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Effect of Dose Size on the Pharmacokinetics of Intravenous Hydrocortisone During Endogenous Hydrocortisone Suppression

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The pharmacokinetics of hydrocortisone were examined following single intravenous doses of 5, 10, 20, and 40 mg hydrocortisone, as the sodium succinate salt, to healthy male volunteers. Endogenous hydrocortisone was suppressed by administration of 2 mg dexamethasone the night before hydrocortisone injection. Plasma samples obtained serially during 8 h after hydrocortisone injection were assayed by reverse-phase HPLC using a fixed wavelength (254 nm) ultraviolet detector. Initial concentrations of hydrocortisone in plasma were proportional to dose size. The subsequent decline in hydrocortisone concentrations was biphasic, and individual data sets were adequately described in terms of the pharmacokinetic two-compartment open model. Values of pharmacokinetic parameters were similar from the 5, 10, and 20 mg doses. Following the 40 mg dose, the overall elimination rate constant decreased, while the distribution volume, V_{dss}, *and plasma clearance increased, in comparison with the values obtained from lower doses. Changes in the pharmacokinetics of hydrocortisone at high doses may be related to drug concentration-dependent changes in the binding of hydrocortisone to plasma proteins. Previously reported dose-dependent changes in some pharmacokinetic parameters following oral hydrocortisone are attributed to absorption rather than distribution or elimination effects.*

KEY WORDS: intravenous hydrocortisone; blood concentrations; pharmacokinetics; endogenous hydrocortisone suppression.

INTRODUCTION

Hydrocortisone, the principal endogenous glucocorticoid in man, is used therapeutically for a variety of indications (1). Two factors which have prevented accurate description of hydrocortisone bioavailability are the insufficient sensitivity and specificity of available analytical methods and interference due to endogenous circulating hydrocortisone.

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High pressure liquid chromatography (HPLC) methods have recently been described, using both ultraviolet absorbance and fluorescence detectors, which are capable of selectively measuring plasma hydrocortisone concentrations as low as 3 ng/ml (2–7). Circulating levels of endogenous hydrocortisone, which vary diurnally between 40 and 200 ng/ml, have recently been shown to be suppressed to concentrations between 6 and 14 ng/ml following oral administration of dexamethasone (8). A 2 mg dose of dexamethasone given at 11 p.m. results in uniform suppression of endogenous circulating hydrocortisone until at least 8 p.m. the following day.

A previous study in this laboratory (7) suggested a possible lack of dose proportionality in plasma hydrocortisone concentrations following 10, 30, and 50 mg oral doses of hydrocortisone. Similar results were observed following oral cortisone acetate (9).

In order to carry out meaningful bioavailability and pharmacokinetic studies with oral hydrocortisone formulations, it is necessary first to establish whether the distribution and elimination characteristics of hydrocortisone exhibit dose dependency following parenteral doses, i.e. in the absence of absorption effects. In the present study, the pharmacokinetics of hydrocortisone were examined following single 5, 10, 20, and 40 mg intravenous doses to healthy male volunteers.

EXPERIMENTAL

Materials

Standard solutions of 0.025–15 μg/ml hydrocortisone (Sigma Chemical Co., St. Louis, Mo.) were prepared in 60% aqueous methanol. The internal standard for the HPLC assay, Δ^4-pregnen-17α,20β,21-triol-3,11-dione (Sigma Chemical Co.) was prepared as a 4 μg/ml solution in methanol. Dichloromethane and methanol (Burdick and Jackson Laboratories, Muskegon, Mich.) were used as received. Distilled water was further purified using a Barnstead PCS water purification system (Barnstead Sybron Corp., Boston, Mass.). Methanolic and aqueous solutions were stored at −10 and 5°C, respectively, and were brought to room temperature before use. All other solvents and chemicals were reagent grade quality, and were used as supplied.

Subjects

After giving informed consent, six male volunteers (age 21–29 years, height 175–196 cm, weight 64–82 kg) underwent physical examinations which included blood and urine analysis. All vital signs and laboratory values were within normal limits.

Protocol

Each subject received single intravenous doses of 5, 10, 20, and 40 mg hydrocortisone sodium succinate (A-Hydrocort, Lot #11-573-AF, Abbott Laboratories, Chicago, Ill.). The solution for injection contained 50 mg hydrocortisone equivalents per ml. Accurate delivery of dosage volumes of 0.1–0.8 ml was assured by injecting these small volumes from a 1.0 tuberculin syringe. Injection time did not exceed 5 s for any dose.

