Metabolic Clearance and Production Rates of Human Luteinizing Hormone in Pre- and Postmenopausal Women

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ABSTRACT Metabolic clearance rates and production rates of human luteinizing hormone (HLH) were determined in pre- and postmenopausal women by the constant infusion technique. Highly purified HLH-\(^{131}\)I was infused into the fasting subjects at a constant rate. Serial plasma samples were obtained and the radioactive hormone was precipitated by a double antibody technique. Plasma HLH-\(^{131}\)I levels reached equilibrium by 4 hr after the infusion started. Metabolic clearance rates were: \(24.4 \pm 1.8\) (mean \(\pm\) se) ml/min in five normal premenopausal women; \(23.3 \pm 1.1\) ml/min in five normal women taking norethindrotriol and mestranol; and \(25.6 \pm 4.1\) ml/min in four postmenopausal women. Endogenous plasma HLH levels measured in the same subjects by radioimmunoassay immediately before infusion were \(32.0 \pm 9.6\) mU/ml in the normal women, \(16.8 \pm 3.2\) mU/ml in the women on oral contraceptives, and \(99.2 \pm 23.2\) mU/ml in the postmenopausal women. The corresponding HLH production rates were: \(734 \pm 170\) mU/min in the normal women; \(387 \pm 86\) mU/min in the women on norethindrotriol and mestranol; and \(2400 \pm 410\) mU/min in the postmenopausal women. The metabolic clearance rate did not change after ovariectomy in one

women, but the production rate rose from 583 to 1420 mU/min. Based on previously reported bioassay values for pituitary content and urinary excretion of HLH, the estimated turnover of HLH in the pituitary is about once per day and less than 5% of the total HLH produced appears in the urine in a biologically active form.

INTRODUCTION

Most of the information available on human gonadotropin physiology has been derived from bioassay data on extracts of large volumes of plasma or urine. The quantity of these fluids required for bioassay has prevented sequential studies at short intervals in a single patient. The recent preparation of highly purified human luteinizing hormone (HLH) (1) and the development of a sensitive radioimmunoassay (2) with which HLH can be easily quantified in small samples of plasma or serum have now made it possible to estimate the pituitary secretion or production rate (PR) of HLH.

The purpose of the present study was to estimate the PR of HLH using the plasma clearance of HLH-\(^{131}\)I and the endogenous plasma HLH level. Initial studies on plasma HLH-\(^{131}\)I levels after a single injection showed a rapid multiexponential disappearance curve, never quite reaching a clearly linear slope during the time HLH-\(^{131}\)I blood levels could be reliably determined. Therefore, we have utilized the constant infusion (to equilibrium) method of Tait (3, 4) to estimate the metabolic clearance rate (MCR) or HLH-\(^{131}\)I from plasma.
METHODS

Preparation of HLH-\(^{131}\)I. Highly purified HLH (1) was labeled to specific activities of 50-150 \(\mu\)c/\(\mu\)g with \(^{131}\)I by a modification of the method of Greenwood et al. (5). Exposure of the hormone to Chloramine T was limited to 15-20 sec before addition of sodium metabisulphite to avoid unnecessary damage to the hormone. HLH-\(^{131}\)I at these specific activities contains about one \(^{131}\)I atom per five molecules of HLH. The relative abundance of \(^{131}\)I per total iodide in so-called "carrier-free \(^{131}\)I" has been found to range from 5 to 44% (6). Therefore, the ratio of total iodide atoms to HLH molecules was about 1:1. Immediately after iodination the mixture was passed through a Sephadex G-75 column. The fraction selected for studies showed essentially no damaged HLH-\(^{131}\)I or free \(^{131}\)I by hydrodynamic flow chromatoelectrophoresis on Whatman 3 MM paper (Fig. 1). The HLH-\(^{131}\)I was sterilized by Millipore filtration, cultured, and pyrogen tested. The interval between iodination and testing was 2-5 days.

