry birds.

The acute oral LD_{50} values for various bird species are mallards 1.485 mg/kg ; Pheasant 167mg/kg; black birds over 100mg/kg and chicken 525mg/kg (7).

Because little information is available in this field, this study was conducted to investigate the acute toxicity of Malathion in pigeons, also oarganophospherous insecticide toxicity in pigeons which has been used as a model for such toxicity.

MATERIALS AND METHODS

A total of 30 wild pigeons (Rock dove) of both sexes were purchased from the local market in Basrah Province within body weight average 300-400g. The birds were reared in separated cages of 100x100x80 cm^3 at the Poultry Disease Unit, College of Veterinary Medicine in Basrah University under suitable conditions, water and feed were supplied *ad libitum.*

Malathion (Cairo, Egypt) was obtained from local market in Basrah province. The Malathion was further diluted in distilled water to obtain the desired concentration for oral dosing by a gavage needle. The solution was prepared and used immediately. The doses of the Malathion were used according to the active ingredients of substance (8).

Fourteen pigeons were used in primary trial to determine the maximum tolerated dose by using different doses until reaching to 3,525mg/kg B.W. in which birds showed clinical signs of acute toxicity.

After determining maximum tolerated dose for malathion in previous experiment the other 16 birds were divided into two group (A,B). Group A was treated with , single dose of malathion (3.525 mg/kg B.W corrected to the nearest mg/kg B.W. of each bird) and signs of toxicity and death were observed in pigeons. Group B administrated distilled water only and served as control.

Twenty one days later all remaining birds were killed by decapitation. About 3 ml blood sample were collected for each bird in a sterile test tubes. Serum was obtained by until analysis. Brain, ◌centrifugation of blood at 3000 rpm for five minuets and kept at -4C spinal cord, sciatic nerve, liver, kidney, and heart samples were also collected for the histopatological examination. Tissue samples were kept in neutral buffered formalin and treated according to (9) to obtain. 5 µm thickened slides, stained with Haematoxylin and Eosin.

Estimation of (ALT) activity was determined according to (10) by using transminases-Kitb (Biomerieu - XSq^{r} made in France)

Data were subjected for statistical analysis as mean ± standard deviation (SD), and 0.05) (11). <difference between these means were considered significant at (P

RESULT AND DISCUSSION

The maximum tolerated dose of Malathion in the present study was 3.525mg/kg. Oral treatment of group A with maximum tolerated dose resulted in death of two pigeons within two hours after Malathion administration. The insecticide caused clinical signs which appeared within two hours before the death. The signs included salivation, lacrimation, gasping, frequent defecation, drooping of wings, tremors, convulsion and recumbancy compared with control pigeons. These results were in agreement with (2) who reported that the exposure of hens to malation in dipping solution caused symptoms like tremors, convulsion.

The 6 survived birds in treated group A exhibited same toxic symptoms in addition to vomition in 3 birds and slight salivation in all. These results were in line with (12) who reported the salivation and vomition in cockerels dosed orally with Malathion.

The clinical toxic signs were attributed to the organophosphorus toxicity mechanism in mammals and birds which inhibit the target enzyme cholinesterase which leads to accumulation of acetylcholine at the never ending and neuromuscular junctions and to cholinergic overstimulation manifested as muscarinic, nicotinic, and central nervous system effects (13).

In this study the signs of unsteady gait persisted for 40 hours in survived birds, this result was in agreement with that of (2),who observed that malathion action persisted longer than 24 hours due to continuous releasing of malathion from the tissue storage site as part of homeostatic mechanism.

Histopathological sections prepared from different organs of control group B did not reveal any pathological alteration, where as moderate to severe changes have been noticed in organs of group A. These changes included vacuolation of never fibers of spinal cord and sciatic never as shown in (fig. 1,2).

