Table II—Comparison of Glucocorticoid Activity with Some Features of Steroid Structure (10)

Treatment	Hepatic Glycogen, mg/g	Thymus Weight, g/100 g of Body Weight	O-3–O-11, Å	O-11–O-17, Å	O-3–Mean Plane C-5–C-17, Å
Cortisone	0.67 ± 0.24 NS[a]	0.44 ± 0.003 NS	6.551 (6)[b]	5.195 (6)	1.30
Control water	0.71 ± 0.10	0.48 ± 0.02			
6α-Methylprednisolone	12.30 ± 1.8*	0.24 ± 0.03*	6.593 (4)	5.219 (4)	1.95
Control water	0.71 ± 0.10	0.48 ± 0.02			
Fludrocortisone	11.53 ± 0.33*	0.11 ± 0.01*	6.816 (5)	5.210 (5)	2.43
Control water	0.59 ± 0.10	0.39 ± 0.02			
Dexamethasone acetate	61.99 ± 4.10*	0.08 ± 0.01*	6.822 (5)	5.321 (5)	2.57
Control water	0.59 ± 0.10	0.39 ± 0.02			

[a] NS = not significant. * = $p < 0.005$. [b] Standard deviation.

distances between oxygen functions must also play significant roles. It can be seen in Table II that the oxygen–oxygen distances also follow those of the O-3–mean plane (C-5–C-17). There is actually a better correlation between the O-11–O-17 distances and glucocorticoid activity than with the one just mentioned.

The effect of the oxygen–oxygen distances on the catatoxic activity of I and dexamethasone cannot be very significant since I, which does not have oxygen functions on either C-11 or C-17, is nonetheless a potent catatoxic steroid. Superimposition of the molecule of fludrocortisone on that of dexamethasone (Fig. 1*a*) indicates that C-16 substitution is a decisive factor for strong catatoxic activity. This is further supported by the fact that I (itself a potent agent) and dexamethasone have a C-16 substituent in the α-configuration. Removal of this substituent results in a loss of catatoxic potency.

The present findings established a relationship between A-ring conformation and distances between oxygen functions and glucocorticoid activity. However, such a correlation was not demonstrated for catatoxic activity. Compound I and dexamethasone, both potent catatoxic steroids, have widely differing A-ring conformations (Fig. 1); dexamethasone and fludrocortisone (Table I), with almost identical A-ring conformations, show great differences in catatoxic activity.

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COMMUNICATIONS

Medazepam pKa Determined by Spectrophotometric and Solubility Methods

Keyphrases □ Medazepam—pKa determination, spectrophotometric and solubility methods compared □ UV spectrophotometry—pKa determination, medazepam, compared to solubility method □ Solubility—medazepam, pKa determination, compared to spectrophotometric method □ Tranquilizers—medazepam, pKa determination

To the Editor:

The spectral behavior of medazepam shows a marked pH dependence. The peak at 253 nm in acid solutions decreases above pH 3.5 while a new peak appears at 234 nm. The absorptivity and the maximum wavelength of medazepam shift with increasing pH. No variation with pH is observed at the isosbestic point at 243 nm.

All UV absorption spectra were taken at 37° using a spectrophotometer[1] with thermostated cell holders. The pKa was calculated from four series of spectra of pH-varied solutions according to the method of Albert and Serjeant (1) at the wavelengths of 233, 253, and 286 nm (Fig. 1) using:

$$\text{pKa} = \text{pH} - \log\frac{A-B}{B-C} \qquad \text{(Eq. 1)}$$

where A is the absorbance of a solution at pH 1.0, B is the absorbance in buffered solution, and C is the absorbance at pH 10.0. The distance $A - B$ is representative for the concentration of the unionized form of the base, whereas $B - C$ is representative for the ionized

[1] Unicam SP 800.

Table I—Comparison of pKa Values of Medazepam

pKa	Determination by	Reference
4.4	Spectroscopy	2
8.7	Polarography, pK_1	3
4.4	Spectroscopy, pK_1	3
6.17	Spectroscopy	This study
6.19	Solubility	This study

form using the Henderson–Hasselbalch equation.

In the region where the pH approximates the pKa, buffer solutions differing by 0.5 pH unit were used. All pH values of the mixtures were measured potentiometrically with a glass electrode, calibrated with accurate standard buffer solutions. Calculation of all results yielded a pKa of 6.17 ± 0.02. The findings disagreed with previous results (2, 3) (Table I).

