lower population of susceptible precursors in the retina would account for the present low incidence of tumor production. Detailed studies of nine intraocular retinoblastoma-like neoplasms obtained to date will be published soon.

The advice of Dr. C. L. Schepens in the preparation of this report is acknowledged. David F. Dobies, provided editorial assistance.

From the Wesley C. Bowers Laboratory of Pharmacology and Experimental Pathology, Department of Retina Research, Retina Foundation, Boston, Mass. 02114. This work was supported by the Massachusetts Lions Eye Research Fund, Inc., by Public Health Service Research Grants EY-00227 and CA-12180, and by grants from Research to Prevent Blindness, Inc. S. Kobayashi was the recipient of a Grant-in-Aid from Fight for Sight, Inc., New York. Manuscript submitted for publication July 19, 1973; manuscript accepted for publication Aug. 28, 1973.

Key words: retinoblastoma-like tumor, human adenovirus Type 12, C-D-strain rats, adenovirus carcinogenesis.

REFERENCES


An in vitro evaluation of tetrahydrotriamcinolone. JOHN F. BIGGER, HARRY A. ZINK, PAUL F. PALMBERG, AND BERNARD BECKER.

A major side effect of corticosteroid therapy for ocular inflammation is the increased intraocular pressure which occurs in susceptible individuals. The therapeutic efficacy of corticosteroids, and the intraocular pressure effect, have been shown to vary with the relative potency of the drug used, its concentration, frequency and duration of usage, and its ocular penetration.

Attempts to dissociate the anti-inflammatory and ocular pressure effects of corticosteroids have led to the evaluation of several new compounds in recent years. One such compound is tetrahydrotriamcinolone (THTA), a derivative of triamcinolone. THTA has been shown to only rarely elevate intraocular pressure, however, its potency as an anti-inflammatory drug is controversial.1, 2

Recently, Bigger, Palmberg, and Becker3 reported that differences in individual sensitivity to corticosteroids could be detected using a peripheral blood lymphocyte transformation inhibition assay and that the systemic sensitivity correlated closely to the results of ocular testing. The purpose of the present study was to ascertain whether differential sensitivity of lymphocytes to THTA occurs, and to determine the potency of THTA relative to prednisolone-21-phosphate.

The method of assay used in the present study has been reported previously3 and will be only briefly described. Suspensions of lymphocytes are prepared from freshly drawn heparinized venous blood and incubated for 48 hours in the presence of phytohemagglutinin-P (PHAP) after preincubation with various concentrations of the corticosteroid compound to be tested. Following this incubation tritiated thymidine is added and the lymphocyte suspensions are allowed to incubate for an additional sixteen hours at which time they are harvested for scintillation counting.

In the present study, for each patient we prepared two unstimulated blanks, six PHAP-stimulated controls, quadruplicates of each of four concentrations of prednisolone-21-phosphate with PHAP, and quadruplicates of each of five concentrations of tetrahydrotriamcinolone (THTA) with PHAP. In each case the prednisolone-21-phosphate or THTA was added to the cell suspension and allowed to incubate at 37° C. for one hour prior to the addition of PHAP.

For each patient two dose-response curves for the inhibition of tritiated thymidine incorporation into PHAP-stimulated lymphocytes were obtained from the same sample of blood; one for inhibition by prednisolone-21-phosphate, and the second for inhibition by THTA. Counts per minute incorporation corrected for the nonstimulated blank was plotted vs. log concentration of the drug. Regres-
Fig. 1. Distribution of Iso values by categories of topical dexamethasone nonresponder (NN), high-responder (GG), and open-angle glaucoma (OAG) for testing with prednisolone-21-phosphate and tetrahydrotriamcinolone (THTA).

Fig. 2. Distribution of Iso values for prednisolone-21-phosphate inhibition plotted against the Iso values for tetrahydrotriamcinolone (THTA) for the 15 individuals.

Table I. Drug concentration to inhibit lymphocyte transformation by 50 per cent (I50)

<table>
<thead>
<tr>
<th>No. of patients</th>
<th>Mean</th>
<th>S.E.M.</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prednisolone-21-phosphate:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NN</td>
<td>6</td>
<td>99.3</td>
<td>9.7</td>
</tr>
<tr>
<td>GC</td>
<td>2</td>
<td>44.5</td>
<td>2.5</td>
</tr>
<tr>
<td>OAG</td>
<td>7</td>
<td>38.8</td>
<td>4.3</td>
</tr>
<tr>
<td>THTA:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NN</td>
<td>6</td>
<td>73.5</td>
<td>8.2</td>
</tr>
<tr>
<td>GC</td>
<td>2</td>
<td>32.5</td>
<td>6.5</td>
</tr>
<tr>
<td>OAG</td>
<td>7</td>
<td>23.1</td>
<td>3.3</td>
</tr>
</tbody>
</table>

*Applanation ocular pressure < 20 mm. Hg after six weeks of topical application of 0.1 per cent dexamethasone drops four times a day.
†Applanation ocular pressure > 31 mm. Hg during six weeks of topical testing.

