Study, to estimate the effect of new schiff base (R-CH=NAr) on the reproductive hormones of female Rats.

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Abstract

The present study is designed to investigated the effect of prepared chemical base (R-CH=N-Ar) which consist of (camphr andglycin) on reproductive hormones of female rats. Twenty four wistar albino female rats used in this study, divided intothree groups: the first group was treated orally with olive oil as a control group, the second and third groups treated orally with schiff base dissolved in olive oil at two doses (50 mg/kg and 100 mg/kg) respectively for 21 day. The result showed that Schiff base did not affect level of LH hormone in second and third groups compared with control group while, FSH level increased significantly ($p<0.05$) in third group only compared with the first and second group. The result indicated a significant increase ($p<0.05$) in level of progesterone and estrogen hormones in seconedand third groups compared with the control group, also non-significant different in levels of both hormones between second and third groups.

Key words: Camphor, Glycin, LH, FSH, Progesterone, Estrogen, Rats.

تأثير قاعدة شيف (R-CH=NAr) على هرمونات التكاثر في أناث الجرذان المختبرية

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الخلاصة:

صممت الدراسة الحالية لتحديد تأثير قاعدة شيف والمحضرة من الكافور والكلايسين على هرمونات التكاثر في أناث الجرذان. حيث استخدم 24 من أناث الجرذان البيض وقسمت إلى ثلاثة مجموعات: المجموعة الأولى جرعت زيت الزيتون واعترفت مجموعة سيطرة اما المجموعتين الثانية والثالثة فقد تم معاملتهما بقاعدة شيف فمويا بعد إذابتها بزيت الزيتون وبجرعتين (50 ملغم/كغم و 100 ملغم/كغم).
على التوالي ولمدة 21 يوم، أوضحت النتائج ان قاعدة شيف لم تؤثر في مستوى الهرمونات اللوتينية LH في المجموعة الثانية والثالثة مقارنة مع مجموعات السيطرة بينما ازداد مستوى الهرمون المحفز لجلوية (FSH) في المجموعة الثالثة فقط مقارنة مع المجموعتين الأولى والثانية، واظهرت النتائج وجود زيادة معنوية في مستوى الهرمون البروجيستروني والاستروجين في المجموعات الثانية والثالثة مقارنة مع المجموعتين الأولى والثانية. فيما لم يكن هناك فرق معنوي في مستوى كلا الهرمونين بين المجموعتين الثانية والثالثة.

الكلمات المفتاحية: الكافور، الكلاسيسين، الهرمون اللوتيني، الهرمون المحفز للجريبات، هرمون البروجيستروني، هرمون الاستروجين، الجرذان.

**Introduction:**

Schiff base (R-CH=N-Ar) consist from camphor with glycin [1]. This response between them have strong activity exercises for example antimicrobial [2]. Antifungal and anticancer [3]. Camphor is anatural product derived from the wood of the camphor laurel (Cinnamomum camphora L.) tree through steam distillation and purification by sublimination [4] and also produced synthetically from vinyl chloride and cyclopentadiene passing through intermediate dehydronorbornyl chloride [5]. There are many pharmaceuti-cal applications for camphor such as topical analgesic, antiseptic, antispasmodic and anti-inflammatory [6, 7]. Recently investigations have show that camphor containing compounds have uterotrophic [8] antitussive [9] nicotinic receptor blocking [10]. Estrogenic activities [11, 12]. Anchorstructur in this Schiff base is glycin, it is major amino acid in mammals and other animals, it is synthesized from serine, threonine, choline and hydroxproline [13]. On the other hand Glycin plays an important role in metabolic regulation, antioxidative reactions and neurological function [14] many studies have focused on the role of glycin on fertility [15]. Some of these studies found increased the level of Estrogen and progesterone hormone [16] and however other studies found no difference in menstrual cycle length or hormone levels with glycin [17].

The present study aimed to evaluate the effects of new Schiff base (R-CH=N-Ar) which consist of (camphor and glycin) on the level of the reproductive hormones: luteinizing hormone (LH), Follicle stimulating hormone (FSH), progesterone and estrogen hormones of female rats.
Material and method

- synthesis of Schiff base:

Camphor and glycin procured from sigma-Aldrich, fluka and BDH used without filtration. Camphor (20mmol, 3.04g) was dissolved in 20 ml of methanol than glycin add to the solution (20 mmol, 1.5g) in 20 ml of methanol and Acohc ml. The reaction mixture obtained was refluxed for 9h. Upon cooling, the white crystalline powder Schiff base.