The different dosages were administered to each subject at least one week apart, in a randomized block design. Subjects received no food from 8 p.m. the previous day until 12 noon on the day of hydrocortisone administration. No liquids were permitted from 11 p.m. the previous day until 8 a.m. the next day, when 180 ml of water was ingested immediately following the hydrocortisone injection. No further fluids were permitted until 12 noon on that day. All subjects received 20 ml of dexamethasone elixir containing 0.1 mg dexamethasone per ml (Decadron elixir, Lot # A 3240, Merck Sharp and Dohme, West Point, Pa.) together with 180 ml of water at 11 p.m. on the day preceding each hydrocortisone dose.

On the study day, hydrocortisone sodium succinate was injected into a forearm vein at 8 a.m. Blood samples (10 ml) were taken from the contralateral forearm vein into heparinized blood collection tubes (Vacutainer, Becton-Dickinson, Rutherford, N.J.) immediately before and then serially from 10 min until 8 h after dosing. Subjects were ambulatory throughout the sampling period. Plasma was obtained by centrifugation, and stored at −20°C until assayed. Assays were routinely carried out within one month, and no degradation of hydrocortisone occurred during this period.

The concentrations of hydrocortisone in plasma were determined by the HPLC-UV method recently developed in this laboratory (7). The peak height ratios from endogenous hydrocortisone in suppressed plasma were subtracted from the ratios for hydrocortisone added to the plasma in the construction of standard curves, and the ratios obtained from predose plasma samples were subtracted from all postdose values in order to obtain circulating drug levels due to administered hydrocortisone. The mean recovery efficiency for hydrocortisone from plasma was 82% and the coefficients of variation for replicate determinations for hydrocortisone concentrations between 5 and 3000 ng/ml were uniformly below 10%.

Data Analysis

Initial estimates of hydrocortisone pharmacokinetic parameters were obtained from individual data sets using standard graphical procedures. Improved estimates of parameter values were obtained by nonlinear

regression analysis of individual nonweighted data using the program NREG (10) on a Univac 1110 digital computer. Model-independent estimates of areas under plasma drug concentration versus time curves from zero to infinite time ($AUC^{0\to\infty}$) were calculated using the trapezoidal method (11). Areas for the portions of plasma curves following the last measurable data point were estimated by dividing the hydrocortisone concentration at that sampling time by the overall elimination rate constant β.

Pharmacokinetic parameter values were examined for dose and subject effects by two-way analysis of variance (ANOVA). When significant dose effects were observed, differences between individual treatments were examined by means of Tukey's test (12).

RESULTS

The mean plasma levels of hydrocortisone resulting from the four different doses are described in Fig. 1. Similar patterns of biphasic decline were observed from all four doses. Drug levels declined rapidly until 0.5–1.0 hr after dosing, and subsequently at a slower rate. During the slower rate of decline, the individual slopes were clearly monoexponential.

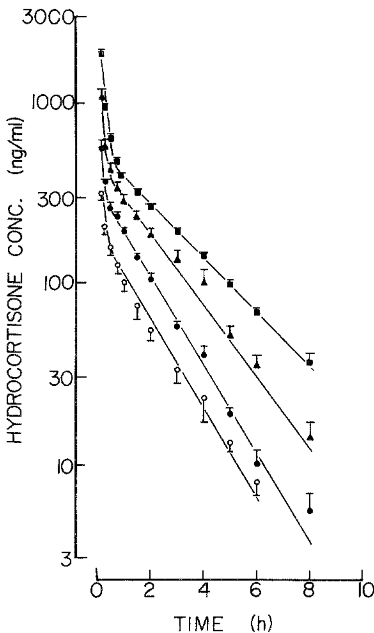

Fig. 1. Mean (±1 SE) plasma concentrations of hydrocortisone following intravenous doses of 5 mg (○), 10 mg (●), 20 mg (▲), and 40 mg (■) hydrocortisone sodium succinate. The drawn lines are constructed from Eq. (1) using the means of individual pharmacokinetic parameters from each dose, following nonlinear regression analysis.

