Effect of DMPA and A Polyherbal Drug on Histomorphometry of Uterus in Adult Female Rats

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Abstract: Adult female albino rats were treated with injectable contraceptive (Demedroxyprogesteroneacetate-DMPA) and then treated with a polyherbal drug containing herbal plants extracts which are proved to have uterotonic effect. Animals were divided into various groups. Experimental groups were treated with DMPA. Uterine tissues were removed from all groups and subjected to morphological and histomorphometrical studies. Results were compared and tabulated.

Keywords: Estrous cycle – DMPA – Uterus – Histomorphometry

1. Introduction

Reproductive cycle in female of any species is subject to vary with any stress applied on them. The stress can be in any form – physical or chemical, external or internal. In the present study contraceptive was taken as chemical stress and given internally. Prolonged usage of contraceptives leads to disturbance in reproductive cycle which in turn affects the morphology and morphometry of reproductive tissue. The main aim of the present work is to study the changes occurring in the estrous cycle and uterus in adult female albino rats and the effect of a polyherbal drug on the affected uterus.

2. Methodology

Adult female albino rats weighing about 110-120gms were selected for the present study. (CPCSEA.No.01/02/2000), (CPCSEA.No.01/010/2003)). Animals were divided into four groups of 6 each as in the Table 1. Group I animals were maintained with ad.libitum till the end of the experiment. Group II – IV were given with injectable contraceptive (DMPA) for one complete reproductive cycle (1 reproductive cycle lasts for four consecutive days). DMPA was given intramuscularly at a dose of 0.3 mg/day/rat. Group III animals were treated with the polyherbal drug was given orally at a dose of 500mg/kg body wt/day/rat. This was continued for one complete reproductive cycle. Group IV animals were left free without any drug administration after the treated with DMPA. The polyherbal drug consists of the following medicinal plants – Abroma augusta, Vitis quadrangularis, Crocus sativa, Cinnamomum camphora and Saraca indica). All these are believed to have uterotonic effect. The drug was tested and authenticated by Department of Pharmacology, University of Madras, for its components and absence of steroid. Every day at regular intervals, animals of all the groups are subjected to vaginal swab (Long, J.A.A and Evan, H.M. 1922). A small sterilized cotton probe was inserted into the vagina and mucus was taken. Smeared on a plain slide and stained with H&E. At the end of experimental period, animals were sacrificed by euthanasia and perfused. Uterine tissues were removed by careful dissection and preserved in 10% formalin. All the tissues were subjected to routine H&E staining (Bancroft, D.J.1982). Observations were made on morphological & histomorphometrical studies.

3. Observations

Observations were made on the following aspects and discussed.

1. Vaginal cytology
2. Morphology
3. Histomorphometry of uterus
4. Progesterone and estrogen hormone levels.

Vaginal cytology: Group I animals show normal reproductive cycle (estrous Cycle) with all the four stages (Allen, 1922, Astwood, 1939, Mandl, 1951). Group II – IV animals had a prolonged diestrous stage which was confirmed by the presence of exfoliated desquamated vaginal smear cells Group III animals after the treatment of polyherbal drug showed recovery to the normal reproductive cycle whereas the Group IV animals though they had a slow pace of recovery, they duration each stage of estrous cycle differs from control group. Results were tabulated and compared. (Table.2, Fig.2)

Morphology and Histomorphometry: The uterus and uterine tubes found to be shrunken in Group II, Group II and Group IV. Animals of Group III returned back to normal in comparison with Group I and Group IV animals though recovering to normal, the results are not in same readings with Group I animals. (Fig.3). In Group II animals, the endometrium undergoes atrophy. The endometrial thickness found to be reduced. The number of endometrial glands and the size of the endometrial glands also found to be reduced. (Table.3; Fig.4). Group III after the treatment of polyherbal drug showed slow recovery to normal. The thickness of endometrium found to be increased and there was an increase in the size and the number of endometrial glands. Group IV animals though they showed a recovery, the morphometric measurements in relation to endometrial thickness and the endometrial glands are not as same reading as control group.

4. Discussion

Control group of animals showed normal estrous cycle with the four stages – Proestrus, estrus, metestrus and diestrous. DMPA treated group of animals showed a prolonged diestrous stage which is in agreement with Bhowmik, T and Mukherjea, M. (1988). Uterus undergone atrophic condition in Group II animals which was confirmed by the endometrial thickness and the endometrial glands readings. (Lee, R.A., 1969) Group III animals showed recovery to normal and was proved by the reading and compared with control. Group IV animals were showing a recovery but the result obtained were not on par with the control group.

5. Results

From the observations made, it can be concluded that prolonged exposure to injectable contraceptive leads to disturbances in the reproductive cycle (Erdelyl, G.J., 1976), (Ferin, M. 1999) which in turn affects the uterine functions and anatomy. This leads to changes in the Histomorphometry of uterus. Administration of a polyherbal drug tried to bring back the reproductive cycle to normal. Also the morphometric readings of uterus also showed normal in close relation to control group. Withdrawal DMPA also leads to recovery of reproductive organs to normal but the readings did not match with the control group.

6. Acknowledgement

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References

[12] Siddha material medica, Vol.3(674-677)

Duration of Estrous Cycle

<table>
<thead>
<tr>
<th>Group</th>
<th>Proestrus</th>
<th>Estrous</th>
<th>Metestrous</th>
<th>Diestrous</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>12.8±0.3</td>
<td>12.8±0.3</td>
<td>23.2±0.5</td>
<td>59.3±0.7</td>
<td>106.5±0.8</td>
</tr>
<tr>
<td>II</td>
<td>9.2±0.5</td>
<td>9.5±0.7</td>
<td>33.2±0.4</td>
<td>77.8±0.8</td>
<td>111.2±0.8</td>
</tr>
<tr>
<td>III</td>
<td>12.3±0.5</td>
<td>12.2±0.6</td>
<td>26.0±0.5</td>
<td>58.9±0.4</td>
<td>111.5±0.5</td>
</tr>
<tr>
<td>IV</td>
<td>10.2±0.6</td>
<td>10.5±0.4</td>
<td>29.3±0.3</td>
<td>69.5±0.6</td>
<td>120.0±0.6</td>
</tr>
</tbody>
</table>

Diameter of Endometrial Tissues (in microns)

<table>
<thead>
<tr>
<th>Group</th>
<th>Endometrial Wall</th>
<th>Endometrial Glands</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>876±0.8</td>
<td>132±0.3</td>
</tr>
<tr>
<td>II</td>
<td>364±1.3</td>
<td>65±0.3</td>
</tr>
<tr>
<td>III</td>
<td>850±0.6</td>
<td>120±0.4 ***</td>
</tr>
<tr>
<td>IV</td>
<td>760±0.6</td>
<td>84.5±0.1</td>
</tr>
</tbody>
</table>
Duration of Estrous Cycle

**ENDOMETRIAL WALL THICKNESS & ENDOMETRIAL MORPHOMETRY**

***P ≤ 0.001***

**Diestrous Stage**

**Estrous Stage**

**Endometrial Wall**

**Group I**

**Group II**

**Group III**

**Group IV**

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