EFFECTS OF TESTOSTERONE PROPIONATE, 5α-DIHYDROTESTOSTERONE PROPIONATE AND OESTRADIOL BENZOATE ON SERUM LEVELS OF LH AND FSH IN THE CASTRATED ADULT MALE RAT

By

ABSTRACT

The influence of treatment with various doses of testosterone propionate, 5α-dihydrotestosterone propionate or oestradiol benzoate on serum levels of LH and FSH (measured by radioimmunoassay) and on weights of ventral prostates and seminal vesicles was investigated in castrated, adult, male rats. For depression of the high, castrate levels of serum gonadotrophins with either of these steroid esters, the inhibition curves were different for LH and for FSH. Serum LH was kept at levels encountered in intact, adult, male rats by lower doses of steroid ester than was serum FSH. Oestradiol benzoate was the most potent suppressor of the serum gonadotrophins among the steroid esters tested, testosterone propionate the least. Treatment with low doses of oestradiol benzoate, however, resulted in serum FSH levels significantly above those of castrates treated with vehicle only. Finally, administration of a synthetic LH-releasing factor to testosterone propionate, 5α-dihydrotestosterone propionate or oestradiol benzoate treated, castrated, adult, male rats resulted in a further release of both LH and FSH. The latter effect was more pronounced in oestradiol benzoate treated castrates than in testosterone propionate or 5α-dihydrotestosterone propionate treated castrates.
Following gonadectomy in male rats, serum levels of LH and FSH will increase rapidly (Gay & Bogdanove 1969; Gay & Dever 1971; Swerdloff et al. 1972, 1973; Swerdloff & Walsh 1973; Dufy-Barbe & Franchimont 1972; Kalra et al. 1973). Administration of testicular steroids to such animals may be used to suppress serum levels of these gonadotrophins (Gay & Bogdanove 1969; Gay & Dever 1971; Swerdloff et al. 1972, 1973; Swerdloff & Walsh 1973; Dufy-Barbe & Franchimont 1972; Kalra et al. 1973). Testosterone is in this respect a potent suppressor, but little is known about the potency of other steroids, such as 5α-dihydrotestosterone and oestradiol which are also secreted by the testis. In order to study the relative effects of testicular steroids on serum levels of LH and FSH, we have examined the doses of testosterone propionate (TP), 5α-dihydrotestosterone propionate (5α-dhTP) or oestradiol benzoate (EB) to keep circulating LH and FSH in the castrated, adult, male rat at levels comparable with those found in intact male rats of the same age. Furthermore, we have investigated the effect of such substitution doses of testicular steroids on weights of accessory sex organs.

The release of LH and FSH from the pituitary gland is mediated via the hypothalamus which secretes gonadotrophin releasing factor(s). It was suggested (Naftolin et al. 1972) that androgens act on the hypothalamus after being converted to oestradiol-17β. Swerdloff et al. (1972), however, showed that 5α-dihydrotestosterone, which cannot be converted to oestradiol-17β, also inhibits LH and FSH secretion in castrated, adult, male rats. These investigators concluded that the androgens may act directly on the hypothalamic-pituitary axis. It has been reported (Debeljuk et al. 1972) that in normal, adult, male rats treated with relatively high doses of TP the circulating levels of LH and FSH are lower than in EB treated animals following administration of a LH-releasing factor. Since no data exist on effects of steroids on LH and FSH release after injection of gonadotrophic releasing factor to castrated rats, serum levels of LH and FSH were determined in TP, 5α-dhTP or EB treated castrated rats subsequent to administration of a synthetic LH-releasing factor.

MATERIALS AND METHODS

Animals

Four months old male rats (R × U strain) weighing 350–450 g were used throughout the experiments. The animals were kept under controlled lighting (14 hours light and 10 hours darkness) and temperature (20–22°C)-conditions. Laboratory chow and tap water were provided ad libitum.

