

found indole to have an antiseptic effect ranging from 2 to 10 times that of carbolic acid, the efficiency depending upon the kind of bacteria exposed to its action. *B. coli* was one of the most resistant organisms but failed to grow in the presence of 0.1% indole. Gordon and McCleod believe that indole production may well be the mechanism by which *B. coli* maintains its dominant position in the fecal flora. During a study of the production of indole by fecal bacteria, information was desired concerning the effect of indole upon the growth of bacteria.

The medium used for the tests was 2% Witte's peptone water which had been adjusted to yield a pH of 7.6 after autoclaving. A solution of indole 1-1000 was prepared and filtered by pressure through a Seitz filter. Various amounts of this solution were added to 50 cc. portions of sterile peptone water. The flasks of medium were then inoculated with the organism to be studied by transferring a loopful of a 24-hour culture in 2% peptone water. All of the test cultures were incubated at 37° C. for 24 hours after which counts were made by the plating method.

The results obtained are shown in Table I. The growth of all of the microorganisms studied was hindered by the presence of indole in the culture medium. *Escherichia coli* was the most resistant of the organisms studied. Nevertheless its growth was prevented in the presence of 0.1% indole. *Salmonella paratyphi* was apparently very susceptible to the action of indole. Each result shown in the table was obtained by the analysis of separate cultures.

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Effect of Echinid Egg-Waters on the Surface Potential Difference of the Sperm.

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The egg-waters of the sand-dollar (*Echinarachnius parma*), starfish (*Asterias forbesii*), and sea-urchin (*Arbacia punctulata*), have been tested by electrophoresis for their effects on the sperm of each of these 3 species. Egg-water was routinely prepared by allowing freshly obtained eggs to stand a half-hour or more in contact with 0.5 M. NaCl adjusted with NaOH to an approximate pH of 8.2.

This solution was filtered and mixed in equal volumes with suspensions of fresh sperm in 0.5 M. NaCl of pH 8.2. In 2 experiments the egg-water and sperm suspensions were both made with sea-water instead of NaCl solution. Later experiments showed that the exposure to 0.5 M. NaCl of pH 8.2 did not kill the sperm or the eggs from which the egg-waters were derived.

For agglutination the egg-water used was the same as in the corresponding electrophoresis experiment, but the sperm were suspended in sea-water. The agglutination technique was essentially that of Lillie and Just.¹

In the earlier experiments precautions to obtain the sperm and eggs free from perivisceral fluid were not taken; in the later experiments such precautions were taken. Differences in result due to such precautions were not noted.

Electrophoresis was conducted in a Northrop-Kunitz microcataphoresis cell² of a type described elsewhere.³ Observations were made on sperm whose heads were in the focal plane.

Each of the sperm species studied was found to be negatively charged. In Table I are given the mean cataphoretic velocities with their probable errors, and the surface potential differences calculated from these velocities making the assumptions of Northrop and Cullen.⁴ The values in Table I are all for control sperm suspensions before contact with egg-water.

TABLE I.

Sperm species.	Suspending medium†	Cataphoretic velocity μ/sec. per volt/cm.	Sperm surface p.d., millivolts
Sand-dollar	0.5 M. NaCl, pH 8.2	1.71 ± .03	21.6 ± .38
Star-fish	" " " "	1.77 ± .05	22.3 ± .63
Sea-urchin	" " " "	2.10 ± .03	26.5 ± .38
Sand-dollar	Sea-water	1.29*	16.3
Star-fish	" "	1.44*	18.1
Sea-urchin	" "	1.75*	22.0

† See Loeb, J., *Am. Nat.*, 1915, xlix, 257.

* 4 determinations only.

The effects of the egg-waters on the sperm surface potential differences are given in Table II.

Sand-dollar egg-water caused a specific *increase* in the negative

¹ Lillie, F. R., and Just, E. E., "Fertilization" in Cowdry, E. V., *General Cytology*, Chicago, 1924.

² Northrop, J. H., and Kunitz, M. J., *J. Gen. Physiol.*, 1924-5, vii, 729.

³ Mudd, S., Lucké, B., McCutcheon, M., and Strumia, M., *Colloid Symposium Monographs*, New York, 1928, vi, 131.

⁴ Northrop, J. H., and Cullen, G. E., *J. Gen. Physiol.*, 1921-2, iv, 638.

TABLE II.

Egg-water species	Sperm species	Sperm P.D. increased. No. expts.	Sperm P.D. reduced. No. expts.	Average change
Sand-dollar	Sand-dollar	7	1	% +20
" "	Sea-urchin	2	4	— 2
" "	Star-fish	4	4	— 2
Sea-urchin	Sand-dollar	10	1	+33
" "	Sea-urchin	7	2	+13
" "	Star-fish	8	2	+20
Star-fish	Sand-dollar	4	3	0
" "	Sea-urchin	0	7	—14
" "	Star-fish	4	3	+ 6

A mean increase in the negative charge is indicated in column 5 as +, a reduction in the negative charge as —.

charge of sand-dollar sperm and was without significant effect on the sperm of sea urchin or starfish. In correlation with this the sand-dollar egg-water caused specific isoagglutination of sand-dollar sperm, to an average dilution of the egg-water of about 1:50.

Sea-urchin egg-water *increased* the negative charge of sea urchin sperm and caused reversible sperm agglutination of the iso-type in an average dilution of the egg-water of about 1:1300. However, sea-urchin egg-water caused an even greater increase in the negative charge of star-fish and sand-dollar sperm, but caused agglutination only of the hetero-type, and this only in high concentrations of the egg-water.

Star-fish egg-waters regularly *decreased* the negative p.d. of the highly charged sea-urchin sperm, showed a somewhat questionable mean *increase* in charge of homologous sperm and showed no mean effect on sand-dollar sperm. Agglutinative effects with star-fish egg-waters were slight.

The only conclusions we care to draw from the work at its present stage is that echinoderm egg-waters contain substances which may combine with substances in the sperm surfaces, and that such combinations can be detected by cataphoresis.