USE OF URINARY ESTRONE AND OTHER PARAMETERS IN MONITORING GONADOTROPIN THERAPY

by

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Introduction

The success of gonadotropin therapy for induction of ovulation and conception depends to a great deal on the accuracy and reliability of parameters used for monitoring this therapeutic regime.

Chemical estimation of ovarian steroids, though feasible and commonly employed in developed countries, is still not very common in our country. The changes in the end organs such as cervix, vaginal epithelium and endometrium and basal body temperature (BBT) are commonly used to assess the follicular response to gonadotropic stimulation.

The object of this study was to establish normal patterns of urinary estrone in normally menstruating women with special emphasis on the pre-ovulatory ranges and evaluate its use along with other parameters in women treated with gonadotropins.

Material and Methods

Serial urinary estrone estimations were carried out in 16 women with normal ovulatory cycles. These women were asked to keep their BBT records and were repeatedly called for vaginal smear cytology and cervical mucus fern and Spinnbarkeit tests on as many days as possible.

Urinary estrone was done by Brown's method (Brown et al, 1968) using Alumina chromatography. It was possible to carry out two to three determinations per day within five to six hours, so that decision on therapy could be made by the evening of the same day.

Results

A total of 195 urine samples in normal women were assayed. Data were pooled vertically so that values recorded on a particular day were superimposed. The means and ranges of estrone values found during normal menstrual cycle are shown in Figure 1. The mean values are low during the first week, about 5.35 ugs. on day seven, then rise rapidly to a peak around day 14, the mean value being 17.48 ± 1.28 (95% confidence limits being 14.91 to 20.05). The estrone value then falls with no definite rise during the mid-luteal phase as demonstrated by Brown.
It is found that the estrone peak coincides with the BBT dip ± one day in 92% cycles and with maximum cervical mucus response in 88% cycles. As far as KPI is concerned, it coincides with it in 60% of the cycles. In the remaining, the KPI peak usually precedes the estrone peak. In cases where typical pre-ovulatory or ovulatory smear patterns are obtained, this finding can be very useful in timing the fertile period.

Gonadotropin treatment for the induction of ovulation was given in properly selected and worked up cases of amenorrhea (primary or secondary) with an in-
fertility problem. Since HMG is very expensive and not easily available, it was not possible for us to have a large series.

Table II shows the type of therapy given and the number of cycles. There were 10 patients, 4 of primary amenorrhea and 6 of secondary amenorrhea treated for 20 cycles. HMG followed by HCG was given in 12 cycles, HMG alone in 3 cycles and Clomid followed by HCG in 5 cycles.

The parameters used for monitoring the dosage of HMG therapy at the time of HCG administration are shown in Table III.

In our earlier treated cycles we depended mostly on KPI, cervical mucus and BBT for assessing degree of follicular activity, since urinary estrone was not yet standardized. During the last one year daily or alternate day urinary estrones are regularly carried out on all such patients, including those treated with Clomid. Urinary pregnanediol was carried out during the second half of the cycle for luteal function.

Figure 3 shows a patient of primary amenorrhea monitored by KPI in vaginal smears and cervical mucus. It took cervical mucus five days to change from...
Figure 4 shows the results with gonadotropin therapy in a patient with secondary amenorrhea. She was monitored by K.P.I., cervical mucus, urinary estrone and B.B.T.

Unfortunately, urinary estrone values were not available immediately and one had to depend mostly on KPI and cervical mucus changes. The KPI did not go up satisfactorily, the maximum KPI was about 37%. Neither did the cervical mucus with the increasing HMG therapy. The timing of HCG was thus delayed, resulting in an ovulatory cycle (confirmed by secretory endometrium), but no conception. Actually the BBT dip could be observed even before HCG was given indicating ovulation occurrence on HMG alone. Perhaps urinary estrone value if available in time could have indicated the optimum timing of HCG administration.

Figures 5 and 6 show two cases of primary amenorrhea where large amounts of H.M.G. failed to stimulate the ovarian follicular activity as evidenced by the K.P.I. The second one showed lowering of KPI in spite of continued treatment and hence the treatment was stopped. Another reason for stopping the treatment was the cost of the drug.
A fairly reliable assessment of ovarian function during the pre-ovulatory period is possible by serial urinary estrone estimations. The pre-ovulatory phase is without doubt the most important period during HMG therapy. It is also well recognized that physical changes in cervical mucus and KPI of vaginal smears can be used as semi-quantitative indicators of estrogenic activity. Thus, theoretically the optimum time for inducing ovulation by HCG administration should be the day when estrogen secretion or KPI and cervical mucus reach the pre-ovulatory titre or values similar to that obtained in a normal menstrual cycle. The average estrone values reached just before HCG in our series was 14.5 ugs., whereas the average KPI for the same purpose was 71% as noted in Table IV.

**Table IV**

<table>
<thead>
<tr>
<th>Average Estrone Values Before HCG Injection</th>
<th>14.5 ugs.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Average K.P.I. Values Before HCG Injection</td>
<td>71%</td>
</tr>
<tr>
<td>The cervical mucus show a +++ score</td>
<td></td>
</tr>
</tbody>
</table>

Table V shows correlation of estrone peak to other parameters during gonadotropin therapy. It shows that estrone peak coincided with the day of the BBT dip and maximum cervical mucus (++++) response in 87.5% cycles but coincided with the KPI peak on the same day in only 62.5% cycles which is similar to what happens in normal cycles.

