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CULTURAL CHARACTERISTICS OF ZOOGLEA-FORMING BACTERIA ISOLATED FROM ACTIVATED SLUDGE AND TRICKLING FILTERS [1]

By ELSIE WATTIE, *Associate Bacteriologist, United States Public Health Service, Stream Pollution Investigations, Cincinnati, Ohio*

The isolation of a zooglea-forming bacterium, tentatively designated as *Zooglea ramigera*, from activated sludge has been described by Butterfield (*3*), who also reviewed the literature relating to such bacteria and their functions in sewage purification. Publications of the series of "Studies of Sewage Purification" emanating from the Stream Pollution Investigations Station of the United States Public Health Service at Cincinnati, Ohio, and papers of other laboratories interested in the same field have presented a large amount of data on the function of this type of bacteria in sewage purification processes. Very little information has been produced concerning the bacterial characteristics essential for purposes of description or of identification of species.

The studies of Butterfield et al. (*4*) indicate that the floc-forming organisms present in activated sludge will develop under aeration an activated sludge in sterile synthetic or sterilized normal sewage. A pure culture sludge, developed by the fill-and-draw-method, will oxidize about 50 percent of the 5-day biochemical oxygen demand (B. O. D.) during a 5-hour aeration period.

The purification accomplished by a pure culture activated sludge and a normal activated sludge has been found to be very similar by Ruchhoft and his associates (*11*). The rate and extent of total purification accomplished during a given period is influenced by the quality and quantity of activated sludge and the substrate in the aeration mixture.

The distinctive characteristic of these bacteria which has been emphasized is that they have the ability to grow in a floc, or colony, in a liquid medium even when they are subjected to agitation produced by the aeration sufficient to maintain aerobic conditions. This

[1] From the Division of Public Health Methods, National Institute of Health.

floc-forming ability is dependent on a gelatinous matrix or capsule which can be demonstrated about each cell. This gelatinous coating of the individual cells apparently becomes the binding agent in the floc.

The purpose of the gelatinous mass which binds the bacteria together is explained by Whitehead and O'Shaughnessy (*16*). From their experiments they concluded that the fine particles floating in sewage were held by the jelly-like mass and used by the bacteria either for food or as particles for attachment. Butterfield (*3*) used similar inorganic particles in his studies for bacterial floc attachment. However, the addition of cotton fibers or other inert particles for floc attachment in the development of pure culture sludges in synthetic sewage has been proven unnecessary. Within 48 hours after inoculating, well-developed flocs were formed in sterile synthetic sewage containing no inert material, as shown in figure 1. Microscopic examinations show the flocs are composed entirely of bacterial cells bound together by a gelatinous matrix.

The ability of zooglea-forming bacteria, isolated from activated sludge, to clarify sterile sewage either under aeration or while the sewage is quiescent has been studied by Heukelekian and Schulhoff (*9*). The characteristics of the organisms isolated are not given. Their results indicate that sterilized sewage inoculated with a floc-forming organism isolated from activated sludge will be clarified more quickly under aeration than when remaining quiescent. In either case, clarification did not exceed that of aerated raw sewage.

Dienert (*6*) isolated several types of bacteria from the zoogleal masses of activated sludge and trickling filters. He classified them as clarifying, reducing, and oxidizing organisms. The clarification of sewage was produced by a large coccus, but it was not obtained in pure culture. The rate of clarification was decreased after the sludge flocs had been mashed or pressed between glass slides and the zoogleal masses dispersed. The oxidizing zoogleal bacterium isolated was a small, Gram-negative coccus, enclosed in a jelly-like mass. This organism did not ferment sugar. Dienert was not successful in isolating from the zoogleal film the organism causing nitrification but reported the breaking down of NH_3 without the formation of HNO_3.

Heukelekian and Littman (*8*) isolated 14 zooglea-forming organisms from activated sludge. All the cultures appeared to be similar. Morphologically and culturally they were indistinguishable from the zoogleal bacteria isolated by Butterfield (*3*).

Gilcreas (*7*) stated that large rod-shaped zoogleal bacteria are present in sewage and the film on the stones of a trickling filter is composed principally of zoogleal and filamentous bacteria.