Experiment I. - Animals were gonadectomized under light ether anaesthesia. Daily, subcutaneous injections of TP, 5α-dhTP or EB in sesame oil (0.04 ml/100 g b. w.)
started immediately after operation and were continued for the next six days. These steroids were injected between 2 and 4 p.m. Control animals received sesame oil only (0.04 ml/100 g b.w.). Steroid esters were purchased from Steraloids, Pawling, New York, and were used without further purification. Twenty-four hours after the last injection of steroids, blood samples were drawn under ether anaesthesia by puncturing the ophthalmic venous plexus. If used 500 ng synthetic LH-releasing factor (Beckman, Palo Alto, California), dissolved in 0.1 ml 0.9% sodium chloride solution, was administered via the jugular vein immediately thereafter. Control animals received 0.1 ml 0.9% sodium chloride solution only via this route and 15 and 45 min after the injection of LH-releasing factor or saline, the animals were bled again by puncturing the ophthalmic venous plexus under light ether anaesthesia. The animals were then sacrificed with chloroform, the ventral prostate and seminal vesicles were dissected free and weighed.

Experiment II. – In one group of gonadectomized animals treated with 5 µg 5α-dhTP/100 g b.w./day for ten days, blood samples were taken under light ether anaesthesia between 2 and 3 p.m. three days after castration. Daily treatment with 5α-dhTP was then continued and blood samples were drawn on day 5, 7 and 10 after removal of the gonads. As controls served castrated animals subjected to the same type of blood withdrawal but treated with sesame oil only (0.04 ml/100 g b.w.). In each of these treatment groups four animals were used. Blood samples from experiments I and II were allowed to clot overnight at 4°C. Serum was stored at −20°C until assayed for gonadotrophins.

Radioimmunoassay

Serum levels of LH and FSH were measured using double antibody radioimmunoassays. Antisera against ovine LH and ovine FSH were obtained by immunizing rabbits with NIH-LH-S17 or NIH-FSH-S9 (Drs. Uilenbroek and Dullaart, Departments of Endocrinology, Growth and Reproduction and Anatomy, Erasmus University, Rotterdam). For both radioimmunoassays the procedures as described by Niswender et al. (1968) were followed. Sensitivity, accuracy and specificity of both radioimmunoassays have been described previously (Welschen et al. 1974, in press). In the radioimmunoassay of rat LH, NIAMD rat LH 1-1 was used for the preparation of the iodinated derivative (Greenwood et al. 1963). The biological activity of this preparation was 1.0 unit NIH-LH-S1/mg as measured by the ovarian ascorbic acid depletion test and the FSH contamination was less than 0.04 unit NIH-FSH-S1/mg as measured by the human chorionic gonadotrophin augmentation test. Iodination was performed with 125I (Philips-Duphar, Petten, The Netherlands). Serum LH levels were expressed on basis of a reference standard preparation (NIAMD rat LH RP-1) which had a biological activity of 0.03 unit NIH-LH-S1/mg and FSH contamination of 0.56 unit NIH-FSH-S1/mg. NIAMD rat FSH 1–1 with a biological activity of 100 unit NIH-FSH-S1/mg and a LH contamination of less than 0.002 unit NIH-LH-S1/mg was used for iodination with 125I (Greenwood et al. 1963) in the radioimmunoassay of rat FSH. Serum FSH levels were expressed on basis of a reference preparation NIAMD rat FSH RP-1 with a biological activity of 2.1 unit NIH-FSH-S1/mg and a LH contamination of 0.02 unit NIH-LH-S1/mg. All serum samples from one single experiment were assayed in duplicate at two levels per assay in order to eliminate inter-assay variations. Statistical significance of the obtained data was determined by Student’s t-test.
RESULTS

Effect of treatment with steroid esters on serum levels of LH and FSH

The effects of daily administration of various doses of TP, 5α-dihydrotestosterone propionate (5α-dhTP) or EB on serum levels of LH and FSH in castrated, adult, male rats are depicted in
Fig. 3.
Effect of treatment with various doses of oestradiol benzoate/100 g b.w./day during seven days on levels of serum FSH and LH in castrated, adult, male rats. Mean data ± sem from three or more animals are shown. Open bars: FSH; Cross bars: LH.

