Effects of Cortisone and Hydrocortisone on the Numbers of Thoracic Duct Lymphocytes

By Gerald F. Hungerford, Ph.D., William O. Reinhardt, M.D. and Choh Hao Li, Ph.D.

It is well established that adrenocorticotropic hormone (ACTH) and certain adrenal cortical steroids may decrease the number of circulating blood lymphocytes.\textsuperscript{1-3} It has also been reported\textsuperscript{4} that the blood lymphocytopenia after ACTH treatment may, in part, be the result of decreased delivery of these cells to the blood stream through the thoracic duct. Attempts to demonstrate that an adrenal steroid material or an adrenal cortical extract is capable of lowering the number of lymphocytes in thoracic duct lymph\textsuperscript{5, 6} have not been successful. In a discussion of the reasons for this discrepancy, it was stated,\textsuperscript{4} as a partial explanation, that adequate dose levels, time intervals and appropriate compounds may not have been studied. The recent availability of hydrocortisone has made it possible to test this steroid material for a possible effect on thoracic duct lymphocytes. The following report compares the effect of the synthetic steroids, cortisone and hydrocortisone, on the lymphocyte content of thoracic duct lymph in normal and in hypophysectomized rats. The data presented indicate that hydrocortisone is markedly more effective than is cortisone in producing a thoracic duct lymphocytopenia. The data on control animals are presented solely for comparison with the data for treated animals. The effects of hypophysectomy and of control injections on the numbers of thoracic duct lymphocytes in the rat have been presented and discussed previously.\textsuperscript{4}

Experimental Methods

Normal male rats of the Long-Evans strain approximately 60 days of age were employed. They were maintained on a standard regimen and housed under similar conditions. Hypophysectomized rats of the same age group were employed one to two days postoperatively. Thoracic duct lymph was collected from the jugular lymph sac in the neck by a technic previously described,\textsuperscript{4} using sodium pentobarbital anesthesia. A drop of heparin was added to each sample of lymph to prevent clotting. A 90 minute sample of lymph was collected from each rat. The volume of lymph was measured in a tuberculin syringe, and a white blood cell count was made on each sample. The total number of lymphocytes was calculated for each sample and then converted to 24 hour values. These values were also expressed relative to 100 Gm. body weight per 24 hours. The cortisone (11-dehydro-17-hydroxy-corticosterone-21-acetate) and hydrocortisone (17-hydroxy-corticosterone-21-acetate) employed were synthetic products in saline suspension with 0.9 per cent benzyl alcohol as preservative.

From the Department of Anatomy of the School of Medicine and the Department of Biochemistry, University of California, Berkeley, Calif.

Assisted in part by grants from the National Institutes of Health, United States Public Health Service, and Merck and Company, Rahway, N. J. Acknowledgement is made to the Institute of Experimental Biology for assistance rendered, and to Dr. J. M. Carlisle and Dr. E. Alpert of Merck and Company, for supplies of cortisone and hydrocortisone. Gratitude is expressed to Mrs. Lorre Keffer for technical assistance.

Submitted July 1, 1952; accepted for publication September 2, 1952.
THORACIC DUCT LYMPHOCYTES

RESULTS

Effects of Cortisone and Hydrocortisone in Normal Rats

Four hours after administering 10 mg. of hydrocortisone to normal rats, the total number of thoracic duct lymphocytes was significantly decreased (P < .01) when compared with normal or saline controls, and when calculated in terms of lymphocyte count, total cells or total cells per 100 Gm. body weight/24 hours.

Four hours after administering cortisone to normal rats in the same manner,

TABLE 1.—Effect of Cortisone and Hydrocortisone on the Rate of Flow and Cell Content of Thoracic Duct Lymph in Normal and Hypophysectomized Rats