A plot of the Iso values for inhibition by prednisolone-21-phosphate and THTA, with respect to each individual's ocular testing classification is shown in Fig. 1. The distribution of the Iso values for prednisolone-21-phosphate was similar to that reported previously with differential separation of the OAG and GG groups from the NN individuals. It should be noted that a similar differential responsiveness occurs to THTA, with the OAG and GG patients demonstrating an increased sensitivity to this drug as compared to the NN patients.

The mean Iso and standard error of the mean (S.E.M.) for each of the three groups for both prednisolone-21-phosphate and THTA, with statistical comparisons is shown in Table I. The mean Iso for the OAG group is lower than for the NN group for both compounds tested, a difference which is statistically highly significant (p < 0.001). Because of the small number of GG patients they were not included in a statistical comparison. The mean Iso values obtained for prednisolone testing are identical to those recently obtained for a much larger series of patients except that the mean Iso for the NN group is 20 per cent higher in the present small group (Bigger, unpublished data).

A plot of the Iso for THTA inhibition vs. the Iso for prednisolone-21-phosphate inhibition for each patient is shown in Fig. 2. Linear regression analysis revealed a correlation coefficient (r) of 0.925. This demonstrates a highly significant statistical correlation between the Iso values for the two compounds (p < 0.001). Based on the ratios of Iso values for both compounds tested, it was possible to calculate a relative potency.
ratio for THTA of 1.4 times that of prednisolone-21-phosphate.

THTA has been shown in previous studies to elevate intraocular pressure only rarely, although it penetrates into the aqueous humor quite readily. Clinically, it has been shown to be effective in suppressing the allergic reaction to topical glaucoma medications. However, in a rabbit xenograft model it was relatively ineffective as a suppressor of graft rejection. In a study of topical application to skin, THTA was shown to be one-sixth to one-eighteenth as effective as triamcinolone.

In the present study we have determined a relative potency ratio for THTA as compared to other corticosteroid compounds. Using dose-response studies in human lymphocytes we find THTA to be 1.4 times as potent as prednisolone; or 5 times as potent as hydrocortisone, the usual reference drug. (This figure is based on an average potency ratio of 3.5 for prednisolone compared to hydrocortisone.) To further test this in vitro assay for comparing relative potency we have also derived by similar techniques, potency ratios of 4 for medrysone and 52 for fluorometholone (Zink, unpublished data). These ratios compare well with those for anti-inflammatory potency summarized by Nelson.

In addition to establishing a relative potency value for THTA, we have also demonstrated that lymphocyte transformation is inhibited by THTA with the same differential sensitivity previously reported for prednisolone. The lymphocytes from individuals with open-angle glaucoma and corticosteroid ocular hypertension are more sensitive to THTA than are lymphocytes from ocular non-responders. There is, therefore, no evidence from the present study to suggest that THTA acts differently from a weak glucocorticoid such as prednisolone, and the low incidence of THTA-induced ocular hypertension may reflect only the low potency of this compound.

The tetrahydrotriamcinolone used in this study was provided by The Squibb Institute for Medical Research, Princeton, N. J. We thank Mrs. Mamie Penny for her technical assistance.

From the Department of Ophthalmology, Washington University School of Medicine, 660 S. Euclid, St. Louis, Mo. 63110. Supported by Grants EY01167, EY00336, and EY00604 from the National Eye Institute, Bethesda, Md. Manuscript submitted for publication June 18, 1973; manuscript accepted for publication July 6, 1973. Reprint requests: Dr. Bigger, Marietta Eye Clinic, 633 Cherokee St., Marietta, Ga. 30060.

Key words: open-angle glaucoma, corticosteroids, tetrahydrotriamcinolone, lymphocyte transformation, phytohemagglutinin, intraocular pressure.

REFERENCES


The resistance of teleost ocular tissues to oxygen toxicity. J. Russell Hoffert, D. A. Baeyens, and P. O. Fromm.

In contrast to mammalian retinal tissue, the freshwater teleost retina is unique in its resistance to the toxic actions of hyperbaric oxygen. Various sulphydryl group-containing compounds and many biologic reducing agents have been repeatedly shown to have a moderating effect on oxygen toxicity. Two such compounds, known to be readily produced by ocular tissues, are reduced glutathione and ascorbic acid. The teleost retina presents an unusual situation in that it is normally enveloped by superatmospheric oxygen tensions. These high oxygen tensions are generated by a countercurrent oxygen multiplier located in the choroid which contains a rete mirabile.

The present studies were undertaken to determine if the marine teleost retina is similar to that of freshwater trout in its resistance to oxygen toxicity and if other teleost ocular tissues are likewise resistant to the toxic effects of hyperbaric oxygen. In addition to ocular tissues the response of homogenates of hepatic tissue to hyperbaric oxygen was investigated. The effect of hyperbaric oxygen on the pseudobranch, a tissue implicated in the function of the choroidal countercurrent oxygen multiplier, was also studied.

White grunts (Haemulon plumieri) weighing approximately 82 grams were obtained from coral ledges in the vicinity of Pigeon Key, Fla. Experimental animals were maintained in recirculated sea water in a 200 gallon aquarium at the Uni-