Figs. 1–3. Treatment with 20 µg TP during 7 days gave suppression of circulating LH to levels below those found in castrated controls (P < 0.001), but not significantly different from the concentrations found in intact controls. For suppression of FSH to levels not significantly different from those of intact control animals, 35 µg TP was required. 5α-dhTP in a dose of 5 µg gave significant suppression of LH below castrate levels (P < 0.005) with no significant lowering of FSH. Twice this dose of 5α-dhTP gave FSH levels significantly lower than those of castrates (P < 0.001) and not significantly higher than those of intact controls (P < 0.10). A dose of 2.5 µg 5α-dhTP caused no significant elevation in serum FSH over castrate control levels (P < 0.10).

Treatment with 0.3 µg EB lowered serum LH levels to those of intact control rats and a dose of 0.5 µg EB suppressed serum LH significantly (P < 0.01) compared to intact control animals. Serum FSH, however, was not significantly higher in the former group of rats than in intact controls (P > 0.10). Administration of 0.01 or 0.05 µg EB elevated serum FSH significantly above castrate control levels (P < 0.001 and P < 0.005 respectively). Figs. 4 and 5 depict LH and FSH serum levels respectively in a longitudinal study on castrated, adult, male rats given daily injections of sesame oil containing 5 µg 5α-dhTP.
Fig. 4.
Levels of serum LH in castrated, adult, male rats in a longitudinal study. Mean data ± SEM from four animals are shown. Solid line: castrated animals treated daily with 5 µg 5α-dihydrotestosterone propionate/100 g b.w. (subcutaneous) for ten days; dotted line: castrated animals treated daily with sesame oil (subcutaneous) only for ten days.

Table 1.
Effect of different doses of testosterone propionate (µg/100 g b.w./day for seven days) on organ weights (mg/100 g b.w.) in castrated, adult, male rats (n = 3).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Ventral prostate (mean ± sd)</th>
<th>Seminal vesicles (mean ± sd)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intact, controls</td>
<td>73.7 ± 6.4x</td>
<td>82.2 ± 5.9x</td>
</tr>
<tr>
<td>Castrate, sesame oil</td>
<td>21.7 ± 4.0*</td>
<td>40.3 ± 1.5*</td>
</tr>
<tr>
<td>Castrate, 25 µg TP</td>
<td>74.6 ± 12.0x</td>
<td>62.4 ± 7.5x*</td>
</tr>
<tr>
<td>Castrate, 30 µg TP</td>
<td>82.5 ± 5.8x</td>
<td>81.6 ± 17.8x</td>
</tr>
<tr>
<td>Castrate, 35 µg TP</td>
<td>88.0 ± 10.9x</td>
<td>98.1 ± 4.4x*</td>
</tr>
</tbody>
</table>

* Mean significantly different (P < 0.05 one sided) from mean of intact controls.

x Mean significantly different (P < 0.02) from mean of castrated animals treated with sesame oil.
Levels of serum FSH in castrated, adult, male rats in a longitudinal study. Mean data ± sem from four animals are shown. Solid line: castrated animals treated daily with 5 µg 5α-dihydrotestosterone propionate/100 g b.w. (subcutaneous) for ten days; dotted line: castrated animals treated daily with sesame oil (subcutaneous) only for ten days.

**Fig. 5.**

Table 2.
Effect of different doses of 5α-dihydrotestosterone propionate (µg/100 g b.w./day for seven days) on organ weights (mg/100 g b.w.) in castrated, adult, male rats (n = 3).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Ventral prostate (mean ± so)</th>
<th>Seminal vesicles (mean ± so)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intact, controls</td>
<td>68.1 ± 7.3x</td>
<td>73.9 ± 7.5x</td>
</tr>
<tr>
<td>Castrate, sesame oil</td>
<td>26.0 ± 5.7*</td>
<td>42.2 ± 4.0*</td>
</tr>
<tr>
<td>Castrate, 2.5 µg 5α-dhTP</td>
<td>41.3 ± 6.1x*</td>
<td>51.5 ± 6.4*</td>
</tr>
<tr>
<td>Castrate, 5 µg 5α-dhTP</td>
<td>41.7 ± 7.9*</td>
<td>58.6 ± 7.3x</td>
</tr>
<tr>
<td>Castrate, 7.5 µg 5α-dhTP</td>
<td>51.5 ± 9.4x</td>
<td>67.8 ± 9.1x</td>
</tr>
<tr>
<td>Castrate, 10 µg 5α-dhTP</td>
<td>72.9 ± 7.2x</td>
<td>74.3 ± 17.6x</td>
</tr>
<tr>
<td>Castrate, 20 µg 5α-dhTP</td>
<td>58.0 ± 8.3x</td>
<td>81.7 ± 11.0x</td>
</tr>
<tr>
<td>Castrate, 30 µg 5α-dhTP</td>
<td>69.5 ± 2.2x</td>
<td>77.3 ± 7.9x</td>
</tr>
</tbody>
</table>

* Mean significantly different (P < 0.05 one sided) from mean of intact controls.

