Developmental Studies on Emetic Response to Tartar Emetic and Copper Sulfate in the Cat.* (25690)

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In a recent investigation of development of the emetic apparatus in the cat(1), we showed that emetic response first appears in animals weighing between 150 and 250 g (about 3 to 10 days old). Emetic stimulants studied were intravenous lanatoside C (.06 to .80 mg/kg) and total body X-irradiation (12,000 r). While incidence of vomiting with various doses of lanatoside C increased with age of animals, the pattern of response in all positive tests appeared to be much the same at all ages. To provide a more adequate means of comparing sequential chain of events in emetic response in young kittens and adults, and to compare the effects of tartar emetic and copper sulfate with lanatoside C, we carried out developmental studies of emetic effects of the latter drugs with the aid of slow-motion cinematography.

Materials and methods. Kittens and cats varying from newborn to adults were tested with oral copper sulfate in 1% aqueous solution at doses of 8 and 80 mg/kg and oral tartar emetic in 1% aqueous solution at 5 and 50 mg/kg. Over 250 animals were used. Since exact ages of a few of the younger animals were known, the animals were grouped according to following approximate weight means(g): 85, 100, 150, 200, 300, 400, 500, 600, 1000, and 2000 or over. A small amount of red food coloring was mixed with tartar emetic to facilitate recognition of the vomitus. Food was withheld from animals for 15 to 18 hours (over night) before each test, and no test meal was given. Following each test the animal was placed in cage with coarse wire mesh floor and observed 6 to 12 hours or until emetic response occurred. A large sheet of white filter paper was placed beneath each cage. Where no vomiting occurred, the filter paper was inspected for vomitus 24 hours after the test. The chi square method was applied to data from emetic tests to determine whether or not data for the group of animals weighing 150 g and under were significantly different from those of animals weighing 200 g or more for each dose with each drug. Similarly the chi square test was applied to data from groups weighing 200 g or more tested with the low dose of each drug as compared with the high dose. Motion pictures of emetic response in animals representative of the various weight groups were taken by a Bell and Howell model 700H camera on Kodak tri-X film at 64 frames/second. The camera is modified to take a 400 foot magazine and is equipped with electric motor drive.

Results. Results of drug tests are summarized in Table I. Incidence of vomiting with oral copper sulfate at 8 mg/kg (Table I) increased from zero in the first 3 groups to a maximum of about 90% in 600 and 1,000 g animals, then decreased to 66% in animals weighing 2 kg or over. Latency was several hours in the single 200 g animal which responded, and averaged 8 minutes in 6 animals of the 300 g group. Latency increased to around 40 minutes in 400 to 600 g animals then decreased to 32 minutes and 23 minutes respectively in 1 and 2 kg groups. At 80 mg/kg (Table I) incidence was zero in animals of 100 g or less but increased to 100% in all groups above 200 g. Average latency in this series is much shorter than in the 5 mg series, but again the longest average latency was observed in the 500 g group.

With 5 mg/kg tartar emetic (Table I) incidence was zero in all 12 animals of 150 g or less, increased from 50% to 100% between 200 and 400 g, then decreased to final value of 77% in young adults. Latency varied from 7 to 87 minutes with no indication of any trend associated with age. At 50 mg/kg tartar emetic (Table I) incidence was zero in 7 newborn animals of less than 100 g, 25% in the 100 g group and 100% in all older groups.

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TABLE I. Changes in Incidence of Vomiting to Copper Sulfate and Tartar Emetic in a Series of Kittens and Cats Weighing 85 g to over 2 kg.

Wt group, g	No. animals tested	Incidence of vomiting, %	Avg latency, min.	No. animals tested	Incidence of vomiting, %	Avg latency, min.
	$CuSO_4$—8 mg/kg			$CuSO_4$—80 mg/kg		
2000 or more	12	66	23	5	100	8
1000	10	90	32	10	"	6
600	11	91	41	10	"	11
500	11	82	49	5	"	21
400	10	60	39	10	"	10
300	10	60	8	3	"	15
200	10	10	Several hr	8	"	15
150	8	0				
100	4	0		6	0	
85	4	0		5	0	
	Tartar emetic—5 mg/kg			Tartar emetic—50 mg/kg		
2000 or more	9	77	34	4	100	15
1000	6	67	7	10	"	9
600	10	90	40	10	"	11
500	1	100	13	7	"	10
400	10	100	28	10	"	13
300	11	73	87	6	"	42
200	4	50	24	6	"	20
150	4	0				
100	2	0		4	25	98
85	6	0		7	0	

Latency period was appreciably shorter on the average than with the 5 mg/kg dose but showed no clear-cut trends or correlation with age.

