and no visible exudate. By the fourth or fifth day following the operation some of the plastic coats began to come loose along the edges. These edges were then repainted with the same solution of the plastic and allowed to dry before the animal was returned to the cage. In this way, with relatively little attention, the wounds were kept sealed and protected until they were completely epithelialized. The changes which took place could be observed and measurements made without disturbing them in any way.

The plastic acted as a splint which prevented contraction of the wound. The marked contraction which occurs in unsplinted wounds[1,2,6] was not observed under these conditions. That contraction can occur after the wound is completely filled with granulation tissue may be demonstrated by removing the plastic coat at that time (8th day post-op.). On the following day, the diameter of the wound is reduced and a papilla of granulation tissue 2 to 4 mm high appears in the center of it.

A further advantage of the method is that the effect of aqueous solutions may be determined without having to incorporate the solution into an ointment which would complicate interpretation of the results observed. An example is given in the preceding paper on the effects of a derivative of chromatin.

The use of the plastic dressing made it possible to study the uptake of radioactive phosphorus (P^{32}) and radioactive sulfur (S^{35}) by the wound during the healing process. The data obtained in these experiments will not be presented here but are mentioned as an additional example of the usefulness of the plastic dressing.

Although the plastic solution produces no reaction in skin wounds of the rat and rabbit, it was found unsuited to the study of human skin wounds since it stimulated the formation of copious amounts of serosanguinous exudate. It may have some practical use, however, since it has been found that when applied to cuts, bruises, and small burns of the hand on the second or third day there is no exudation. Since the plastic does not contract, as does collodion for example, an entire finger may be coated. It has been found to adhere and protect the wound for 3 to 4 days under conditions (K.P. duty) where a bandage dressing was of little value. By the proper combination of normal and isobutyl polymers almost any degree of flexibility may be obtained.

Summary. The use of methacrylate polymers as a wound dressing in rodents is described and its advantage in the experimental study of wound healing indicated.

14844

Origin of Cardiac Disorders in Thiamine-Deficient Cats.

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Some of the cardiac disorders reported in thiamine deficiency may be neurogenic rather than myogenic in origin.[1,2] The following observations suggest a dual origin of the electrocardiographic signs in the thiamine-deficient cat.

Method. The method used for producing specific thiamine deficiency in cats has been reported previously in connection with a study of the neurological findings.[3] During the development of the deficiency and during the recovery period, limb lead electrocardiograms were taken in 37 unanesthetized animals. The electrocardiograms were recorded with an ink-

[1] McEachern, D., and Brophy, D., PROC. SOC. EXP. BIOL. AND MED., 1942, **51**, 75.

[2] Weiss, S., and Wilkins, R. W., *Ann. Int. Med.*, 1937, **11**, 104.

[3] Everett, G. M., *Am. J. Physiol.*, 1944, **141**, 439.

TABLE I.
Incidence of Abnormal Electrocardiographic Findings in Thiamine-deficient Cats.

Abnormality	No. abnormal	Total No. of cats	% abnormal
(1) Prompt response to thiamine			
Bradycardia*	22	37	59
Tachycardia*	11	37	30
Sinus irregularity	32	37	87
Excitement syndrome	17	18‖	95
Ectopic rhythms	5	37	14
(2) Slow response to thiamine			
QRS prolongation†	8	16‡	50
Other QRS changes†	5	16‡	31
T wave changes†	32	37	87
Inversion of T_2	16	37	43
Prolonged systole-cycle constant*	20	27§	74

* Beyond normal limits for entire group, prior control.

† Compared with prior control on same animal.

‡ Excluding those without satisfactory prior controls.

§ Excluding those with low-voltage T-waves.

‖ Excluding those observed only at rest.

writing oscillograph fed by amplifiers having a suitably long time-constant.

Results. Table I summarizes the abnormal electrocardiographic findings. The disorders may be divided into two groups on the basis of their response to thiamine therapy:

1. *Disorders promptly reversible by thiamine injection.* During thiamine deficiency, sinus bradycardia began as early as the second week. Tachycardia was less frequent and began late. Sinus irregularity was common; it was sometimes related to respiration but more often not. Cycle durations varied widely and randomly from beat to beat and bore no simple numerical relation to each other. The irregularity was greatest at lowest heart rates and disappeared when rates were high, as when the animal was excited by handling, or after atropinization, or during the brief spontaneous tonic convulsive seizures characteristic of cats in the critical stage of thiamine deficiency. Irregularity sometimes occurred with heart rates within the normal range, but not in animals anesthetized with pentobarbital (40 mg/kg) even when heart rate was low. In all but one animal examined during the critical stage of thiamine deficiency, handling of the animal or spontaneous seizures resulted in an "excitement syndrome": a brief period of tachycardia was followed by severe bradycardia and irregularity, sometimes with escaped beats of ventricular or low nodal origin Recovery often required many minutes.

