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Research Papers

Toxicity and tissue distribution studies on the hydrochloride, bismuth iodide complex and a resinate of emetine

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The hydrochloride, the bismuth iodide complex and a resinate of emetine have been compared on mice, cats and dogs for toxicity, on cats for emetic activity and on rats for distribution of emetine in the tissues. Emetine hydrochloride, injected subcutaneously, was as toxic as emetine hydrochloride or emetine bismuth iodide administered orally, but only one-sixth as emetic. Given orally emetine resinate was considerably less toxic and emetic than the other preparations. The patterns of emetine distribution in the tissues of rats were similar for all the preparations, but the tissue concentrations were much lower with emetine resinate. In infected rats the resinate was as amoebicidal as the hydrochloride or the bismuth iodide complex. It is suggested that emetine resinate might be better than emetine bismuth iodide for the treatment of intestinal amoebiasis.

EMETINE is potentially very toxic (Klatskin & Friedman, 1948), yet it has retained its pre-eminence in the treatment of amoebic infection despite the challenge of several new amoebicides (Woodruff, Bell & Schofield, 1956; Woodruff, 1959; Adams, 1960; Bell & Woodruff, 1960; Carneri, Coppi, Almirante & Logemann, 1960; Dooner, 1960; Wilmot, Powell, McLeod & Elsdon-Dew, 1962).

Professor W. H. Linnell suggested to us that a study of emetine resinates might be rewarding. Several resinates were made, and one was compared with emetine hydrochloride and the bismuth iodide complex.

Materials and methods

Emetine hydrochloride B.P. (71% emetine base*). An aqueous solution of the hydrochloride was used for subcutaneous administration to mice, rats, cats and dogs, and a solution in 0·5% sodium carboxymethylcellulose for oral administration to mice and rats. For oral administration to cats and dogs, the hydrochloride as powder was presented in hard gelatin capsules.

Emetine bismuth iodide B.P. (28% emetine base*).

Emetine resinate. Emetine adsorbed from aqueous solution on a polystyrene sulphonic acid ion exchange resin (Permutit Zeo Karb 225, SRC 16) gave a resinate containing 23% emetine base† after drying.

Both emetine bismuth iodide and resinate were presented as suspensions in 0·5% sodium carboxymethylcellulose for oral administration to mice and rats and as powders in hard gelatin capsules for oral dosing to cats and dogs. The bismuth iodide was administered to dogs also as enteric coated tablets.

From Glaxo Research Ltd., Greenford, Middlesex

* Determined by Kjeldahl nitrogen and B.P. assay for emetine.
† Determined by Kjeldahl nitrogen.

TOXICITY

Acute systemic toxicity in mice. Groups of 10 female mice (A2G strain, body weights 18–22 g) were dosed subcutaneously with the hydrochloride or orally with all three preparations. Group mortalities were recorded 14 days later and the LD50 values were calculated by the method of de Beer (1945).

Subacute toxicity in dogs. Mongrel dogs of either sex were dosed according to the schedules in Table 2. When possible, dosing was continued for 56 days, but some of the animals died earlier or became so ill that they had to be destroyed.

ACUTE EMETIC ACTIVITY IN CATS

Adult cats of either sex which had been fasted overnight received single subcutaneous doses of the hydrochloride or single oral doses of all three preparations, and a positive score was recorded for each animal that vomited within 24 hr. The dose (mg base/kg) that should produce emesis in 50% of the cats receiving it (ED50) was calculated for each preparation. No animal was dosed more than once weekly.

TISSUE EMETINE LEVELS IN RATS

Each of 96 female rats of the WAG strain (initial body weight approximately 100 g) received the equivalent of 100 μg emetine base (approximately 1 mg/kg) once daily for up to 10 days. The hydrochloride was administered subcutaneously and all three preparations orally; 24 animals received each treatment.

Three rats from each group were killed with coal gas 24 hr after receiving their first, second, third, sixth or tenth doses; the remaining rats were killed 5, 12 or 19 days after their last doses. The emetine concentrations in the heart, kidneys, liver, lungs and spleen were determined on the bulked organs from each group of 3 animals by the spectrophotofluorometric method of Davis, Dodds & Tomich (1962).

