TABLE II.
Comparative Titrations of Toxoplasma Infected Tissues in Mice and Developing Chick Embryos. 10-day chick embryos inoculated via the yolk sac. Mice inoculated intracerebrally.

Passage	Inoculation: Tissue	Inoculation: Amt, ml	Titration in:	LD_{50} titer	LD_{50} doses per g of indicated tissue, × 1000
335	Mouse brain	.5	Embryos	$10^{-5.0}+$	200+
		.03	Mice	$10^{-4.5}$	1,000
336	" "	.5	Embryos	$10^{-5.0}$	200
		.03	Mice	$10^{-4.0}$	330
344	" "	.5	Embryos	$10^{-4.5}$	64
		.5*	Mice	$10^{-4.5}$	64
15	Y.S.-C.A.	.5	Embryos	$10^{-4.0}$	20
		.03	Mice	$10^{-3.0}$	33

* Mice in this case injected intraperitoneally.

that neutralization tests could be performed in the chick embryo.

The embryonated hen's egg may be an adjunct for the primary isolation of toxoplasma and although inoculation of eggs with the laboratory-adapted strain gave uniformly good results the probability is that for the initial recovery of strains the mouse is still the animal of choice.

Summary. A laboratory-adapted strain of toxoplasma was successfully propagated in the embryonated hen's egg. Uniform mortality was obtained and the majority of deaths occurred between 5 and 6 days after inoculation. The concentration of organisms in the chick embryo gave LD_{50} titers of $10^{-4.5}$ to $10^{-5.0}$ which approximated that usually attained in mice. Repeated passage through eggs resulted in no modification of pathogenicity for animals.

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Histochemical Differentiation Between Esterases.*

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Strikingly different properties of esterases (lipases) obtained from various species[1,2] have been reported by a number of workers. The differences include substrate specificity,[3] stereochemical specificity,[4] behavior towards activators and inhibitors, and pH optima.[5] Owing to multiple overlapping of differences and similarities between enzymes obtained from various sources, it is still undecided whether the various effects are due to the existence of several well defined individual enzymes or to the presence of unidentified accompanying substances.

With a histochemical method for the visualization of sites of lipase activity, similar differences between the esterases of various organs were found.

Experimental. Organs of 8 freshly killed white mice (5 males and 3 females) were fixed in chilled acetone, dehydrated, embedded

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and stained according to the technique previously published.[6,7] In addition to the original substrates, a new one, G-9096-CJ (Atlas Powder Co., Wilmington, Del.), a stearic ester of modified mannitan, was used. This new substrate seems to be hydrolyzed more rapidly than the related Tweens, but the localization and the relative intensity of the reaction in various organs is the same with all substrates. Pieces of practically all organs (liver, kidney, spleen, lung, heart, stomach, intestine, pancreas, salivary glands, testis, uterus, ovary, urinary bladder, etc.) were included in a single paraffin block, and serial sections were incubated with the substrate to which the substances to be tested were added.

The following substances were selected for their known effects on esterases: cholate,[8,9] quinine,[10-13] arsanilate,[1,10,11,13] eserine,[4,14] pilocarpine,[4] urethane,[15] caprylate,[16] n-butyraldehyde,[17] acetophenone,[17] hexylresorcinol[18] and NaCl.[19]

Results. Since a strict quantitative evaluation of histochemical reactions of this type is impossible, increase or decrease in the in-

TABLE I.
Effect of Various Substances on Lipase (Esterase) Activity.

	Liver	Pancreas	Kidney	Intestine	Stomach	Testis: Interstitial cells	Testis: Spermatic elements	Lung
Quinine, .005 to .01 M	1—	2— to 3—	1— to 2—	3—	2— to 3—	0 to 1—	0 to 1—	0 to 3—
Arsanilate, .0002 M	3—	1—	3—	3—	1—	3—	0 to 1—	2— to 3—
Taurocholate, .02 M	2— to 3—	2+ to 3+	3—	3—	2+ to 3+	2—	2—	2— to 3—
Pilocarpine, .01 M	0	0	0 to 1—	0 to 1—	0 to 1—	0 to 1—	0 to 1—	0 to 1—
Urethane, 1 M	0	0	0	0	0	0	0	0
Eserine, .002 to .005 M	0 to 1—	0 to 1—	0 to 2—	0 to 2—	0 to 2—	0	0	0
Caprylate, .005 M	2— to 3—	1—	3—	2—	1—	2—	2—	2—
n-Butyraldehyde, .005 M	0	0	0	0	0	0	0	0
Acetophenone, .005 M	0	0	0	0	0	0	0	0
Hexylresorcinol, .0005 M	0	0	0	0	0	0	0	0
NaCl, 1.2 M	0	0	0	0	0	0	0	0

The symbol 3— stands for complete suppression of enzyme activity.

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tensity of the reaction will be indicated only by plus and minus signs, with the arbitrary degrees 1, 2 and 3. (Table I). The pattern of response was markedly uniform in all animals, with the exception of the lung which behaved erratically towards quinine (no inhibition, one case; slight to moderate inhibition, 3 cases; complete suppression of the reaction, 4 cases).

The histochemical results are in good agreement with previous findings on the *in vitro* behavior of hepatic and pancreatic lipase towards quinine and arsanilate and on the similarity between the pancreatic and gastric enzymes, both being activated by bile salts.[9] On the other hand, the effect of several substances, markedly active in test tube experiments, such as urethane, butyraldehyde, acetophenone, hexylresorcinol and NaCl, cannot be observed under the conditions of the histochemical experiment. An interesting new finding is the difference in sensitivity to arsanilate between the enzyme of the interstitial cells (testis) and that of the spermatic elements. Another curious observation was a marked diffusion of the reaction around the sites of activity in the pancreas and the stomach, but nowhere else, when cholate was added to the substrate. In some cases such large numbers of lead sulfide granules were embedded in the protecting collodion membrane and on its surface as to make the exact localization of the enzyme virtually impossible. This finding may indicate an increased diffusibility of pancreatic and gastric lipase in the presence of bile salts.

Summary. Differences in the behavior of esterases (lipases) from various sources towards activator and inhibitor substances, similar to those found previously in *in vitro* experiments, can be observed also in tissue sections stained for lipase.

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Biological Activity of Crystalline Procaine Penicillin *In vitro* and *In vivo*.*

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In a recent communication Salivar, Hedger, and Brown[1] described the preparation and chemical properties of crystalline procaine penicillin. The present report deals with the biological activity and with the absorption and excretion of this form of penicillin and of other relatively insoluble salts of penicillin.

Materials and Methods. Unless otherwise specified, crystalline procaine penicillin G and dihydro-F, prepared from crystalline sodium penicillins G and dihydro-F, respectively, were used throughout this study. The potencies of these preparations were determined by the Oxford cup plate method;[2] the per cent G, non-G penicillin by a modification of the N-ethyl piperidine method.[3] Sensitivity determinations were carried out by a quantitative broth dilution technic. The method of Tompsett, Schultz, and McDermott,[4] using *Strepto-*

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3 *Federal Register*, April 4, 1947, v. **12(67)**, p. 2222, 141.5 (f).

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