Results of chi square tests revealed that differences in incidence between groups (150 g and under; 200 g and over) were highly significant for copper sulfate at 8 mg/kg, and at 80 mg/kg and for tartar emetic at 5 mg/kg and at 50 mg/kg. Also the differences in incidence between high and low doses of copper sulfate for animals of 200 g or over and between high and low doses of tartar emetic were highly significant. All of the above are significant beyond $p = .001$. Differences between successive small groups, as between the 1 kg (90%) and 2 kg (66%) groups with 8 mg/kg copper sulfate are probably not significant.

The sequence of events leading to the emetic act, as determined through direct observation and study of the motion picture films, appeared very much the same in young animals as in adults. It is noteworthy, however, that the reaction in adults is generally much more vigorous than in young animals, especially those representing earliest stages in vomiting. In fact, while vomiting in many adult animals was of a "projectile" type, some of the youngest animals following mild retching merely expelled stomach contents into the mouth. While some variation was observed in events leading to the emetic act in all age groups, the most general sequence observed was as follows: active licking of lips, quick voluntary shaking of head, increased swallowing activity with frequent spasmodic extension of head and neck, yawning, squinting, irregular respiratory movements becoming rapid and fairly regular, a cry, or rather shrill mewing, spasmodic contraction of abdominal wall muscles with simultaneous extension of head and neck, weak at first but becoming stronger and regular with opening of mouth and protrusion of tongue at maximal extension of head. In vomiting it appears that the diaphragm contracts as the initial movement and is followed by contraction of body wall muscles with extension of neck and head, opening of mouth and expulsion of stomach contents.

We noted previously(1) that only a very small percentage of younger animals in which vomiting was not elicited exhibited definite retching movements or movements suggestive of vomiting. In the present study, a number of "negative" animals in the younger groups exhibited enhanced lick-

FIG. 1. Typical position assumed by older kittens and cats at time of emetic act. The subject is 600 g kitten responding to 50 mg/kg tartar emetic.

ing of lips, salivation and increased rate of respiration; but definite "retching" movements with coordinated contraction of diaphragm and body wall muscles with extension of head and neck and opening of mouth were observed only in very few such cases. In a few "negative" animals in younger groups, however, it appeared that contraction of diaphragm occurred spasmodically followed by contraction of the body wall muscles without extension of head and neck. These movements were difficult to distinguish from weak efforts at "mewing."

Our previous statement(1) that vomiting occurs only when an animal is in a "standing" position requires some qualification for emesis occurs only when the animal is supporting itself on all 4 feet and is never seen in reclining position or when prostrate. The emetic act generally occurs with the subject in a prone, sitting, or crouching position and in many cases the animal crouches very low as emesis occurs (Fig. 1). The youngest animals exhibiting a positive response (200 to 300 g) often appear to suffer severe malaise and prostration and have difficulty maintaining a crouching position, but invariably make an effort to extend the forelimbs and bring anterior part of body at least into a standing or crouching-sitting position when the emetic act occurs. The single case in which a younger animal (100 g) vomited was unfortunately not observed at the instant of response.

It was observed repeatedly that tartar emetic results in shivering in many animals, especially at higher dose level, a short time after administration of the drug. This reaction was not observed with copper sulfate or in the earlier investigation with lanatoside C.

Comment. Chi square tests showed clearly that the differences in incidence of vomiting in younger groups (85 to 150 g, from about 1 to 3 days of age) as compared with older groups (200 g or over, about 7 days of age or older) were highly significant and that the differences in incidence between lower and higher doses of each drug for animals of 200 g or over were likewise highly significant. Only one out of 22 animals in the younger groups responded to these higher dose levels thus confirming the fact(1) that between 150 and 200 or 250 g stages is a critical period in development of the emetic mechanism in the cat.

Since we obtained different doses by administering different volumes, it might be argued that the results could be explained to some extent on the basis of different volumes or on absolute amount of drug administered. However, at a given dosage on a mg basis at a constant volume, the results would be complicated to a greater extent by considerations of varying drug concentration than by varying volume. We believe the gastric and/or duodenal receptors are more evenly stimulated by varying volume and maintaining concentration than the converse.