Results

TOXICITY

Acute systemic toxicity in mice. The LD50 values are given in Table 1. Emetine hydrochloride and bismuth iodide were equally toxic by the oral

TABLE 1. ACUTE TOXICITY OF EMETINE PREPARATIONS TO MICE

Preparation	Emetine hydrochloride		Emetine bismuth iodide	Emetine resinate
Route	subcutaneous	oral	oral	oral
Doses (mg base/kg)	Mortalities per group of 10 mice			
20	0	0	0	–
25	0	1	0	–
30	2	5	2	–
35	9	9	7	–
40	10	10	9	–
145	–	–	–	–
218	–	–	–	–
290	–	–	–	–
1370	–	–	–	1
2280	–	–	–	4
LD50 (mg base/kg)	32	30	33	>2280

TABLE 2. SUBACUTE TOXICITY OF EMETINE PREPARATIONS TO DOGS

Sex of dog	Compound	Route	Treatment mg base/kg/day	Initial weight	Final weight	Fate	Cumulative dose mg base/kg
Female	Emetine hydrochloride (solution)	Subcutaneous	1·4	7·5	5·3	Died on day 9	7·0
Female	Emetine hydrochloride (solution)	Subcutaneous	1·4	6·0	4·2	Died on day 8	7·0
Female	Emetine hydrochloride (solution)	Subcutaneous	1·0	3·4	3·4	Died on day 7	5·0
Male	Emetine hydrochloride (solution)	Subcutaneous	1·0	3·7	3·6	Died on day 13	10·0
Female	Emetine hydrochloride (in gelatin capsules)	Oral	1·0	5·0	4·1	Died on day 16	11·0
Female	Emetine hydrochloride (in gelatin capsules)	Oral	1·0	3·3	3·2	Died on day 13	10·0
Female	Emetine bismuth iodide (in enteric coated tablets)	Oral	1·7	8·0	7·7	Very ill on day 15: Killed	17·0*
Male	Emetine bismuth iodide (in enteric coated tablets)	Oral	1·7	8·5	8·3	Very ill on day 15: Killed	17·0*
Female	Emetine bismuth iodide (in gelatin capsules)	Oral	1·0	4·3	3·0	Died on day 16	11·0
Female	Emetine bismuth iodide (in gelatin capsules)	Oral	1·0	3·4	3·2	Died on day 10	7·0
Male	Emetine resinate (in gelatin capsules)	Oral	1 for 2 weeks 1 for 2 weeks 4 for 2 weeks 8 for 2 weeks	2·7	8·0	Appeared normal on day 56	148·0
Female	Emetine resinate (in gelatin capsules)	Oral	2 for 2 weeks 4 for 2 weeks 8 for 2 weeks 16 for 2 weeks	2·6	9·1	Appeared normal on day 56	296·0

‡ Some tablets appeared unchanged in the faeces.

route and as toxic as the hydrochloride by the subcutaneous route. Orally the resinate exhibited one-eightieth the toxicity of the hydrochloride.

Subacute toxicity in dogs. The results are given in Table 2. Dogs were highly sensitive to the toxicity of emetine administered as hydrochloride or bismuth iodide. All four dogs injected subcutaneously with hydrochloride died after cumulative doses of 5 to 10 mg emetine base/kg, and two receiving it orally died after 10 or 11 mg/kg. Each of the two dogs dosed with enteric coated tablets of the bismuth iodide had to be killed after receiving a total dose of 17 mg emetine base/kg. Several enteric coated tablets were found in the faeces of these animals.

In contrast, two dogs dosed with resinate showed no ill effects: they grew well and appeared perfectly healthy after 56 days, by which time one had received a total of 148 and the other 296 mg emetine base/kg.

ACUTE EMETIC ACTIVITY IN CATS

From Table 3 it may be seen that emetine hydrochloride and bismuth iodide given orally were equally emetic and both were approximately 120 times more emetic than the resinate. The emetic activity of the hydrochloride by the oral route was 6 times that by the subcutaneous route.