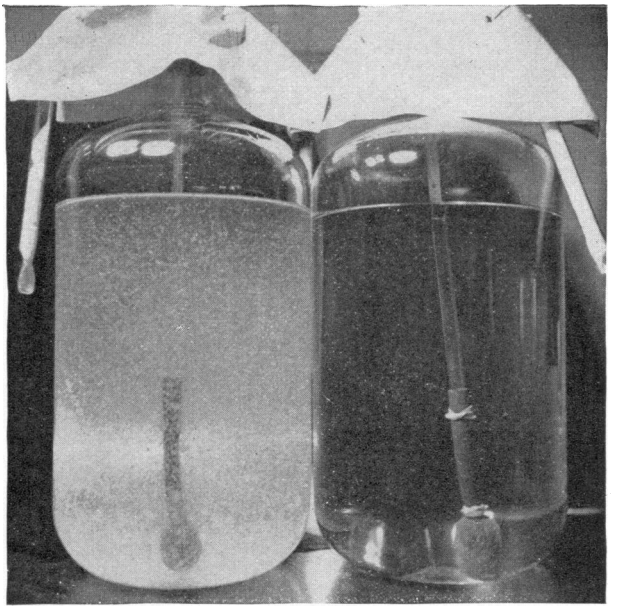

FIGURE 1 (a).—Eight-liter culture bottles of synthetic sewage. Bottle on right sterile, bottle on left with 48-hour growth under aeration of zooglea No. 86.

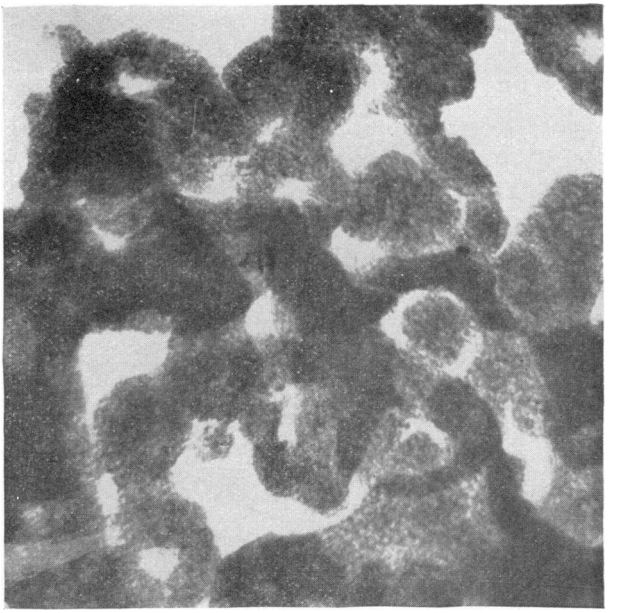

FIGURE 1 (b).—Floc of zoogleal culture, magnification 1426X.

A floc-forming organism was isolated by Buswell and Suter (*2*) from flocs present in chlorinated water supplies of various Illinois cities. Microscopic examination showed that these flocs were composed of small capsulated cocci or small rods, 0.3 to 0.5 microns in diameter, and larger organisms as well as filamentous bacteria. The organism isolated was nonmotile and formed nitrates from ammonia. This would indicate that the organism is not related to the activated sludge or trickling filter floc-forming organisms being discussed here, but to the family of Nitrobacteriaecae isolated by Winogradsky.

A zooglea organism, Nitrocystis, has been isolated by Winogradsky (*17*) from activated sludge, as one of the dominant organisms and is mentioned by Bergey (*1*). Other writers have studied briefly and reported zoogleal growths in sewage purification processes. It appears that a number of strains, possibly types of zoogleal bacteria, have been isolated and studied to some extent. Bergey (*1*) in his "Manual of Determinative Bacteriology" lists one species, *Zooglea ramigera*. Intensive effort at the Stream Pollution Investigations Laboratory of the United States Public Health Service has been directed to the isolation and to the study of the cultural characteristics of the zoogleal bacteria found in activated sludge and in trickling filters.

METHODS OF ISOLATION OF ZOOGLEAL CULTURES

Two methods of isolating zoogleal organisms were followed. The first method was similar to the technique used by Butterfield (*3*)—repeated washing and "teasing" of a zoogleal floc picked from activated sludge, or the film washed from the stones of a trickling filter. Through the use of this method, question might arise as to the failure to select a predominant zoogleal floc from a sludge sample.

In the second procedure, the sample of sludge was mixed thoroughly and a 10 ml. portion was placed in a sterile 30 ml. glass-stoppered bottle containing sterile glass beads. If the sludge used was heavy and dense, it was diluted 1:10 with sterile dilution water before removing the portion to be shaken. The bottle was then shaken 10 minutes at high speed on a shaking machine, breaking up the zoogleal flocs and freeing the organisms previously held in the gelatinous masses. When portions of the shaken and unshaken sludge were examined microscopically, a marked difference was noted. A much greater number of free bacterial cells was observed in the shaken sample, but complete dispersion was not obtained since some small flocs remained.

The shaken sludge sample was planted in serial dilution in standard lactose broth as soon as possible after shaking. It has been observed that the bacterial cells freed from their gelatinous mass quickly unite

again if allowed to remain quiescent after shaking. Tubes were incubated 48 to 120 hours at 20° C. before examining for typical zoogleal flocs.

