Hydrocortisone Choleresis in the Dog

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ABSTRACT Hydrocortisone sodium succinate (Solu-Cortef; Upjohn Co., Kalamazoo, Mich.) has been found to induce choleresis in unanesthetized fasting dogs fitted with Thomas duodenal cannulae for direct quantitative collection of bile. In all experiments, bile flow increased (average, 68%) 15–20 min after beginning hydrocortisone by infusion in association with an equivalent increment in the output of sodium, potassium, chloride, and bicarbonate. In five animals, the choleretic response occurred independently of, and apparently additive to, the effect of simultaneously administered sodium taurocholate. The fluid added to the bile resembled an ultrafiltrate of plasma. Erythritol clearance increased in proportion to flow, suggesting an effect at the hepatocellular rather than ductal level and probably independent, therefore, of endogenous secretin release. Hydrocortisone and its metabolites were excreted in amounts too small to induce choleresis osmotically. Simultaneous administration of sulfobromophthalein sodium blocked the choleretic response without preventing hydrocortisone excretion. The data suggest that a previously ill-defined mechanism of canalicular bile formation, not mediated by bile salt excretion, may be operative in choleretic response to a variety of agents.

INTRODUCTION

The physiologic effects of the acute administration of adrenocortical steroids on the normal liver are not well-defined. Studies in man (1–4) have yielded conflicting data with respect to the influence of cortisol and other glucocorticoids on bile composition and flow. Both choleretic and noncholeretic responses have been observed after administration of cortisone and hydrocortisone. These disparate results may reflect differences in methods of bile collection or patient selection rather than differing responses to the same hormone.

A marked sensitivity of hepatobiliary secretory mechanisms to a variety of other steroid compounds has been amply demonstrated. Active secretion of primary bile salts, for example, has long been known to provide a major impetus for bile formation in most species (5). In contrast, some steroids (estrogens, androgens, secondary bile acids) have deleterious effects upon hepatic excretory function (6–9).

Systematic evaluation of the influence of adrenocortical steroids on biliary composition and flow in normal experimental animals appears to be lacking (10–12). In the studies presented herewith, the effects of intravenous hydrocortisone on bile formation during constant taurocholate infusion have been explored in unanesthetized dogs equipped with duodenal cannulae. The results indicate that hydrocortisone is highly active in stimulating a choleresis that is canalicular in origin and independent of bile acid secretion.

METHODS

All studies were made in five healthy unanesthetized adult dogs (16–30 kg) which had undergone cholecystectomy and installation of a Thomas duodenal cannula at least 8 months before the present studies. Before each experiment the animal under study was fasted for 24 hr. The common bile duct was then catheterized under direct vision with a No. 6 olive-tipped ureteral catheter. Thereafter, the dog was maintained in the upright position with the aid of a sling. Bile samples (3.5–4 ml) were collected by gravity in graduated centrifuge tubes.

Throughout all studies, a 1.3% solution of sodium taurocholate (Nutritional Biochemicals Corporation, Cleveland, Ohio) was infused (Bowman pump, Bowman Electrical Products Corp., Chicago, Ill., at a constant rate of 10–12 μEq/min) through a No. 17 Deseret polyethylene catheter in a foreleg vein. Pipenzolate methylbromide[1] (Piptal) was also given intravenously (2 mg every 5 min during the 1st hr and every 10 min thereafter) to diminish spontaneous

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variations in bile flow (13). Each experiment consisted of a control period of 60 min, a period of about 1 hr when 200 mg of hydrocortisone sodium succinate (Solu-Cortef) in 50 ml of normal saline were administered intravenously, and two 60-min recovery periods. When plasma samples were obtained, they were drawn through a polyvinyl catheter (Vx 044 BD) in one of the saphenous veins.

On three occasions sodium taurocholate-24-^{14}C (Tracerlab Div., LFE Electronics, Richmond, Calif.) was added in tracer amounts to the taurocholate infusion and infused at a rate of about 0.35 μCi/min. Biliary excretion of the labeled bile salt was calculated from the product of the concentration (dpm/ml) and the bile flow (ml/min). Since the hepatic extraction of sodium taurocholate at this rate of infusion has been shown to approximate 100% (14), the hepatic plasma flow was estimated by calculation of the biliary clearance, C_B, of the tracer by the formula $C_B = BV/A$ where C_B = biliary clearance, ml/min; BV = biliary output of the tracer, dpm/min; and A = the plasma concentration of the labeled bile salt, dpm/ml.

In four experiments hydrocortisone-4-^{14}C (New England Nuclear Corp., Boston, Mass., SA, 0.1 mCi/0.0697 mg of hydrocortisone) was infused with the unlabeled hydrocortisone at a rate of 0.1 μCi/min. The recovery of the tracer was measured in bile and urine samples which were collected simultaneously. Urine samples were obtained through a Bardex Four Wing Malecot catheter which had been passed into the urinary bladder under local anesthesia.