**Table V**

<table>
<thead>
<tr>
<th>No. of Cycles</th>
<th>With BBT</th>
<th>With KPI Cervical Mucus Evaluated</th>
<th>Same Day</th>
</tr>
</thead>
<tbody>
<tr>
<td>8</td>
<td></td>
<td>87.5% 62.5%</td>
<td>87.5%</td>
</tr>
</tbody>
</table>

The KPI peak in most cases precedes the estrone peak. Probably the reason for this is that after the HCG administration the estrone values continue to rise as a result of synergistic action of the two gonadotropins, HMG and HCG, whereas the vaginal smear which also reflects progestational activity probably shows early changes of regression by fall of KPI due to pre-ovulatory secretion of progesterone (Johansson, 1969).

Table VI shows the KPI and estrone patterns immediately after HCG administration.

![Image](https://via.placeholder.com/150)

Table VI shows the KPI and estrone patterns immediately after HCG administration. Figure 7 shows a patient of primary amenorrhea with steep rise of KPI with HMG treatment in relation to urinary estrone. KPI was about 80% and urinary estrone 12.5 ugs. when HCG was given. Cervical mucus took six days to change from + to ++++. After HCG, KPI rose to 94% and then came down, whereas estrone continued to rise to 32 ugs. In this

**Table VI**

<table>
<thead>
<tr>
<th>No. of Cycles</th>
<th>K.P.I. Fall Rise</th>
<th>Urinary Estrone Fall Rise</th>
</tr>
</thead>
<tbody>
<tr>
<td>16*</td>
<td>60% 40%</td>
<td>12.5% 87.5%</td>
</tr>
</tbody>
</table>

*In one patient there was no follow-up and in three cycles ovarian response to the HMG was poor and hence HCG was not given.
amenorrhea who responded to Clomid with ovulatory cycles for about a year but did not conceive. Eventually she was treated with gonadotropins resulting in pregnancy in the very first cycle. HCG was given when estrone was 14 ugs. and KPI was 75% and cervical mucus ++++. The estrone curve again shows rise after HCG, whereas KPI tends to come down.

Most investigators are not agreed on the necessity of monitoring ovarian function closely in cases treated with Clomid or Clomid with HCG. However, if the objective is to diagnose ovulation time as closely as possible, assessment of pre-ovulatory estrogenic activity by direct hormone assays and by end organ effect may indicate the optimum time for coitus or for administration of HCG in such cases.

Figure 9 is a case of secondary amenorrhea treated with Clomid and HCG.
USE OF URINARY ESTRONE IN MONITORING THERAPY

HCG was given when cervical mucus reached ++ to +++ score, KPI reached 65% and urinary estrone was 14 ugs. After HCG the KPI came down, whereas the estrone values continued to rise to 26 ugs. and ovulation seems to have occurred on that day and thereafter the titres came down.

Figure 10 shows another case of secondary amenorrhea treated with clomid and HCG. In this case both KPI and estrone went up after HCG for a day or so coinciding with BBT dip, and then came down. Estrone was 14 ugs. and KPI 52% when HCG was given.

While the pre-ovulatory phase and dosage of HMG can be fairly well controlled by serial estrone assays, karyopyknotic index and cervical mucus changes, the parameters for ovulation and luteal activity are assessed by biphasic shift of BBT, pregnanediol or progesterone secretion, endometrial biopsy and progestational changes in vaginal smears. Of all these parameters, the adequacy of corpus luteal function can be best assessed by luteal hormone assays, or the degree of regression and progestational changes in vaginal smears. Hormone assays take time and endometrial biopsy is not usually done in each and every cycle. Vaginal smear pattern during the second half of the cycle can thus be used to indicate whether further administration of HCG is necessary.

The number of patients and treated cycles are very small but results in properly selected cases of amenorrhea appear quite satisfactory. There are 11 ovulatory cycles out of 12 and 4 single pregnancies in 5 patients. Mild degree of overstimulation like pain in the lower abdomen and moderate enlargement of the ovaries were noted in 7 out of 12 cycles.

**TABLE VII**

Results of Gonadotropin Therapy

<table>
<thead>
<tr>
<th>Type of Therapy</th>
<th>No. of Patients</th>
<th>No. of Cycles</th>
<th>No. of Ovulation</th>
<th>No. of Pregnancies</th>
</tr>
</thead>
<tbody>
<tr>
<td>HMG + HCG</td>
<td>5</td>
<td>12</td>
<td>11</td>
<td>4</td>
</tr>
<tr>
<td>Only HMG</td>
<td>3</td>
<td>3</td>
<td>Nil</td>
<td>Nil</td>
</tr>
<tr>
<td>Clomid + HCG</td>
<td>4</td>
<td>5</td>
<td>4</td>
<td>Nil</td>
</tr>
</tbody>
</table>

*Two patients had more than one type of therapy.*
It is obvious that the objective of using these various parameters with gonadotropin therapy is to find the correct dose and timing of HMG and HCG for an individual subject which will produce changes in endogenous ovarian hormones which are as close to normal ovulatory cycle as possible. Thus, the chances of normal single pregnancies are likely to improve.

Acknowledgements

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References