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Excretion of nitrofurantoin in dog hepatic bile

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Summary

1. After the intravenous administration of nitrofurantoin sodium to dogs at nitrofurantoin doses of 1·5–24·0 mg/kg, a substantial amount of nitrofurantoin is excreted in bile. The bile to blood drug ratios were about 200. A marked hydrocholeretic effect which correlated directly with the amount of nitrofurantoin administered was also observed.

2. The excretion of nitrofurantoin in bile and the hydrocholeretic effect were linear with the dose of drug over the range 1·5–12·0 mg/kg. Maximum increases in hepatic bile flows were usually from 5–10 ml/0·5 h, while average control bile flow was 1·6 ml ± S.D. 0·6/0·5 hours. The lowest dose at which the hydrocholeretic effect was still detectable was 0·09 mg/kg.

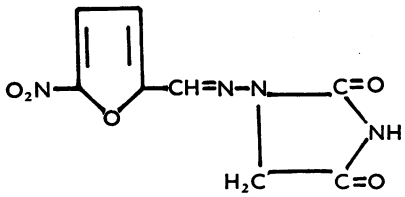
3. Apparent saturation of the biliary excretion system for nitrofurantoin and the hydrocholeretic mechanism occurred after a dose of 24·0 mg/kg. Saturation of the urinary system for nitrofurantoin excretion was noted after a dose of 6·0 mg/kg.

4. Biliary nitrofurantoin recoveries ranged from 16·5% ± S.D. 4·2 to 22·6% ± S.D. 4·7 for the 6 h period after doses of 1·5, 3·0, and 6·0 mg/kg. Urinary nitrofurantoin recoveries for the same interval ranged from 24·1% ± S.D. 6·6 to 36·2% ± S.D. 8·3.

5. In comparison to values obtained in normal dogs, only about one-tenth of the drug excretion in bile and about one-fifth of the hydrocholeretic effect were obtained after intravenous drug administration to dogs with hepatic impairment induced by CCl_4.

Introduction

The clinical effectiveness of nitrofurantoin, 1-(5-nitrofurfurylideneamino) hydantoin, in urinary tract infections in man and animals is well established (Carroll, 1953 ; Paul & Paul, 1964 ; Paul & Paul, 1966). This drug is readily absorbed and quickly distributed into most body fluids, and renal excretion of unchanged nitrofurantoin and enzymatic degradation are the principal mechanisms of elimination (Paul & Paul, 1964 ; Paul & Paul, 1966). The liver is a site for nitrofurantoin degradation (Paul, Paul, Bender, Kopko, Harrington, Ells & Buzard, 1960 ; Buzard, Conklin, O'Keefe & Paul, 1961), but little information is available concerning its biliary excretion. Results are now presented regarding the excretion of nitrofurantoin in dog hepatic bile. Some of these results were presented at the FASED Meeting, Atlantic City, New Jersey, April 1970.



1-(5-Nitrofurfurylideneamino)hydantoin

Methods

Collection of specimens

Adult male Beagles (10–16 kg), maintained under conditions approved by the American Association for Accreditation of Laboratory Animal Care, were anaesthetized with aqueous pentobarbitone sodium (35 mg/kg i.v.). Both femoral veins were isolated; one for the collection of blood and the other for hydration and for the administration of nitrofurantoin sodium (Furadantin Sodium, Eaton Laboratories) and supplemental doses of the anaesthetic. After the cystic duct was ligated at the common bile duct and the gall bladder emptied, the common bile duct was cannulated with a 16 gauge gavage needle attached to polyethylene tubing (PE-200), about 4–6 cm from the junction of the common bile duct with the cystic duct and main hepatic duct. Each ureter was cannulated (PE-90) about 6–8 cm from the hilus of the kidney.

Blood, bile, and urine were always collected before drug administration. Nitrofurantoin sodium dissolved in 5% dextrose was administered as a single intravenous injection (0·04–24·0 mg/kg). A nitrofurantoin sodium solution in 5% dextrose was also administered intravenously ((1·5 mg/kg)/h) by infusion (Harvard Apparatus) at a constant rate of 0·764 ml/minute. Drug solutions were prepared (commercially available vials) by dissolving the drug in 15 ml of 5% dextrose to obtain a nitrofurantoin concentration of 12 mg/ml (pH 9·0). All drug doses are presented as mg/kg of nitrofurantoin.