During three studies erythritol-^{14}C (Amersham-Searle) was administered at a rate of about 0.3 μCi/min after an initial priming dose of 3 μCi. An equilibration period of at least 60 min was followed by the usual control and hydrocortisone infusion periods. Further collection of plasma and bile samples was continued for only a single recovery period. Clearance values during all three periods were calculated as for bile salt as noted above.

Measurements of sulfobromophthalein sodium (BSP, Hynson, Westcott & Dunning, Inc., Baltimore, Md.), transport maximum (T_m), and relative storage capacity (S) during control, hydrocortisone infusion, and recovery (R_1) periods were made on five occasions (four dogs) 1 hr after the BSP infusion (5–10 mg/min) had begun. In addition, all dogs received an initial prime of 8 mg/kg. Plasma samples were drawn every 5 min, and bile samples collected every 10 min. T_m (mg/min) was taken as the biliary output (mg/min) of BSP as long as the plasma concentration remained in excess of 3 mg/100 ml. The relative storage capacity, S (mg/mg per 100 ml), was estimated as follows: $S = ([I - \Delta pPV] - T_m)/\Delta p$ where I = infusion rate of BSP, mg/min; Δp = the change in plasma BSP concentration per min, mg/100 ml per min; and PV = the plasma volume in hundreds of millimeters. On one occasion (Dog R‡—Table IV), hydrocortisone-4-^{14}C was added to the hydrocortisone infusion, and biliary clearance of radioactivity was also measured.

Analytical techniques. Sodium, potassium, and chloride concentrations in bile were determined with a Technicon autoanalyzer (Technicon Co., Inc., Tarrytown, N. Y.). Total carbon dioxide content was measured with a Natelson gasometer, and bicarbonate concentrations were estimated by subtracting 1.2 mmole, the concentration of carbonic acid at an assumed P_{CO_2} of 40 mm Hg. Bile acid concentrations were calculated as the difference between the sums of these cations and anions (15). pH was measured with a Beckman Zeromatic (Beckman Instruments Inc., Fullerton, Calif.) pH meter in bile samples immediately after collection in corked tubes. Osmolality was estimated by freezing point depression in a Fiske Osmometer (Fiske Associates, Uxbridge, Mass.).

Radioactivity in plasma and bile samples was measured with a Packard Tri-Carb (Packard Instrument Co., Inc., Downers Grove, Ill.) model 3003 liquid scintillation spectrometer. All plasma samples were treated as previously described (15). Bile samples containing sodium taurocholate-24-^{14}C were diluted 2–500 times with physiologic saline. A 1 ml aliquot of this dilution was then added to 10 ml of Bray's solution for counting. Bile samples containing erythritol-^{14}C as well as those with hydrocortisone-4-^{14}C were treated as previously described (16) except that 20 μl of 30% hydrogen peroxide were added after bleaching with sodium hypochlorite (Chlorox, Texo Corp., Cincinnati, Ohio) to eliminate extraneous scintillations caused by reaction between sodium hypochlorite and Bray's solution. Quenching was estimated by addition of an internal standard.

Chromatographic analysis of bile and urine samples containing hydrocortisone-4-^{14}C was made with a solvent system of 1-butanol (Fisher certified, Fisher Scientific Company, Pittsburgh, Pa.) methanol (Fisher histologic grade), and deionized water in the proportions of 2:1:1, v/v/v, which appeared to give the best separation of free cortisol and cortisol metabolites from bile and other constituents of canine bile and urine. Aliquots of bile (0.3–0.5 ml), urine, or methylene chloride extracts of urine (0.5–1.5 ml), and hydrocortisone hemisuccinate or hydrocortisone-4-^{14}C solutions (0.005–0.01 ml) were applied across 10-cm strips on 23 cm 3 MM Whatman paper strips which were developed by descending chromatography at 4°C for 18 hr.. After air drying, each paper was cut in three equal longitudinal strips with one to be examined directly under ultraviolet light (long and short wave), one to be sprayed with Kritchevsky's reagent before examination under ultraviolet light, and one for strip counting in a Packard (Packard Instrument Co., Inc.) Radiochromatogram Scanner, model 385.

Sulfobromophthalein sodium (BSP) concentrations in plasma and bile samples and the plasma volume were determined by methods previously described in detail (17).

Statistical significance of changes in bile composition and flow was evaluated with the Student's *t* test.