Measurement of HLH-\(^{131}\)I. HLH-\(^{131}\)I was precipitated from plasma or solution by a double antibody system (7). Aliquots of 1 ml were treated with excess rabbit anti-HLH at 4°C for 24 hr. A sheep anti-rabbit globulin was added in excess, and the mixture was in-ubated for an additional 24 hr. The samples were centrifuged, and the supernatant was removed by suction. Total and precipitated radioactivity were determined on duplicate aliquots in a well-type scintillation counter. Greater than 97% of the fraction used for studies was precipitable in a double antibody system with excess antibody on the day of iodination.

Measurement of endogenous HLH. Plasma HLH was determined by radioimmunoassay as previously described (2).

Plasma HLH disappearance studies. Preliminary studies were performed to determine the feasibility of using the disappearance curve of HLH-\(^{131}\)I from plasma after a single injection to determine the MCR and PR of HLH. The disappearance of total and antibody precipitable radioactivity from plasma after a single bolus injection of HLH-\(^{131}\)I was investigated in four patients: three with chromophobe adenomas (a 45 yr old woman, a 62 yr old woman, and a 44 yr old man) and a 40 yr old woman with carcinoma of the breast. All patients were given five drops of saturated potassium iodide solution every 6 hr for 24 hr before study to inhibit \(^{131}\)I uptake by the thyroid. 12-25 \(\mu\)c of HLH-\(^{131}\)I which contained the equivalent of 1.6-3.2 \(U\) were injected intravenously in 0.5-1.0 ml of saline. Serial 10-ml heparinized venous blood samples were obtained over a 24 hr period. The patients were kept at bed rest and received a liquid diet during the study. Urine was collected periodically for 24 hr.

Because the HLH-\(^{131}\)I disappearance curve after the single injection appeared multipexponential, the MCR was calculated according to the formula for any system of pools (3, 8):

\[
MCR = \frac{\int_0^\infty x \cdot dt}{\int_0^\infty x^2 \cdot dt}
\]

FIGURE 1 Scans of paper chromatoelectrophoretograms of HLH-\(^{131}\)I after iodination. Origin is near cathode, and anode is to the right. A is the total iodination mixture showing HLH-\(^{131}\)I, "damaged HLH-\(^{131}\)I," and free \(^{131}\)I. B is the fraction used for clearance studies immediately after passage through a G-75 Sephadex column. C is the same fraction as B after incubation with excess anti-HLH for 1 hr, showing that the material adhering to the origin is immunoreactive. D is the B fraction as prepared for infusion.

\(^1\) Obtained from Iso-Serve, Cambridge, Mass.
\(^2\) In terms of International Reference Preparation of Human Menopausal Gonadotropin No. 2.
against plotting the measured and hypophysectomy. This is to show undetectable.

The radioactivity.

HLH was determined biologically active.

0.

fallof zero was the result of incomplete hypophysectomy. This was confirmed by the finding of biologically active gonadotropin in the urine after surgery.

Continuous HLH-

infusion studies. Three groups of subjects were used for these studies: (a) five normal premenopausal women studied at random times during the menstrual cycle; (b) five normal premenopausal women taking norethindrone and mestranol (5 mg daily on day 1 through 20 of cycle) for contraceptive purposes; and (c) four postmenopausal women. The latter group included two normal women and two women with carcinoma of the breast with no evidence of extensive hepatic or other metatases at the time of study. The subjects were again prepared with five drops of saturated potassium iodide every 6 hr for 24 hr before study. The infusion tests were initiated between 6:00 and 10:00 a.m. with the subjects fasting and at bed rest. A 10 ml heparinized sample of blood was drawn immediately before the test for endogenous HLH determination. A total dose of 5-25 µc of HLH-\textsuperscript{131}I containing 0.8-3.2 U of H.lH was given to each subject. One-fifth of the total dose was given directly intravenously as a priming dose 20 min before starting the infusion. The remainder of the HLH-

\textsuperscript{131}I was infused in a 1% albumin solution in saline at a constant rate of 1.25-1.30 ml/min over a 3-6 hr period.

The HLH-\textsuperscript{131}I was measured in the infusion solution by antibody precipitation. Only the immunoreactive radioactivity (80-95%) was used in calculating the rate of infusion. The concentration of antibody precipitable

where \( R \) is the total HLH-\textsuperscript{131}I injected as a single dose, and \( x' \) is the plasma concentration of antibody precipitable radioactivity. The integral \( \int_{t_0}^{t} x' \cdot dt \) was determined by plotting the measured plasma HLH-\textsuperscript{131}I concentration against time after administration and by numerically measuring the area under the disappearance curve (8). The production rate (PR) of HLH was then calculated as

\[
PR = MCR \cdot \dot{x}
\]

where \( \dot{x} \) is the concentration of endogenous unlabeled HLH determined by radioimmunoassay (3).