Following the intramuscular injection of thiamine (0.5 to 2.0 mg/kg) in the critical stage of deficiency, a relative tachycardia appeared in all but one animal. It coincided with the first improvement in neurological signs (20 minutes at the earliest) and persisted for as long as 24 hours, after which the rate subsided to normal or below. Recovery both from bradycardia and from disorders of impulse formation and rhythm was complete in most animals within 3 days, at which time neurological recovery was also complete. In 3 animals with persistent neurological signs (ataxia and ophthalmoplegia), recovery from the above cardiac disorders was delayed.

Sections of the brain stem of 2 animals sacrificed in the critical stage of deficiency showed vascular lesions restricted to the midbrain, with associated chromatolysis (*cf.* Weiss and Wilkins[2]).

A neurogenic (central nervous) origin of the above described disorders seems plausible. Their prompt reversibility by thiamine therapy demonstrates the specificity of thiamine deficiency in their production.

2. *Disorders not promptly reversible by thiamine injection.* T waves became flattened or inverted in at least one lead in most animals, the change being evident as early as the 10th day (a small percentage of normal cats showed inversion of T_1 or T_3, but T_2 was always normally upright). Inversion was usually terminal, was associated with a flat and prolonged ST segment, and was independent of wide variations in heart rate. It persisted under pentobarbital anesthesia or full atropinization. Such stability was seen also in the abnormally prolonged systole-cycle constant ($K = QT/\sqrt{\text{cycle}}$) and in the altered QRS complexes (prolongation, slurring, and notching).

Alterations in this category were slow to disappear (3 days to 2 months) following thiamine treatment, despite prompt neurological recovery. Their persistence suggests myocardial damage, and the combination of

signs points to greater involvement of the left ventricle. However, neither gross nor microscopic pathology was evident in hearts of sacrificed animals. Early appearance and late disappearance of signs did not parallel changes in body weight.

Summary. Electrocardiographic changes in cats with acute thiamine deficiency fall into two categories: (1) disorders of rate, regularity, and impulse formation, which yield promptly to thiamine treatment; (2) changes in the ventricular complex, which respond more slowly to treatment.

14845

An Electrophoretic Study of Tetanus Toxoid Prepared on Mueller's Peptone-Free Medium.

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Recent advances in the study of the complexity of tetanus toxin have been summarized by Smith[1] and Petrie.[2] Smith compared the toxicity of five different tetanus toxins in mice, guinea pigs, and rabbits. She showed that the ratio of the LD_{50} for the rabbit and mouse and the ratio for the rabbit and guinea pig (weight for weight) may show extraordinary and significant variations, whereas the ratio of the lethal dose for the mouse and guinea pig is more or less constant. Her findings indicate that tetanus toxin contains two or more toxic factors and that the mouse and the guinea pig are relatively more sensitive to one and that the rabbit is relatively more sensitive to the other factor. Petrie studied the variable interactions of tetanus toxins and tetanus antitoxins with the object of throwing light upon the cause of the discrepancies that are met when samples of tetanus antitoxin are assayed in different laboratories. His results indicate that crude tetanus toxins are not homogeneous solutions of toxin and toxoid molecules. Whether they are used as antigens for the hyperimmunization of horses or as test-toxins, crude tetanus toxins are to be regarded as mixtures in varying proportions of the "primary" toxin or toxoid molecule and of an antigenic variant of this which is formed during the later stages of the culture growth; preparations of tetanus serum likewise consist of mixtures of varying proportions of the corresponding antitoxins. Differences in mutual neutralizing affinities of these components in a reacting system account for the discrepancies in the titer of tetanus sera when different test-toxins are used for the assays.

The crude tetanus toxins referred to in these papers were prepared from culture media containing Witte, Riedel, or Difco peptone and beef or horse-meat infusion. The tetanus toxoid studied in the present paper was derived from tetanus toxin, prepared on a medium containing only low molecular nutrient materials. The toxoid could be purified and concentrated without chemical treatment.

Materials and Methods. The tetanus toxin was produced on a peptone-free medium according to the method of Mueller and coworkers.[3] It contained approximately 20,000 guinea pig MLD per cc and had an L^+ of 0.04 cc. The conversion of the hydrolysate tetanus toxin to toxoid was also carried out according to Mueller's procedure.[4] The antigenicity was appraised by injecting 0.4 cc of the toxoid subcutaneously into each of 20 mice. At the end of 10 days, the mice were

[1] Smith, M. L., *Bull. of the Health Organization*, 1942-3, **10**, 104.

[2] Petrie, G. F., *Bull. of the Health Organization*, 1942-3, **10**, 113.

[3] Mueller, J. H., Schoenbach, E. B., Jezukawicz, J. J., and Miller, P. A., *J. Clin. Invest.*, 1943, **22**, 315.

[4] Schoenbach, E. B., Jezukawicz, J. J., and Muller, J. H., *J. Clin. Invest.*, 1943, **22**, 319.