After injection of the drug, bile and pooled urine from both kidneys were collected at 0·5 h intervals, usually for a period of 6 h, commencing immediately after a 1 h control period, and the volumes recorded. Blood samples were obtained in heparinized syringes. Hydration was maintained by an intravenous infusion (Harvard Apparatus) of 5% dextrose at a constant rate of 0·764 ml/min throughout each evaluation, including the control period.

To produce hepatic impairment, a mixture of carbon tetrachloride and vegetable oil (1:1) was administered orally at 0·25 ml/kg, once daily to dogs for 2 days. On the third day, the mixture was administered at 8.00 a.m. and again at 4.00 p.m. and the animals then prepared on the fourth day for the collection of hepatic bile and ureteral urine. A solution of nitrofurantoin sodium was administered intravenously (3·0 mg/kg) and bile and urine collected for 3 hours. At 3·5 h, the drug was administered again and the fluids collected for another 3 hours. Blood samples were also collected at selected intervals.

Drug analysis

Urine (1·0 ml) was analysed for nitrofurantoin by the nitromethane-hyamine procedure (Conklin & Hollifield, 1965) and blood (0·5 ml) by the procedure of Conklin & Hollifield (1966).

Nitrofurantoin in bile (0·5 ml) was determined as follows. Bile (0·5 ml) and 1·0 ml of water were mixed in a test tube, followed by the addition of 2·5 ml of a saturated ammonium sulphate solution (pH 6·0). The contents were mixed, and 8·0 ml of practical grade nitromethane were added. These contents were mixed vigorously for 1 min and centrifuged (2,000 r.p.m.) for 10 minutes. Then, 6·0 ml of the nitromethane (top layer) were removed and transferred to another test tube containing 40 mg of an aluminium hydroxide adsorbent (Seramox, Warner—Chilcott Laboratories Division). The contents were mixed for 1 min and centrifuged for 10 min at 2,000 r.p.m. After 4·0 ml of the solvent were transferred to another tube, 0·5 ml of 0·04 M hyamine hydroxide in methanol (Packard Instrument Co., Inc.) was added. The contents were mixed and allowed to stand for at least 1 minute. The concentration of the nitrofurantoin-hyamine complex was then determined spectrophotometrically at 400 nm. Nitromethane subjected to the procedure with either water or bile served as a control. Pure nitromethane was used to set the instrument to zero balance. The absorbance value of all samples was determined within 5 min after the addition of the hyamine solution.

The total bile solids were determined gravimetrically, after 0·5 ml of bile was transferred to a 5 ml tared beaker, together with 2 ml of washings and taken to dryness in an oven 120° C for 16 hours.

Bilirubin determinations were made according to the Technicon method, N-12a.

Analysis of nitrofurantoin in bile

A standard curve for nitrofurantoin as determined by the described procedure is linear to 100 μg. Spectrophotometric absorbances obtained at 400 nm when the drug was recovered from water standards were about 10% lower than those representing drug recovered from either dog gall bladder (107·0% $\pm$ S.D. 3·3) or hepatic bile (109·9% $\pm$ S.D. 4·5) over the range 5–100 μg. Therefore, a bile reference standard curve was used to calculate biliary concentrations of nitrofurantoin. The lowest drug concentration detectable with accuracy by this method is 10 μg/ml.

