EFFECT OF ACUTE MORPHINE ADMINISTRATION ON HORMONAL MODULATION AND THEIR RELATION TO MALE REPRODUCTION

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Abstract

Twenty-four adult male Sprague-Dawley rats weighing 130-140 grams were used in this study. In experiment I, twelve intact adult sexually active male rats were subdivided into two equal groups. In group A, six animals were injected with 0.2 ml saline 60 min. before experimentation, while, in group B, animals were injected with 10 mg/kg b. w. morphine sulphate 60 min. before experimentation. In experiment II, twelve male rats were used to study the effect of acute morphine administration in castrated male rat (castrated two weeks earlier). Animals were subdivided into two equal groups. In group A, animals were injected with 0.2 ml saline 60 min. before starting the experimentation, while, in group B, animals were injected with 10 mg/kg b. w. morphine sulphate 60 min. before experimentation. In both experiments, exploratory behaviour parameters (sniffing, rearing and investigating movement) were recorded for both latency and frequency, sexual behaviour parameters (mounting, intromission and ejaculation) were also recorded. At the end of 30 minute-behavioural test, all animals were sacrificed and trunk blood samples were collected. LH and FSH and prolactin were assayed in blood serum. Epididymal spermatozoa were collected and examined for sperm concentration (million / ml), sperm viability (%), sperm motility (%) and abnormal forms (%).

The obtained results indicated that, in experiment I, acute morphine administration resulted in a non-significant (P<0.05) changes in all of the exploratory behaviour, inhibition in all parameters of sexual behaviour, significant (P<0.01) decrease in LH and FSH, and significant (P<0.01) increase in prolactin level. There was a significant (P<0.01) decrease in sperm viability and motility, while, there were non-significant changes in sperm concentration and abnormal forms. In experiment II, acute morphine administration in castrated male rats resulted in a non-significant change on the exploratory behaviour; a complete inhibition of the sexual behaviour, and non-significant change in LH, FSH and prolactin level. It is concluded that, acute morphine administration affects significantly male reproduction, while, castration does not enhance the effect of morphine administration.
INTRODUCTION

The effect of opioids on different aspects of sexual function may be due to central mechanisms that may underlie these effects which had not yet been determined. However, the role of opioids in the modulation of hormonal or neurotransmitter systems that may subserve different sexual aspects has been examined only within the last decade (Pfaus & Gorzalka, 1987 and Agmo & Paredes, 1988). Acute morphine administration has been claimed to increase or does not affect brain endogenous opioids peptides (EOPS) (Smyth, 1983).

The present study is an attempt to clarify the effect of acute administration of morphine on gonadotropins and prolactin levels, and its relation to male sexual performance on both sexually active male rats.

MATERIALS AND METHODS

Twenty-four adult male Sprague-Dawley rats weighing 130-140 grams were used in this study. Animals were fed mixed cereal diet together with green fodder and dried skimmed milk (waynforth, 1980).

Experiment 1

Twelve intact male rats were used to study the effect of acute morphine administration in sexually active male rats. Animals were subdivided into two equal groups: Group (A) control group, six intact adult sexually active male rats were injected i.m. with 0.2ml saline 60 minutes before experimental conduction. Group (B), six intact adult sexually active male rats injected i.m. with 10mg/kg b.w. morphine sulphate 60min. before conduction of experiment. The dose of morphine was decided according to Wells (1968) and Agmo & Paredes (1988).

In both groups, after 60 minutes of injection, exploratory behaviour parameters (sniffing, rearing and investigating movement) were recorded for both latency (the time elapsed from the beginning of the experiment until end of acting performance by the rat) (expressed in seconds) and frequency (the number of acts performed by the rat per 30 minutes). According to Clark et al. (1988), sexual behaviour parameters (mounting intromission and ejaculation) were also recorded for both latency (expressed in minutes) and frequency (Meyerson et al., 1988). At the end of 30 minute-behavioural test, animals of both groups were sacrificed, and trunk blood samples were collected; serum was separated and kept at 20°C until hormonal assay. LH and FSH hormones were assayed by direct RIA according to the method of Davidson and Henry (1974), and prolactin was assayed following the method of Djursing (1981).
Epididymal spermatozoa were collected and examined for sperm concentration (million/ml), sperm viability and motility. Abnormal forms were determined from film stained with eosin & negrosin (Blom, 1983).