Initial hydrocortisone levels were closely related to dose size. Mean peak values from the 5, 10, 20, and 40 mg doses, obtained at 10 min after dosing, were respectively 312, 573, 1095, and 1854 ng/ml.

By 8 hr postdosing, mean plasma hydrocortisone levels from the 10, 20, and 40 mg doses had fallen to 6, 14, and 36 ng/ml, respectively. Hydrocortisone levels from the 5 mg dose could not generally be detected beyond the 6 hr sampling time.

The biphasic decline in plasma hydrocortisone levels is consistent with the pharmacokinetic two-compartment open model, and individual data sets were therefore analyzed in terms of Eqs. (1) and (2) (ref. 13):

$$C = [D/V_1(\alpha - \beta)][(k_{21} - \beta)\, e^{-\beta t} - (k_{21} - \alpha)\, e^{-\alpha t}] \quad (1)$$

$$\genfrac{}{}{0pt}{}{\alpha}{\beta} = 0.5[(k_{12} + k_{21} + k_2) \pm [(k_{12} + k_{21} + k_2)^2 - 4k_{21}k_2]^{1/2}] \quad (2)$$

where D is the administered dose, V_1 is the apparent drug distribution volume in the central body compartment, and k_{12}, k_{21}, and k_2 are first-order rate constants for drug transfer between the central and peripheral compartments (k_{12}, k_{21}) and for drug elimination, respectively.

The results of pharmacokinetic analysis are given in Table I. The high coefficients of determination, r^2, indicate excellent description of the observed data by the model. Mean parameter values were used in Eq. (1) to construct the curves which are superimposed on the means of the observed hydrocortisone levels in Fig. 1.

The values of α, k_{12}, and k_2 were independent of dose size, while β and k_{21} were significantly reduced following the 40 mg dose as compared to the 5 and 10 mg doses. The overall elimination half-life, $t_{1/2\beta}$, was also significantly longer following the 40 mg dose, compared to the 5 and 10 mg doses. No differences were observed in the values of any rate constants between the 5, 10, and 20 mg doses. The mean elimination half-lives of 1.3–1.7 hr from these doses are similar to previously reported values (14–17).

Areas under hydrocortisone plasma profiles were dose-related. Mean ratios of $AUC^{0\to\infty}$ values between the 10 and 5 mg and the 20 and 10 mg doses were 2.0 and 1.95, respectively. Doubling the dose from 20 to 40 mg, however, yielded a mean $AUC^{0\to\infty}$ ratio of 1.6.

After correction for the administered dose, area values from the 5, 10, and 20 mg doses, and also those from the 20 and 40 mg doses, were indistinguishable, whereas the corrected area from the 40 mg dose was significantly smaller than those from the 5 and 10 mg doses.

Dose related changes in corrected area values were similar to changes observed in the apparent distribution volumes of hydrocortisone. The mean volumes of the central compartment V_1, ranged from 6.9 to 8.8 liters, and

Table I. Mean Values of Pharmacokinetic Parameters for Hydrocortisone at Four Dose Levels

Parameter	Value ($n = 6$)								P	Tukey's test
	5 mg		10 mg		20 mg		40 mg			
$\alpha (h^{-1})$	6.3	(1.5)	8.7	(2.7)	7.6	(1.1)	6.8	(0.5)	>0.05	—[a]
$\beta (h^{-1})$	0.56	(0.18)	0.55	(0.08)	0.46	(0.08)	0.37	(0.02)	<0.05	5 10 20 40
$k_{12} (h^{-1})$	3.0	(0.9)	4.9	(1.9)	4.3	(0.8)	3.8	(0.3)	>0.05	—
$k_{21} (h^{-1})$	2.3	(0.7)	2.3	(0.6)	1.5	(0.2)	1.2	(0.2)	<0.025	5 10 20 40
$k_2 (h^{-1})$	1.6	(0.4)	2.2	(0.6)	2.2	(0.6)	2.2	(0.4)	>0.05	—
$t_{1/2el}$(h)	0.46	(0.11)	0.38	(0.11)	0.33	(0.07)	0.33	(0.04)	>0.05	—
$t_{1/2\beta}$(h)	1.3	(0.3)	1.3	(0.2)	1.7	(0.2)	1.9	(0.1)	<0.005	40 20 10 5
V_1 (liters)	8.5	(2.1)	7.0	(2.2)	6.9	(1.9)	8.8	(1.8)	>0.05	—
V_{dss}[b] (liters)	20.7	(7.3)	20.8	(4.3)	26.0	(4.1)	37.5	(5.8)	<0.001	40 20 10 5
C_{pl}[c] (ml/min · m^2)	209	(42)	218	(23)	239	(44)	294	(34)	<0.001	40 20 10 5
r^2[d]	0.998	(0.002)	0.999	(0.007)	0.999	(0.004)	0.999	(0.003)	>0.05	—
$AUC^{0 \to \infty}$ (ng h/ml)	410	(80)	790	(100)	1480	(310)	2290	(260)	<0.001	40 20 10 5
$AUC \cdot \beta$ (ng/ml)	220	(70)	430	(80)	630	(90)	840	(110)	<0.001	40 20 10 5
AUC/D (10^{-3} kg h/ml)[e]	6.1	(0.1)	5.9	(0.8)	5.4	(0.6)	4.3	(0.4)	<0.025	5 10 20 40

