Effects of the Carbon Tetra Chloride on the Body Weight and Ovaries, Uterus, Testes of the Rabbit

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ABSTRACT

Aims: To investigate the effects of carbon tetra chloride on the body weight and ovaries, uterus, testes of the rabbit for 30 days.

Methodology: The rabbit were randomly divided into two equal groups (control and one treated groups), and the animals were treated as follow: 1- Control group C. (n = 15) received Distilled water 1ml /kg b.w four times a week intraperrturnium for 30 days. 2- Treatment group. (n=15) were treated with carbon tetrachloride (1 mg / kg b.w mixing with equal volume of olive oil (1ml/ kg b.w) four times a week intraperturnium for 30 days. The body weight of the animals detected at the day 1th, 7th, 14th, 21th and 30th of the experimental period. At 30 days animals sacrificed and ovaries, uterus, testes were removed and taken for histopathological study.

Results: Treatment with carbon tetra chloride causes a significant decrease in body weight. Also a significant decreased of ovarian, testicular weight and non-significant increase of uterus weights.

Conclusion: Carbon tetra chloride causes decrease body weight, degeneration testes, ovaries followed by effect on fertility.

Keywords: Rabbits, Carbontetrachloride, Ovaries, Uterus, Testes.

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INTRODUCTION

Carbon tetra chloride was originally synthesized by the French chemist Henri Victor Regnault in 1839 but now produced from methane.2,3

\[\text{CH}_4 + 4 \text{Cl}_2 \rightarrow \text{CCl}_4 + 4 \text{HCl}\]

It is haloalkane used extinguish fires cleaning solvent, a refrigerant, and in lavalamps.4,5 CCl4 leads to generation of free radicals that causes cell injury and necrosis.6,9 It is also known to be hepatotoxic and nephrotoxic to experimental animals.7 Acute exposure to carbon tetrachloride through any route cause gastrointestinal and neurological effects in the first 24 hours, liver damage occur after 24 hours and kidney damage can occur within 1-6 days in association with liver failure.4 Chronic CCl4 treatment induces hepatic fibrosis7, renal9, pulmonary11 and testicular injuries12 and cardiac tissue damage13 in experimental animals.8 Tissue damage by CCl4 depends on the amount of dosage and duration of exposure of the experimental animals to this toxicant.14 Carbon tetrachloride also cause degeneration growth, decrease body weight and irritation of eyes, skin.15 CCl4 toxicity can affect the pituitary gland causing a reduction of FSH and LH leading to degeneration follicle in female. Also causes inhibition of testosterone with degeneration and decrease sperm count and testicular atrophy in male in experimental animals.16,17 CCl4 causes advance cirrhotic state that’s lead to hypogonadism and gonadal dysfunction in male and female.18,19

The aim of the present study was to investigate the effects of Carbon tetrachloride on the body weight and weight of ovaries, uterus, testes with histological changes of ovaries, uterus, testes in rabbits.

MATERIALS AND METHODS

Thirty adult rabbit were obtained from market of Kut. Animals were placed at the animal house, College of science, Wasit University and fed with pellet during experimental periods; temperature was 25˚C. Female was left in a separate cage with one male for the each cage ratio 2:1. Male and female couples were kept together in mating cage for four weeks. The rabbit were randomly divided into two equal groups (control and one treated groups), and the animals were treated as follow:

1: Control group C. (n = 15) received Distilled water 1 ml /kg b.w four times week intraperturnium for for 30 days
2: Treatment group. (n=15) were treated with carbon tetrachloride (1 mg / kg b.w mixing with equal volume of olive oil (1ml/ kg b.w) four times week intraperturnium for 30 days.

