Study of the effect of decorticated and defatted Castor Seeds (Ricinus communis Linn.) on Testosterone level and testicular architecture of male mice

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Abstract

The aim of this study is to investigate the effect of decorticated and defatted castor seeds (Ricinus communis) on the testes of male mice hormonally and histopathologically at two consecutive spermatogenic cycles. Eighteen albino Swiss male mice were used as animal model, they divided in to three groups equally, 1st group served as control group, while the 2nd group was treated orally with 1.76 mg/kg. B.W. from watery suspension of decorticated and defatted castor seeds extract for 38 days (single spermatogenic cycle). The 3rd group also treated by the same protocol of the 2nd group but was allowed a recovery period (free from the treatment) of another 38 days (double spermatogenic cycles). The measured parameters were: the ratio of testicular weight to the body weight, Levels of testosterone hormone and histopathological study of testicular architecture. The results of the present study showed that the treatment with decorticated and defatted castor seeds caused a significant decrease (P<0.05) in the ratio of weight of testis to the body weight and the Levels of testosterone hormone, in treatment group and these effects have been sustained in the recovery group. The histopathological sections of testis of treated mice showed sever changes in testicular architecture in both periods of the experiment. This was attributed to the lipolytic and cytotoxic activity of ricin, a toxic protein present in castor seeds. In conclusion, the decorticated and defatted castor seeds have negative hormonal and histopathological effects on the testis of mice and this effect was irreversible at the level of two spermatogenic cycles.

Keywords: Castor Seeds, Testosterone

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Introduction

Many trails have been done to investigate the antifertility effect of many medicinal plants in male animals. (Deshpande, et al., 1980); (Zhen, et al., 1995); (Lohiya, et al., 2002); (Mdhluil, and Van der Horst, 2002) and (Coutinho, 2002)Some of these plants had spermicidal effects; others caused reduction in the sperm counts and altered the mobility of the sperms. Some of them caused testicular change and altered hormone levels. (Bhargava, 1984) and (Madhusudhana, et al., 1997)Castor bean plant Ricinus communis (Linn), belongs to the family Euphorbiaceae. Different parts of the plant have been reported to have several medicinal values. (Chiej, 1984). Different parts and extractx of Castor bean has been examined to detect its capability to be used as antifertility agent among different species of animal females (Okwuasaba, et al., 1991); (Al-Tahan, 1994); (Makonnen, et al., 1999-); (Okwuasaba, et al., 1999); (Isichei, et al., 2000); (Das, et al., 2000) and (McNeil, et al., 2003). Sandhyakumary, et al., (2003) and Raji, et al., (2006) studied the antifertility effect of different alcoholic extracts of Ricinus communis on male rats. Both of them found a reversible negative impact on male reproductive functions. Al-Tahan, and Jasim, (2011) concluded that the decorticated and defatted castor seeds have an injurious effects on sperm characters of male mice in particular on sperms mobility of the examined sperms and these effects have continued for two consecutive spermatogenic cycles. Here we are try to investigate if there were a possible effect of the decorticated and defatted castor seeds extract on the testicular architecture and testosterone level and explore if this effect will be reversible or not.

Materials and Methods

This study has been achieved in the animal house of the college of veterinary medicine / Baghdad University, during January – March in 2008. Decorticated and defatted castor seeds extract has been prepared as previously mentioned (Al-Tahan, and Jasim, 2011) and (Al-Tahan and Al-Shaha, 1990). Eighteen albino Swiss male mice were used as animal model, they divided in to three equal groups; 1st group served as control group and received distilled water only, the 2nd group has been treated orally with 1.76 mg/kg (Al-Tahan, 1994), from the watery suspension of decorticated and defatted castor seeds extract for 38 days (single spermatogenic cycle). The 3rd group also treated by the same protocol of the 2nd group but was allowed a recovery period (free from the treatment) of another 38 days (double spermatogenic cycles). After treating periods, animals were weighed, and after sacrifice tests were removed and weighed by sensitive balance after being cleaned from the accessory connective and adipose tissues. Testicular weight to body weight ratio was calculated as in the following equation:

\[
\text{Testicular wt-to-body wt ratio} = \frac{(\text{Wt. of testis (gm) / Wt. of animal (gm)}) \times 100}{1}.
\]