The disappearance of endogenous unlabeled HLH from plasma was also determined on one patient at the time of hypophysectomy for palliation of carcinoma of the breast. This patient had previously had a bilateral ovariectomy and the plasma HLH level was elevated. Plasma HLH was measured in serial blood samples taken during the surgery. Because plasma HLH levels did not fall to undetectable levels, the residual HLH concentration after hypophysectomy was subtracted from each sample to show the disappearance curve. The failure of the plasma HLH to fall to zero was the result of incomplete hypophysectomy. This was confirmed by the finding of biologically active gonadotropin in the urine after surgery.
HLH-\(^{131}\)I in the infusion solution did not change throughout the test.

10-ml heparinized blood samples were drawn every 20-30 min for the last 2 hr of the infusion (Fig. 2). Four or more samples were used to calculate the mean plasma HLH-\(^{131}\)I at equilibrium. As a control for non-specific degradation of HLH-\(^{131}\)I by plasma, the HLH-\(^{131}\)I was placed in plasma and incubated at 37°C. The loss of immunoreactivity was less than 2% from 0 to 6 hr and was only 5% at 24 hr.

The plasma MCR defined as the volume of blood cleared completely and irreversibly of HLH-\(^{131}\)I in unit time was calculated after the method of Tait (3, 4) according to the following formula:

\[
MCR = \frac{r}{x'c}
\]

where \(r\) is the rate of infusion of HLH-\(^{131}\)I in cpm per minute and \(x'c\) is the plasma HLH-\(^{131}\)I level after equilibrium has been reached in cpm per ml. The production rate (PR) of LH or secretion into the plasma is again calculated as

\[
PR = MCR \cdot i.
\]

RESULTS

Plasma HLH disappearance studies. The disappearance curve of HLH-\(^{131}\)I from plasma after a single injection showed a multiexponential type of fall with an initial half-time between 30 and 60 min (Figs. 3 and 4). The divergent total plasma \(^{131}\)I curve was interpreted as showing the return of degradation products of HLH-\(^{131}\)I back into the plasma. The antibody precipitable radioactivity fell from a mean of 0.027% of the injected dose per ml of plasma at 20 min to 0.00058% at 24 hr. The per cent of the total plasma radioactivity which was antibody precipitable fell from a mean of 0.027% at 20 min to 31% at 24 hr. Urinary excretion of the total injected \(^{131}\)I ranged from 62 to 99.5% over the first 24 hr with fairly constant excretion of 10-15% per hour for the first 5 hr. Antibody precipitable radioactivity in the urine was always less than 0.1% of the total.

The type of disappearance curve of HLH-\(^{131}\)I from plasma suggested that the HLH-\(^{131}\)I was dis-
tributed in more than three mathematical compartments. The semilogarithmic plot of the curve showed no definitely linear segment during the first 24 hr although most of the labeled hormone had been degraded during this time as shown by the total plasma \(^{131}\text{I}\) and urinary \(^{131}\text{I}\) excretion. The MCR's and PR's of HLH calculated from the single injection studies are shown in Table I. Although the MCR and PR could be calculated by the single injection technique, the disappearance curve of HLH\(^{131}\text{I}\) was complex, and the area under the curve had to be mechanically or numerically integrated. Therefore, the constant infusion technique was used for subsequent studies.