RESULTS

Water and electrolyte excretion. Within 15–20 min after the start of a hydrocortisone infusion, a marked increase in bile flow was observed in all five dogs (Fig. 1). The increment, which ranged maximally from 38 to 82% (mean 68%) in excess of control valves persisted for at least 1–2 hr after termination of the infusion. The increments in flow were statistically significant at the 0.5% level in each animal. In association with this choleresis, the bile concentrations of sodium and potassium fell (Table I). Bicarbonate concentrations either increased slightly or remained unchanged while chloride concentrations rose. Consequently, the calculated concentrations of bile acids fell in every instance. The changes in concentration paralleled the changes in bile flow, usually appearing during the infusion and tending to persist thereafter.

The biliary outputs of all electrolytes, with the exception of taurocholate, were markedly increased during

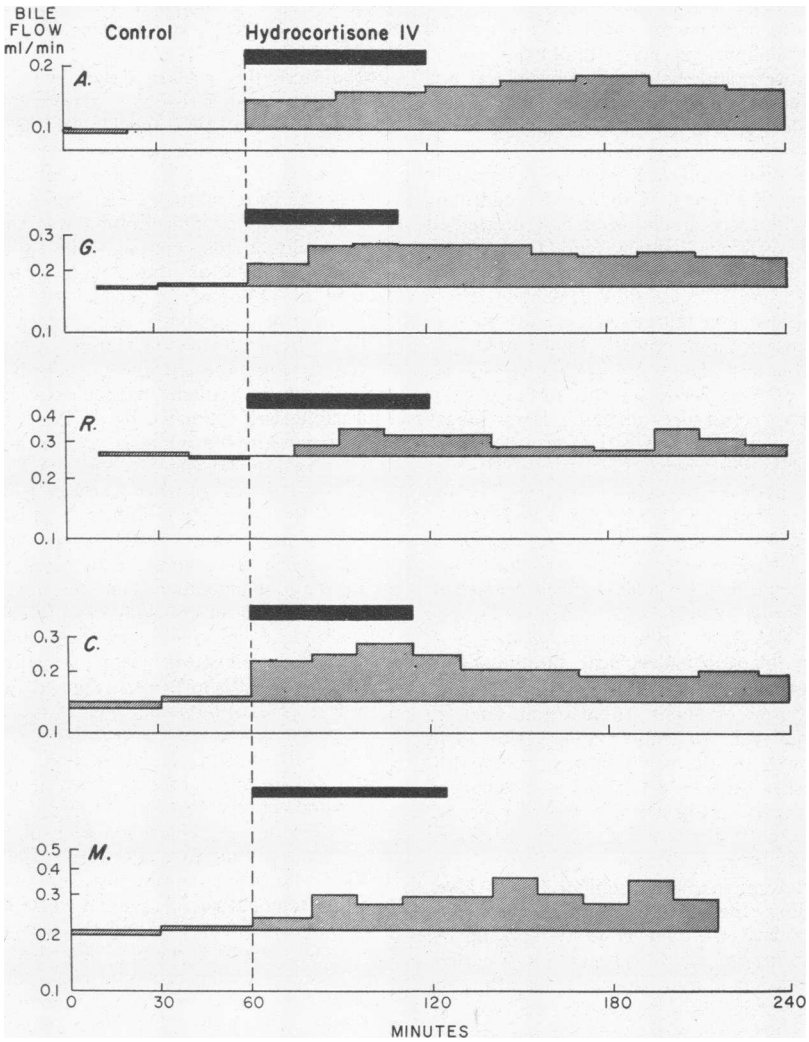

FIGURE 1 Effect of hydrocortisone on bile flow in the dog. Bile flow (ml/min) is plotted logarithmically on the ordinate against time (min) for each of five dogs (A, G, R, C, and M) before, during, and after intravenous administration of hydrocortisone. Control bile flow is plotted on the left; cross-hatched areas indicate deviations from the mean control values throughout the course of each experiment. Bile flow increased significantly in every experiment.

administration of hydrocortisone and for 1 hr thereafter (H and R_1, Table II) when bicarbonate output had returned to control levels. All increments were statistically significant at the 0.1–0.5% levels. There was no significant change in biliary pH or osmolality (Table I).

Bile acid excretion. Computation of bile acid excretion, as the difference between cation and anion excretion, failed to show significant change during hydrocortisone choleresis. This finding was to have been expected during the maintenance of a constant infusion of sodium taurocholate. A variable discrepancy between the amount of bile acid administered and the amount excreted in the bile (11.75–22.21 μEq/min in the control periods, Table II) was observed in these as in earlier experiments (13). These differences may be accounted for by variations in endogenous bile acid synthesis or entry via the enterohepatic pathway. It seems likely that synthesis tended to be suppressed by the relatively high plasma concentrations maintained throughout and that inconstant reabsorption of bile acids present in the intestinal contents at the beginning of each experiment was chiefly responsible. The fact that output tended to be constant, except for changes to be ex-

