temporarily lead to varying degrees of anoxia of the heart muscle.

The changes in the tonus of the autonomic nerves (in mammals particularly the sympathetic nerves) which has been mentioned above can also directly influence the final deflection of the electrocardiogram. In favor of the presence of such changes in tonus is the appearance of A-V rhythm with changes in posture (Fig. 3) and the remarkable fact that pronounced abnormalities of the P-R interval disappear or diminish when the subject is erect.[10,11,12] A change of the tonus of the sympathetic nervous system will modify the form of the T waves and the ventricular gradient. The sympathetic nerves accelerate the repolarization process. Under normal conditions the subendocardial layers are depolarized earlier and repolarized later than the subepicardial muscular layers. This is probably due to the high intraventricular pressure. A faster repolarization of the inner layers would lead to a depression and inversion of the T waves.

The importance of a change in the tonus of the sympathetic nervous system is demonstrated by the absence of positional electrocardiographic alterations after the administration of sympathicolytic drugs.[5,6,13] The frequent ineffectiveness of ergotamine shown in this report (Fig. 4) had to be expected, because the immediate changes appearing on standing are not due to a change of sympathetic tonus as frequently maintained. There is furthermore no proof that the sympathetic system alone is involved in the electrocardiographic changes which appear on continued standing.

Summary. The immediate alterations in the eletrocardiogram caused by assuming the erect posture and those appearing during standing, were studied in 80 patients.

The genesis of the changes which appear immediately on standing and disappear immediately when the supine posture is resumed is separated from those which are seen during prolonged standing. The latter is ascribed to changes in the tonus of the autonomic nervous system which may act on the heart muscle directly or via the coronary arteries.

A-V rhythm may appear on standing due to the change of the tonus of the autonomic nerves.

Administration of ergotamine preparations does not always prevent the appearance of immediate changes in the electrocardiogram on change of posture.

Our thanks are due to Dr. Henze of the Sandoz Drug Company for the supply of Dihydro-Ergotamine 45.

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A Comparison of the Acute Toxicity of Two Forms of Thiamine.*

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Recently there has become available for investigational use a new form of vitamin B_1, thiamine mononitrate. Considering the wide therapeutic use and the parenteral toxicity of thiamine hydrochloride,[1-13] it was thought

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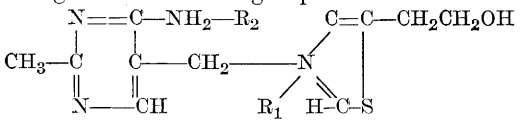
[1] Steinberg, C. L., *Am. J. Digestive Dis.*, 1938, **5**, 680.

TABLE I.
Properties of Two Forms of Thiamine.

Property	Thiamine Hydrochloride	Thiamin Mononitrate
Melting point	245-248°C	196-200°C dec.
Molecular wt	337.26	327.36
Units per mg	333	343
Solubility	100 g per 100 g water	2.7 g per 100 g of water

advisable to compare the hydrochloride and the mononitrate in regard to their toxicity in animals.

Comparison of the structural formulas of both forms of thiamine shows where the changes in substituent groups have been made.



	R_1	R_2
Hydrochloride	Cl	HCl
Mononitrate	NO_3	—

Table I shows the chemical and physical characteristics of both compounds.[14-15] From these data one can conclude that, although the 2 forms are approximately equal in potency, their solubility characteristics would favor the use of the hydrochloride when large doses are to be administered parenterally. However, the solubility of the mononitrate increases to 18.5 g per 100 cc when the pH of the solutions is adjusted to 4.0. Aqueous solutions at this pH are stable for one year,[14] while aqueous solutions of the hydrochloride at pH 2.7 to 3.0 begin to show a loss of potency in about 6 months.[16]

Acute toxicity studies of thiamine hydrochloride have shown that, although large oral doses were toxic, these doses were much larger than those used therapeutically.[17-20] However, the same is not true of parenterally administered doses of the drug. Early studies indicated toxic symptoms both in animals and humans[1,2,18-20] and recently Haley and Flesher[21] found that rabbits developed symptoms of toxicity after intravenous injections of 200 to 300 mg per animal. This work has been extended to dogs by Smith *et al.*[22] who have reported similar results.

Experimental. Acute toxicity of the hydrochloride was determined by intraperitoneal injection in mice and intravenous injection in rabbits and of the mononitrate by intraperitoneal and intravenous injection in mice and intravenous injection in rabbits. In the mouse experiments, a total of 150 animals weighing 22-42 g were used and the concentration of the drugs was 50 mg/cc. The dosage ranged from 0.04 to 0.07 cc intravenously and from 0.17 to 0.32 cc intraperitoneally. Death occurred within 5 minutes after intraperitioneal and within 30 seconds after intravenous injection in mice. In the rabbit experiments

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20 Molitor, H., and Sampson, W. L., *Merck Jahresbericht*, 1936, **50**, 51.

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TABLE II.
Toxic Doses of Two Forms of Thiamine.