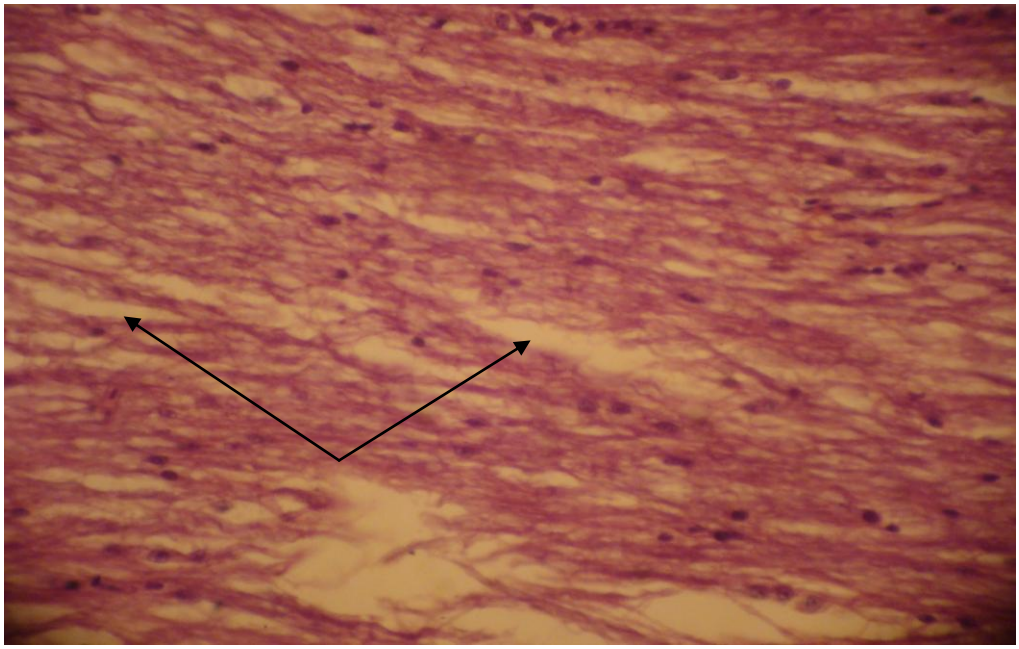

Fig (1) Spinal cord, longitudinal section , vacuolated nerve fibers. H&E 400x

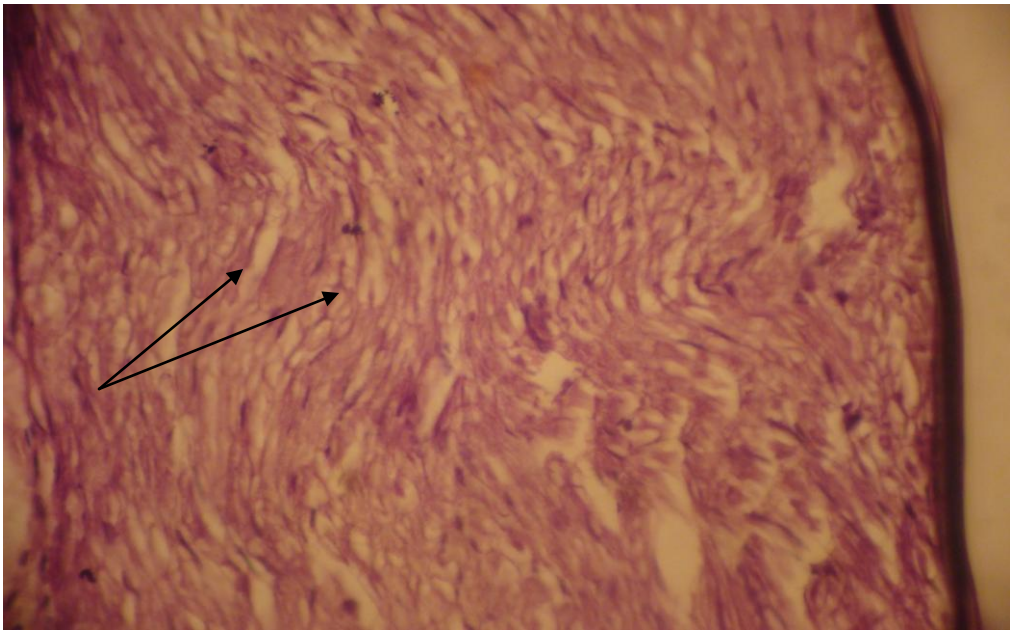

Fig (2) Sciatic nerve longitudinal section , vacculation of nerve fibers. H&E 400x

These observations were in accordance with that of (14) and (15) who observed that insecticides caused nervous system lesion as well as skeletal abnormalities in various species of birds. On the other hand, these results were contradicted with that of (16) who did not report any histopathological changes in the spinal cord or sciatic never, of chickens that received single oral dose of organophosphorus (Coumaphos).

The brain of the treated group revealed meningitis associated with lymphocytic infiltration fig (3).