Bile samples collected from dogs after intravenous administration of nitrofurantoin sodium and nitrofurantoin standards were examined by ascending paper chromatography and co-chromatography. The samples were subjected to the described analytical procedure and the nitromethane extracts spotted on paper and treated according to procedures reported previously (Conklin & Hollifield, 1965). The spot representing nitrofurantoin in bile travelled further under acid conditions (R_F 0·46) than under base conditions (R_F 0·16). These R_F values are in excellent agreement with those reported elsewhere for the drug in plasma, blood, and urine (Buzard & Conklin, 1965; Conklin & Hollifield, 1965; Conklin & Hollifield, 1966). Similar R_F values were obtained with the extracts of bile samples from dogs given nitrofurantoin and with the extracts of control bile samples to which the drug was added. Nitromethane elution of the spots representing nitrofurantoin, followed by the addition of the hyamine solution, gave an absorbance spectrum characteristic of the nitrofurantoin-hyamine complex. After a chromatographic comparison with control bile samples, no evidence was found indicating the presence of metabolites related to nitrofurantoin in the nitromethane extracts.

Bile samples collected from dogs given nitrofurantoin were analysed by the described procedure. Subsequently, known amounts of nitrofurantoin were added

to duplicate samples and the samples were again analysed by the same procedure. The analyses yielded the anticipated increase in drug concentration with a mean of 96·1% ±S.D. 3·8 of the added material being recovered after the addition of the known amounts of nitrofurantoin which ranged from 10 to 100 μg. The nitromethane extracts from this experiment were also subjected to paper chromatography using both the acid and base systems to verify co-chromatographically (Hollifield & Conklin, 1968) the presence and position of the nitrofurantoin. Agreement was obtained between the chromatography data acquired before and after the addition of nitrofurantoin to the bile samples.

Results

Excretion of nitrofurantoin in bile

Nitrofurantoin rapidly appeared in bile in concentrations greatly exceeding those found in blood within 30 min of the intravenous administration of nitrofurantoin sodium (Table 1). The analysis of samples collected at 5 min intervals established that the greatest concentration of nitrofurantoin in blood occurred within 5 min while concentrations of nitrofurantoin in bile reached a maximum within 10–15 minutes. Bile to blood drug concentration ratios of about 200 were obtained during this 30 min interval. When the drug dose was increased 4-fold from 3·0 to 12·0 mg/kg, the concentration of drug in the blood at either 30 or 60 min increased about 6-fold while the amount of nitrofurantoin excreted in bile within 60 min increased only about 2-fold, suggesting that the biliary excretion system for nitrofurantoin can be saturated (Table 2).

Half-lives of the drug in the blood increasing from 10 to 26 min were obtained over the dose range 1·5–24·0 mg/kg. The drug-dose relationship observed in bile for the 6 h period after dosage is linear from 1·5 to 12·0 mg/kg (Table 3). Apparent saturation of the system for nitrofurantoin excretion in bile for the 6 h

TABLE 1. *Initial biliary excretion of nitrofurantoin in dogs after an intravenous dose of nitrofurantoin sodium*

Time after dose (min)	Nitrofurantoin conc. (μg/ml)* Blood	Bile	Bile to blood conc. ratio*
0–5	6·4±1·2	10·7± 1·4	1·7± 0·2
5–10	4·8±0·8	543·5±121·4	112·7± 6·5
10–15	3·2±0·5	697·1± 92·3	223·7±54·8
15–20	2·9±0·4	597·5± 64·2	207·1±42·8
20–25	2·3±0·3	511·5± 62·9	226·4±37·5
25–30	2·1±0·3	439·5± 39·7	207·8±24·7

Conditions: nitrofurantoin sodium as a solution in 5% dextrose (pH 9·0) was administered as a single intravenous injection at a nitrofurantoin dose of 3·0 mg/kg. Bile was collected for 5 min intervals and blood obtained at the end of each collection interval. * Average value ± standard deviation, based on three dogs.

TABLE 2. *Effect of intravenous doses of nitrofurantoin sodium on the initial biliary excretion of nitrofurantoin in dogs*

Nitrofurantoin dose (mg/kg)	Nitrofurantoin conc. (μg/ml)* Blood (60 min)	Bile (0–60 min)	Dose excreted (%)* Bile (0–60 min)
3·0	1·0±0·2	554·5±62·6	21·1±2·1
6·0	1·8±0·5	619·2±56·6	15·2±1·1
12·0	6·4±1·2	720·0±30·4	8·8±1·2

Conditions: nitrofurantoin sodium as a solution in 5% dextrose (pH 9·0) was administered as a single intravenous injection. Bile was collected for a 60 min interval and blood obtained at the end of the interval. * Average value ± standard deviation, based on three dogs/dose.

period was observed at 24·0 mg/kg. In contrast, apparent saturation of the urinary system for nitrofurantoin excretion (0 to 6 h) occurred after a dose of 6·0 mg/kg, with marked inhibition noted after either 12·0 or 24·0 mg/kg.