Upon examination, tubes of the highest dilution showing floc formation were used for further purification. Two types of flocs were observed—the fingered type and the round, solid, compact type. Plantings were made from these high dilution tubes, using standard nutrient agar diluted 1:3 with sterile dilution water. After 96 hours' incubation at 20° C., colonies were picked from the agar plates to standard lactose broth. Planting and picking from dilute agar was repeated several times to insure absolute purity of the culture. Tests were made to determine the ability of the isolated pure cultures to develop, under aeration, an activated sludge in sterile synthetic media of the same composition as used by Butterfield, Ruchhoft, and McNamee (*4*), and in sterilized domestic sewage.

Samples of activated sludge used in the isolation of zooglea-forming organisms were obtained from various sources. Activated sludges used were from the North and South plants at Lancaster, Pa., Calumet Sewage Treatment Works of the Sanitary District of Chicago, Ill., and the experimental activated sludge plant at this laboratory. Trickling filter zooglea-forming bacteria were isolated from film-covered stones picked from the experimental trickling filter at this laboratory and from the municipal trickling filters at Dayton, Ohio, and at Osgood, Ind.

The experimental trickling filter at the Stream Pollution Investigations Station was constructed in three sections to provide for sampling at various depths. The filter was fed settled domestic sewage at an average rate of 3 million gallons per acre per day. In this experimental unit zooglea-forming organisms were observed after the filter had been in operation about 48 hours.

Flocs were more numerous in the film washed from the stones taken from the top section than from the center or bottom sections. Fingerlike flocs appeared in the film covering the stones a few inches below the surface of an experimental contact filter after the filter had been in operation 7 days, being fed twice daily with raw domestic sewage.

The film-covered stones selected at random for examination were removed from the trickling filter with sterile forceps and placed in sterile petri dishes. Extraneous matter was washed gently from the surface of the stones with sterile dilution water. The attached film was then scraped from the stones and placed in a sterile bottle containing 20.0 ml. of sterile dilution water. During the period between removing the stones from the filter and scraping off the film, care was taken to keep the surface of the stones sufficiently moist to prevent drying of the film. When a portion of the washed film was examined microscopically, fingerlike flocs were observed, similar to those found

in activated sludge flocs. The same two methods were followed in the isolation of floc-forming organisms from the washed film of the trickling filter stones as were used for activated sludge.

The only zooglea-forming cultures retained for further study were those that would produce, under aeration sufficient to maintain aerobic conditions, an activated sludgelike floc in sterile synthetic sewage and in sterilized domestic sewage.

The purification efficiency of a pure culture sludge developed by the various zoogleal organisms isolated in sterile synthetic sewage or sterilized domestic sewage was measured by the total oxidizable material removed from the supernatant. This percentage of the 5-day biochemical oxygen demand of the oxidizable material present in the substrate after 3 or 5 hours of aeration has been calculated for some of the organisms studied.

Isolations of 14 zooglea-forming bacteria have been made from activated sludge and 4 from the films washed off stones taken from trickling filters. For convenience in comparison, the characteristics of the zooglea-forming organisms studied are summarized in table 1.

CULTURES ISOLATED FROM ACTIVATED SLUDGE

Culture 50 was isolated from sludge of the experimental activated sludge plant of the Stream Pollution Station, Cincinnati, Ohio. This culture was originally Z–1, reported by Butterfield (*3*) and is listed by Bergey (*1*). Pure culture sludges of about 2,000 p. p. m. suspended solids, developed by this culture in sterile synthetic sewage, will oxidize during a 3-hour aeration period an average of 73.3 percent of the 5-day biochemical oxygen demand of the substrate. Under similar conditions, with sludges of about 792 p. p. m., but using sterilized domestic sewage, an average of 79.3 percent of the oxidizable material was oxidized. This culture differs from culture 88, *Zooglea ramigera*, isolated by Soriano (*14*), in respect to flagella, appearance of growth on agar slant, and liquefaction of gelatin.