TABLE I

*Effect of Hydrocortisone on Biliary Electrolyte Composition**

Dog	Period	pH	Na^+	K^+	Cl^-	HCO_3^-	Tau^-	Osmolality
					mEq/liter			*mOsm/kg*
A	C	7.78	200	7.8	63	21	125	286
	H	7.80	185	7.2	71	27	95	285
	R_1	7.88	175	6.6	78	31	73	282
	R_2	7.80	181	6.6	78	29	80	287
G	C	7.80	163	6.1	58	26	86	253
	H	7.72	152	5.6	68	26	63	258
	R_1	7.66	155	5.6	69	26	66	262
	R_2	7.66	154	5.5	69	27	63	259
R	C	7.89	177	6.3	62	29	92	275
	H‡	7.83	167	6.0	69	29	75	271
	R_1	7.90	163	5.9	71	29	69	270
	R_2	7.90	168	6.8	72	32	70	273
C	C	7.90	183	6.3	65	29	96	283
	H	8.00	172	5.9	77	33	68	273
	R_1	7.78	157	5.4	72	28	62	263
	R_2	7.88	155	5.4	75	23	62	264
M	C	7.90	192	7.0	76	34	89	288
	H	7.90	185	6.7	80	37	76	281
	R_1	8.00	178	6.1	83	39	62	284
	R_2	7.90	172	5.6	82	43	52	282

* Values for biliary electrolyte concentrations (mEq/liter), including taurocholate (Tau^-), were obtained in five experiments during a 60 min control period (C), infusion of hydrocortisone (H), and the last 30 min of each of two subsequent 60-min periods (R_1 and R_2).

‡ Includes samples only from final 30 min.

pected as a result of dead space effects and depletion of the endogenous bile acid pools during complete collection of bile, suggests that the hormone had no direct effect upon biosynthesis or intestinal absorption.

In order to define more precisely the hepatic excretion and clearance of bile acids, sodium taurocholate-4-^{14}C was added to the taurocholate infusion at the beginning of the experiment in each of three dogs. The biliary output of radioactivity remained constant throughout in conformity to the indirect determinations. Since the extraction of taurocholate by the canine liver is close to 100% when it is administered in doses (10–12 μEq/min) comparable with those employed in these studies (14), it may be inferred that the hepatic clearance of labeled taurocholate may be used as a reliable estimate of hepatic blood flow. The constancy of labeled taurocholate clearances under these circumstances throughout each of these three experiments therefore indicates little or no change in hepatic perfusion and in consequent delivery of bile acids or other substances to the liver as a result of such a change.

It should be stressed that the choleresis produced by hydrocortisone appeared to be superimposable upon that attributed to taurocholate excretion (Fig. 2). The extent to which this response was strictly additive cannot be defined at this point owing to the intrinsic variance in bile flow, unpredictable differences in control values for flow from animal to animal and from time to time in the same animal, and uncertainties regarding the hydrocortisone dose-response relationship. In two experiments, taurocholate was infused at successively higher rates (10, 45, and 90 μEq/min) to determine the bile flow-taurocholate output relationship. Bile flow increased with each increment in taurocholate infusion to reach an equilibrium after about 60 min at each rate (shown as closed circles in Fig. 2). These measurements were then repeated during constant hydrocortisone infusion (4 mg/min; open circles in Fig. 2). In one animal (C) the taurocholate infusion was successively reduced from 90 to 45 to 10 μEq/min. In the other (R) the taurocholate infusion was reduced to 10 μEq/min and then increased successively to 45 and 90 μEq/min. Each measurement

TABLE II

*Effect of Hydrocortisone on Bile Flow and Biliary Electrolyte Excretion**

Dog	Period	Bile flow	Na^+	K^+	Cl^-	HCO_3^-	Tau^-
		ml/min			*μEq/min*		
A	C	0.094	18.51	0.73	5.87	1.92	11.75
	H	0.144	26.43	1.03	10.13	3.88	13.70
	R_1	0.162	28.27	1.07	12.56	5.02	11.83
	R_2	0.144	26.06	0.95	11.23	4.19	11.50
G	C	0.166	27.06	1.01	9.63	4.20	14.29
	H	0.270	40.98	1.50	18.38	6.96	17.00
	R_1	0.230	35.80	1.29	15.87	5.95	15.20
	R_2	0.230	35.50	1.27	15.87	6.24	14.50
R	C	0.241	42.61	1.52	14.97	7.01	22.21
	H	0.333	55.55	1.99	22.84	9.80	24.92
	R_1	0.295	48.15	1.75	21.12	8.55	20.36
	R_2	0.260	43.68	1.77	18.72	8.45	18.20
C	C	0.140	25.60	0.88	9.06	4.11	13.42
	H	0.245	42.03	1.45	18.83	8.02	16.62
	R_1	0.209	32.80	1.13	15.13	5.95	12.95
	R_2	0.188	29.13	1.01	14.19	4.34	11.65
M	C	0.207	39.64	1.45	15.70	6.98	18.41
	H	0.273	50.33	1.83	21.67	10.04	20.60
	R_1	0.307	54.45	1.85	25.43	12.09	19.00
	R_2	0.295	50.80	1.65	24.20	12.70	15.40