Note: Standard deviations are shown in parentheses. The p value indicates the significance level of treatment effects, calculated by ANOVA. Differences between individual treatments are calculated by Tukey's test.

[a] Not calculated.

[b] $V_{dss} = V_1(1 + k_{12}/k_{21})$.

[c] Plasma clearance $= D/AUC^{0 \to \infty}$.

[d] $r^2 = (\sum obs^2 - \sum dev^2)/\sum obs^2$.

[e] D = dose per kg body weight.

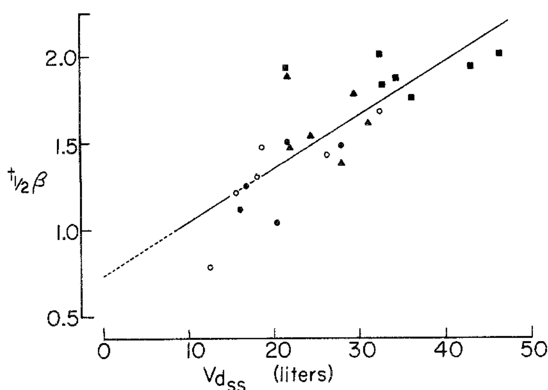

Fig. 2. Relationship between the overall elimination half-life, $t_{1/2\beta}$, of hydrocortisone vs the apparent volume of distribution at steady state, V_{dss}, for hydrocortisone doses of 5 mg (○), 10 mg (●), 20 mg (▲), and 40 mg (■). The linear regression equation for the line is (SD in parentheses): $t_{1/2\beta} = 0.029$ (± 0.005) $Vd_{ss} + 0.79$ (± 0.14), $r = 0.771$.

were independent of dose. Mean values of V_{dss} were similar from the 5, 10, and 20 mg doses (20.7 to 26.0 liters), but significantly increased to 37.5 liters from the 40 mg dose. The high correlation between V_{dss} and $t_{1/2\beta}$ is shown in Fig. 2.

The mean plasma clearance of hydrocortisone increased from 209 to 239 ml/min following the 5, 10, and 20 mg doses, and to 294 ml/min following the 40 mg dose. Since the value of β decreased following the 40 mg dose, compared to the lower doses, the increased plasma clearance must be related to increases in overall drug distribution following the 40 mg dose. There were no significant subject effects in the analysis of variance of hydrocortisone pharmacokinetic parameters.

DISCUSSION

A factor which has hindered the study of hydrocortisone pharmacokinetics has been that of differentiating between endogenous hydrocortisone and administered compound. One approach, which has been pursued in this laboratory and elsewhere, is that of suppressing endogenous hydrocortisone by administering dexamethasone (8). Dexamethasone is thought to act by blocking release of adrenocorticotrophic hormone (18).

Administering 2 mg dexamethasone at 11 p.m. suppresses endogenous hydrocortisone plasma levels to 11 ng/ml, or less, at least until 6–8 p.m. the following day, and the hydrocortisone levels during that time are quite stable (8). The levels of endogenous hydrocortisone that were obtained

immediately before hydrocortisone administration varied from 7 to 11 ng/ml, and these values were subtracted from total plasma levels following hydrocortisone administration.

The dosed hydrocortisone could further suppress endogenous levels by a direct feedback mechanism. However, as the plasma drug levels were generally far greater than the suppressed, endogenous levels, additional suppression would have a negligible effect on the net plasma levels of hydrocortisone, and on the results of the study.

