lignin in sandy loam soil definitely reduced the amount of nitrate nitrogen recovered from either dried blood or ammonium sulfate. The effect was much more marked with the dried blood than with the ammonium sulfate.

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Administration of Streptomycin in Peanut Oil and Beeswax and in Solvecillin

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One of the most effective methods for delaying the absorption and excretion of penicillin, with the prolongation of effective serum levels, is by intramuscular or subcutaneous injections of the compound suspended in sterile peanut oil and beeswax, as first proposed by Romansky and Rittman (*7*) in 1944. Fixed oils themselves delay absorption, but the addition of beeswax enhances these effects. Since then their observations have been amply confirmed by various investigators (*1, 4, 6, 9*). Freund and Thomson (*3*) have also proposed the administration of penicillin in water-in-oil emulsion for slower absorption, using as a vehicle a lanolin-like substance prepared from oxycholesterins and cholesterol esters commercially available under the name of "solvecillin."

Neither of these methods has been reported upon in relation to the administration of streptomycin. Since the indications for slow absorption with prolonged therapeutically effective serum levels are the same as in penicillin therapy, the purpose of this investigation was to study the absorption and excretion of streptomycin suspended in peanut oil and beeswax and in solvecillin administered by intramuscular injection.

Single doses of streptomycin[1] suspended in 2 cc. of sterile peanut oil and 4 per cent beeswax were administered by intramuscular injection to adults of approximately the same body weight. The local reactions were quite mild and similar to those produced by intramuscular injections of similar amounts of streptomycin dissolved in sterile saline solution. One subject showed a delayed reaction occurring 7 days after injection and characterized by a generalized urticaria.

Emulsions in solvecillin were prepared by dissolving streptomycin in 1.4 cc. of sterile saline solution and adding the solution to 3.1 cc. of previously warmed solvecillin followed by thorough emulsification. Single doses of the compound in a total dose of 4.5 cc. of menstruum were likewise administered by intramuscular injection to adults of approximately the same body weight. Only mild local reactions resulted.

At intervals of 1, 2, 3, 4, 6, and 24 hours thereafter blood and urine were collected for assay purposes, each specimen of urine being measured and the total excretion of streptomycin calculated on the basis of units/cc. All assays were conducted according to the method of Stebbins and Robinson (*8*), using *Staphylococcus aureus* (SM strain).

TABLE 1

Intervals*	Subject No. 1				Subject No. 2				Subject No. 3			
	Serum	Urine			Serum	Urine			Serum	Urine		
	(units/cc.)	Vol. (cc.)	Units/cc.	Total units	(units/cc.)	Vol. (cc.)	Units/cc.	Total units	(units/cc.)	Vol. (cc.)	Units/cc.	Total units
1	0	154	11	1,694	0	75	17	1,275	Trace	430	1.6	688
2	0	40	35	1,400	0	75	20	1,500	"	175	10.0	1,750
3	0	140	48	6,720	0	200	4.8	960	0			
4	0	45	35	1,575	0	205	1.8	369	0	350	10.0	3,500
6	0	80	37	2,960	0	200	6.0	1,200	0			
24	0	660	6.5	4,290	0	1,150	2.0	2,300	0	1,020	6.1	6,222
Totals		1,119		18,639		1,905		7,604		1,975		12,160
Per cent† .				7.4				3.4				4.8

* Hours after administration of streptomycin.
† Per cent of injected dose of streptomycin excreted in the total 24-hour urine.

Table 1 shows the serum levels and urinary excretions observed in three subjects following single intramuscular injections of 250,000 units of streptomycin suspended in 2 cc. of sterile peanut oil and 4 per cent beeswax. It will be observed that only one sub-

[1] Streptomycin sulfate kindly supplied by the Abbott Laboratories, North Chicago, Illinois.

ject (No. 3) showed a trace of the compound in serum collected 1 and 2 hours after treatment. The total urinary excretion of the compound over a period of 24 hours varied from 3.4 to 7.4 per cent of the amount administered. Similar doses dissolved in 2 cc. of sterile saline solution and administered to three adults by intramuscular injection showed serum levels of 1.5, 3.0, and 2.5 units/cc., respectively, at the end of 6 hours, with total excretions of 22.0, 24.0, and 48.0 per cent of the amounts administered in the 24-hour urine (*5*).

TABLE 2

Intervals	Subject No. 4				Subject No. 5				Subject No. 6			
	Serum (units/cc.)	Urine			Serum (units/cc.)	Urine			Serum (units/cc.)	Urine		
		Vol. (cc.)	Units/cc.	Total units		Vol. (cc.)	Units/cc.	Total units		Vol. (cc.)	Units/cc.	Total units
1	5.0	225	20	4,500	3.0	42	115	4,830	2.5	143	55	7,865
2	5.0	110	120	13,200	3.0	27	183	4,941	2.5	87.5	133	11,638
3	4.0	50	200	10,000	4.0	29	168	4,872	2.0	46	140	6,440
4	3.5	160	80	12,800	2.0	57	100	5,700	2.0	41	113	4,633
6	2.5	170	80	13,600	Trace				Trace	121	28	3,388
24	0	1,260	51	64,260	0	625	21	13,125	0	1,495	23	34,385
Totals		1,975		118,360		780		33,468		1,933.5		68,349
Per cent ..				24				6.6				14

Table 2 shows the serum levels and urinary excretions observed in three additional subjects following single intramuscular injections of 500,000 units of streptomycin suspended in 2 cc. of sterile peanut oil and 4 per cent beeswax. It will be observed that the serum level of the compound in one subject (No. 4) was 2.5 units/cc. at the end of 6 hours but that only traces of the compound appeared in the serum of the remaining two subjects at the end of this period. The total urinary excretion in 24 hours varied from 6.6 to 23 per cent of the dose administered. Similar doses dissolved in 2 cc. of sterile saline solution and administered to three adults by intramuscular injection showed serum levels of 1.0, 2.5, and 2.5 units/cc., respectively, at the end of 6 hours, with total excretions of 36.0, 41.0, and 75.0 per cent of the amounts administered in the 24-hour urine (*5*).