- Animal and experimental design:

Twenty-four wistar albino female rats (Rattusnorvegicus) at age (70-80 day old) weighting (200-230 g) were obtained from the Animal house of Department of biology, college of science, university of Thi-qar, Iraq. Animals were housed in standard plastic cages and kept in room temperature of (24±2 C°) with natural light cycle (12h light/12h dark) and access to water and food ad libitum. Animals divided into 3 groups (8 rats of each group): the first group was treated orally with olive oil as a control group, the second and third groups treated orally with schiff base dissolved in olive oil at two doses (50 mg/kg and 100 mg/kg) respectively for 21 day. Animals in all groups received value itself (0.4 ml/rat) from materials.

Blood collection and hormonal assay:

At the end of experiment, blood samples collected by cardiac puncture and put in tubes without EDTA, then centrifuged at 3500 (rpm) for 15 minutes to obtain serum and stored it at -20C° until assay for hormones. The level of luteinizing hormone (LH), Follicle stimulating hormone (FSH), progesterone and estrogen hormones measured by using Immulite 1000 device in Siemens kits, German.

2-4 statistical analysis:

Data were analyzed by using the SPSS (version 17.0). Analysis of variance (one-way ANOVA) was performed to test for any significant differences
among groups and independent sample t-test was used to calculate statistical significance between the control group and each treated group. The level of significance was set as $p<0.05$ for all statistical tests (18).

Results:

The result showed that Schiff base did not affect level of LH hormone in second and third groups compared with control group while, FSH level increased significantly ($p<0.05$) in third group only compared with the first and second group table (1) and fig (1). The result indicated a significant increase ($p<0.05$) in level of progesterone and estrogen hormones in second and third groups compared with the control group, also non-significant different in levels of both hormones between second and third group stable (1) and fig (2).

Table (1): the effect of Schiff base (R-CH=N-Ar) on LH, FSH, progesterone and estrogen hormones of female rats.

<table>
<thead>
<tr>
<th>Hormones</th>
<th>control</th>
<th>50mg/kg</th>
<th>100mg/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>mean</td>
<td>Std</td>
<td>mean</td>
</tr>
<tr>
<td>LH (u/l)</td>
<td>0.1000</td>
<td>±0.00</td>
<td>0.1000</td>
</tr>
<tr>
<td>FSH (u/l)</td>
<td>0.1400</td>
<td>±0.018</td>
<td>0.1383</td>
</tr>
<tr>
<td>PROGESTRON (u/l)</td>
<td>20.09</td>
<td>±5.03</td>
<td>60.4100*</td>
</tr>
<tr>
<td>ESTRO (u/l)</td>
<td>18.18</td>
<td>±1.31</td>
<td>23.783*</td>
</tr>
</tbody>
</table>

*mean significant difference at ($p<0.05$) level.
Figure(1): the effect of Schiff base (R-CH=N-Ar) on LH and FSH hormones of female rats.
Figure (2): the effect of Schiff base (R-CH=N=Ar) on progesterone and estrogen hormones of female rats.

Discussion

The mixing between camphor and glycin in new Schiff base recording significantly increased in progesterone and estrogen levels into two dose (50, 100 mg/kg) and in level of FSH hormone when use high dose (100 mg/kg) in female rats which agree with many studies such as (17, 26) were showed significantly increased in these hormones by using (camphor only) and also agree with (19, 20) which found significantly increased in progesterone and estrogen hormones by using (glycin only) but disagree with (21) which founded significantly decreased in FSH hormone.

Camphor is consider as an endocrine disruptor and an estrogen agonist (22) and also progesterone and estrogen were governed by coactivators and repressers of steroid receptors (23) on the other hand glycin has protective effect on estradiol hormone (24) the main reason to increase progesterone and estrogen is induce glycin increase SHBG synthesis and secretion (25).

Therefore synergism between two substance (camphor + glycin) in new Schiff base showed stronger effective in reproductive hormone (progesterone and estrogen) than effective glycin or camphor alone, and reduced the impact demper for glycin in FSH hormone.

On the other hands showed in this research non significant for LH hormone with low and high dose of Schiff base which agree with (27) which found no effect on LH hormone.

A possible explanation of not affected LH hormone, may be due to the increased in the level of total estrogen which produces a negative feedback mechanism leading to un elevated levels of gonadotrophic releasing hormone (GnRH) of the hypothalamus which will stop the secretion of LH.

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