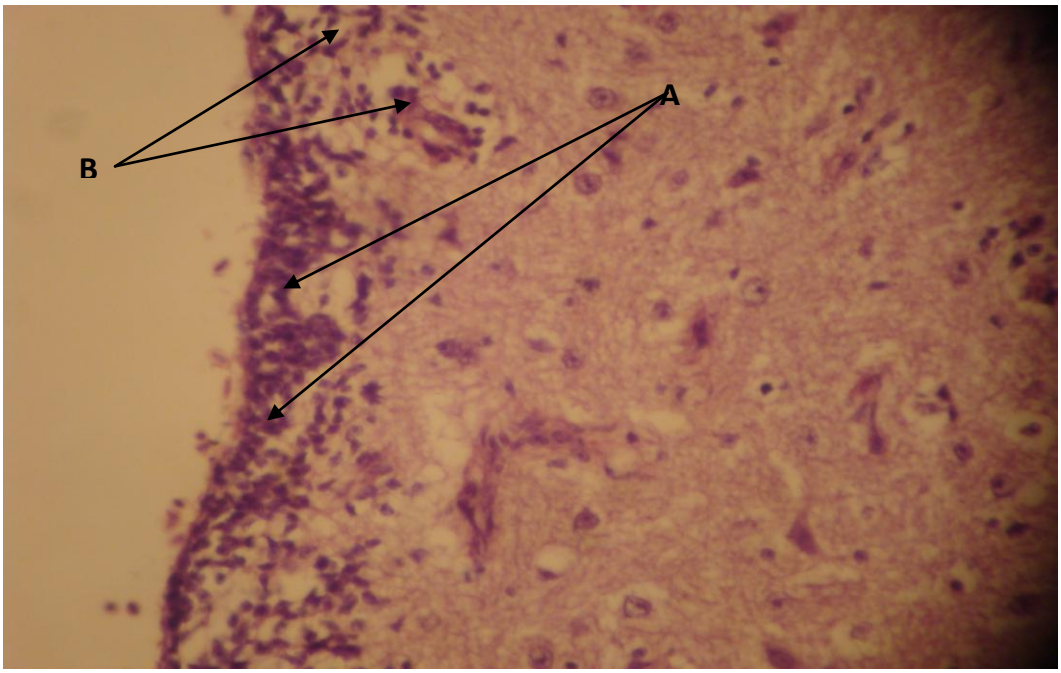

Fig (3) Brain, a) meningitis b) lymphocytic infiltration. H&E 400x

The brain histopathological results were in agreement with that (2) who published that inoculation of organophosphorus (endosulfan) in developing chick resulted in histopathological lesion in brain tissue. In addition (17and13), also mentioned that nervous system including whole brain tissue are affected by organophosphorus poisoning.

The brain changes were in disagreement with that reported by (18), who found that brain tissue was not affected with organophosphous as neurotoxic substance which is inline with (2) observations who mentioned that malathion at usual concentration did not produce any changes in brain tissues .

These differences may be attributed to the used organophosphorous type, its dose, route and duration of exposure, species involved, toxicokinetic aspects of insecticide, tissues examined and sampling time (19 and 2).

Histopathological examination of heart showed moderate myocardial fibrosis (fig 4).

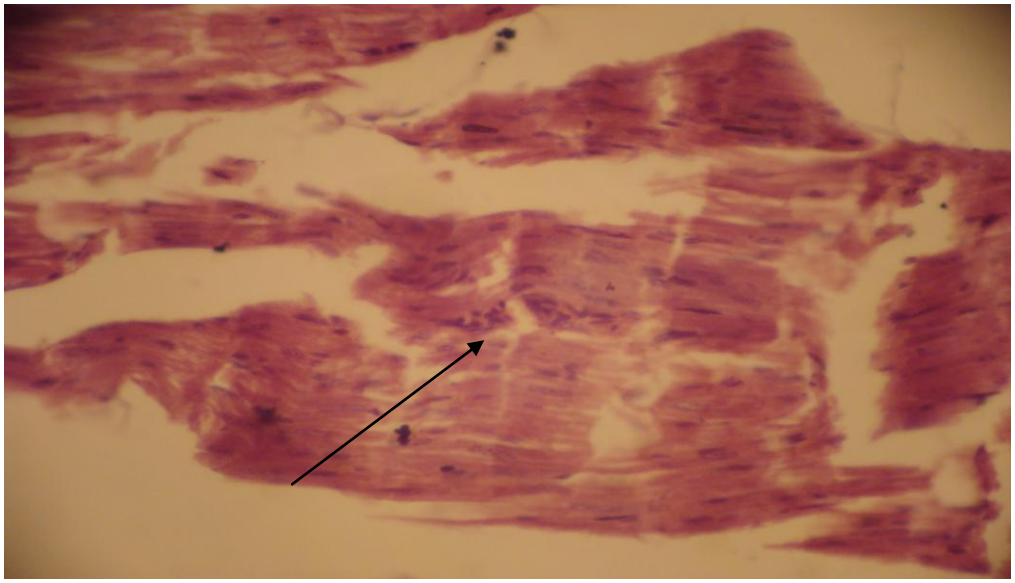

Fig (4) Heart, minimal fibrosis. H&E 400x

(20) Also reported that, Malathion causes lesions in the heart while (21) have demonstrated that organophosphorus has no specific microscopic change have been identified, in heart of treated chickens with organophosphorus insecticides. However the difference in observations may be attributed to differential action of different type of pesticide in different species of animals (2).