The disappearance curve of endogenous HLH from the plasma of the patient during hypophysectomy was not entirely satisfactory because blood was given to the patient during the procedure. However, before blood administration, the HLH

<table>
<thead>
<tr>
<th>Age</th>
<th>Sex</th>
<th>Diagnosis</th>
<th>MCR (ml/min)</th>
<th>Plasma (ml)</th>
<th>PR (mU/min)</th>
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</thead>
<tbody>
<tr>
<td>45</td>
<td>F</td>
<td>Chromophobe adenoma</td>
<td>19.6</td>
<td>&lt;4.5</td>
<td>-</td>
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<tr>
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<td>F</td>
<td>Chromophobe adenoma</td>
<td>19.8</td>
<td>42.4</td>
<td>839</td>
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<tr>
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<td>F</td>
<td>Carcinoma of breast*</td>
<td>19.6</td>
<td>28.0</td>
<td>549</td>
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<tr>
<td>44</td>
<td>M</td>
<td>Chromophobe adenoma</td>
<td>20.3</td>
<td>7.4</td>
<td>150</td>
</tr>
</tbody>
</table>

* Patient previously ovariectomized.
fell with a half-time of 30–60 min. Fig. 5 shows a comparison of the disappearance curves of HLH-\(^{131}\)I after a single injection and of endogenous HLH at hypophysectomy.

**Continuous HLH-\(^{131}\)I infusion studies.** Plasma HLH-\(^{131}\)I levels appeared constant by hr 3 or 4 of the infusion in 13 of 14 subjects. The mean and SD of the last four or more values used in calculating the MCR are shown in Table II. In only one subject (subject 3) was there greater than 5% variation of any sample from the mean during the last hour of infusion. To further examine for evidence that equilibrium had been reached, the plasma HLH-\(^{131}\)I levels were expressed as a percent of the final concentration and inspected for the presence of an upward or downward trend. Only subject 3 continued to show a slight upward trend of plasma HLH-\(^{131}\)I levels over the last hour of infusion. However, her values appeared sufficiently near equilibrium to include with the others (see Table II). The MCR's were: 24.4 ± 1.8 (mean ± SD) ml/min in the five normal premenopausal women; 23.3 ± 1.1 ml/min in five

**Table II**

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age (yr)</th>
<th>Weight (kg)</th>
<th>Height (cm)</th>
<th>Body surface area (m(^2))</th>
<th>Day of menstrual cycle</th>
<th>Precipitable cpm infused</th>
<th>Blood level at equilibrium (cpm)</th>
<th>MCR (ml/min)</th>
<th>Plasma LH (mU/ml)</th>
<th>PR (mU/min)</th>
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<td>1</td>
<td>29</td>
<td>64</td>
<td>155</td>
<td>1.61</td>
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<td>20,027</td>
<td>723 ± 5</td>
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<td>2</td>
<td>24</td>
<td>58</td>
<td>173</td>
<td>1.68</td>
<td>19</td>
<td>15,306</td>
<td>603 ± 7</td>
<td>25.4</td>
<td>28.0</td>
<td>711</td>
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<tr>
<td>3</td>
<td>26</td>
<td>58</td>
<td>172</td>
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<td>889 ± 52</td>
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<td>16.8</td>
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<td>4</td>
<td>20</td>
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<td>154</td>
<td>1.41</td>
<td>20</td>
<td>14,074</td>
<td>763 ± 22</td>
<td>18.4</td>
<td>68.8</td>
<td>1266</td>
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<tr>
<td>5</td>
<td>25</td>
<td>55</td>
<td>157</td>
<td>1.53</td>
<td>24</td>
<td>15,306</td>
<td>550 ± 10</td>
<td>27.8</td>
<td>33.6</td>
<td>935</td>
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</table>