Animal	Form	Route of administration	Dose mg/kg	Mortality ratio	LD_{50} mg/kg	Standard error
Mouse	Mononitrate	Intravenous	80	0/5	84.24	±1.14
			84	4/5		
			86	3/5		
			88	3/5		
			90	3/5		
			92	5/5		
,,	,,	Intraperitoneal	380	1/5	387.3	±1.65
			385	3/5		
			390	2/5		
			395	4/5		
			400	3/5		
,,	Hydrochloride	,,	310	3/5	329.8	±3.93
			320	3/5		
			330	3/5		
			335	4/5		
			340	4/5		
			350	4/5		
			Animal wt, kg	Total dose, mg	Intrav. lethal dose, mg/kg	Avg intrav. lethal dose, mg/kg
Rabbit	Mononitrate	Intravenous	3.66	500	136.61	112.58
			4.30	475	110.46	
			3.22	375	116.46	
			4.44	437.5	98.53	
			4.66	470	100.85	
,,	Hydrochloride	,,	1.704	180	105.63	117.45
			1.818	220	121.01	
			1.591	200	125.70	

the dosage was 1 cc (50 mg/cc) per minute until death, which occurred within 10 minutes. The rabbits used weighed 3.22 to 4.44 kg. Table II gives the mortality figures for mice including the LD_{50} which is the dose calculated to kill 50 per cent of the mice, according to the method of Miller and Tainter;[23] Table II also lists the intravenous dose required to kill all of the rabbits injected.

The symptoms of toxicity observed were: restlessness, labored respiration, vasodilatation, cyanosis, muscular twitching, clonic convulsions and death by respiratory paralysis. This respiratory paralysis was of central origin because electrical stimulation of both the muscle of the diaphragm and the phrenic nerve showed that the muscle was still capable of contraction. Visual signs of anoxia were a gradually deepening bluish coloration of the ears and all other body areas where the fur was thin enough to permit direct observation of the skin. In all animals cardiac arrhythmias were seen upon opening the thoracic cage. Auricular/ventricular rates of from 2:1 to 5:1 were common.

In conjunction with these toxicity studies more than 100 unanesthetized rabbits, weighing between 3.11 and 5.33 kg, were injected intravenously with solutions of thiamine hydrochloride ranging from 10 to 100 mg/cc. This routine testing over the period of one year showed that, although fatalities were seldom observed, clonic convulsions were produced in 80% of the animals when the total dose was above 300 mg per animal. These convulsions usually occurred after the animal had been returned to its cage. No visual signs of anoxia, cyanosis of the blood in the ear veins or of the skin in the scrotal region, were seen. However, no blood samples were

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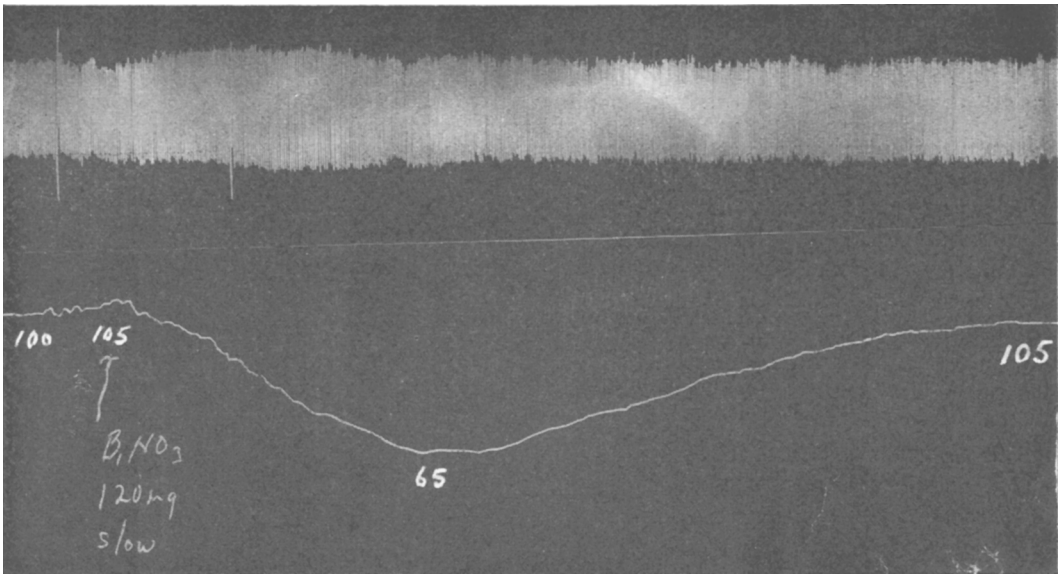

FIG. 1.
Top, Respiration. Bottom, Blood Pressure.

obtained for the determination of oxygen content so that partial anoxia cannot be entirely ruled out as a causative factor in the convulsions.