A large amount of nitrofurantoin is excreted in urine after either oral or parenteral drug dosage (Conklin & Hailey, 1969; Conklin, Sobers & Wagner, 1969). In our study, urine drug recoveries ranging from 24·1% ± S.D. 6·6 to 36·2% ± S.D. 8·3 of the dose were found within 6 h after doses of 1·5, 3·0, and 6·0 mg/kg. Bile drug recoveries within the same 6 h period ranged from 16·5% ± S.D. 4·2 to 22·6% ± S.D. 4·7 of the dose. Combined bile and urine nitrofurantoin recoveries after these doses account for about 50% of the dose administered, in the absence of enterohepatic recycling (Table 3).

Relatively steady concentrations of drug in the blood of 1·5 μg/ml ± S.D. 0·1 and 2·0 μg/ml ± S.D. 0·1 were attained within 1 h in two dogs by a constant intravenous infusion of nitrofurantoin sodium ((1·5 mg/kg)/h). These equilibrium concentrations of drug in the blood established an almost constant rate of drug excretion in bile (1·6 mg ± S.D. 0·1/0·5 h, 3·5 mg ± S.D. 0·4/0·5 h) while maintaining a relatively constant state of choleresis (3·5 ml ± S.D. 0·1/0·5 h, 4·9 ml ± S.D. 0·2/0·5 h), respectively, in each of the animals. The ratios of the concentration of drug in bile to those in blood were 241 ± S.D. 17 and 263 ± S.D. 39, respectively. The averages presented for each animal are based on values determined at 0·5 h intervals during the equilibrium period (1–3 h). These results show that nitrofurantoin transfer into bile takes place against a concentration gradient under the described conditions of drug equilibrium.

Action of nitrofurantoin on bile flow

Immediately after the intravenous administration of nitrofurantoin sodium, bile flow increased in direct proportion to the amount of drug administered. To obtain a reasonable estimate of this choleretic effect, all of the values representing it have been adjusted, using the bile flow in each animal for the 0·5 h interval just before drug administration as a control value.

A maximum choleretic effect ranging from about 5 to 10 ml for the first 0·5 h after dosage was encountered over a dose range of 1·5–24·0 mg/kg, in comparison to an average control bile flow of 1·6 ml ± S.D. 0·6/0·5 h (Table 4). As shown in Table 4, the choleretic effect was still detectable after a dose as low as 0·09 mg/kg but was not detectable at 0·04 mg/kg. Onset of maximum bile flow was related directly to drug dose. It occurred within 20 min after doses at 1·5 and 3·0 mg/kg but not until 90 min after the 24·0 mg/kg dose. A return to a relatively constant

TABLE 3. *Biliary and urinary excretion of nitrofurantoin in dogs after intravenous doses of nitrofurantoin sodium*

Nitrofurantoin dose (mg/kg)	Hepatic bile 0–6 h		Ureteral urine 0–6 h	
	Total drug* (mg)	Dose recovered* (%)	Total drug* (mg)	Dose recovered* (%)
1·5	3·3 ± 1·2	16·5 ± 4·2	5·1 ± 1·0	25·4 ± 3·0
3·0	9·0 ± 1·4	22·6 ± 4·7	14·4 ± 3·0	36·2 ± 8·3
6·0	17·0 ± 1·9	20·0 ± 1·8	20·1 ± 3·4	24·1 ± 6·6
12·0	31·0 ± 7·6	18·3 ± 5·5	17·3 ± 9·0	10·0 ± 4·9
24·0	51·4 ± 15·7	15·3 ± 5·2	10·9 ± 8·3	3·1 ± 2·1

Conditions: nitrofurantoin sodium as a solution in 5% dextrose (pH 9·0) was administered as a single intravenous injection. * Average value ± standard deviation, based on at least five dogs/dose.

rate of flow required about 3 h after doses at 1·5, 3·0, and 6·0 mg/kg. This return was not evident at 6 h after the dose of 24·0 mg/kg. The choleretic response observed for the 6 h period after dosage was linear from 1·5 to 12·0 mg/kg, with apparent saturation of the mechanism at 24·0 mg/kg.

