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INVESTIGATIONS

THE UNIVERSITY OF CHICAGO
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INVESTIGATIONS REPRESENTING
THE DEPARTMENTS

ZOÖLOGY ANATOMY PHYSIOLOGY NEUROLOGY
BOTANY PATHOLOGY BACTERIOLOGY

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THE SELF-PURIFICATION OF STREAMS

THE SELF-PURIFICATION OF STREAMS

EDWIN O. JORDAN

It has long been a popular belief that "running water purifies itself," and that a polluted stream again becomes pure after flowing for some distance from the point of defilement. This widespread belief appears to be based on the evidence of the senses and to depend largely upon naked-eye inspection of the flowing stream. The most casual observer can often detect evidences of recent pollution at or immediately below the point where fouling of the water occurs, but farther down stream these indications of contamination become less striking, and farther still every trace of them vanishes.

Such observations are, however, open to misinterpretation. The mixing of the befouled water with the purer water of the river becomes more complete the greater the distance from the source of pollution, and when the increase in the volume of the stream from tributaries, and especially from underground sources, is also taken into consideration, a sufficient explanation is afforded for the changes observed. The thorough mingling of the polluting matters with the total volume of water in the stream, combined with a continuous and often rapid increase in the volume of the stream itself, may dilute sewage to such a degree that it seems as if the polluting substances had been materially lessened or had altogether disappeared. It is quite evident, however, that, while such dilution may improve the general appearance of a river water, it does not by any means follow that noxious elements introduced with the pollution are destroyed. Typhoid bacilli, for example, that enter a sewage-polluted river may persist in the river water, so far as the unaided senses can determine, for many miles below the point of entrance, although the water of the stream may to all outward appearances have quickly regained its pristine purity. Dilution in itself can neither remove nor destroy dangerous substances; it may, in fact, mask their presence.

A necessity arises, therefore, for a more searching study of the conditions existing in a sewage-polluted river. One method of investigation that has often been applied to this study consists in a determination of the chemical changes that occur in the flowing water. The amount of organic matter in water, especially the organic nitrogenous matter, can be determined by analytical processes of great delicacy and precision. It is found that fresh sewage or sewage-polluted water contains a relatively high proportion of organic nitrogenous matter in an unstable condition. "Organic nitrogen," or "albuminoid ammonia," and "free ammonia" are present in great abundance in the water of streams freshly contaminated with sewage. The putrescible nature of these substances, due to their liability to offensive bacterial decomposition, imparts to polluted water many of its characteristic qualities. Analyses have shown that a diminution in the amount of these organic substances occurs during the pas-

sage of the water down stream, and that hand in hand with the lessening of "organic nitrogen" and "free ammonia" in the water there is to be noticed first an increase in "nitrites" and then in "nitrates," until eventually a large part of the unstable organic nitrogen entering the river in sewage becomes oxidized into the stable mineral condition of nitrate. When the nitrogen has reached the state of nitrate—that is to say, when nitrification is complete—the river water may be said to be chemically purified, and, unless fresh organic matter is introduced or is formed by algal growth, is no longer capable of giving rise to offensive decomposition products.

This process of chemical purification is an important one, and is most influential in restoring a polluted stream to its original condition. Since the oxidation of the free ammonia and nitrites, as well as of the organic nitrogen, is due to bacterial activity, there is a falling off in the number of bacteria as their food-supply lessens; and thus it happens that in a general way the lines of chemical and bacterial purification follow a parallel course. In a freshly polluted river the number of bacteria found in one cubic centimeter of the water may be as high as several millions, while in the same river, after oxidation has taken place and the nitrogen has passed into the fully oxidized condition, the bacteria may number only a few thousand. It need hardly be pointed out perhaps that dilution tends to reduce the proportion both of organic matter and of bacteria in a given quantity of water. The evidence is, however, entirely convincing that, in addition to this relative diminution, there is also an absolute loss in the quantity of organic nitrogen and an absolute destruction of bacterial life in the flowing stream.