TABLE 3

Intervals	Subject No. 7				Subject No. 8				Subject No. 9			
	Serum (units/cc.)	Urine			Serum (units/cc.)	Urine			Serum (units/cc.)	Urine		
		Vol. (cc.)	Units/cc.	Total units		Vol. (cc.)	Units/cc.	Total units		Vol. (cc.)	Units/cc.	Total units
1	1.0	190	18	3,420	5.5				7.0	65	230	14,950
2	1.0	52	13	676	5.0				4.5	35	400	14,000
3	2.5	108	70	7,560	4.5	158	150	23,700	2.5	120	150	18,000
4	2.0	190	27	5,130	3.0				2.0	185	40	7,400
6	1.5	165	23	3,795	2.5				1.5	225	45	10,125
24	0	910	14	12,740	0	650	45	29,250	0	340	60	20,400
Totals		1,615		33,321		808		52,950		970		84,875
Per cent ..				13				21				34

As expected, the absorption of streptomycin administered in water-in-oil emulsions (solvecillin) was much more pronounced than that following intramuscular injections of the compound suspended in peanut oil and beeswax. As shown in Table 3, the serum levels in three subjects at the end of 6 hours following intramuscular injections of 250,000 units in solvecillin were 1.5, 2.5, and 1.5 units/cc., respectively, and closely similar to the results observed following the intramuscular injection of 250,000 units dissolved in 2 cc. of sterile solution. The total excretions in the 24-hour urine varied from 13 to 34 per cent of the amounts of streptomycin administered.

It appears, therefore, that streptomycin suspended in peanut oil and beeswax is not absorbed from the muscles as readily as penicillin. Similar results have been observed in the administration of streptomycin suspended in peanut oil and 3 per cent beeswax to guinea pigs by intramuscular injection (*2*). This slow absorption probably results in a greater fixation or inactivation of streptomycin in the tissues, accounting for the low serum levels and reduced urinary excretions. Maintenance of minimum effective serum levels over a period of 6 hours requires the injection of more than 500,000 units per dose. Absorption after intramuscular injections of the compound emulsified in solvecillin is more pronounced and similar to that following injections of the compound dissolved in saline solution, but serum levels following intramuscular injections of 250,000 units in solvecillin were not

as high as those following intramuscular injections of 500,000 units suspended in peanut oil and beeswax.

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Effect of Di-Isopropyl Fluorophosphate (DFP) on the Action Potential of Muscle

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Di-isopropyl fluorophosphate (DFP) abolishes the nerve action potential, but this effect is reversible for a limited period of time. DFP is a strong inhibitor of cholinesterase. A striking parallelism between the reappearance of the action potential and cholinesterase activity in nerve can be demonstrated during the recovery from DFP poisoning. The inhibition of cholinesterase by DFP can also be reversed *in vitro* during a period of time comparable to that in the experiments on nerves (*1*). These observations are consistent with the concept that cholinesterase activity and, consequently, the release of acetylcholine are essential events in the conduction of the nerve impulse.

It is generally considered that the mechanisms of the axonal and end-plate potentials are basically identical (*4*). The role of acetylcholine in the end-plate potential is supported by the relation between the activity of cholinesterase and the action potential of the electric organ, which may be considered analogous to the end-plate potential (*5*). Further support is found in the persistence of the high cholinesterase concentration at the motor end plate after complete degeneration of the axon (soleplate) (*2, 3*).

No facts are available concerning the chemical mechanism involved in the action potential of muscle, although many physiologists believe that the electrical manifestations of nerve and muscle are fundamentally identical. Muscle fibers and nerves are the only tissues which contain specific cholinesterase (*6*). This makes possible the assumption that acetylcholine plays a role in both tissues. However, the presence of an enzyme alone does not permit an interpretation of its function. We have therefore tested whether DFP abolishes the action potential of muscle as it does that of nerve.

Frogs (*Rana pipiens*) were curarized with crystalline *d*-tubocurarine chloride (Squibb). After curarization, the sartorius muscle was excised and mounted in a specially constructed chamber. The action potentials evoked by single electrical stimuli were recorded by means of a cathode-ray oscillograph. When not stimulated, the muscle was kept immersed in a Ringer solution containing 0.1 mg./cc. of curarine. This solution was then replaced by an identical solution containing DFP. Under the effect of DFP the action potential of the muscle rapidly disappears. With a concentration of 2 mg. of DFP/cc., the action potential is abolished in as little as 8 minutes. With a concentration of 1 mg. of DFP/cc., the abolition takes longer (20–30 minutes). After washing with the curarine Ringer solution (without DFP) the reappearance of the response is observed.

These experiments present the first evidence that acetylcholine may play a role in the muscle action potential. They are consistent with the idea that the natures of axon, end-plate, and muscle action potentials are basically identical.

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Derivation, Interpretation, and Application of the Second Law of Thermodynamics

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The second law of thermodynamics is commonly derived by means of an extended series of differential equations not easy to follow and involving so many assumptions and limitations that the result is not altogether convincing. Its physical interpretation is given in a wide variety of statements. That a number of the fundamental relations of thermodynamics may be simply and directly derived from it appears to have been entirely overlooked. It is here derived as a by-