Two dogs were prepared as described earlier for the collection of bile and urine and a 5% dextrose solution was infused intravenously for 7 h at 0·764 ml/minute. One hour was allowed for a stable bile flow to be attained. Only minor fluctuations in bile flow were evident during the succeeding 6 hours. The average bile flow for this interval was 1·8 ml ± S.D. 0·1/0·5 h and 1·4 ml ± S.D. 0·3/0·5 h, respectively, in the two animals. In comparison, the average control bile flow obtained in the dogs used in this study was 1·7 ml ± S.D. 0·6 for the 0·5 h interval just before drug was administered. Additionally, no change in bile flow was observed in dogs within 1 h of the intravenous injection of 5% dextrose solutions with a pH or volume similar to that of the drug solutions used in the study.

As reported by Cook, Bianchi, Hambourger & Green (1950), control hepatic bile flows ranging from (0·12 to 0·33 ml/kg)/h were obtained in anaesthetized dogs prepared surgically in a manner similar to that described in this report. In agreement with this, an average control bile flow of (0·25 ml/kg)/h ± S.D. 0·08 was obtained in the dogs used in this study.

Six dogs were prepared as described earlier and hepatic bile and ureteral urine were collected at 0·5 h intervals for 6 h after a 1 h interval to stabilize bile flow. A solution of 5% dextrose was infused intravenously at a constant rate of 0·764 ml/min during the 7 h period. Bile flow was relatively constant from 0 to 3 h and then decreased slightly during the next 3 hours. The average bile flow per 0·5 h in 6 dogs was 1·9 ml ± S.D. 0·1 from 0 to 3 h and 1·5 ml ± S.D. 0·1 from 3 to 6 hours. On the basis of results from three dogs, the concentration of bile solids and the total amount of bile solids excreted per 0·5 h interval in each animal decreased only slightly from 0 to 3 hours. During the succeeding 3 h the amount of bile solids gradually declined, while the concentration of bile solids remained essentially constant.

After administration of nitrofurantoin, a decrease below the control value was noted in the concentration of bile solids, with a trend for the concentration to return to its control value with time. The amount of total solids increased transiently during the first 0·5 h after dosage, then promptly and abruptly decreased during the next 0·5 hour. From 1 to 3 h, the bile solids excreted continued to decrease while, during the remainder of the 6 h period (3–6 h), there was a more

TABLE 4. *Hepatic bile flow in dogs after intravenous doses of nitrofurantoin sodium*

Nitrofurantoin dose (mg/kg)	Control flow before dose (ml/0·5 h)*	Choleretic effect after dose (ml/0·5 h)*	Nitrofurantoin dose (mg/kg)	Control flow before dose (ml/0·5 h)†	Choleretic effect after dose (ml/0·5 h)†
0·04	2·0±0·1	Not detectable	1·5	2·1±0·9	4·6±1·1
0·09	1·9±0·9	0·6±0·1	3·0	1·6±0·5	6·8±1·5
0·18	1·1±0·2	1·3±0·2	6·0	2·0±1·1	9·7±1·1
0·37	1·4±0·7	1·7±0·2	12·0	1·4±0·4	10·1±1·5
0·75	1·5±0·4	2·9±0·2	24·0	1·5±0·7	9·6±3·0

Conditions: nitrofurantoin sodium as a solution in 5% dextrose (pH 9·0) was administered as a single intravenous injection. * Average value ± standard deviation, based on at least three dogs/dose. Choleretic effect for the first 0·5 h after dose, adjusted for the bile flow obtained for the 0·5 h just before drug dosage. † Average value ± standard deviation, based on at least five dogs/dose. Choleretic effect for the first 0·5 h after dose, adjusted for the bile flow obtained for the 0·5 h just before drug dosage.

gradual decrease. Typical patterns of total bile flow and total solids excretion in untreated dogs and in dogs given nitrofurantoin at 3·0 mg/kg are illustrated in Fig. 1.