The body weight of the animals detected at the day 1th, 7th,14th, 21th and 30th days of the experimental period by electrical balance. After eight day until end of experiment period every day the females were examined for signs of pregnancy by palpating the abdomen. At end of experiment period rabbit weighted and sampling ovaries, uterus, testes weighted by sensitive balance then preserved in 10% formalin buffer solution until preparation of histopathological section. Tissue was cut at 7-8µm and embedded in paraffin and takes sections of ovaries, uterus, testes were...
stained with hematoxylin–Eosin stain (H&E) for histopathological study.21

Statistical Analysis
Data were expressed as mean ± standard error of mean and were compared by one way ANOVA followed by LSD. P value more than 0.05 was considered as statistically significant.23

RESULTS AND DISCUSSION
Table 1 showed a significant decrease of body weight in carbon tetrachloride compared with the control along time of the experimental periods. Except one day non-significant between carbon tetrachloride compared with the control. Carbon tetrachloride is readily absorbed from the gastrointestinal tract causes vomiting and gastrointestinal pain apparent following acute exposure,23,24 but in chronic exposure causes irritation to the gastrointestinal tract with cell injury and necrosis followed by degeneration growth, decrease body weight gradually.25,26

Data in table 2 demonstrated that there was significant decrease in testicular weight with carbon tetrachloride when compared with control group. CCl4 toxicity cues inhibition of testosterone may cause a decrease in the number and function of somatic and germinal cells of testis followed by a testicular weight reduction.27,28

The result of Table 3 showed a significant decrease in the weight of the ovaries in CCL4 treated group when compared with that of normal control. CCl4 toxicity affected the pituitary gland causing a reduction in level of FSH and LH leading to inhibition of follicles and hypogonadism.29

Table 4 indicated that, there was no significant increase in the weight of the uterus of CCL4 treated group when compared with that of normal group. Hypertrophy and hyperplasia of endometrium and myometrium followed by enlargement of uterus.30

Table 1: Effect of Carbon tetrachloride on the body weight of the adult rabbit in the different periods

<table>
<thead>
<tr>
<th>Time</th>
<th>Control Weight rabbit Kg</th>
<th>Carbon tetrachloride Weight rabbit Kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 day</td>
<td>1.96 ± 0.017 a</td>
<td>1.96 ± 0.006 a</td>
</tr>
<tr>
<td>7 day</td>
<td>2.2 ± 0.029 a</td>
<td>1.60 ± 0.052 b</td>
</tr>
<tr>
<td>14 day</td>
<td>2.5 ± 0.097 a</td>
<td>1.44 ± 0.033 b</td>
</tr>
<tr>
<td>21 day</td>
<td>2.11 ± 0.077 a</td>
<td>1.20 ± 0.034 b</td>
</tr>
<tr>
<td>30 day</td>
<td>2.15 ± 0.08 a</td>
<td>1.15 ± 0.68 b</td>
</tr>
</tbody>
</table>

- The value represent Mean (gram) ± Standard Error. The different small letters show significant effect while the same small letters show insignificant effect between different groups.

Table 2: Effect of CCl4 on testicular weight in adult male rabbit

<table>
<thead>
<tr>
<th>Groups</th>
<th>Control</th>
<th>Carbon tetrachloride</th>
</tr>
</thead>
<tbody>
<tr>
<td>After 30 day of treatment</td>
<td>1.86 ± 0.054 a</td>
<td>0.34 ± 0.043 b</td>
</tr>
</tbody>
</table>

- The value represent Mean (gram) ± Standard Error
- The different small letters show significant effect

Table 3: Effect of CCl4 on ovarian weight in adult female rabbit

<table>
<thead>
<tr>
<th>Groups</th>
<th>Control</th>
<th>Carbon tetrachloride</th>
</tr>
</thead>
<tbody>
<tr>
<td>After 30 day of treatment</td>
<td>0.80 ± 0.057 a</td>
<td>0.38 ± 0.015 b</td>
</tr>
</tbody>
</table>

The value represents Mean (gram) ± Standard Error
- The different small letters show significant effect

Table 4: Effect of CCl4 on uterus weight in adult female rabbit

<table>
<thead>
<tr>
<th>Groups</th>
<th>Control</th>
<th>Carbon tetrachloride</th>
</tr>
</thead>
<tbody>
<tr>
<td>After 30 day of treatment</td>
<td>0.45 ± 0.068 a</td>
<td>0.55 ± 0.029 a</td>
</tr>
</tbody>
</table>