* Values for bile flow (ml/min) and electrolyte output (μEq/min), including taurocholate, Tau^-, were obtained as noted in Table I.

was made after 60 min of equilibration. It may be seen in Fig. 2 that hydrocortisone always increased bile flow above levels produced by taurocholate regardless of the rate of taurocholate infusion or antecedent bile flow rate.

TABLE III

Recovery of Hydrocortisone-4-^{14}C from Bile and Urine

Dog	Weight	Bile	Urine
	kg		
G	16.8	41%	27%
R	21.9	50%	37%
C	28.1	12%	26%
M	29.5	38%	31%

Hydrocortisone excretion. The output of radioactivity in urine and bile was determined in four dogs after administration of hydrocortisone-4-^{14}C (dog R, rapid single intravenous injection of 200 mg of cortisol and 1 uCi of labeled compound; dog M, intravenous infusion, 60 min, 5 μCi of labeled compound alone; dogs G and C, intravenous infusion, 60 min, 200 mg of cortisol and 5 μCi of labeled cortisol). The cumulative recoveries in urine and bile during a 4 hr period in R and during 3 hr after termination of infusion in the others are presented in Table III. It may be seen that radioactivity was largely eliminated in both urine and bile. Labeled material appeared in the bile in measurable amounts within 30 min in each study. The plasma radioactivity tended to "level off" for a period of approximately 1 hr shortly after completion of injection or infusion and declined slowly thereafter. Calculation of biliary clearances during this "equilibrium period" yielded mean values of 56.0 ml/min for dog R, 45.0 ml/min for dog M, 41.0 ml/min for dog G, and 14.0 ml/min for dog C in association with bile flows of 0.23, 0.18, 0.34, and 0.17 ml/min, respectively. It should be noted that bile flow increased significantly in all except the animal (dog M, see above) that received a tracer dose only of the steroid.

TABLE IV

BSP Transport Maximum and Relative Storage Capacity

Dog	Control						
	PV	I_{BSP}	V	P_{BSP}	Δp	T_m	S
		mg/min	*ml/min*	*mg/100 ml*	*mg/100 ml per min*	*mg/min*	*mg/mg per 100 ml*
G	715	9.02	0.35	4.27	0.092	4.16	46
C	1000	5.68	0.29	3.59	0.007	4.40	162
M	1200	9.57	0.35	5.33	0.052	5.83	60
R	1006	10.09	0.33	4.27	0.068	5.66	55
R‡	1006	8.74	0.44	3.28	0.026	5.72	105

* Abbreviations: PV = plasma volume; I_{BSP} = infusion rate of BSP; V = bile flow; $P_{BSP.}$ = plasma concentration of BSP at start of period; Δp = rate of increase in BSP plasma concentration throughout period; T_m = mean maximal biliary transport of BSP during each period; and S = relative storage capacity of BSP during each period.
‡ Dog R also received hydrocortisone 4-^{14}C; see Fig. 4.

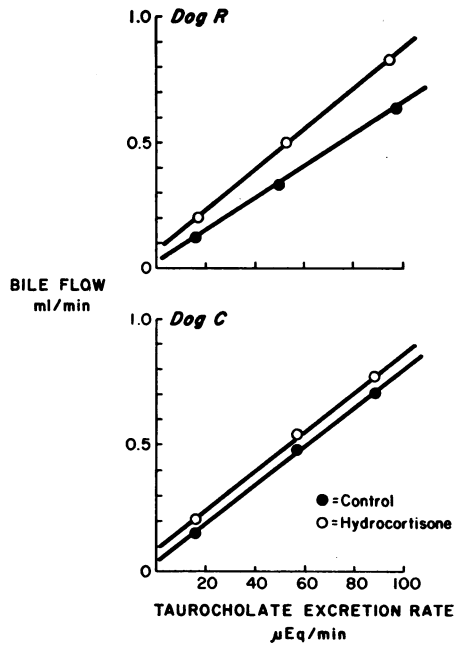

FIGURE 2 Effect of hydrocortisone on the changes in bile flow with taurocholate excretion in the dog. Taurocholate excretion was maintained at a constant value after intravenous administration of sodium taurocholate (1.3%) for 60 min at three successively different rates (10, 45, and 90 μEq/min). Mean values for bile flow at each level in two dogs (R and C) were obtained in the absence of (closed circles) and during (open circles) administration of hydrocortisone infusion. See text for further detail. Hydrocortisone choleresis was always superimposed upon that attributable to the bile salt.