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No. of rats</th>
<th>Body wt.</th>
<th>WBC</th>
<th>per 24 hr.</th>
<th>per 100 Gm./24 hr.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Lymph flow</td>
<td>Total cells</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Lymph flow</td>
<td>Total cells</td>
</tr>
<tr>
<td>Normal Rats</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>None</td>
<td>28</td>
<td>252</td>
<td>31,500</td>
<td>21.0</td>
<td>642 (0.8) (38)</td>
</tr>
<tr>
<td>0.9% NaCl 0.5 ml.</td>
<td>10</td>
<td>240</td>
<td>22,600</td>
<td>23.8</td>
<td>521 (2.3) (47)</td>
</tr>
<tr>
<td>Cortisone 10.0 mg.</td>
<td>7</td>
<td>207</td>
<td>25,300</td>
<td>19.7</td>
<td>451 (1.6) (32)</td>
</tr>
<tr>
<td>Hydrocortisone 10.0 mg.</td>
<td>9</td>
<td>203</td>
<td>13,450</td>
<td>19.2</td>
<td>248 (1.3) (20)</td>
</tr>
<tr>
<td>Hypophysectomized Rats</td>
<td>10</td>
<td>230</td>
<td>52,600</td>
<td>17.6</td>
<td>917 (0.9) (60)</td>
</tr>
<tr>
<td>None</td>
<td>8</td>
<td>176</td>
<td>36,300</td>
<td>13.8</td>
<td>506 (0.8) (56)</td>
</tr>
<tr>
<td>Cortisone 10.0 mg.</td>
<td>8</td>
<td>177</td>
<td>25,400</td>
<td>8.0</td>
<td>215 (0.6) (35)</td>
</tr>
<tr>
<td>Hydrocortisone 10.0 mg.</td>
<td>8</td>
<td>177</td>
<td>25,400</td>
<td>8.0</td>
<td>215 (0.6) (35)</td>
</tr>
</tbody>
</table>

Mean values presented with standard errors of the means in parentheses. Body weight in Gm.; WBC per cu. mm.; lymph flow in ml.; total cells in millions. Lymph collection started 4 hours after intraperitoneal injection of cortisone or hydrocortisone, and 2 hours after saline injection.

the total cells per 100 Gm. body weight/24 hours were not markedly different from the ratio in the normal or saline control rats, although a decrease was noted in the absolute lymphocyte count. This decrease was not nearly as marked, however, as that produced by hydrocortisone.

Effects of Cortisone and Hydrocortisone in Hypophysectomized Rats

Whereas the depression of lymphocytes in thoracic duct lymph of hypophysectomized rats produced by cortisone is probably significant, that produced by
hydrocortisone is approximately 50 per cent greater and of high significance (P < .01). It is of interest that in addition to decreasing the absolute cell count, hydrocortisone produces a highly significant decrease in lymph flow in hypophysectomized rats as compared to the effect of cortisone. A similar effect on lymph flow has been noted with ACTH preparations in hypophysectomized rats.4

DISCUSSION

It was recently reported4 that single injections of Adrenal Cortex Extract (Upjohn) 1.0 ml., cortisone 2.5 mg. and desoxycorticosterone glucoside 5.0 mg., were ineffective in altering the level of thoracic duct lymphocytes in normal rats. A similar experience with the effect of particular doses of adrenal cortical extract on thoracic duct lymphocytes of cats has been recorded by Valentine, Craddock and Lawrence.2

The present experiment compares the effect of cortisone and hydrocortisone at the 10 mg. dose level. The data presented are interpreted to indicate that at these dose levels, hydrocortisone is much more effective than is cortisone in decreasing thoracic duct lymphocytes in either normal or hypophysectomized rats.

It is of interest that a preliminary study4 of the relative activity of cortisone and hydrocortisone, using as a test the 4 hour blood eosinophil depression in the Long-Evans rat, has shown that hydrocortisone is demonstrably more eosinopenic than is cortisone, in either normal or hypophysectomized rats, when tested at comparable dose levels and time intervals.

These results lead to the hypothesis that the thoracic duct lymphocytopenic effect produced by ACTH stimulation of the adrenal cortex is more likely dependent on the release of hydrocortisone (or a substance with similar properties) than on the release of cortisone.

SUMMARY

The effects of cortisone and hydrocortisone on the levels of thoracic duct lymphocytes have been studied in normal and hypophysectomized rats under standardized conditions. It is shown that:

1. Four hours after its administration, hydrocortisone causes a marked thoracic duct lymphocytopenia, the effect being even more marked in hypophysectomized than in normal rats.

2. At comparable dose levels, hydrocortisone is demonstrably and significantly more effective in producing a thoracic duct lymphocytopenia than is cortisone.

REFERENCES


Effects of Cortisone and Hydrocortisone on the Numbers of Thoracic Duct Lymphocytes

GERALD F. HUNGERFORD, WILLIAM O. REINHARDT and CHOH HAO LI