**Experiment II**

Twelve male rats were used to study the effect of acute morphine administration in castrated male rats. Animals were subdivided into two equal groups. Group I. Control castrated group, six castrated rats (operated upon two weeks earlier) were injected i.m. with 0.2 ml saline. Group II. six male castrated rats (castrated two weeks earlier) were injected i.m. with 10 mg/kg b.w. morphine sulphate.

After 60 minutes of injection, both exploratory and sexual behaviour parameters were recorded, blood samples were collected for hormonal assay as in experiment I.

**Statistical analysis**

Student "t" test was applied to compare between the treated and the control groups in each experiment (Snedecor and Cochrans, 1967).

**RESULTS**

**Experiment I**

Acute morphine administration (10 mg/kg b.w. i.m) resulted in a statistically non-significant changes ($P<0.05$) in all of exploratory behaviour (Sniffing, rearing and investigating movement) in both latency and frequency (Table 1).

**Table 1. Effect of morphine treatment on exploratory behaviour in male rats.**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group A (control)</th>
<th>Group B (treated)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Latency (sec.)</td>
<td>Frequency</td>
</tr>
<tr>
<td>Sniffing</td>
<td>2.00±0.00</td>
<td>1.00±0.58</td>
</tr>
<tr>
<td>Rearing</td>
<td>7.00±0.26</td>
<td>4.00±0.37</td>
</tr>
<tr>
<td>Investigating Mov.</td>
<td>150.00±0.37</td>
<td>8.00±0.26</td>
</tr>
</tbody>
</table>

Effects of acute morphine administration on masculine sexual behaviour are shown in Table 2. There was a statistically significant ($P<0.01$) inhibition in all parameters of sexual behaviour.
Table 2. Effect of morphine treatment on sexual behaviour of male rats.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group A (control)</th>
<th>Group B (treated)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Latency (min)</td>
<td>Frequency</td>
</tr>
<tr>
<td>Mounting</td>
<td>5.23±0.07</td>
<td>6.83±0.31</td>
</tr>
<tr>
<td>Inrountion</td>
<td>5.55±0.11</td>
<td>8.83±0.31</td>
</tr>
<tr>
<td>Ejaculation</td>
<td>14.90±0.42</td>
<td>—</td>
</tr>
</tbody>
</table>

* Significant at level P<0.01

Serum FSH and LH levels were decreased significantly (P<0.01) in treated group more than those of the control groups as shown in Table 3, while, the prolactin level was increased significantly (P<0.01).

Table 3. Effects of acute morphine administration on serum hormonal levels.

<table>
<thead>
<tr>
<th>Hormone</th>
<th>Group A (control)</th>
<th>Group B (treated)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FSH</td>
<td>2.5 ± 0.13</td>
<td>0.61 ± 0.02</td>
</tr>
<tr>
<td>LH</td>
<td>2.3 ± 0.13</td>
<td>0.80 ± 0.04</td>
</tr>
<tr>
<td>Prolactin</td>
<td>5.8 ± 0.21</td>
<td>10.5 ± 0.19</td>
</tr>
</tbody>
</table>

Effects of acute morphine administration on seminal parameters are shown in Table 4. There were significant inhibition in both sperm viability and motility (P<0.01), while, there were non-significant (P<0.05) changes in sperm count and abnormal forms.

Table 4. Effects of morphine treatment on seminal parameters of male rats.

<table>
<thead>
<tr>
<th>Characters</th>
<th>Group A (control)</th>
<th>Group B (treated)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sperm cell count</td>
<td>90.33 ± 1.44</td>
<td>85.83 ± 2.56</td>
</tr>
<tr>
<td>Viability %</td>
<td>79.33 ± 1.26</td>
<td>13.33 ± 1.10*</td>
</tr>
<tr>
<td>Motility %</td>
<td>75.83 ± 1.23</td>
<td>11.67 ± 0.68*</td>
</tr>
<tr>
<td>Abnormal forms %</td>
<td>16.50 ± 0.62</td>
<td>14.67 ± 0.62</td>
</tr>
</tbody>
</table>