It may properly be asked: What is the precise sanitary significance of these changes that have just been described? Are the rate of oxidation of nitrogenous organic matter and the death-rate among river bacteria to be taken as legitimate criteria of an increasing wholesomeness of the water? The question is difficult to answer. Under ordinary conditions in this country the chief water-borne disease is typhoid fever. Stripped of all technicalities, the real question at issue on the sanitary side in the self-purification of streams is this: How far can typhoid bacilli travel in a flowing stream?

It is probably true that the increasing freedom of river water from typhoid germs as the point of pollution is left behind corresponds roughly with the increase in nitrates and the diminution of free ammonia and nitrites in the water; but there is no necessary connection. The observation that there is often coincidence between the state of the nitrogenous constituents in a water and the wholesomeness of that water is due to the fact that in a recently polluted water not only free ammonia and nitrites are likely to be present, but also typhoid bacilli. In a water in which pollution is more remote both free ammonia and typhoid bacilli are more likely to have disappeared.

The actual number of bacteria of all kinds in a river water is possibly a more warrantable standard of the degree of purification that has occurred than the state of the chemical substances in such a water. There is no escape from the conclusion that the duration of life of the ordinary sewage bacteria, when these are introduced into

water, measures the probable duration of life of the typhoid bacillus with greater accuracy than does the progressive oxidation of organic nitrogenous substances. At the same time it must be remembered that both methods are inferential only. There is no reason for believing that either such amounts of "organic nitrogen" or of "nitrites," or such numbers of bacteria as are ordinarily found in a polluted river, are in themselves directly harmful. Their presence and abundance simply furnish indications, more or less cogent, for gauging the probability of occurrence of typhoid bacilli.

In connection with the writer's study of the conditions attending the opening of the Chicago Drainage Canal, it became necessary to review the evidence for the self-purification of streams and to consider the practicability of applying to the study of the problem other methods than those just cited. The direct and ideally preferable method would consist in determining the proportion of typhoid bacilli in sewage, and then tracing the fate of these bacilli in the water course; but this is unfortunately not applicable, owing to the practically insurmountable difficulty of rapidly isolating and identifying the typhoid bacillus in the presence of large numbers of sewage and water bacteria. Several existing methods for the isolation of the typhoid bacillus from water were tested and proved entirely inadequate for this purpose.

Another method was accordingly employed, which has yielded results of interest and value. This consists in a determination of the relative abundance of *B. coli communis* in the river water at various points. *B. coli*, as is well known, occurs in large numbers in fresh sewage, and its presence can easily be detected by appropriate tests. The peculiar importance attaching to the fate of this microbe in a flowing stream lies in its close biological relationship to the typhoid bacillus, and in the fact that, like the typhoid bacillus, it enters river water with sewage. It is, moreover, invariably present in sewage in much larger numbers than the typhoid bacillus. Nearly all of the colon bacilli and probably all of the typhoid bacilli found in sewage pass directly into the sewage from the human body. Conditions, therefore, that affect the abundance of the colon bacillus in water are likely to affect that of the typhoid bacillus also.

It is not necessary to rehearse the general circumstances of the investigation, since these have been set forth with sufficient detail elsewhere.¹ References to the accompanying figure (Fig. 1) will show the principal points of collection of water samples. The methods employed for the detection of the colon bacillus must, however, be briefly considered. The necessity for handling a large number of water samples in a limited time led to the use of some method which could be applied with a fair degree of exactness to routine work. Both the carbol-broth method² and the fermentation-tube method³ have been used. In much of the earlier work the cultures isolated by these two methods were worked out in detail, so that complete identification

¹*Journal of Experimental Medicine*, Vol. V (1900), p. 271.

²*Journal of Hygiene*, Vol. I (1901), pp. 295-320.