Further determinations of the pKa were carried out by the solubility method reported previously (4). The plot of the hydrogen-ion activity against solubility is shown in Fig. 2. The relationship yields an ordinate intercept of $-K$, which can be converted into pKa = $-\log K$. In the equation:

$$[H^+] = \frac{KS}{S_0} - K \qquad \text{(Eq. 2)}$$

K is the acid dissociation constant of the base, S_0 is the low solubility of the free base, and S is the total solubility at a particular pH.

Solubility determinations were carried out at 37° by incubating excess amounts of medazepam with buffer solutions in closed vials in a temperature-controlled water bath for 48 hr. The samples were intermittently shaken, the supernate was filtered at 37° and diluted with buffer solution, and the concentrations of medazepam were determined by UV spectrophotometry at 37°. The pH of the supernate was measured with a glass electrode at 37°.

All determinations were repeated, with a longer equilibrating time, without any deviation in the results. The ordinate intercept of K was 6.5×10^{-7} and yielded a pKa of 6.19. This finding was in good agreement with the value from the spectrophotometric determination.

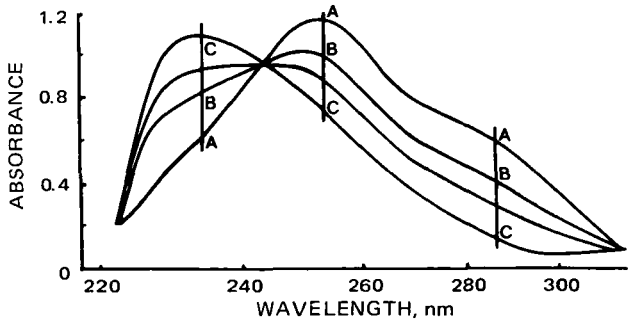

Figure 1—*UV spectra of medazepam in acid (pH 1, points A) and alkaline (pH 12, points C) solutions and at pH 6.0 (points B).*

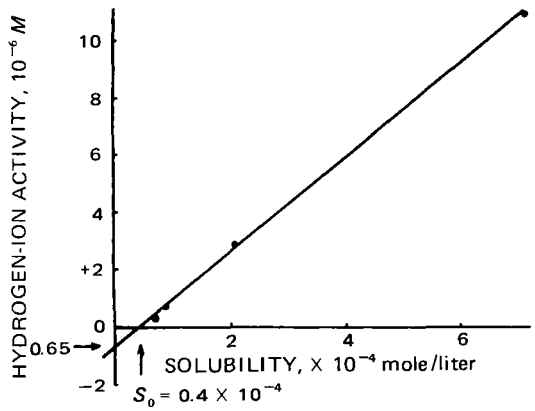

Figure 2—*Determination of* −K *by the method of Green (4) from the relationship between hydrogen-ion activity [H⁺] and solubility,* S, *of medazepam;* S_0 *is the low solubility of the free base.*

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Differences in Ocular Penetration of Pilocarpine in Rabbits of Different Ages

Keyphrases □ Pilocarpine—differences in ocular penetration related to age, rabbits □ Ocular penetration—pilocarpine, related to differences in age, rabbits □ Age, rabbits—effects on ocular penetration of pilocarpine

To the Editor:

In topical ophthalmic drug therapy, generally no distinction is made between age groups. In most cases, from the neonate to the geriatric, the same dosage is administered. The potential hazards of such an approach were recognized by France and France (1), who observed toxic side effects when administering the usual adult dose of phenylephrine to neonates.

It has been our contention for some time that the administration of equal doses of topical ophthalmic drugs to widely varying age groups should result in substantial differences in the amount of drug reaching the aqueous humor. The theoretical aspects of these considerations were discussed recently (2). Such an approach involves both therapeutic and toxicity considerations.

The present communication demonstrates, for the first time, that differences do exist in the penetration of topically applied ophthalmic drugs as a function of age. Such differences may be due to size, weight, tear flow, rate of drainage, and/or structure and function of the various ocular tissues. A more detailed examination of such differences is warranted so that ophthalmic dosage regimens can be placed on a more rational basis.

Identical doses of 25 μl of 1×10^{-2} *M* tritiated pilocarpine solution were instilled into the eyes of 20- and