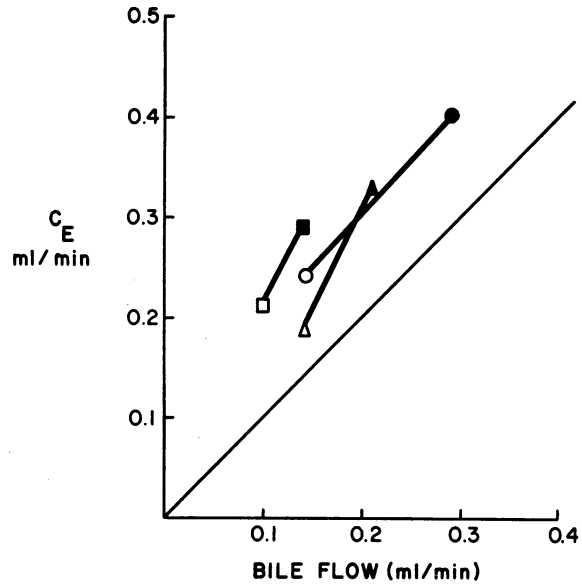

FIGURE 3 Biliary clearance (C_E) of erythritol-^{14}C from plasma before and after administration of hydrocortisone in the dog. Bile/plasma concentration ratios exceeded unity in all three dogs (open square—A, open triangle—C, and open circle—G) so that the mean clearance values all lie above the diagonal of identity. The maximal increment in bile flow after administration of hydrocortisone (closed square, triangle, and circle) was associated with roughly equivalent change in erythritol clearance.

Bile and urine samples obtained during the "equilibrium period" in two dogs (M and G) were subjected to paper chromatography and strip radioscanning to evaluate metabolic changes in hydrocortisone-4-^{14}C during the stay in the body. The radioactivity in samples of both bile and urine was found to move preponderantly as a single band, having an R_f of 0.50. In urine, a small quantity was also detectable close to the advancing front with an R_f of 0.82. Hydrocortisol-4-^{14}C hemisuccinate added to control bile was found to move in the same chromatographic and solvent system with an R_f of 0.88.

*during Control, Hydrocortisone Infusion, and Recovery**

Hydrocortisone					Recovery				
V	P_{BSP}	Δp	T_m	S	V	P_{BSP}	Δp	T_m	S
ml/min	*mg/100 ml*	*mg/100 ml per min*	*mg/min*	*mg/mg per 100 ml*	*ml/min*	*mg/100 ml*	*mg/100 ml per min*	*mg/min*	*mg/mg per 100 ml*
0.34	8.82	0.072	4.31	58	0.32	11.85	0.092	3.84	49
0.33	4.10	0.023	4.33	49	0.32	5.15	0.015	4.22	86
0.34	5.4	0.042	5.40	87	0.28	10.30	0.050	4.69	87
0.30	9.00	0.043	5.10	106					
0.46	4.07	0.026	6.11	92	0.46	5.31	0.026	6.03	61

Most, if not all, of the substance excreted in bile was, therefore, a derivative of the infused steroid. The small amount of unaltered steroid appearing in the urine was concentrated by methylene chloride extraction to permit more precise chromatographic identification. The bile acids (R_f 0.70) interfered with extraction from bile.

Erythritol excretion. To assess the effect of hydrocortisone upon the excretion of small, metabolically inert molecular species such as creatinine, urea, and mannitol, the clearance of erythritol-^{14}C was measured before, during, and after administration of the steroid in three dogs. The average clearance values during 1 hr of control measurements ranged from 0.19 to 0.24 ml/min (Fig. 3) in association with bile/plasma concentration ratios of 1.35 in dog C, 1.71 in dog G, and 2.13 in dog A. These findings conform to earlier observations made in dogs receiving taurocholate at the same infusion rate (10 μEq/min) (16). The clearances increased 33–72% during hydrocortisone choleresis in all three animals. Maximal clearances (measured over 15 min or more) ranged from 0.29 to 0.41 ml/min (Fig. 3). These changes were proportional to those in bile flow so that bile/plasma