That animals in younger groups where incidence was very low or zero (150 g or less) in many cases received a larger amount of fluid as well as a larger absolute amount of drug than animals in older groups where the incidence was fairly high argues against the view that lack of response in these younger animals is due only to the small amount of drug received. For example, the 6 animals in the 100 g group tested with larger dose of copper sulphate each received about 8 mg of drug in .8 cc of fluid (incidence 0) while animals in the 300 g group given the small dose of copper sulphate received about 2.4 mg of drug in .24 cc of fluid (incidence 50%). Thus it appears that neither amount of fluid nor absolute amount of drug can be the determining factor in production of an emetic response. Rather, presence or absence of response depends here on functional maturity of the emetic apparatus itself.

There were no notable differences in premonitory signs or the actual emetic act itself between copper sulfate, tartar emetic, lanatoside C, or total body X-irradiation (except for shivering reaction seen with tartar emetic).

Summary. Incidence of vomiting to copper sulfate (8 mg/kg and 80 mg/kg in 1% solution) and tartar emetic (5 mg/kg and 50 mg/kg in 1% solution) increased significantly as shown by chi square tests, from animals weighing 150 g or less to a group weighing 200 g or more. A significant increase in incidence was also noted between animals (200 g or over) tested with low and high doses of these 2 drugs. While incidence increased to higher dose levels in the latter group no significant increase in incidence to the higher dose levels was found in the younger group (150 g or less). The above findings indicate that the period between 150 g and 200 g stages is a critical period in development of emetic response to copper sulfate and tartar emetic. The emetic act in older animals is more vigorous than in younger groups but the sequential chain of events leading to vomiting is much the same in young and old animals.

1. Brizzee, K. R., Vitale, D., *Am. J. Physiol.,* 1959, v196, 1189.

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Identification of *Listeria monocytogenes* by the Fluorescent Antibody Technic. (25691)

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Reports of human infections due to *Listeria monocytogenes* have increased in the past decade, due either to a greater number of such infections or to improved technics and awareness. *Listeria* is associated with many disease syndromes(1-3). Infections due to this organism are generally not distinct clinical entities. King and Seeliger(4) reported, within an 8 year period, 100 isolations of *Listeria monocytogenes* in the United States, only 14 of which have been reported in the literature as case histories. The organism was originally described(5) as a Gram-positive rod with a tendency to palisade formation typical of diphtheroids. Because of this similarity to diphtheroids, the organism is undoubtedly often discarded as a contaminant. More recently it has been reported that pleomorphism is predominant and that the coccoid to diplococcoid form is a more typical property(6). It is easily decolorized, and on blood agar the colonies may resemble streptococci. It is often difficult to isolate, requiring incubation at 4°C for several months(7). Because of increasing importance of this organism in clinical bacteriology and the inherent difficulties in isolating and identifying it, use of the fluorescent antibody technic(8) was investigated to provide a rapid and specific method for its detection and positive identification.

Materials and methods. Somatic antisera. Strains ATCC 7648, Type I (Schultz); ATCC 7644, Type II (Gibson); KC224 (Type III); Seeliger 5214 (Type IVa)(1); and Seeliger 1071/53 (Type IVb) were used to prepare somatic antigens, essentially according to method of Paterson(9). Pairs of mature albino rabbits were injected IV every 4 days with 0.5; 1; 2; 3; 3; and 3 ml of antigen adjusted to a McFarland 3, and bled 5 days after last injection. *Flagellar antisera.* Strains ATCC 7644; ATCC 4428 (Type I); and 5214 were used to prepare the flagellar antigens according to method of Paterson(10). It was found necessary to use an additional inoculum of 1.5 ml of antigen to obtain a good immune response. *Whole cell antisera.* Strains ATCC 4428, ATCC 7644, and 5214 were inoculated in trypticase soy broth and incubated at 37°C for 18 hours. Kolle flasks of trypticase soy agar were inoculated with 0.5 ml of broth culture and incubated at 37°C. The growth was removed with 0.5% formalinized saline and the cells washed 4 times with normal saline. Pairs of albino rabbits were injected IV with 0.1; 0.2; 0.4; 0.8; and 1.0