³SMITH AND BROWN, "Report on Mohawk and Hudson Rivers," *Thirtieth Annual Report of the State Board of Health of New York*, 1893, p. 680.

of the bacterial species was secured. The exigencies of the later work brought about the partial abandonment of this procedure and compelled the adoption of a rough field method by means of which large numbers of water samples could be treated with a limited margin of error. The dextrose fermentation tube proved the most available for this purpose, experiments with neutral-red⁴ and other methods not yielding, on the



FIG. 1

whole, as satisfactory results. The fermentation tube was used in this way to some extent in routine work in 1899–1900, and was employed as the sole method in a more extended series of tests in the autumn of 1901. The interpretation adopted for the changes produced by inoculating water into the fermentation tube was as follows: Positive reactions—*i. e.*, those indicating the presence of *B. coli*—were regarded as those tubes showing gas production amounting to over 20 per cent. of the tube length, the tubes yielding on absorption with NaOH a gaseous residue (H) appreciably in excess of the CO₂ absorbed; negative reactions were those showing (a) no gas production, or (b) gas production less than 10 per cent. of the tube length; the doubtful class was made to include (a) those tubes yielding only 10–20 per cent. of gas, and (b) those yielding more than 20 per cent., but with an appreciable excess of CO₂.

⁴ E. E. Irons, *Journal of Hygiene*, Vol. II (1902), p. 314.

Any method susceptible of rapid application, like the one outlined, leads to occasional misinterpretation, but the following data indicate that where a large number of water samples are treated the error is not unduly large. The organisms were isolated from a series of fermentation tubes and studied in detail, and complete identification was arrived at. The procedure is illustrated in Table I:

TABLE I

Date collected	Date examined	Source ⁵	Sample number	Amount of water	GAS PRODUCTION ⁶		Absorption H-CO ₂	Culture. Number of org. iso.	Interpretation	Organisms isolated
					24 hours	48 hours				
Oct. 23	Oct. 23	Three	1039	1 c.c.	10	30	2-1	1039 x	+	+ B. coli
25	25	Four	1044	1-100 c.c.	16	32	1-1	1044 y	?	- B. cloacæ
28	28	Eight	86	1 c.c.	25	25	4-1	86 y	+	- B. proteus
30	30	Two	2032	1-1000 c.c.	38	43	2-1	2032 x	+	+ B. coli
Nov. 4	Nov. 4	One	2039	1-10 c.c.	36	62	1-2	2039 x	?	- B. cloacæ
4	4	Four	1060	1-10 c.c.	20	85	2-1	1060 x	+	+ B. coli
8	8	Two	2048	1-1000 c.c.	42	67	3-1	2048 z	+	+ B. coli
19	19	Six	180	1 c.c.	5	95	1-1	180 z	?	+ B. coli
19	19	Seven	179	1 c.c.	20	45	1-1	179 z	?	- B. cloacæ
Oct. 11	Oct. 11	Three	1019	1-10 c.c.	5	8	-	1019 x	-	- B. proteus
Nov. 20	Nov. 20	Two	2065	1-1000 c.c.	20	35	3-1	2065 x	+	+ B. coli
22	22	One	2071	1 c.c.	5	15	No absp.	2071 x	?	- B. proteus
16	16	Four	1082	1-10 c.c.	15	25	4-1	1082 z	+	+ B. coli
Dec. 5	Dec. 5	One	2091	1-10 c.c.	82	82	2-1	2091 x	+	+ B. coli
16	16	One	2109	1 c.c.	90	94	2-1	3109 x	+	+ B. coli
27	27	One	2127	1-10 c.c.	5	22	2-1	{ 2127 y 2127 y	+	{ + B. coli B. proteus

Sixty-three tubes were examined in this way, with the following results:

- 39 Interpretation positive. Typical B. coli isolated.
 7 Interpretation positive. B. coli not isolated.
 9 Interpretation doubtful. B. coli not isolated.
 4 Interpretation doubtful. Typical B. coli isolated.
 4⁷ Interpretation negative. B. coli not isolated.