At doses of 1·5–6·0 mg/kg, at least 50% of the nitrofurantoin recovered in bile is excreted within the first 0·5 h after dosage. In agreement with this, about 45% of the total choleretic effect also occurs within the same interval. The transitory rise noted in the amount of bile solids excreted may be partially explained by the flushing out of the dead space in the bile duct system produced by the rapid excretion of nitrofurantoin. A similar effect on excretion of bile pigment was previously observed by Cantarow, Wirts, Snape & Miller (1948). Since the

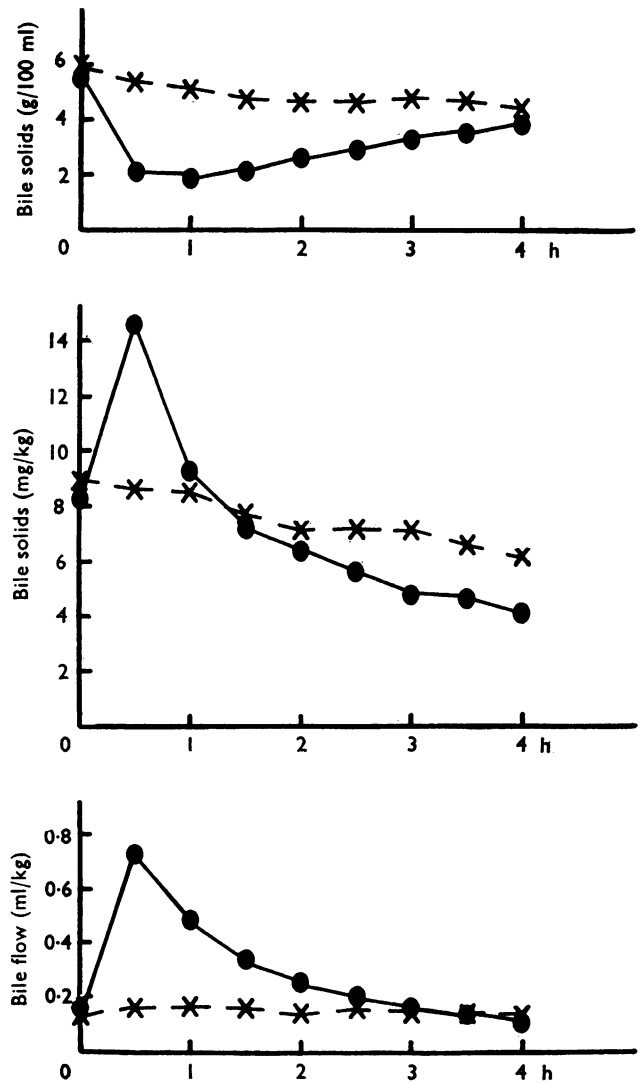


FIG. 1. Patterns of total bile flow and total solids excretion in untreated dogs and in dogs given an intravenous injection of a nitrofurantoin sodium solution at a nitrofurantoin dose of 3·0 mg/kg. Results are based on an average from at least three dogs. ×, Untreated dogs; ●, treated dogs.

weight of excreted nitrofurantoin represented only about 2–3% of the bile solids total measured within the first 0·5 h after these drug doses, its contribution was considered negligible. The gradual deterioration in bile flow and excretion of total solids noted in both untreated and treated dogs during the latter portion of the experimental interval (3–6 h) is probably associated with the interruption of the usual cycle of bile excretion into the duodenum by the described surgical procedure. Therefore, the marked changes in bile flow and total solids excretion observed within the first 3 h of drug administration are attributed essentially to nitrofurantoin.

