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Virulence, Electrophoresis, and Conversion Characteristics of Bact. phaseoli sojense, S and R.

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The occurrence of "Rough" and "Smooth" strains among bacteria pathogenic for plants has been reported, and the conversion of R to S forms by passage through plants described. The conditions associated with conversions of R to S and of S to R strains have been further investigated.

S to R Conversion: The culture of Bact. phaseoli sojense used in these experiments grows best at a temperature of 30° C. At this temperature, R and S strains are stable even after 5 months' incubation. When held at higher temperatures (up to 37.5° C.) S strains are transformed (not completely) to R. Growth in nutrient broth of hydrogen ion concentration between pH = 5.2 and 8.8 is without effect in changing the colony form. Cultivation in broth containing 1 to 20% homologous (S) anti-serum (rabbit) results in conversion of S to R (not completely); but in broth containing the heterologous (R) anti-serum, normal serum or no serum, no change is effected. If rapid serial transfers be made in broth containing 10% anti-S serum, the conversion of S to R forms is rapidly effected and is carried almost but not quite to completion. Similar conversion is effected by growth in peptone water containing from 2 to 15% peptone and is probably complete in the 10-15% solutions. Rapid transfer in nutrient broth or on agar slants or plates was without demonstrable effect.

R to S Conversion: Normal or rapid transfers in broth or on agar slants or plates did not effect conversion. Cultivation in the presence of homologous (R) anti-serum results in rapid but not complete R to S changes and serial transfer in the presence of 10% of this serum in the broth results in nearly complete transformation of the culture; whereas growth in the broth alone or in the broth containing normal or heterologous (S) anti-serum effects no change. A striking and nearly or entirely complete conversion of R to S is effected by growth on glucose agar. This conversion does not occur in glucose broth or other fluid glucose media (except when rapidly transferred), nor in solid or fluid media containing other sugars.

<sup>&</sup>lt;sup>1</sup> Cf. Sharp, C. G., Bot. Gaz., 1927, lxxxiii, 113; also Link, G. K. K., Chap. xliv, and Falk, I. S., Chap. xlii, in "The Newer Knowledge of Bacteriology and Immunology," edited by E. O. Jordan and I. S. Falk, Chicago, 1928.

TABLE I.

Virulence for Manchu Soy Bean Plants Infections in Leaf Punctures.

Strain	3 days	% infection in 4 days	5-7 days	Average Size of Lesions
R	31	49	61	mm. 1.37
RD S	52 5 <b>7</b>	64 78	92 98	1.65 1.72

57		78			98	
TABLE II. Agglutination of S, R, R D Types by S Immune Sera.		Control	111		Control	111
		5120	++1		5120	111
		2560	++1 1		2560	+1 ] ]
	1280	++++	Agglutination of R, RD, S Types by R Immune Sera	1280	+	
	640	++ ++ ++ ++		640	+++1	
	320	++ +++ +++ ++		320	+++++++++++++++++++++++++++++++++++++++	
	160	+++ +++ +++		160	+++ +++ +++	
	80	++++ +++ ++++ ++++ ++++ ++++ ++++ ++++ ++++		80	+++ +++ +++ +++ +++ +++ +++ +++ +++ ++	
	Aggluti	40	+++ +++ +++	Agglut	40	+++
	20	+++ +++ +++		20	+++	
		10	+++ +++ +++		10	+++
		Final serum dilutions	R C R		Final serum dilutions	3 3 5 5 8 7 8 8

Although stable, the R colonies differ in morphology when grown on solid media containing different sugars.

The R strain which has been converted to an S strain by growth on solid glucose media gives an unusually large colony and is subsequently stable and characteristically an S culture when carried on media containing or lacking glucose. The R to S transformation is effected in the presence of 0.3 to 20.0% glucose; but the colony is smaller in the presence of the highest concentrations of sugar. The effectiveness of the glucose is apparently not dependent upon impurities, as evidenced by experiments upon 5 commercial brands of the sugar of varying degrees of purity.

The R strains which have been converted to S by growth on glucose (dextrose)-containing medium, called the RD strains, show virulence, agglutination and electrical characteristics more like the typical S than the typical R strains. Tables I and II illustrate some experiments.

In agglutination tests with homologous and heterologous sera, results like those in Table II are obtained.

The electrophoretic charges or potential differences for 10 strains of each R, R D and S cultures are illustrated in Table III.

TABLE III. Electrophoretic Potentials in  $\mu/\text{sec./volt/cm.}^2$  (Migrations to the anode.) Strain R 2.66 R D 4.04

In one experiment, S and R D cultures were found to be virulent for mice and the R cultures were not virulent. But it has been difficult to duplicate this experiment. Hence its significance is still uncertain.

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## Experimental "Food-Poisoning" in Monkeys with Living Paratyphoid Bacilli.

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Previous experiments in this laboratory have shown: (1) that the symptoms of food-poisoning were not reproduced when enormous

<sup>&</sup>lt;sup>2</sup> Using the "slide cell" method of Falk, I. S., Jensen, L. B., and Mills, J. H., J. Bact., 1928, xv, 421.