The satisfactory application of this method to the problem of the self-purification of streams depends upon ascertaining in each case the dilution at which the test gives uniformly positive and that at which it gives uniformly negative results. For illustration, sixty-nine separate examinations of .001 of a cubic centimeter of the Mississippi river water at Grafton were made in October-December, 1901, and in no case was the presence of B. coli detected in this quantity of water. Unless the negative limit is ascertained on each day, it is apparent that the results lose almost all their significance. The mere fact that B. coli is "present" in 1 c.c. of water is without meaning

⁵ See Fig. 2, p. 11.

⁶ Percentage of tube length.

⁷ These were all cases showing less than 10 per cent. of

gas in which gas-forming organisms — not B. coli — were isolated. In addition, some twenty tubes were examined which showed growth in the closed arm, but yielded no gas-producing organism of any kind.

unless at the same time it is shown to be absent in a small amount (.1 c.c.) of the same water. On the other hand, any attempt to reduce the findings to an exact numerical basis, even if it were possible, would be of doubtful value. No sanitary importance can be attached to slight variations in the colon content, such a difference, for instance, as between thirty and forty per cubic centimeter being probably devoid of significance; but the greater diversity revealed by the decimal dilution is unquestionably fraught with meaning. The method here used has the advantage of recording the more con-

TABLE II
ILLINOIS RIVER (AVERYVILLE)
Presence of Bacteria of the Colon Group

DATE	SERIAL No.	.001 c.c.	.01 c.c.	.1 c.c.	1 c.c.	DATE	SERIAL No.	.001 c.c.	.01 c.c.	.1 c.c.	1 c.c.
		+ - ?	+ - ?	+ - ?	+ - ?			+ - ?	+ - ?	+ - ?	+ - ?
Oct. 19	2013	1	2	1	Nov. 27	2079	1	3
	21	1	2	1 1		29	2081	2	2
	22	1	1 1	1 1		30	2083	1	1 1
	23	3	1 1	Dec. 2	2085	1 1	1
	24	3	2		3	2087	1	2
	25	3	2		4	2089	1	1 1
	26	2	2		5	2091	1	2
	28	3	1 1		6	2093	1	2
	29	1	2	1		7	2095	1	2
	30	2	2		9	2097	1	1 1
	31	2	2		10	2099	1	2
Nov. 1	2035	1 1	2		11	2101	1	1 1
2	2037	2	2		12	2103	1	1 1
4	2039	1 1	1 1		13	2105	1	1 1
5	2041	2	1 1		14	2107	1	2
6	2043	2	1 1		16	2109	1	1 1
7	2045	2	1 1		17	2111	1	1 1
8	2047	2	2		18	2113	1	1 1
9	2049	2	1 1		19	2115	1	2
11	2051	1 1	1 1		20	2117	1	2
12	2053	1	1 1	1		21	2119	1	1 1
13	2055	1	1 1	1		23	2121	1	1 1
14	2057	1	1 1	1		24	2123	1	1 1
15	2059	1	1 1	1		26	2125	1	2
16	2061	1	1 1	1		27	2127	1	2
18	2063	2	1		28	2129	1	1 1
19	2065	2	2		30	2131	1	1 1
20	2067	1	1 2						
21	2069	1	2 1		No. days examinat's	3	49	60	41
22	2071	1	1 1 1		No. days B. coli f'nd	0	3	38	34
23	2073	2	2		No. determinations	3	72	114	54
25	2075	2	1 1		No. positive results	0	4	49	39
26	2077	1	1 1		Per cent. pos. results	0%	6%	43%	72%



siderable differences and fluctuations in the colon content without obscuring the issue by a pretense to greater accuracy than can be obtained by any existing methods.

The mode of employment of the method may be shown by the record of fermentation-tube work upon the water of the Illinois river at Averyville (Table II).