Septal hepatic fibrosis and aggregation of lymphocyte were the main historpathological alteration have been noticed in present study (fig, 5).

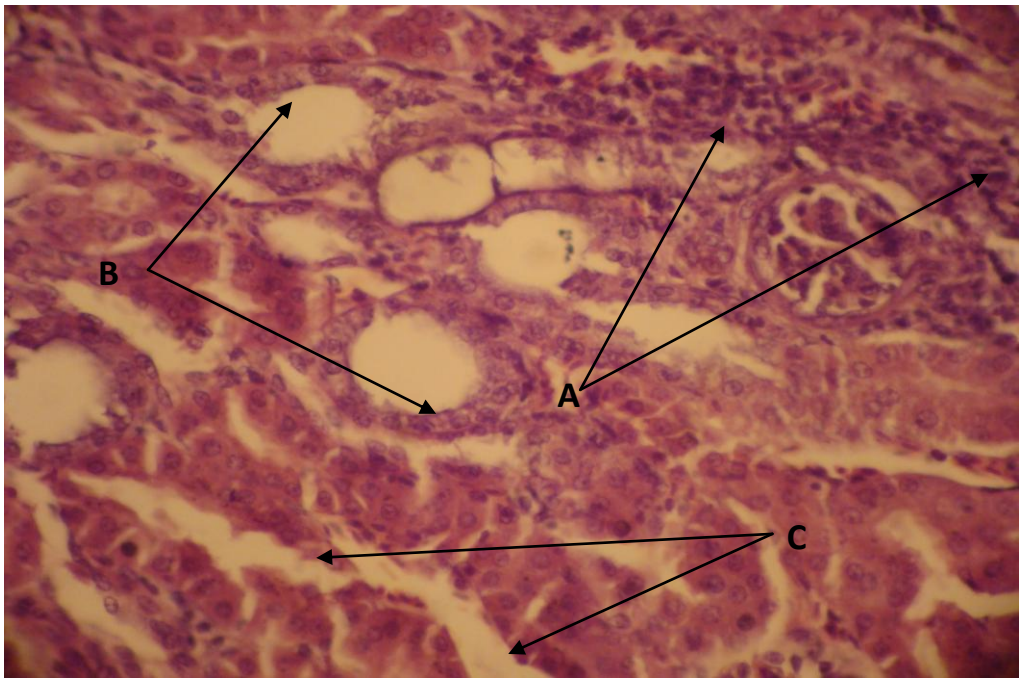

Fig (6) kidney,a) cortical fibrosis with inflammatory cells b)regenerating renal cortical tubules c)dilated cortical tubules . H&E 400x

These observations were in accordance with that of (1), who reported that histopathological lesions of degeneration and necrotic change with infiltration of lymphomononuclear cells in liver of chicks that received malathion. (22), observed the retarding of liver weight with congestion and granular degeneration of hepatocyte upon the effect of endosulfan in chicks.

Oral administration of malathion in pigeons produced histopathological lesions of toxicity in kidneys including, cortical fibrosis with inflammatory cells, regenerating renal cortical tubules and dilated cortical tubules as shown in fig,6.