* Significant at level P<0.05

Experiment II

Effect of acute morphine in castrated male rats on the exploratory behaviour parameters are tabulated in Table 5.
Table 5. Effect of morphine treatment on exploratory behaviour of castrated male rats.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group I (control)</th>
<th>Group II (treated)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Latency (sec.)</td>
<td>Frequency</td>
</tr>
<tr>
<td>Mounting</td>
<td>2.00 ± 0.00</td>
<td>48.17 ± 0.40</td>
</tr>
<tr>
<td>Intromission</td>
<td>8.00 ± 0.37</td>
<td>54.00 ± 0.58</td>
</tr>
<tr>
<td>Ejaculation</td>
<td>144.00 ± 0.93</td>
<td>8.00 ± 0.37</td>
</tr>
</tbody>
</table>

* Significant at level P<0.01

Acute morphine administration at a dose rate of 10 mg /kg b.w. resulted in complete inhibition of sexual behaviour parameters compared to control group (Table 6).

Table 6. Effect of morphine treatment on sexual behaviour of castrated male rats.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group I (control)</th>
<th>Group II (treated)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Latency (min.)</td>
<td>Frequency</td>
</tr>
<tr>
<td>Mounting</td>
<td>0.90 ± 0.90*</td>
<td>1.33 ± 1.33*</td>
</tr>
<tr>
<td>Intromission</td>
<td>0.95 ± 0.95*</td>
<td>1.67 ± 1.67*</td>
</tr>
<tr>
<td>Ejaculation</td>
<td>2.50 ± 2.50*</td>
<td>2.50 ± 2.50*</td>
</tr>
</tbody>
</table>

n = 6 * Significant at level P<0.05

Changes in hormonal level (FSH, LH & PRL) in both groups I & II are tabulated in Table 7. These changes were non-significant.

Table 7. Effects of morphine treatment on levels of castrated male rats.

<table>
<thead>
<tr>
<th>Hormone</th>
<th>Group A (control)</th>
<th>Group B (treated)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FSH</td>
<td>9.90 ± 0.14</td>
<td>9.70 ± 0.14</td>
</tr>
<tr>
<td>LH</td>
<td>10.36 ± 0.22</td>
<td>10.20 ± 0.15</td>
</tr>
<tr>
<td>Prolactin</td>
<td>1.40 ± 0.03</td>
<td>1.50 ± 0.06</td>
</tr>
</tbody>
</table>

DISCUSSION

The effects of morphine administration at a dose rate of 10 mg/kg b.w in both sexually active and castrated groups showed that there were no significant changes (P>0.05) in exploratory behaviour parameters, while, the parameters of sexual behaviour were significantly decreased. These results are in agreement with the acute effects of opioids on sexual behaviour in male rats by other investigators (Hetia, 1977; Mumford & Kumar, 1979; Pfaus & Gorzaika, 1987; Agmo & Paredes, 1988).
The inhibitory effects of morphine on male rats sexual behaviour may be due to a direct effect of morphine on central opioid receptors, decreasing sexual motivation (the initiation of sexual contact with a female) (Agmo & Paredes, 1988), or may be due to inhibitory effect on gonadotropin releasing hormone (GnRH) (Gabriel et al., 1986 and Masotto & Negro-Vilar, 1988). Meites (1962) and Bruni et al. (1977) stated that, acute morphine administration, also, causes hyperprolactinemia which, in turn, can also, inhibit male copulatory behaviour by inhibiting both sexual arousal and erectile functions (Kalra et al, 1983 & Doherty et al., 1989).

Complete inhibition of sexual activities of castrated male rat after administration of 10 mg/kg b.w. may be due to elimination of certain permissive or facilitative role played by gonadal hormones antagonizing the inhibitory action of opioids on male sexual behaviour, or it may be due to increased number of opioid receptors in the brain after castration (Hahn & Fishman, 1979 & 1985 and Pfaus & Gorzalka, 1987).

In treated group, there were no significant changes in sperm cell concentration and sperm morphological abnormalities in relation to control group, while, the sperm motility and viability were significantly decreased. These results were in accordance with those of Davies (1983). These effects may be due to acute hormonal changes (marked decrease in FSH & LH and prolatin increase) which may, in turn, mediate to decrease in serum and intratesticular testosterone levels that affect the physiological functions of accessory sex organs (seminal vesicles and prostate), and may lead to decrease the sperm motility and viability (Ahmed et al., 1987).