(A. Al Janabi, 2007). Blood samples were obtained via cardiac puncture from each anesthetized animal using disposable insulin syringes. Samples were centrifuged at 2500 RPM for 15 minutes, Levels of the hormone testosterone were measured by using Radio-immunoassay (RIA) kit, after treating the samples with \(^{125}\text{I} \) (Labeled testosterone tracer), by employing Gamma Counter the connection between \(^{125}\text{I} \) with testosterone hormone were measured in ng/ml unit (Al-Janabi, 2007). Tests were excised and cleared off the attached fat and connective tissue. Testis preserved in formalin 10% and Histological sections were prepared for histological study (Luna, 1968). For comparison among groups, the one way ANOVA method was followed by utilizing means and standard error.

Results and discussion

The results of our work revealed to irreversible negative impact of decorticated and defatted castor seeds on testosterone level and ratio of testis weight to the body weight both of two parameters were decreased significantly (P< 0.05) (table1). This effect was credited to presence of the lectins in particular the toxic moiety (Ricin) as a major continent of the decorticated and defatted castor seeds (Al-Tahan, 1994); (Al-Tahan and Al-Shaha, 1990) and (Gruenwald, 2000). Lipolytic activity of ricin on neutral lipids has been mentioned (Gruenwald, 2000) and (Lumbard et al., 2001). Cholesterol is one of the neutral lipids (Lumbard et al., 2001), cholesterol is considered as a precursor in androgenic biosynthesis process (Murray et al., 2003). Consequently the disturbance in testosterone hormone level (table1) was negatively reflects on the ratio of testis weight to body weight (table1) (Raji, et al., 2006). Testosterone is necessary for the development, growth and normal functioning of the testis and male accessory reproductive glands (Kamitchouing et al., 2002) and (Setty, et al., 1977). The abnormal histopathological view of the testis that treated with decorticated and defatted castor seeds 1.76 mg/kg in treatment group (Figure,2) and even sustained severely in the recovery group (figure,3) display a thickness of basement membrane of the seminiferous tubules, vacuolation of Sertoli cells and mono-nuclear cells and cellular debris inside the lumen of the seminiferous tubules in compare with normal histopathological section of normal testis of control group (figure1) , Sertoli cells represent a good target to ricin molecule due to presence of the glycoconjugates on its’ surface (Arenas, et al., 1998) and (Gheri, et al., 2004). Reduction of Sertoli cells led to depletion in its'
function of secreting of androgen binding protein (ABP) which enhance accumulation of testosterone and dihydrotestosterone in high concentrations within the seminiferous tubules and the interstitium of the testis (Cunningham, 2002). This find out led to adverse effects on the structure of the testis and that might due to reduction of serum testosterone levels (Prins, et al., 1991) which consequently caused this persistent and negative effect of decorticated and defatted castor seeds on the testis even on the recovery group.

### Table (1): Effect of decorticated and defatted castor seeds suspension on Testosterone level and testicular weight/body weight ratio in response to different duration of treatment.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Testicular weight / body weight ratio (%)</th>
<th>Testosterone level (ng/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control Group</td>
<td>0.28 ± 0.014 A</td>
<td>0.016 ± 0.0122 A</td>
</tr>
<tr>
<td>Treatment group treated with decorticated and defatted castor seeds <em>Ricinus communis</em> (1.76 mg/kg) at 38th day</td>
<td>0.22 ± 0.010 B</td>
<td>0.010 ± 0.0054 B</td>
</tr>
<tr>
<td>Treatment group at 76th day (Recovery group)</td>
<td>0.21 ± 0.009 B</td>
<td>0.011 ± 0.0067 B</td>
</tr>
</tbody>
</table>

P < 0.05 (n=6). M ± SE

*Difference in letters refers to significant differences among groups.

**References**