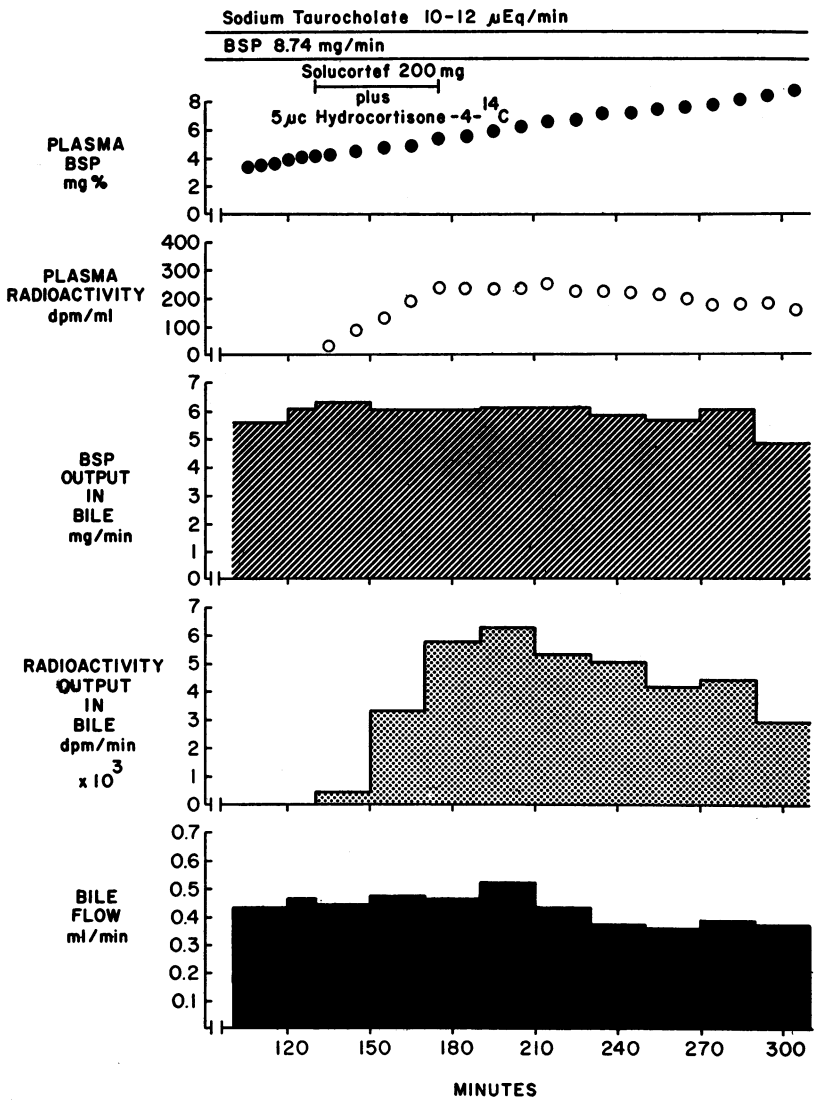

FIGURE 4 Effect of sulfobromophthalein sodium (BSP) on the choleretic response to hydrocortisone. Sodium taurocholate and BSP were infused throughout. Administration of labeled hydrocortisone was followed by the appearance of radioactivity in plasma and bile without change in bile flow or maximal BSP excretion.

TABLE V

*Composition of Maximal Increment in Bile Flow (ΔF) during Hydrocortisone Choleresis**

Dog	ΔF	Maximal increment				Composition				
		Na^+	K^+	Cl^-	HCO_3^-	Na^+	K^+	Cl^-	HCO_3^-	Osm
		μEq/min				*mEq/liter*				*mOsm/kg*
A	0.068	9.76	0.34	6.69	3.10	143	5.0	98	46	296
G	0.104	13.92	0.49	8.75	2.76	134	4.7	84	27	278
R	0.092	12.94	0.47	7.87	2.79	141	5.1	86	30	292
C	0.105	16.43	0.57	9.77	3.91	156	5.4	93	37	323
M	0.102	14.81	0.40	9.73	5.11	145	3.9	95	50	298

* Composition (mEq/liter) of fluid added to bile in hydrocortisone choleresis was computed by dividing the maximal increment in output (μEq/minute) of each electrolyte by the maximal increment in flow. Osmolality (Osm) of the increment was estimated as the sum of sodium and potassium incremental concentrations multiplied by two.

concentration ratios remained relatively constant (1.42—G, 1.55—C, 2.04—A). Although taurocholate infusion was kept constant throughout, the response was similar to that observed when taurocholate infusion was increased two- to threefold in other studies (13).

Sulfobromophthalein sodium (BSP) excretion. The biliary excretion of BSP was evaluated on five occasions before, during, and after administration of hydrocortisone infusion as a guide to the effect of the hormone upon hepatocellular function. Values for relative storage capacity (S) and maximal transfer rates (T_m) of BSP are presented in Table IV. No obviously consistent change was observed. Maximal transport decreased slightly in three experiments and increased somewhat in two. Storage capacity varied widely, increasing markedly in one experiment, decreasing in a second, and changing relatively little in three more. In one experiment (Fig. 4), hydrocortisone-4-^{14}C was added to the infusion of Solu-Cortef. The presence of BSP appeared to have no clearly demonstrable effect upon biliary clearance (29.0 ml/min) of the isotope. It is noteworthy, however, that the choleresis associated with maximal excretion of BSP was not further augmented, as expected, during administration of hydrocortisone (Table IV and Fig. 4).