The significant decrease in FSH and LH levels were found in line with the results of Van Vuigt et al. (1984), Gabriel et al. (1986), Miller et al. (1986) and Kalra et al. (1988). The inhibitory effect of acute morphine administration on serum FSH and LH levels of intact rats may be due to inhibitory effect of opioid administration on hypothalamic gonadotropin secretion (Masotto & Negro-Vilar, 1988), or due to the testosterone on pituitary cells (Kaynord et al., 1990, Abass et al., 1991 and Salem et al., 1991).

As regards to serum prolactin levels, there was more increase in prolactin level in treated group than in control group. These results are in agreement with those of Bruni et al., (1977), Van-Vuigt & Meites (1980) and Ahmed et al. (1989).
This is due to reduction in dopamine release and turnover in hypothalamic tuberoinfundibular dopaminergic neurons which, in turn, cause increase prolactin level (Van Loon et al., 1980 & Forman et al., 1981).

It is concluded that, acute morphine administration reduces the sexual behaviour, FSH, LH, sperm motility and viability, however, it increases prolactin level. It does not affect exploratory behaviour, sperm concentration and sperm abnormalities in intact rats. On the other hand, acute morphine administration inhibits sexual behaviour, and it does not affect both exploratory behaviour and FSH, LH and prolactin in castrated rats.
REFERENCES


تأثير الجرعة الحادة من المورفين على الموتىات الهرمونية
وتعلقها بالتكاثر في الذكور

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تم استخدام في هذه الدراسة 24 نكرا من الفئران البيضاء، تحوزن أوزانها من 200-150 جم قسمت إلى مجموعتين تحتوي كل منها على تسعة عشر فأراً.

التجربة الأولى:

تم استخدام ثلاثة عشر فأراً قسمت بدورها إلى مجموعتين متساويتين حفظت المجموعة (أ)، 7 ملilitراً من محلل فيسيولوجياً مقطع، بينما حفظت المجموعة الثانية (ب)، 10 ملilitراً من زن الفئران سلفات المورفين. هذا وقد تم الحقن في المجموعتين قبلاً.

إجراء التجربة بحوالي 40 دقيقة.

التجربة الثانية:

تم استخدام آخر عشر فأراً لدراسة تأثير الجرعة الحادة لسلفات المورفين على ظهور الفئران البيضاء، الفيسية (تقسيم الفئران قبل التجربة). تم قصس الفئران إلى مجموعتين متساويتين، حفظت المجموعة الأولى (أ)، 7 ملilitراً من محلل فيسيولوجياً مقطعاً بينما حفظت المجموعة الثانية (ب)، 10 ملilitراً من زن الفئران سلفات المورفين. هذا وقد تم الحقن في المجموعتين قبل أجراء التجربة بحوالي 40 دقيقة.

هذا وقد تم أخذ القياسات الأثرية محق كل تجربة. وفيما: الإشادة المشتركة وال الاستكافاوية. وعندما تتبناي الفئران تقياس مستوى نمو الهرمونات وتقياس مستوى FSH والهرمونات البالغة للفئران، وقد أتى زن الفئران ونسبة الأشكال الكاذبة في الفئران.

وقد لا تتأثر الجرعة الحادة لسلفات المورفين على الفئران السلبية غير معنى على النشاط الاستكشافي وتشتيت كل ساعات النشاط البيئي، ولكنه على نفس معنى في مستويات الهرمونات لدى الفئران البالغة. وهذا بسبب زيادة معيثية في مستويات الهرمون البالغattributes / دراسة المناعة، أما بالنسبة للسلوط الوراثية في برنامج الجين، فقد أتى الفئة الحادة لسلفات المورفين إلى تشتيت معيثية وحركة الفئران، مع عدم الإثارة عادةً على الهرمونات. وبسبب الأشكال الكاذبة، أما بالنسبة للتجربة الثانية، فقد كان الزيادة في مستويات الفئران على زن الفئران في مكانة غير معنى بالنسبة للنهاية الاستكشافي. ومضى الهرمون البالغة للفئران، والذي يمثل الفئران سلفات الهرمون البالغة، وتملك الفئران على زن الفئران في مكانة رقمي معينة. هذا بالإضافة إلى أن نجحت منذ البداية بإجابة.