* Mean significantly different (P < 0.05) from mean of castrated animals treated with sesame oil.
Table 3.
Effect of different doses of oestradiol benzoate (µg/100 g b. w./day for seven days) on organ weights (mg/100 g b. w.) in castrated, adult, male rats (n = 3).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Ventral prostate (mean ± sd)</th>
<th>Seminal vesicles (mean ± sd)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intact, controls</td>
<td>68.1 ± 7.3x</td>
<td>75.9 ± 7.5x</td>
</tr>
<tr>
<td>Castrate, sesame oil</td>
<td>26.0 ± 5.7*</td>
<td>42.2 ± 4.0*</td>
</tr>
<tr>
<td>Castrate, 0.01 µg EB</td>
<td>24.2 ± 5.4*</td>
<td>54.8 ± 2.5x*</td>
</tr>
<tr>
<td>Castrate, 0.05 µg EB</td>
<td>25.5 ± 2.5*</td>
<td>56.1 ± 3.0x*</td>
</tr>
<tr>
<td>Castrate, 0.10 µg EB</td>
<td>24.2 ± 2.8*</td>
<td>60.7 ± 6.4x</td>
</tr>
<tr>
<td>Castrate, 0.50 µg EB</td>
<td>28.6 ± 6.4*</td>
<td>65.0 ± 3.5x*</td>
</tr>
</tbody>
</table>

* Mean significantly different (P < 0.05 one sided) from mean of intact controls.
x Mean significantly different (P < 0.01) from mean of castrated animals treated with sesame oil.

Five days after castration there was a significant difference in serum LH between castrated animals receiving sesame oil and 5 µg 5α-dhTP in this vehicle (P < 0.05), while the level of serum LH in the castrates after treatment with 5 µg 5α-dhTP was not significantly higher than the intact control level (P > 0.10). The FSH levels were not significantly different between castrates administered sesame oil or 5 µg 5α-dhTP in sesame oil during this longitudinal study.

Effects of steroid esters on weights of accessory sex organs

The effects of various doses of TP, 5α-dhTP or EB on ventral prostate and seminal vesicles weights are summarized in Tables 1–3. When given by daily injections, less 5α-dhTP than TP was required to maintain normal ventral prostate and seminal vesicles weights in castrated, adult, male rats. Administration of ≥ 0.01 µg EB prevented the weight decrease of the seminal vesicles as observed in castrated rats, although the weights were slightly decreased when compared to intact controls. The weights of the ventral prostate in animals treated with either amount of EB, even with 0.5 µg, were however, not significantly different from those of control castrates.

Effects of synthetic LH-releasing factor in TP, 5α-dhTP, or EB treated, castrated, adult, male rats