As shown in Table 5, the concentration of bile solids decreased noticeably in comparison to control values during the 3 h interval after the administration of nitrofurantoin at doses of 1·5, 3·0, and 6·0 mg/kg. Also apparent in the treated dogs is a marked increase in bile flow and the absence of a significant net increase in the output of total solids, in comparison to control values. A small decrease in the excretion of total solids between the control and treated intervals is also evident in the untreated dogs. However, even if this decrease is used as a control for the treated dogs, the small increase noted in total solids indicates that nitrofurantoin apparently produces only a small choleretic effect. These results are interpreted as indicating that the action of nitrofurantoin on bile flow is predominantly a hydrocholeretic effect. A comparison of the total weight of nitrofurantoin excreted in bile and its related hydrocholeretic effect revealed that for every milligramme of nitrofurantoin excreted, there was an increase in bile flow of approximately 1·6 ml.

TABLE 5. *Excretion of total bile solids in dogs following intravenous administration of nitrofurantoin sodium*

Nitrofurantoin dose	Hepatic bile: Total volume* ((ml/kg)/h)		Hepatic bile: Total bile solids* ((mg/kg)/h)		Hepatic bile: Total bile solids* (g/100 ml)	
	Control	Treated	Control	Treated	Control	Treated
1·5 mg/kg	0·35±0·08	0·60±0·04	15·3±4·9	15·1±4·7	4·20±0·78	2·46±0·69
3·0 mg/kg	0·30±0·08	0·72±0·09	16·1±4·3	15·9±3·6	5·75±3·23	2·22±0·69
6·0 mg/kg	0·29±0·05	0·98±0·14	18·9±1·4	19·7±2·6	6·75±1·91	1·98±0·18
Untreated†	0·28±0·03	0·30±0·05	17·8±4·1	15·5±3·8	6·08±0·70	4·95±0·40

Conditions: nitrofurantoin sodium as a solution in 5% dextrose (pH 9·0) was administered as a single intravenous injection. * Average value ± standard deviation, based on three dogs/dose. Control—based on values obtained for the 0·5 h just before drug dosage. Treated—based on average values obtained for the 3 h just after drug dosage or values obtained for the same interval in untreated dogs
† Nitrofurantoin was not administered.

TABLE 6. *Biliary excretion of nitrofurantoin and its hydrocholeretic effect in a dog with hepatic dysfunction and in normal dogs*

Experiment	Hydrocholeretic effect (ml)*	Hepatic bile: Total drug (mg)	Hepatic bile: Dose recovered (%)
Hepatic dysfunction†			
First drug dose (0–3 h)	3·3	0·6	1·3
Second drug dose (3·5–6·5 h)	2·6	0·5	1·2
Normals+ (0–3 h)	14·2±2·8	8·8±1·4	22·3±4·7

Conditions: nitrofurantoin sodium as a solution in 5% dextrose (pH 9·0) was administered at 0 and 3·5 h as a single intravenous injection at a nitrofurantoin dose of 3·0 mg/kg. * Adjusted for the bile flow obtained for the 0·5 h just before drug dosage, 0·9 ml in hepatic dysfunction dog and 1·6 ml±S.D. 0·5 in normals, based on five dogs. † Dog pretreated orally with carbon tetrachloride and vegetable oil. + Average value ± standard deviation, based on at least five dogs.

Effect of hepatic dysfunction

The administration of certain organic solvents results in some degree of hepatic dysfunction (Klaassen & Plaa, 1967; Van Vleet & Alberts, 1968). In particular, evidence has been reported showing that the administration of carbon tetrachloride (CCl_4) impairs hepatic excretory function (Priestly & Plaa, 1970). Nitrofurantoin sodium was administered intravenously (3·0 mg/kg) to a dog pretreated orally with CCl_4 as described earlier. The results in Table 6 show that only about 1% of the dose was recovered in the bile (0–3 h) of the dog pretreated with CCl_4, compared with a corresponding dose recovery of about 22% in normal dogs. In addition, there was only about a 3-fold increase in the hydrocholeretic effect in this dog compared to a corresponding increase of about 9-fold in normal dogs. Urine drug recoveries from this dog agreed with those found in normal dogs (0–3 h) at this dose.