TABLE 3. EMETIC ACTIVITY OF EMETINE PREPARATIONS IN CATS

Compound	Emetine hydrochloride						Emetine bismuth iodide		Emetine resinate			
Route	oral		subcutaneous				oral		oral			
Dose mg emetine base/kg	0·35	0·7	1·4	2·8	4·2	5·6	0·55	1·1	10	20	40	80
No. vomited / No. dosed	2/9	6/8	0/5	2/9	5/5	4/5	2/5	4/5	0/1	0/2	0/4	2/4
ED50*	0·6		3·5				0·7		ca. 80			

* ED50. Dose in mg emetine base/kg that causes emesis in 50% of cats.

TISSUE EMETINE DISTRIBUTION IN RATS

From Table 4 it may be seen that the emetine distribution pattern in the organs examined was similar for all three preparations and that of the hydrochloride was unaffected by the route of administration. The spleen had the highest concentrations of emetine, with the lungs, liver, kidneys and heart in decreasing order. The total amounts of emetine found in these organs during and after each treatment are given in Fig. 1. Cumulation occurred with all treatments and, except in the group which received emetine resinate, emetine was detectable in some organs 19 days after the last dose.

Emetine concentrations of similar order were found in the tissues after the oral administration of emetine bismuth iodide or hydrochloride, or after subcutaneous administration of the hydrochloride. Emetine resinate, however, produced emetine concentrations less than one-third of those produced by the hydrochloride.

TABLE 4. TISSUE EMETINE LEVELS IN RATS DURING AND AFTER 10 CONSECUTIVE DAILY DOSES OF DIFFERENT EMETINE PREPARATIONS

Treatment† and route	Days after beginning treatment	Cumulative dose to group (mg emetine base)	Emetine concentration (μg base per g wet tissue)*					Total emetine in tissues examined	
			Heart	Kidney	Liver	Lung	Spleen	(μg base)*	% of dose)*
Emetine hydrochloride Subcutaneous	1	0·3	1·0	2·6	2·9	4·9	6·8	68·4	22·8
	2	0·6	1·0	3·8	4·5	7·4	9·8	107·0	17·8
	3	0·9	2·1	5·1	5·5	10·5	12·4	148·0	16·4
	6	1·8	2·3	8·8	10·8	16·7	22·8	265·2	14·8
	10	3·0	3·6	11·7	11·7	17·7	21·2	327·2	10·9
	14	3·0	3·2	9·5	8·4	15·1	17·7	251·6	8·4
	21	3·0	1·5	4·0	3·2	7·6	9·8	105·6	3·5
	28	3·0	0·6	1·5	1·5	3·2	5·5	54·7	1·8
Emetine hydrochloride Oral	1	0·3	1·3	3·4	3·9	5·5	6·5	83·7	27·9
	2	0·6	1·6	4·9	6·5	17·9	10·7	156·5	26·1
	3	0·9	1·2	4·9	5·5	9·2	9·2	143·7	16·0
	6	1·8	2·4	8·0	10·7	16·5	19·2	251·5	13·9
	10	3·0	3·3	8·6	9·0	16·7	17·4	245·4	8·2
	14	3·0	1·8	5·7	5·2	10·4	16·1	164·7	5·5
	21	3·0	1·3	2·9	2·5	6·7	8·9	86·6	2·9
	28	3·0	0·4	0·4	0·6	1·5	3·1	21·6	0·7
Emetine bismuth iodide Oral	1	0·3	1·1	3·0	3·3	5·0	4·3	75·2	25·1
	2	0·6	1·5	5·2	6·1	8·8	13·7	126·9	21·2
	3	0·9	1·8	5·8	5·2	11·1	17·1	135·8	15·1
	6	1·8	2·4	8·3	10·1	13·6	20·3	241·0	13·4
	10	3·0	3·1	9·6	10·2	18·2	18·5	292·2	9·7
	14	3·0	1·3	4·9	6·0	17·4	15·3	175·1	5·8
	21	3·0	2·5	2·2	2·2	6·3	7·9	79·7	2·7
	28	3·0	0·0	0·7	0·0	2·2	3·1	15·3	0·5
Emetine resinate Oral	1	0·3	0·3	0·3	0·5	1·1	0·5	12·5	4·2
	2	0·6	0·8	1·3	1·9	2·4	2·4	33·1	5·5
	3	0·9	0·6	1·5	1·8	2·5	3·1	40·2	4·5
	6	1·8	0·6	1·5	2·4	5·0	5·3	63·9	3·6
	10	3·0	1·3	2·9	3·3	5·7	5·4	89·7	3·0
	14	3·0	0·5	0·5	1·3	2·5	2·8	35·3	1·2
	21	3·0	0·0	0·3	0·0	0·5	1·0	4·2	0·1
	28	3·0	0·0	0·2	0·2	0·3	0·6	5·5	0·2

* Group values (3 rats per group).
† Each rat received the equivalent of 100 μg emetine base once daily for 10 days.