Table 4 shows levels of serum LH and FSH following intravenous injection of 500 ng synthetic LH-releasing factor in castrated rats treated during the 7 preceding days with 40 µg TP, 10 µg 5α-dhTP or 1 µg EB per day. 15 min
Table 4.
Effect of 500 ng synthetic LH-releasing factor (LH-RF) on serum LH and FSH levels in intact, adult, male rats and castrated, adult, male rats, treated for seven days with either sesame oil, 40 µg testosterone propionate/100 g b.w./day, 10 µg 5α-dihydrotestosterone propionate/100 g b.w./day or 1 µg oestradiol benzoate/100 g b.w./day. A single injection of LH-RF was given on day 8 at 3 p.m. Effect of injecting saline in intact, adult, male rats is also shown. Blood samples were drawn immediately before and 15 and 45 min after injection of saline or saline containing LH-RF. Three or more animals were used per experiment.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>ng NIAMD rat LH RP-1/ml serum (mean ± sd)</th>
<th>ng NIAMD rat FSH RP-1/ml serum (mean ± sd)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intact controls</td>
<td>80 ± 19</td>
<td>378 ± 42</td>
</tr>
<tr>
<td>15 min after 500 ng LH-RF</td>
<td>209 ± 38</td>
<td>463 ± 42</td>
</tr>
<tr>
<td>45 min after 500 ng LH-RF</td>
<td>108 ± 37</td>
<td>487 ± 49</td>
</tr>
<tr>
<td>15 min after saline</td>
<td>73 ± 4</td>
<td>378 ± 9</td>
</tr>
<tr>
<td>45 min after saline</td>
<td>64 ± 17</td>
<td>377 ± 13</td>
</tr>
<tr>
<td>Control castrates, sesame oil</td>
<td>220 ± 15</td>
<td>958 ± 67</td>
</tr>
<tr>
<td>15 min after 500 ng LH-RF</td>
<td>364 ± 184</td>
<td>1107 ± 99</td>
</tr>
<tr>
<td>45 min after 500 ng LH-RF</td>
<td>303 ± 113</td>
<td>1119 ± 123</td>
</tr>
<tr>
<td>Castrates, 40 µg TP</td>
<td>31 ± 5</td>
<td>406 ± 78</td>
</tr>
<tr>
<td>15 min after 500 ng LH-RF</td>
<td>89 ± 19</td>
<td>464 ± 151</td>
</tr>
<tr>
<td>45 min after 500 ng LH-RF</td>
<td>59 ± 13</td>
<td>465 ± 50</td>
</tr>
<tr>
<td>Castrates, 10 µg 5a-dhTP</td>
<td>n. d.*</td>
<td>508 ± 64</td>
</tr>
<tr>
<td>15 min after 500 ng LH-RF</td>
<td>111 ± 19</td>
<td>642 ± 20</td>
</tr>
<tr>
<td>45 min after 500 ng LH-RF</td>
<td>48 ± 24</td>
<td>673 ± 91</td>
</tr>
<tr>
<td>Castrates, 1 µg EB</td>
<td>21 ± 2</td>
<td>470 ± 63</td>
</tr>
<tr>
<td>15 min after 500 ng LH-RF</td>
<td>399 ± 23</td>
<td>992 ± 115</td>
</tr>
<tr>
<td>45 min after 500 ng LH-RF</td>
<td>242 ± 11</td>
<td>843 ± 295</td>
</tr>
</tbody>
</table>

* n. d.: not detectable.

after injection of this LH-releasing factor (dissolved in saline) there was a significant increase (P < 0.025) in serum FSH and LH levels, compared with the levels found before injections of this LH-releasing factor. However, no significant increase occurred in serum LH and FSH in castrates given saline only and in serum FSH in castrates treated with 40 µg TP for the 7 previous days.

DISCUSSION
The data presented show the effects of daily injections of TP, 5α-dhTP or EB on serum levels of LH and FSH and on weights of ventral prostates and
prostate
hibiting
vesicles
5a-dhTP
5a-dihydrotestosterone
5a-dhTP
reaches
after
castration.
8c
8c
castrates.