Mean ± se

| 24.4 ± 1.8 | 32.0 ± 9.6 | 734 ± 170 |

Premenopausal women on norethindrol and mestranol

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<tr>
<th>Patient</th>
<th>Age (yr)</th>
<th>Weight (kg)</th>
<th>Height (cm)</th>
<th>Body surface area (m(^2))</th>
<th>Day of menstrual cycle</th>
<th>Precipitable cpm infused</th>
<th>Blood level at equilibrium (cpm)</th>
<th>MCR (ml/min)</th>
<th>Plasma LH (mU/ml)</th>
<th>PR (mU/min)</th>
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<tbody>
<tr>
<td>6</td>
<td>22</td>
<td>59</td>
<td>169</td>
<td>1.67</td>
<td>9</td>
<td>30,600</td>
<td>1362 ± 12</td>
<td>22.5</td>
<td>9.6</td>
<td>216</td>
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<tr>
<td>7</td>
<td>23</td>
<td>58</td>
<td>161</td>
<td>1.59</td>
<td>11</td>
<td>11,539</td>
<td>607 ± 4</td>
<td>19.0</td>
<td>22.4</td>
<td>426</td>
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<tr>
<td>8</td>
<td>22</td>
<td>61</td>
<td>168</td>
<td>1.69</td>
<td>22</td>
<td>9,740</td>
<td>410 ± 14</td>
<td>23.8</td>
<td>12.8</td>
<td>305</td>
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<tr>
<td>9</td>
<td>25</td>
<td>63</td>
<td>171</td>
<td>1.73</td>
<td>10</td>
<td>15,153</td>
<td>552 ± 8</td>
<td>27.5</td>
<td>25.6</td>
<td>704</td>
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<tr>
<td>10</td>
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<td>5,953</td>
<td>250 ± 17</td>
<td>23.8</td>
<td>12.0</td>
<td>286</td>
</tr>
</tbody>
</table>

Mean ± se

| 23.3 ± 1.1 | 16.5 ± 3.2 | 387 ± 86 |

Postmenopausal women

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age (yr)</th>
<th>Weight (kg)</th>
<th>Height (cm)</th>
<th>Body surface area (m(^2))</th>
<th>Day of menstrual cycle</th>
<th>Precipitable cpm infused</th>
<th>Blood level at equilibrium (cpm)</th>
<th>MCR (ml/min)</th>
<th>Plasma LH (mU/ml)</th>
<th>PR (mU/min)</th>
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<tbody>
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<td>49</td>
<td>153</td>
<td>1.43</td>
<td>—</td>
<td>18,988</td>
<td>882 ± 33</td>
<td>21.5</td>
<td>98.4</td>
<td>2120</td>
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<tr>
<td>12</td>
<td>39</td>
<td>65</td>
<td>156</td>
<td>1.63</td>
<td>—</td>
<td>20,924</td>
<td>1022 ± 29</td>
<td>20.5</td>
<td>164.0</td>
<td>3360</td>
</tr>
<tr>
<td>13</td>
<td>49</td>
<td>83</td>
<td>154</td>
<td>1.81</td>
<td>—</td>
<td>13,637</td>
<td>601 ± 15</td>
<td>22.7</td>
<td>62.4</td>
<td>1420</td>
</tr>
<tr>
<td>14</td>
<td>39</td>
<td>64</td>
<td>175</td>
<td>1.77</td>
<td>—</td>
<td>10,596</td>
<td>281 ± 10</td>
<td>37.7</td>
<td>72.0</td>
<td>2710</td>
</tr>
</tbody>
</table>

Mean ± se

| 25.6 ± 4.1 | 99.2 ± 23.2 | 2400 ± 410 |

* In terms of the Second International Reference Preparation of Human Menopausal Gonadotropin.

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normal women taking oral contraceptives; and 25.6 ± 4.1 ml/min in the four postmenopausal women. There were no significant differences among groups (P > 0.05). The higher endogenous plasma LH levels in the premenopausal women on no medications were due in part to the various times in the menstrual cycle when the studies were performed. Subject 4 was apparently having an ovulatory LH surge at the time of study. The PR’s were: 734 ± 170 mU/min in the normal premenopausal women; 387 ± 86 mU/min in the five women on norethindronel and mestranol; and 2400 ± 420 mU/min in the four postmenopausal women. The patient who underwent ovariectomy was 49 yr old but was having regular menses before surgery. The preoperative MCR was 24.3 ml/min with a plasma LH of 24.0 mU/ml and a PR of 583 mU/min. The postoperative MCR was 22.7 ml/min with a plasma LH of 62.4 mU/ml and a PR of 1420 mU/min.

DISCUSSION

There are no previously published estimates of the secretion rate of LH. In general, methodology has not been available to permit such studies. Highly purified LH labeled with radioiodine can be used in tracer amounts for metabolic studies in doses that are not supraphysiologic. However, before an iodinated protein can be used as metabolic tracer in vivo, it must be shown to be indistinguishable from the unlabeled protein in that particular system under study.

