potential in Lead II was reduced, and the QRS in Lead III changed from an RS type to an R type, an indication of axis deviation.

A clearly apparent increase in the QRS interval appeared in the record of 19 March 1946. The QRS in Lead II also changed from an RS type to an R

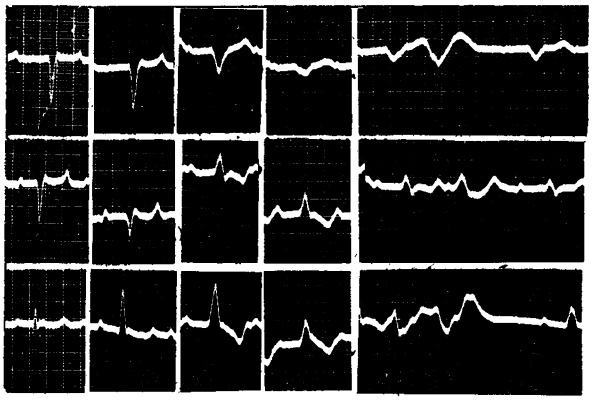

FIG. 1. Electrocardiograms of E541 on dates indicated. (Leads I, II, and III, top to bottom in order.)

type. In the record of 26 March 1946 the potential of the various deflections has decreased and remains so in the subsequent recordings.

In general, the electrocardiograms obtained on this animal appear to show a decreased functional activity of the myocardium in the terminal stages of the deficiency, as indicated by the decrease in the potential of the deflection of the QRS complex and by the increase in duration of the P–R, QRS, and Q–T intervals. The extra systoles which are apparent in the last record indicate dissociation of atrial and ventricular impulses and possibly damage to the conducting tissue. As has been stated, there also was a change in the electrical axis of the heart as the deficiency progressed.

Microscopic studies of heart sections, especially involving the Purkinje network of this and other animals in the study, are being made. It can be stated, even though this work has not been completed, that definite abnormalities have been noted. Atrophy and scarring of the cardiac muscle fibers is clearly indicated. An increase in cellular elements is noted, in some instances strikingly resembling, though smaller than, the Aschoff nodules seen in human endocarditis.

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Influence of Purified Lignin on Nitrification in Soil [1]

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The study reported here was preliminary to a more extensive study planned to investigate the effect of plowed-under crop residues on the nitrifying process in soil. The object of this preliminary study was to determine the effect of purified lignin on the nitrification of dried blood and of ammonium sulfate in soil.

It is an accepted fact that plant residues of high lignin content decompose slowly in soil because of the resistance of lignin to decay; and lignin in soil has been found to interfere with the natural process of breaking down other organic matter. In particular, lignin depresses protein decomposition. Among the authorities who may be cited in support of this contention are Waksman and Iyer (*7*), Waksman and Hutchings (*6*), and Osugi and Endo (*3*).

In the first of these references the statement is made that the action is not toxic but is an interaction between lignin and protein which results in the formation of a complex "humus nucleus" of lignin and protein. In the second reference it is stated that lignin acts as a buffer to absorb NH_3 and combines with protein to form a complex that is highly resistant to decomposition. Smith and Brown (*4*) state that lignin does not possess antiseptic property but that it does decompose very gradually; while Norman (*2*) states that isolated lignin apparently has bacteriostatic action.

EXPERIMENTAL METHOD

The purified lignin employed in this study was an "alkali" lignin prepared by treating ligneous plant residue with sodium hydroxide, followed by electrodialysis of the product. The author is indebted for its preparation to Emmett E. Bennett, of the Massachusetts Agricultural Experiment Station.

The soil employed was Connecticut Valley sandy loam. The plot from which the soil was taken had been used previously for the cultivation of tobacco, but had been allowed to lie fallow during the season in which the soil was taken in the late summer for this investigation.