In a similar experiment involving CCl_4, a bile dose recovery of only about 3% and about a 4-fold increase in hydrocholeretic effect were obtained within 3 h (3·0 mg/kg). Increased concentrations of bilirubin in plasma in this animal provided evidence for hepatic dysfunction. The pattern observed in total bile solids excretion after drug administration was very similar to that observed in normal dogs after the administration of nitrofurantoin. Control bile flows in the two dogs pretreated with CCl_4 were approximately half of the average control bile flow observed in normal dogs. These results suggest that, the system(s) for the biliary excretion of nitrofurantoin and its hydrocholeretic effect in the dog, were at least partially impaired under the described conditions.

Discussion

In comparison to the abundant information available on the renal excretion of many substances, studies concerning hepatic excretion have often been avoided for practical reasons. While the completion of such a study can be most informative, its neglect sometimes can lead to inaccurate conclusions regarding drug elimination and metabolism. The previous knowledge that a large amount of nitrofurantoin is excreted in urine was extremely useful in understanding some of the absorption and elimination aspects of this drug. It is equally significant to learn from the results in this report that a substantial amount of nitrofurantoin is also excreted in bile. In the dog, at least half of an intravenous dose of nitrofurantoin is recovered as parent drug from these two excretion routes within a short time after drug dosage. Since enzymatic degradation of nitrofurantoin by mammalian tissues has also been reported (Paul *et al.*, 1960; Buzard *et al.*, 1961), a large portion of the drug dose administered is accounted for. These results, when coupled with the relatively low concentrations of drug in the blood usually obtained with nitrofurantoin and its short half-life in blood, explain at least partially why significant biological drug residues have not been encountered with nitrofurantoin under clinical conditions (Conklin & Hailey, 1969; Conklin *et al.*, 1969).

Brauer (1959) divided substances which are excreted in bile into three groups on the basis of their bile to blood drug concentration ratios. According to Brauer (1959) and other recent reviewers (Smith, 1966; Stowe & Plaa, 1968), substances with bile to blood ratios of 10–1,000 usually require some active secretory process for their transport from blood to bile. Characteristically these substances compete

for transport, and their transport mechanism can be saturated by excess compound (Smith, 1966; Stowe & Plaa, 1968). Nitrofurantoin appears in dog bile at concentrations greatly exceeding those concurrently found in blood, showing that the transfer from blood to bile takes place against a concentration gradient. High doses of nitrofurantoin saturate the biliary excretion system for this drug. These results suggest that the transfer of nitrofurantoin from blood into bile cannot be completely explained by the process of simple diffusion.

Sperber's view (1959) indicates that there are similarities between the transport mechanisms of the liver and kidney, in that certain organic acids are efficiently secreted renally and hepatically. However, it was also indicated that some compounds which are efficiently secreted by the one mechanism are transferred inefficiently or not at all by the other. Nitrofurantoin is a weak organic acid with a pKa of 7·2 (Paul & Paul, 1964). Buzard, Bender, Nohle, Humphrey & Paul (1962) established that nitrofurantoin is subjected to both tubular secretion and reabsorption in the dog kidney, with evidence that the renal secretion transport system involved is common to the secretion of other weak acids. In particular, selective inhibition of para-aminohippuric acid secretion occurred after nitrofurantoin dosage (Buzard *et al.*, 1962). The results presented in this report suggest that nitrofurantoin is transported into dog bile by an active secretory process. Cook, Lawler, Calvin & Green (1952) reported that para-aminohippuric acid also is actively secreted into dog bile. It is therefore concluded that nitrofurantoin belongs to a select group of compounds which are efficiently secreted by a weak acid transport system in both the liver and kidney.

Although claims have been made that numerous drugs evoke a hydrocholeresis, only a few drugs have been ultimately classified as authentic hydrocholeretics according to the criteria of Wheeler (1968). The observation that nitrofurantoin exhibits a hydrocholeretic effect reveals a new pharmacological aspect, which has not been reported for this series of drugs.

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