Inasmuch as Molitor[20] has shown that thiamine has a pronounced effect upon respiration and little effect upon the blood pressure of the dog, it was decided that a study of this phenomenon should be undertaken. Six rabbits, weighing 3.66 to 5.22 kg were anesthetized with 20 mg/kg of sodium pentobarbital intravenously and 3.5 cc/kg of 20% urethane intraperitoneally and prepared for recording of blood pressure via the carotid artery and respiration by cannulation of the trachea. Respiration was recorded with the Haley respirometer[24] and blood pressure with a mercury manometer. The two forms of thiamine in a concentration of 50 mg/cc in normal saline were injected at a fixed dose of 120 mg alternately. As the rate of injection partially determines the effect on the animal, 2 injection rates were employed; slow (1 mg/second) and fast (12 mg/second). Further, in order to rule out the effect of pH, 2 rabbits were injected with the mononitrate at pH 6.8 and 4 at pH 0.9. The hydrochloride was always at pH 2.7. Fig. 1 shows a typical record of a slow injection and Fig. 2 is typical record of a fast injection.

The results of this work show that slow injections of either form of thiamine had very little effect on respiration but caused a fall in blood pressure averaging 36 mm of mercury. This fall was gradual, requiring 114 seconds to be completed and the recovery required 219 seconds to reach the previous normal level. Fast intravenous injections had a more pronounced effect on respiration, decreasing both the rate and the depth. There was also a blood pressure fall averaging 23 mm of mercury.

Discussion. From the results of the acute toxicity determinations given in Table II there appears to be little difference in the toxic doses of either form of thiamine. Further, Molitor[25] has found that the mouse intravenous toxicity of the hydrochloride is 85 mg/kg which agrees with the 84.24 mg/kg figure for the mononitrate.

From the results herein presented as well as those of previous investigators[20–22] one must conclude that thiamine exerts its principal toxic effect by paralysis of the respiratory center. However, the work of Zaidi[26] on the

[24] Haley, T. J., *J. Am. Pharm. Assn.*, in press.

[25] Molitor, H., personal communication.

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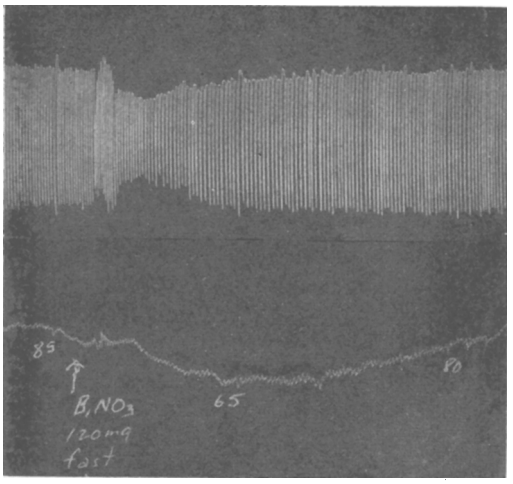

FIG. 2.
Top, Respiration. Bottom, Blood Pressure.

isolated frog heart and Smith *et al.*[22] on the isolated turtle heart indicates that thiamine has also a direct toxic effect on the myocardium. Furthermore, irregularities in the electrocardiogram of dogs after the administration of large doses of thiamine[22] shows that the mammalian heart is affected.

Smith *et al.*[22] found that, even after vagotomy or atropinization, there was a prolonged peripheral blood pressure fall. Haimovici and Pick[27] reported that thiamine caused vasodilatation which counteracted the vasoconstrictor action of nicotine on the perfused frog hind limb preparation. Thus it is probable that thiamine causes vasodilatation by direct action on the peripheral vascular musculature.

Summary. There is little difference in the lethal dose of either thiamine hydrochloride or mononitrate and the symptoms of toxicity are the same for both forms of the drug. The toxic effects of both forms of the vitamin on respiration and blood pressure are due to their thiamine content and not to the pH of the injected solutions or to the substituent groups on the nitrogen of the thiazole nucleus. In lethal doses either form of the vitamin causes death by a direct paralyzing action on the respiratory center followed by cardiac failure.

The author wishes to thank Merck and Co. for the generous supply of thiamine mononitrate used in this study.

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Influence of Nephrectomy and Renal Pedicle Ligation on the Activity of Liver Arginase in Rats.*

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Intermediary nitrogen metabolism has been reported to be impaired in dogs with reduced or abolished kidney function.[1] This deficiency is manifest in the fact that 30 to 75% of fed nitrogen is not transformed into urea.

Consideration was given to the possibility that renal transformation of citrulline into arginine[2,3] might represent a reserve mechanism for the urea cycle in the liver and that curtailment of this renal reserve might be related to the observed deficiency.

The hypothesis that an increase in arginase activity might compensate—at least in part—for the lesser concentration of arginine and ornithin by speeding up the turnover of the urea cycle led to a series of experiments. They include determinations of liver arginase

* Aided by a grant from the Commonwealth Fund.

† James Hudson Brown Junior Fellow.

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