The soil was brought into the laboratory, where it was air-dried and screened through a 40-mesh screen. Following determination of its water-holding capacity, 100-gram quantities were put into glass tumblers and materials added as follows: calcium carbonate; calcium carbonate and mannite; calcium carbonate, mannite, and dipotassium phosphate to furnish

[1] Contribution No. 571 of the Massachusetts Agricultural Experiment Station.

phosphorus and potassium. The quantities of these added substances per 100 grams of soil were: mannite, 1 gram; calcium carbonate, 0.2 gram; and dipotassium phosphate, 0.3 gram. Calcium carbonate was added because the reaction of the soil as taken from the field was about pH 5; mannite, to observe the effect of an added source of energy; and phosphorus and potassium, because of the known lack of these materials in the type of soil employed. The amounts of these several substances were based upon previous experience in this laboratory with this type of soil.

TABLE 1

NITRIFICATION OF DRIED BLOOD AND AMMONIUM SULFATE IN SOIL WITH AND WITHOUT PURIFIED LIGNIN

Treatment	Incubation period	Dried blood series		Ammonium sulfate series	
		Soil without lignin; ppm nitrate nitrogen	Soil with lignin; ppm nitrate nitrogen	Soil without lignin; ppm nitrate nitrogen	Soil with lignin; ppm nitrate nitrogen
	(mos.)				
Calcium carbonate	2	528	416	45	26
	3	728	432	51	40
	4	691	365	46	45
Calcium carbonate and mannite	2	480	297	46	27
	3	485	328	44	29
	4	472	310	41	29
Calcium carbonate, mannite, and dipotassium phosphate	2	272	273	36	25
	3	305	267	38	28
	4	269	237	40	31

Two sets of tumblers were prepared of each mixture. To one set purified lignin was added at the rate of 2 grams/100 grams of soil. To the duplicate set no lignin was added.

Two series of experiments were set up. In one, dried blood (about 12 per cent nitrogen) was added at the rate of 1 gram/100 grams of soil; in the other, an aqueous solution of ammonium sulfate was added at a rate to supply 30 mg. of nitrogen/100 grams of soil. After all of the materials had been thoroughly mixed in the respective tumblers, distilled water was added to 60 per cent of the water-holding capacity of the soil. The ammonium sulfate was added by dissolving it in the water used to moisten the soil. This technique for nitrification study is essentially that employed by Waksman (*5*).

After the water had been added, the tumblers were covered with waxed paper and weighed. During the incubation period the tumblers were weighed twice each week, and water was added to make up any loss. The tumblers were put into dark cupboards and allowed to stand at room temperature. One-third of the tumblers from each category were tested for nitrates at two months, one-third at three months, and the final third at four months. Nitrate determinations were made colorimetrically by the phenoldisulfonic acid method (*1*).

The experiments were so set up that triplicate tumblers were available for each determination. The results, shown in Table 1, were nearly enough alike for each triplicate set of tumblers to justify the use of averages.

RESULTS

In the tumblers containing dried blood the results showed that:

(1) In the presence of calcium carbonate the amount of nitrate in tumblers without lignin was much greater than when lignin was present.

(2) When both mannite and calcium carbonate were present, the same relationship existed as with calcium carbonate alone, but nitrate values for both lignin and nonlignin tumblers were lower than in the comparable tumblers without mannite.

(3) When dipotassium phosphate was added to calcium carbonate and mannite, all nitrate values were much lower than with calcium carbonate alone or with calcium carbonate and mannite. Nitrate values in the absence of lignin were not much greater than in its presence; in fact, they were about the same at the end of the first two months.

(4) Most nitrate values increased from two months to three and then decreased at four months. In some instances the values at four months were lower than those at two months.

(5) In all instances, except the one mentioned in (3), the values in the presence of lignin were definitely lower than in its absence.

In the tumblers containing ammonium sulfate, the nitrate values in the presence of lignin were lower than in its absence. The length of the incubation period exerted little influence, except for the slight influence shown in the presence of calcium carbonate alone, which resembled the effect noted with dried blood but to a much smaller extent. The presence of mannite and dipotassium phosphate did not exert the noticeable effect obtained with dried blood.

The recovery of nitrate nitrogen was relatively much less in the ammonium sulfate series than in that with dried blood. This may have been due to the absorption of NH_3 by lignin, mentioned by Waksman and Hutchings (*6*).

The reason is not apparent for the lower nitrate results obtained when mannite and dipotassium phosphate were added to soil in the dried-blood series. A check of the pH values of the soil in the tumblers gave no information that could explain the phenomenon.

CONCLUSION

It may be concluded that the presence of purified

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.