duration
tuitary-hypothalamic
8c
8c
vestigators
serum
controls
5a-dhTP
reaches
after
castration.
(Figs. 1–3)
T
Serum
levels
of
androgen
in
the
system
are
required
to
maintain
normal
weights
of
ventral
prostates
and
seminal
vesicles
after
castration
were
slightly
higher
than
those
effecting
normal
LH
after
castration
(Tables 1 and 2, Figs. 1 and 2). If the same
amount
of
androgen
reaches
the
target
organs
following
daily
administration
of
either
TP
or
5a-dhTP
in
sesame
oil,
5a-dhTP
appears
to
be
more
potent
than
TP
in
inhibiting
secretion
of
gonadotrophins
after
castration
and
in
keeping
ventral
prostate
and
seminal
vesicles
weights
of
castrated
rats
equal
to
those
of
intact
controls
(Figs. 1 and 2, Tables 1 and 2). However,
the
suppression
curves
for
serum
LH
and
FSH
by
steroid
esters
are
not
parallel
(Figs. 1–3). Other
investigators
(Gay
&
Bogdanove
1969;
Swerdloff
et
al.
1972,
1973;
Swerdloff
&
Walsh
1973;
Dufy-Barbe
&
Franchimont
1972;
Kalra
et
al.
1973;
Mallampati
&
Johnson
1973)
who
have
studied
the
problem
of
steroid
effects
on
the
pituitary-hypothalamic
system
have
used
other
time
schedules
with
regard
to
duration
of
steroid
therapy
and
interval
after
castration
such
treatment
was
started.
The
results
obtained
in
the
current
work
are
in
agreement
with
those
previously
reported,
although
smaller
doses
of
steroid
esters
were
required
in
our
investigation
to
keep
normal
serum
LH
and
FSH
levels
in
castrates.
Since
5a-dihydrotestosterone
cannot
be
converted
to
oestradiol-17β,
the
current
work
and
also
the
data
of
Swerdloff
et
al.
(1972)
decide
that
the
androgens
must
not
necessarily
be
metabolized
to
oestrogens
in
order
to
inhibit
pituitary
production
of
gonadotrophins. The
doses
of
5a-dhTP
or
5a-dihydro-
testosterone
used
in
the
present
investigation
and
by
Swerdloff
et
al.
(1972)
are
high,
however,
when
compared
to
the
production
in
vitro
of
5a-dihydrotestos-
terone
by
the
rat
testis
(Folman
et
al.
1972). Moreover,
in
neither
investigation
the
proof
has
been
delivered
that
5a-dihydrotestosterone
is
the
"active
form"

of
this
androgen
at
the
tissue
organization
tested.
Low
doses
of
EB
gave
a
significant
increase
in
serum
FSH
over
castrate
levels
possibly
indicating
a
stimulatory
feedback
mechanism,
if
circulating
gonadotrophins
are
true
indications
of
pituitary
production
and
secretion
de
these
hormones.
However,
it
must
be
kept
in
mind
that
the
clearance
rates
may
differ
for
the
different
steroids,
employed
in
our
work
and
that
the
use
of
one
daily,
subcutaneous
injection
of
a
steroid
ester
may
result
in
a
variable
concentration
of
that
steroid
in
circulating
blood
over
the
ensuing
24
hours.
This
is
indeed
the
case
when
TP
is
administered
subcutaneously
to
castrated
rats
(unpublished
observations).
Therefore,
we
have
used
a
strict
time
schedule
of
daily
steroid
injections
and
in
the
longitudinal
study
(Figs. 4 and 5) levels
of
serum
LH
and
FSH
on
day
5,
7 and 10 after castration in the adult, male rats treated with 5 \( \mu g \) 5a-dhTP are rather similar.

Administration of LH-releasing factor caused an increase in serum LH and FSH levels in castrated, adult, male rats treated with steroid esters (Table 4). Using the same dose of this LH-releasing principle higher serum levels of FSH and LH were obtained in castrates treated with EB than in castrates treated with either TP or 5a-dhTP (Table 4). In normal, male rats treated with relatively high doses of TP the circulating levels of LH and FSH are lower than in EB treated animals following administration of a LH-releasing factor (Debeljuk et al. 1972). Circulating levels of LH and FSH in the castrates are affected in a different way by the steroid treatments, which might have influenced the hypothalamic secretion of releasing factor(s) and/or modified the sensitivity of the pituitary gland to hypothalamic regulators. The nature and the dose of the steroid employed may play an important role in both aspects. The sensitivity to administration of LH releasing factor appeared to be higher in EB treated castrates than in TP or 5a-dhTP treated castrates.

The oestrogen-androgen interaction at the level of the hypothalamic-pituitary axis may still remain the topic of much debate. Since it is known that C_{19}-steroid 5a-reductase activity will increase in the adrenal gland (Maynard & Cameron 1973) and in the hypophysis (Kniewald & Milković 1973) following castration, it is of interest to measure circulating levels of LH and FSH in adrenalecto-mized, gonadectomized male rats.

ACKNOWLEDGMENTS

This investigation was supported by a grant from the Norges Almenvitenskapelige Forskningsråd.

We are grateful to the National Institute of Arthritis and Metabolic Diseases, Rat Pituitary Program, for gifts of the preparations used in the radioimmunoassay of gonadotrophins.

REFERENCES


Received on January 14th, 1974.