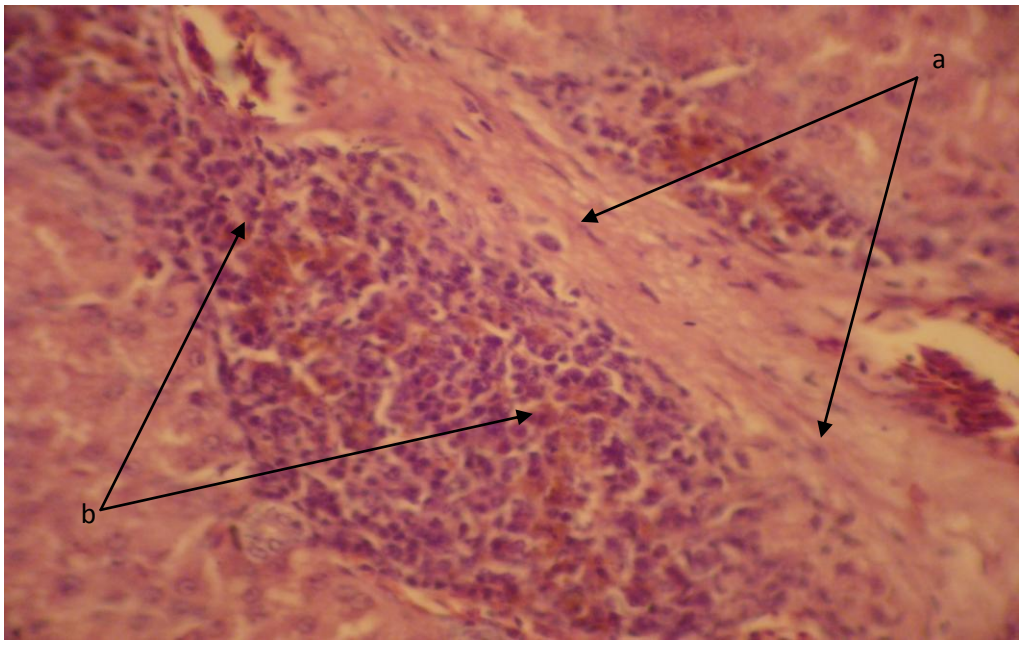

Fig (5) Liver a) Septal fibrosis b) Aggregation of lymphocytes H&E 400x

These results were in agreement with (23 and 24) who observed that malathion cause degenerative changes of epithelium lining of renal tubules .

A perusal of table one indicated that acute toxicity of Malathion resulted in increase of ALT values as compared with control 5.33 and 3.26 respectively. This result was statistically significant at ($p<0.05$). The result of the present study was in line with that of (14), who reported an increase of ALT values in broiler chicken fed ration contained organophosphorus insecticide.

The result was in agreement with that of (25), who mentioned that , oral dose of DDT, resulted in an increase liver protein

Increasing of the the ALT values might be attributed to the liver damage by the organophosphorus dosed birds (26).

Table (1) : progressive increase of ALT (U/L) value in treated pigeons

Group	No. of Samples	Mean	S.D
A	**6**	**5.33***	**±1.50**
B	**6**	**3.26**	**±0.43**

A=Treated group , B=Control group , ALT= Alanine transaminase , S.D= Standard deviation.

0.05.<* = Means significant at P

In conclusion, acute exposure of pigeons to malathion was associated with poisoning signs , pathological lesions and increase of ALT, at maximum tolerated dose in the surviving pigeons.

Further studies are needed to (re) evaluate toxicity of other organophosphorous insecticides using pigeons as suitable animal model for acute organophosphate toxicty studies.

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التغيرات المرضية للتسمم الحاد الناتج عن تجريع الحمام بالمالاثيون

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الخلاصة

إن هدف الدراسة الحالية هو معرفة التأثير المرضي للمبيد الفسفوري المالاثيون في الحمام، وقياس مستوى إنزيم الالينين ترانس امينيز في مصل دم الحمام المعامل. الجرعة الأعلى التي يتحملها الطير كانت 3.525 ملغم/ كغم من وزن الجسم. أعطيت هذه الجرعة إلى مجموعة A اثنان من ثمان حمامات ماتت خلال ساعتين من المعاملة. واظهر الحمام قبل الموت علامات التسمم بعد ساعتين من التجريع والتي تتضمن، إفرازات لعابية، تدمع، صعوبة تنفس، التبرز المتكرر، تهدل الأجنحة، رعشة، تشنج واضطجاع. بينت النتائج تغيرات مرضية تتضمن تنكس الألياف العصبية للحبل ألشوكي والعصب ألوركي، التهاب الدماغ واتشاح الخلايا اللمفية، تليف عضلة القلب، تنكس حويجزي، تجمع خلايا اللمفية في الكبد، ، تليف قشرة الكلية مع تواجد خلايا التهابية،تجدد الخلايا المبطنة للأنابيب الكلوية وتوسع الأنابيب الكلوية في منطقة القشرة .كما سبب المبيد الفسفوري زيادة في مستوى إنزيم الالينين ترانس امينيز وهذه الزيادة كانت معنوية مقارنة مع مجموعة السيطرة. ان المبيد الفسفوري الحشري المعطى فمويا بالجرعة الأعلى التي يتحملها الطير يظهر العلامات السريرية للتسمم، تغيرات مرضية في عدد من الأعضاء التي تم فحصها في الحمام المعاملة بهذه المادة. و زيادة في مستوى إنزيم الالينين ترانس امينيز

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