The values represent Mean (gram) ± Standard Error
- The same small letters show in significant effect
Cross examination of testes rabbits (Fig 1 - A control group) normal size with no congestion while in (Fig 1 - B treated with carbon tetra chloride) degeneration and atrophy testes with congestion. CCl4 causes advance cirrhotic and necrosis by free radicals which lead to testicular atrophy and dysfunction. Histopathological examination of testes rabbits in (Fig 2-A) control group no degeneration in seminiferous tubule and in leydig cell. (Fig. 2-B) control group show normal size blood vessels (Fig. 2-C) treated group with 1mg/kg of CCL4 mixed with 1 ml of olive oil.(I.P) showed degeneration in seminiferous tubule and leydig cell (Fig. 2-D) treated group hyperplasia blood vessels.

CCl4 induce oxidative stress and free radical in testis may cause degeneration with atrophied of leydig cells with tubules except blood vessels cause enlargement. Cross examination of ovaries rabbits in (Fig. 4-A control group) showed normal size with no hemorrhage while in (Fig. 4-B treated with carbon tetra chloride) showed degeneration and atrophy ovary and hemorrhage spot on ovarian surface. It is evidenced that metabolic activation of CCl4 by cytochrome P450 resulted in the production of trichloromethyl radical (_CCl3) and peroxy trichloromethyl radical (_OOCCl3) that, in turn, initiate subsequent lipid peroxidation, responsible for ovarian injuries and degeneration. Free radical by CCl4 which lead hyper atrophy capillary followed by hemorrhage.
Fig 3: Cross examination of ovaries and uterus rabbits (A) control group. (B) Group treated with 1mg/kg CCL4 mixed with 1 ml of olive oil. (I.P)

Fig 4: Cross examination of ovarian rabbits (A) control group. (B) Group treated with 1mg/kg CCL4 mixed with 1 ml of olive oil. (I.P)

Fig 5: Histolopathological examination of ovarian rabbits (under fox 10*0.25)
Histopathological examination of ovaries rabbits in (Fig. 5-A control group) normal size follicles and no degeneration. (Fig. 5-B control group) showed normal size blood vessels. (Fig. 5-C treated group with 1mg/kg CCL4 mixed with 1 ml of olive oil(I.P) atrophy follicles and degeneration. (Fig. 5-D treated group) hyperplasia blood vessels. CCL4 lead to generation oxidative stress, which produces major interconnected changes of cellular metabolism, increases fragmentation, and destruction with necrosis and degeneration germinal cell with layers but blood vessels may hyperplasia.37,38 Gross examination uterus rabbit of in (Fig. 6-A control group) normal shape and no congestion while in (Fig. 6-B treated with carbon tetra chloride) enlargement of uterine horns, body which were swollen and extensive edema of the organ with congestion. CCl4 causes extensive hemorrhages due to hyperplasia and hypertrophy of the capillary which leads congestion. Hypertrophy of mucosa and muscular layer that cause enlargement of uterus.39 Histolopathological examination of uterus rabbits in (Fig. 7-A control group under fox 4*0.10) consist endometrium, myometrium, with normal size. (Fig7-B control group under fox 10*0.25) normal size glands with blood vessels were seen. (Fig. 7-C treated group under fox 4*0.10 .consist of 1mg/kg CCL4 mixed with 1 ml of olive oil,(I.P) proliferation endometrium and myometriumas. (Fig. 7-D treated group under fox 10*0.25) hyper atrophy gland with blood vessels. CCl4 generates free radicals that causes hyperplastic or metaplastic changes of lining of epithelium of the endometrium, which was that of uterine mucous cuboidal type with proliferation endometrium, myometrium. As well as capillaries and glands there was hypertrophy.40,41
CONCLUSION
The present study has demonstrated the induction of Carbon tetrachloride in intraperitoneum in rabbit animal affects body weight, testes, ovaries, uterus. The results also recorded decrease weight of (body, testes, ovaries) while increased in uterus weight induced by carbon tetrachloride.

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REFERENCES