Two series of determinations have been made in this way, one in 1899-1900, elsewhere described,⁸ and the other in 1901. The results may be set forth most clearly in tabular form (Tables III-VII):

TABLE III⁹
Principal Stations on the Illinois River, 1899-1901

COLLECTING STATION	.00001 c.c.		.0001 c.c.		.001 c.c.		.01 c.c.		.1 c.c.	
	No. of days water exam'd	No. of days B. coli found	No. of days water exam'd	No. of days B. coli found	No. of days water exam'd	No. of days B. coli found	No. of days water exam'd	No. of days B. coli found	No. of days water exam'd	No. of days B. coli found
Ill. and Mich. Canal, Lockport.....	28	7	32	28	11	8	4	4	2	2
Illinois river, Morris	3	1	20	11	30	20	23	20
Illinois river, Ottawa.....	22	6	34	19
Illinois river, Averyville.....	1	0	27	4
Illinois river, Wesley City.....	7	1	22	3	26	13
Illinois river, Grafton.....	4	1	35	13

TABLE IV⁹

Illinois River at Averyville and Grafton Compared with Tributaries and with the Mississippi (Grafton) and Missouri (West Alton) Rivers, 1899-1900

COLLECTING STATION	.01 c.c.		.1 c.c.		1 c.c.		5 c.c.	
	No. of days water exam'd	No. of days B. coli found	No. of days water exam'd	No. of days B. coli found	No. of days water exam'd	No. of days B. coli found	No. of days water exam'd	No. of days B. coli found
Illinois river, Averyville.....	1	0	27	4	31	13
Illinois river, Grafton.....	4	1	35	13	38	26	4	2
Mississippi river, Grafton.....	2	0	34	10	35	23	4	3
Desplaines river.....	8	1	5	2
Kankakee river.....	6	3	5	4
Fox river.....	22	2	23	6	13	10
Big Vermilion river.....	5	1	9	3
Sangamon river.....	13	4	25	14	27	21
Missouri river.....	6	3	32	13	31	21

⁸ See footnote 2, p. 5.

⁹ It will, of course, be observed that this method of summarizing the results is not altogether precise. The fact that on certain days and with certain dilutions more than one determination was made obviously implies the examination of a larger quantity of water at those times and the

increased possibility of a positive finding. The tabulation of the results on the basis of the total number of determinations is, however, open to objection on other grounds, and the method I have employed seemed to me on the whole to present fewer disadvantages.

TABLE V¹⁰
1899-1900

	.1 c.c.		1 c.c.	
	No. of days water exam'd	No. of days B. coli found	No. of days water exam'd	No. of days B. coli found
Total, Illinois river (Averyville and Grafton).....	62	17	69	39
Total, tributaries of Illinois river.....	66	21	69	36
Total, Mississippi and Missouri rivers.....	66	23	66	44

TABLE VI
October-December, 1901

COLLECTING STATION	.0001 c.c.		.001 c.c.		.01 c.c.		.1 c.c.		1 c.c.		2 c.c.		5 c.c.	
	No. days water examined	No. days B. coli found	No. days water examined	No. days B. coli found	No. days water examined	No. days B. coli found	No. days water examined	No. days B. coli found	No. days water examined	No. days B. coli found	No. days water examined	No. days B. coli found	No. days water examined	No. days B. coli found
Illinois river, Averyville.....	3	0	49	3	60	38	41	34
Illinois river, Pekin.....	17	4	44	14	44	40	17	17
Illinois river, Grafton.....	52	4	64	6	64	12	53	19	13	5	16	8
Mississippi river, Grafton....	52	0	64	6	62	25	54	25	12	4	16	11
Miss. r., Chain, Ill. shore.....	25	0	41	3	41	8	42	25	7	5
Miss. r., Chain, mid-stream...	27	0	41	4	41	16	42	35	7	6
Miss. r., Chain, intake tower..	27	0	43	11	42	30	43	36	6	6	5	5
Miss. r., Chain, Mo. shore....	27	1	41	8	41	24	41	36	4	4
Missouri r., Bellefontaine....	32	1	44	13	44	31	43	34	8	7