DISCUSSION

These experiments have clearly defined a major choleretic action of hydrocortsione in the dog. Although the administered dose of glucocorticoids was high relative to body weight (equivalent to about 10 mg of hydrocortisone per kg) choleresis began when considerably less than half of the total dose had been infused. In two experiments a similar choleretic response was observed with a total dose of only 50 mg of hydrocortisone by infusion over 1 hr. A response does seem to occur, therefore, at dosage levels which are not greatly in excess of maximal rates of cortisol synthesis reported (18) for the dog. The data are insufficient, however, to warrant the conclusion that adrenal steroids exert an influence upon hepatobiliary function under normal physiologic conditions.

According to the current view (5), bile is secreted by the hepatic cells into the canaliculi by a mechanism dependent, in the main, upon bile salt excretion. A second moiety of fluid and electrolyte is believed to be added by the ducts and ductules under the influence of secretin (13). Additional support for this hypothesis has emerged from recent studies (16, 19) of the biliary clearances of a variety of metabolically inert, lipid insoluble, and high diffusible solutes of relatively small molecular dimensions (including urea, creatinine, mannitol, and erythritol). All appear to be distributed rapidly and equally between the plasma and the hepatic cells, all are cleared at approximately the same rate, and all are correlated linearly with taurocholate excretion. In contrast, secretin choleresis does not affect inert solute clearance in keeping with the inference that the bile salts control canalicular flow, and biliary volume may be modified by fluid transfer more distally.

The maximal increment in bile formed after administration of hydrocortisone resembled that produced in response to the additional excretion of 10–12 μEq of sodium taurocholate per min (Table V). Its composition, computed by dividing the increment in output of each constituent by the increment in flow, was similar to that of plasma water except for a somewhat high bicarbonate concentration. Since studies with isotopically labeled hydrocortisone failed to show any cleavage of the steroid nucleus, only the cationic free or conjugated forms appear to have been present. Consequently, biliary output of the hormone appeared to amount to no more than 1–2 μEq/min. This excretion of hormone did not change even when the expected increase in bile flow

was eliminated during maximal excretion of BSP. In view of this fact and the absence of additional taurocholate excretion, the bile flow increment was probably not the result of an osmotic gradient imposed at the canalicular level by either hydrocortisone or bile salt per se.

It is possible that the changes observed in biliary volume and composition could have been induced by a secondary rise of endogenous secretin levels. In line with this possibility, hydrocortisone is known (20) to induce gastric acid secretion, a potent stimulus to secretin release. With secretin, however, a much more alkaline and more concentrated maximal increment would have been expected (13). Moreover, the erythritol clearance increased with bile flow during the action of hydrocortisone resembling, in this respect, the response to taurocholate rather than the response to secretin (16, 19). The alteration in erythritol clearance strongly suggests that hydrocortisone acts at the hepatocellular level. Although hepatocellular transport and accumulation of BSP were not affected by hydrocortisone in these experiments, maximal BSP excretion did apepar to prevent hydrocortisone choleresis independently of steroid clearance (Fig. 4). These findings are consistent with a mechanism of canalicular choleresis which is independent of the cellular transport of BSP or hydrocortisone as such.

Although canalicular bile formation may depend importantly upon active transport of bile acids and other organic anions, the failure of changes in bile acid output (BV) to affect canine bile flow at very low absolute levels of BV (21) and the positive intercept for mannitol clearance (Cm) when Cm is plotted against BV in the dog (16) indicate that other mechanisms may also play a primary role. The present study suggests that such a mechanism is activated by hydrocortisone. The data are consistent with an increase in bile flow as the result of the addition of an iso-osmotic ultrafiltrate of plasma, possibly secondary to a change in the permeability of an intercellular barrier between plasma and bile. The choleretic responses to hydrocortisone and taurocholate appear to be independent and additive (Fig. 2) and may, therefore, depend upon different canalicular mechanisms. Furthermore, the observed interaction between BSP and hydrocortisone, but not between either and taurocholate, indicates that certain organic anions may not share in toto the secretory pathway for taurocholate, a conclusion in harmony with recent work by Alpert, Mosher, Shanske, and Arias (22) on Corriedale sheep in which BSP transport is impaired independently of bile acid transfer. More work is necessary to define the extent to which biliary hydrocortisone secretion and its choleretic effect may be generally representative of biliary excretion of other organic cations or anions (23). Such a mechanism of taurocholate-independent choleresis may well be involved in producing inexplicable variations in basal bile flow under a wide variety of conditions and in the beneficial response to steroid therapy sometimes observed (24) in patients with intrahepatic cholestasis.

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