Dexamethasone could also compete with hydrocortisone for binding sites on plasma proteins and elsewhere, but this is unlikely. Dexamethasone is less extensively bound to plasma proteins than hydrocortisone, and has a biological half-life in man of approximately 2 hr (19,20). Extrapolation from previous data (20) suggests that dexamethasone levels in plasma resulting from the 2 mg, 11 p.m. dose would be less than 2 ng/ml at the 8 a.m. sampling time the next day. This value is considerably lower than the levels of suppressed endogenous hydrocortisone and is a negligible fraction of the levels resulting from administered compound.

The results of a previous study involving oral doses of hydrocortisone (7) have suggested that the pharmacokinetics of hydrocortisone may exhibit dose dependency in the therapeutic range. Doses of 10, 30, and 50 mg hydrocortisone yielded mean peak plasma levels of hydrocortisone ranging from 192 ng/ml following the 10 mg dose to 396 ng/ml following the 50 mg dose. Lack of dose proportionality in the plasma levels of hydrocortisone led to the suggestion that the absorption of hydrocortisone may decrease, the distribution of hydrocortisone in the body may increase, or both of these may occur, with increasing doses.

In the present study, intravenous doses of 5, 10, 20, and 40 mg hydrocortisone gave rise to mean maximum plasma hydrocortisone levels much higher than those following the oral doses, ranging from 312 ng/ml from the 5 mg dose to 1854 ng/ml from the 40 mg dose. The peak level that was previously obtained following the 50 mg oral dose (7) was thus intermediate between the maximum level following the 5 and 10 mg intravenous doses. Evidence of dose nonproportionality following intravenous hydrocortisone occurred only following the 40 mg dose.

These observations lead us to conclude that the distribution and elimination characteristics of hydrocortisone were unaffected by the oral doses that were employed in the previous study (7). Observed nonlinearity in the pharmacokinetic parameters of oral hydrocortisone is attributable therefore to dose-dependent changes in drug bioavailability.

Drug concentration-dependent changes in the binding of hydrocortisone may explain the apparent increase in V_{dss} following the 40 mg intravenous hydrocortisone dose. At concentrations below 200 ng/ml,

plasma hydrocortisone is tightly bound to transcortin, and the free fraction of circulating drug is only 5% (21). At plasma hydrocortisone concentrations greater than 200 ng/ml, a greater proportion of compound is loosely bound to albumin, and the free fraction increases to approximately 25%.

Following the 5 and 10 mg intravenous doses, plasma levels of hydrocortisone were generally less than 200 ng/ml for almost the entire postdistributive, or β phase. The 20 mg dose yielded plasma hydrocortisone levels greater than 200 ng/ml for approximately 1.5 hr, while the 40 mg dose resulted in levels exceeding 200 ng/ml for 3–4 hr postdosing. As the α phase of plasma hydrocortisone profiles lasted for approximately 1 hr after dosing, there was insufficient time to detect a change in drug distribution or elimination during the β phase. The excellent fit of all individual postdistributive data to a monoexponential function suggests that no such change occurs. However, the changes are possibly obscured by the compensating effects of increased binding to plasma proteins, which causes reduced tissue distribution, and also less efficient biotransformation of lower drug concentrations.

The longer plasma half-life of hydrocortisone at higher drug concentrations is clearly related to an increase in the apparent distribution volume, as shown in Fig. 2. However, this effect is attenuated as reduced binding to plasma proteins at high drug concentrations not only permits more hydrocortisone to enter extravascular fluids, but also increases access of drug to the hepatocyte, which facilitates biotransformation. The low affinity binding of hydrocortisone to plasma albumin has been shown to be less inhibitory to hepatic uptake and biotransformation than the high affinity binding to transcortin (22).

The results of this study show that the distribution and elimination characteristics of hydrocortisone in healthy male volunteers are independent of dose size at intravenous doses of 5, 10, and 20 mg, but are dose-dependent after an intravenous dose of 40 mg. In view of the low circulating levels of hydrocortisone obtained after oral doses, compared to intravenous doses, dose-dependent changes in hydrocortisone distribution or elimination are unlikely to occur following single oral doses of 5–50 g. This conclusion does not extend to possible dose-related changes in the first-pass metabolism of orally administered hydrocortisone, which has yet to be investigated.

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