TABLE VII
October-December, 1901

COLLECTING STATION	.0001 c.c.			.001 c.c.			.01 c.c.			.1 c.c.			1 c.c.			2 c.c.			5 c.c.		
	No. determinations made	No. positive results	Percentage positive results	No. determinations made	No. positive results	Percentage positive results	No. determinations made	No. positive results	Percentage positive results	No. determinations made	No. positive results	Percentage positive results	No. determinations made	No. positive results	Percentage positive results	No. determinations made	No. positive results	Percentage positive results	No. determinations made	No. positive results	Percentage positive results
Illinois river, Averyville..	3	0	0	72	4	6	114	49	43	54	39	72
Illinois river, Pekin.....	25	7	28	75	16	21	77	60	78	17	17	100
Illinois river, Grafton....	72	5	7	117	6	5	125	16	13	71	21	30	15	5	33	17	8	47
Mississippi river, Grafton....	69	0	0	115	6	5	123	29	24	74	28	38	12	4	33	16	11	69
Miss. r., Chain, Ill. shore..	25	0	0	66	3	5	80	8	10	42	25	60	7	5	71
Miss. r., Chain, mid-stream	27	0	0	68	4	6	82	18	22	43	35	81	7	6	86
Miss. r., Chain, intake tower	27	0	0	71	14	20	84	48	57	43	36	84	6	6	100	5	5	100
Miss. r., Chain, Mo. shore..	27	1	4	68	9	13	81	33	41	42	37	88	4	4	100
Missouri r., Bellefontaine..	34	1	3	84	16	19	95	48	50	44	35	79	8	7	88

¹⁰ See footnote 9, p. 9.

The situation of the collecting stations on the lower Illinois, Mississippi, and Missouri rivers is shown by the accompanying map.

If any weight is to be attached to the relative abundance of the colon bacillus in river water, it is clear from the data here presented that the water of the Illinois river undergoes a real and very considerable purification. The colon bacteria which are

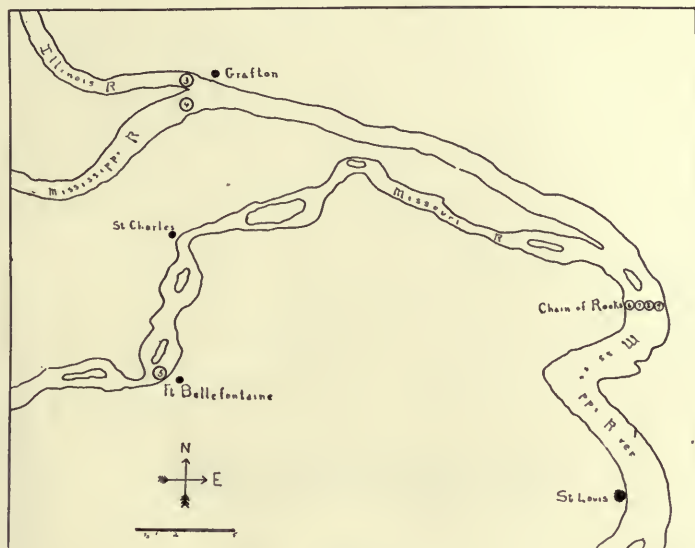


FIG. 2

present³ in such large numbers in Chicago sewage (*cf.* Table III) disappear almost completely in less than 150 miles' flow. At the mouth of the Illinois river, despite the enormous initial pollution and the very large secondary pollution at Peoria (Tables III, VI, and VII), the number of colon bacteria is certainly no greater than the number in the Mississippi river (*cf.* .01 c.c., Table VII), and perhaps not as large as the number in the Missouri river. Since all investigators are agreed that the colon bacillus is more hardy than its relative, the typhoid bacillus, and can live in water for a longer time, there is every reason for supposing that the latter microbe dies out with at least the same rapidity.

If it be true that the fate of the colon bacillus in running water furnishes the most satisfactory indication we can secure at present of the continuance of vitality of the typhoid bacillus, there can be no hesitation as to the conclusions to be drawn from our investigation. Since this near biological relative of the typhoid bacillus perishes speedily and in large numbers in the course of the Illinois river, there is reason to suppose that the typhoid bacillus itself does not long survive exposure to the same conditions.



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