The assumption that LH-131I was cleared from the plasma at the same rate as endogenous LH is critical to the validity of the present study. This assumption seems justified on the basis of studies on the disappearance curve of LH in the serum of monkeys.5 Because highly purified LH was not available in sufficient quantity for extensive clearance studies, a preparation of human menopausal gonadotropin was injected simultaneously with the purified LH-131I into five Macaca mulatta monkeys. The disappearance of antibody precipitable LH-131I was compared with the disappearance of LH determined by radioimmunoassay. The disappearance curve in plasma was indistinguishable for the LH-131I and unlabeled LH. The additional finding of a similar disappearance curve of LH-131I and endogenous LH at the time of hypophysectomy in one patient (Fig. 5) suggests that the lightly iodinated LH is cleared from the plasma at the same rate as unlabeled LH. Unfortunately, a method other than hypophysectomy for acutely suppressing plasma LH levels is not yet available to facilitate endogenous plasma LH half-time determinations.

Another assumption necessary to validate production rate studies is that a steady state is maintained in regard to secretion and degradation of the endogenous hormone during the test period. The degradation of LH-131I must reach a steady rate during the constant infusion studies. Studies of diurnal LH variation have shown no significant change over the course of 24 hr in women except during the LH surge at the time of ovulation.4 Therefore, it appears that steady rates of LH production and degradation were probably maintained throughout the period of these studies. The possible exception is subject 4 who was apparently having an ovulatory LH elevation.

The finding that plasma LH-131I in humans did not have a linear disappearance rate on a semilogarithmic plot after a single injection is of interest. Disappearance curves in the literature for 131I-labeled insulin (9), glucagon (10), growth hormone (11), and thyrotropin (12) after a single injection appear to show an initial linear segment after a mixing period followed by a slower decline. These disappearance curves are in good agreement with the disappearance of appropriate unlabeled hormones in these studies. If a single compartment of distribution is assumed, fractional turnover rates can easily be calculated from these data. In the case of LH, however, there appears to be a definite multieponential disappearance curve. Although the disappearance curves of LH-131I and unlabeled LH again appear to be in good agreement in the one patient at hypophysectomy and in primates, the initial disappearance is not linear and the single compartment assumption cannot be made. A meaningful MCR in a multi-compartmental system should be a function of the total integrated area under the disappearance curve.


curve. Highly complex curves are mathematically difficult to resolve and it becomes necessary to integrate mechanically or numerically the area under the curve. The continuous infusion method (3, 4) provides a simple means of integrating the area under the disappearance curve and is a valid technique for determining the clearance rate in both single and multicompartmental systems.

Another possible interpretation of the disappearance curves after the single injections is that some damaged HLH-\(^{131}\)I was present. Iodination of smaller peptides such as alpha MSH (13) and arginine vasopressin (14) can result in a slowing of the disappearance curve. However, proteins smaller than HLH such as insulin (9), glucagon (10), and growth hormone (11, 15), or equivalent in size and composition, such as thyrotropin (12), can be lightly iodinated without significantly changing the disappearance curve. Furthermore, the fraction used for the present studies showed less than 1% damaged HLH-\(^{131}\)I by chromatoelectrophoresis and only immunoreactive HLH-\(^{131}\)I was used in determining the infusion rates. It is very possible that a small fraction of damaged HLH-\(^{131}\)I might result in the slow disappearance indicated by the late portion of the plasma HLH-\(^{131}\)I curve after the single injection, at a time when most of the labeled hormone had already been degraded. However, the error in clearance determinations of such a small fraction would be almost negligible over the time period of the constant infusion method. We also have found that human menopausal gonadotropin with almost no FSH activity can be iodinated with \(^{125}\)I using Chloramine T, total iodide, and sodium metabisulfite in ratios proportional to those used for the present study with minimal or no loss of biologic LH potency as determined by the rat ventral prostate assay.\(^{5}\)

Tait (3, 4) has described the rationale for using the constant infusion-to-equilibrium method for determining the metabolic clearance rates of steroids. This method has not previously been applied to polypeptide hormones, but is equally applicable when it can be shown that the labeled hormone behaves in an identical manner to the unlabeled hormone in the system under study. The constant infusion approach can be regarded as imitating the