Discussion

An ideal amoebicide should eliminate the parasites, wherever they may be located, without harming the host. Since emetine is toxic, any derivative that liberates the free base will fall short of this ideal.

Studies of distribution in the tissues of rats show that orally administered emetine hydrochloride and bismuth iodide are both well absorbed and that the levels obtained are similar to that after subcutaneous hydrochloride. Further, the bismuth iodide given orally was as toxic to mice as the hydrochloride given orally or subcutaneously, and orally the two compounds were equally emetic to cats. It would therefore seem that neither derivative is ideal for the treatment of intestinal amoebiasis. It should be emphasised, however, that in our experiments on the bismuth iodide the mice and rats were dosed with aqueous suspensions and the cats with gelatin capsules, whereas patients generally receive either gelatin capsules or enteric coated tablets. In dogs emetine bismuth iodide was less toxic than the hydrochloride; this was not surprising, because some of the enteric coated tablets were found in the faeces. If patients likewise eliminate some of their enteric coated tablets of emetine bismuth iodide

it explains why this preparation given in this form is not generally considered as effective as the hydrochloride in the treatment of hepatic amoebiasis, even though Woodruff (1959) has suggested that with a standard course of emetine bismuth iodide sufficient emetine is absorbed from the intestine for the purpose. Where, therefore, apparent paradoxes exist in the correlation of laboratory animal and clinical observations, these may be related to the differences in degree of absorption of emetine from the preparation administered and to species differences, such as that due to absence of a vomiting centre in the rat.

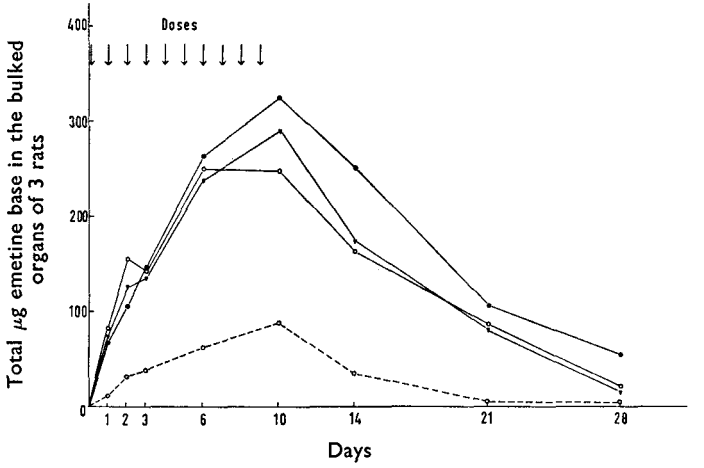

FIG. 1. Total amounts of emetine in the bulked organs of female rats (3 per group) during and after dosing once daily with various preparations of emetine equivalent to 100 μg emetine base per rat per day for up to 10 days. —●— Emetine hydrochloride—subcutaneous. —○— Emetine hydrochloride—oral. —▼—Emetine bismuth iodide—oral. ----○---- Emetine resinate—oral.

Combination of emetine with Zeo Karb 225, SRC 16, reduces its acute toxicity in mice, its subacute toxicity in dogs and its emetic action in cats. The reduced toxicity presumably reflects a lower degree of intestinal absorption, since tissue levels of emetine in rats after the resinate were lower than after the two other preparations.

The low toxicity of emetine resinate in animals is of interest because Muggleton, Heath & Johnson (personal communication) have shown that it is as effective as oral hydrochloride against intestinal *E. histolytica* infections in rats.

Preliminary results of clinical trials (Woodruff, personal communication) appear to confirm that the resinate is both effective and well tolerated by patients with intestinal amoebiasis.

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remaining resinates, including the one described, and Mr. P. G. Box and Miss O. Uvarov administered the drugs in the dog experiments. We thank Professor A. W. Woodruff for permitting us to report briefly on his findings before their publication.

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