HLH secretion of the pituitary by using HLH-\(^{131}\)I. If the labeled hormone is cleared from the plasma at the same rate as the endogenous hormone, the ratio of the rate of HLH-\(^{131}\)I infusion to the final plasma HLH-\(^{131}\)I level at equilibrium will be equivalent to the ratio of the secretion of endogenous HLH to the endogenous plasma HLH level. The similar MCR values found by the single injection and constant infusion techniques suggest that either may be used for MCR and PR determinations. However, the constant infusion technique provides some advantages over the single injection method. The area under the infusion curve is automatically integrated, and factors which influence the MCR can be followed in the same subject (3). The shorter time period for infusion of HLH-\(^{131}\)I reduces nonspecific damage to the iodinated hormone which could result in spuriously low MCR and PR values.

The present data from the constant infusion studies clearly show that there is little difference in the MCR of HLH among three groups of subjects with rather markedly differing plasma HLH levels. The studies before and after ovariectomy in one patient also showed no change in MCR while the plasma HLH more than doubled. These findings indicate that the fluctuations noted in plasma HLH levels are the result of changes in the rate of HLH secretion rather than the rate of HLH clearance. Although subject 4 who had the lowest body surface area also had the lowest MCR, generally the MCR did not correlate (\(P > 0.05\)) with height, weight, or body surface area.

Ryan (16) has found that the HLH content in the pituitaries of five healthy young premenopausal women dying suddenly ranges from 0.162 to 2.209 mg of NIH-LH-S1 per pituitary (mean = 1.445) by the ovarian ascorbic acid depletion assay (17). The HLH content in the pituitaries of four postmenopausal women was 1.191–5.462 mg of NIH-LH-S1 per pituitary (mean = 3.453). Rosenberg and Lewis (18) have found 1 mg of NIH-LH-S1 to be equivalent to 500 U of the 2nd International Reference Preparation of Human Menopausal Gonadotropin by the ovarian ascorbic acid depletion assay. The total pituitary HLH content, therefore, averaged about 725 U in the premenopausal and 1725 U in the postmenopausal women. If the HLH PR's were to be maintained at a relatively
stable level for 24 hr as suggested from the lack of significant diurnal variation except during the ovulatory surge, the premenopausal women would have a secretion of about 500–1100 U/day, and the postmenopausal women would have a secretion of about 3500 U/day. These values would represent a one- to twofold turnover of total pituitary HLH content daily in both groups. This estimated daily pituitary turnover of HLH is similar to the value reported for human thyrotropin (12) and suggests that the secretion of both hormones is an extremely active process with a rather rapid turnover rate in the pituitary.

The amount of HLH-\(^{131}\)I that could be precipitated from the urine in the present study was less than 0.1%. This value would suggest a low renal clearance of undegraded HLH. However, although we have shown that the clearance of HLH-\(^{131}\)I from plasma is similar to unlabeled HLH, we have no evidence that the renal clearance of HLH-\(^{131}\)I is the same as unlabeled HLH. Keller (19) has found that the renal clearance of HLH by bioassay is between 0.03 and 0.26 ml/min in postmenopausal women. When these values are compared to our total MCR's of 18.4–37.7 ml/min from the present study, it appears that less than 5% of the total HLH production actually appears in the urine in the biologically active form. Becker and Albert (20) have reported that the normal values for urinary excretion of HLH by the rat ventral prostate assay (21) are from 0.21 to 0.43 mg equivalents of NIH-LH-S1 per 24 hr in premenopausal women and 2.1 mg equivalents of NIH-LH-S1 per 24 hr in postmenopausal women. After conversion to International Units, according to Rosenberg and Lewis (18) (× 62.5 for rat ventral prostate assay), the values for premenopausal women are 13.1–25 U/24 hr in premenopausal and 131.3 U/24 hr in postmenopausal women. Again, it would appear that less than 5% of the total daily HLH production can be measured in the urine by bioassay. There is no reason to believe that the renal clearance of biologically active HLH is important to hormone activity. It is of interest that urinary gonadotropin values which have been clinically invaluable in the past have probably been derived from a very small fraction of the total HLH production.

ACKNOWLEDGMENTS

The authors wish to thank Doctors C. W. Bardin, M. B. Lipsett, and D. Rodbard for their advice and encouragement.

REFERENCES