Culture 53 was isolated from a sample of sludge received from the South unit of the activated sludge plant at Lancaster, Pa. When the sludge was examined microscopically many fuzzy tree-like protuberances were found, indicating that the sludge at the time of examination was in a poor condition and would settle slowly. Pure culture sludges with suspended solids of about 1,048 p. p. m., during a 5-hour aeration period, oxidized 70 percent of the 5-day biochemical oxygen demand of the substrate, using sterile synthetic sewage and 89.5 percent in sterilized domestic sewage by sludges with 1,351 p. p. m. suspended solids. This culture was reported by Butterfield et al. (*4*) as culture Z–4. Ruchhoft et al. (*11*) have reported that there is a remarkable similarity between the purification accomplished by a

TABLE 1.—*Characteristics of zooglea-forming organisms isolated*

Log No.	Source	Form	Diameter	Gram stain	Capsule	Chains	Photic	H_2S	Voges-Proskauer	Methyl red	Citrate	Indol	Flagella	Spores	NO_3 to NO_2	Gelatin liquefaction	Relation to O_2 [1]	Agar growth [2]	Agar slant [3]	Chromogenesis	Peptonization	Carbohydrates 0.5 percent [4]										
																						Arabinose	Xylose	Glucose	Galactose	Mannose	Sucrose	Lactose	Cellobiose	Raffinose	Melizitose	Dextrin	Mannitol	Salicin
50	Activated sludge	Rod	1.0	−	+	−	−	−	−	+	−	−	Polar	+	−	+	1	1	D	−	−	−	A±		A±			A±				A±	
53	do	do	1.0	−	+	−	−	−	−	+	−	−	do	+	−	+	1	2	G	−	−	+	A+	A±	A+	A±	A±	A±	0	0	0	A±	A±
55	do	do	1.0	−	+	−	−	−	−	+	−	−	Peri	+	−	+	1	2	G	−	−	+	A+	A+	A+	A±	A±	A±	0	0	0	A±	A±
58	do	do	>1.0	−	+	−	−	−	−	+	−	−	do	−	−	+	1	2	G	−	−	−	−	0	0	−	−	−	0	−	−	−	−
60	do	do	>1.0	−	+	−	−	−	−	+	−	−	Polar	−	−	+	1	2	G	−	−	−	A+	0	0	A±	A±	A±	A±	A+	0	0	A±	A±
62	do	do	>1.0	−	+	−	−	−	−	+	−	−	Peri	−	−	+	1	3	G	−	−	+	−	0	A+	−	−	−	0	−	−	−	A±	−
82	do	do	1.0	−	+	−	−	−	−	+	Tr	−	Polar	+	−	+	1	2	G	−	−	+	A+	A±	A+	A±	A+	A±	0	A+	0	A±	A±	A±
83	do	do	1.0	−	+	−	−	−	−	+	+	−	do	+	−	+	2	2	G	B	−	+	A+	A±	A±	A±	A±	0	0	A+	0	A±	0	0
85	do	do	1.0	−	+	−	−	−	−	+	−	−	Peri	−	−	+	1	2	G	−	−	−	A±	0	A+	A±	A±	0	0	A±	0	0	A+	A±
86	do	do	1.0	−	+	−	−	−	−	+	−	−	Polar	+	−	+	1	2	G	O	−	−	A+	A±	0	A±	0	0	0	A±	0	A±	0	0
88	do	do	1.0	−	+	−	−	−	−	+	−	−	Peri	+	−	+	1	1	G	−	−	−	A+	0	A±	0	A±	0	0	A+	0	A+	A+	A±
104	do	do	>1.0	−	+	−	−	−	−	+	−	−	do	−	−	+	1	2	G	−	−	−	−	0	A±	−	−	−	0	−	−	−	A±	−
105	do	do	1.0	−	+	−	−	−	−	+	−	−	do	−	−	+	2	2	G	−	−	−	−	0	0	−	−	−	0	−	−	−	0	−
113	do	do	1.0	−	+	−	−	−	−	+	−	−	Peri	+	−	+	1	2	G	−	−	+	A+	A±	A+	A±	A±	A±	A+	A+	0	0	A±	A±
87	Trickling filter	do	1.0	−	+	−	−	−	−	+	−	−	do	−	−	+	2	2	G	Y	−	−	A±	0	0	A±	A±	0	0	A+	0	A±	A±	0
100	do	do	1.0	−	+	−	−	−	−	+	−	−	Polar	−	−	+	2	2	G	−	−	−	A+	0	0	A+	A±	0	0	A±	0	0	0	0
102	do	do	1.0	−	+	−	−	−	−	+	−	−	do	−	−	+	2	2	G	−	−	−	A+	A+	A+	A±	A+	A±	0	A±	0	0	A+	A±
103	do	do	1.0	−	+	−	−	−	−	+	−	−	do	−	−	+	2	3	G	−	−	−	A+	−	A+	A+	−	−	−	−	−	−	A+	−

[1] Relation to O_2: 1=strict aerobe. 2=facultative anaerobe.
[2] Agar growth: 1=scanty. 2=moderate. 3=abundant.
[3] Agar slant: D=dull. G=glistening.

[4] Carbohydrates: A±=pH 6.9 to 6.5. A+=pH below 6.5. 0=no change in pH.
Chromogenesis: −=no color. B=Brown. O=orange. Y=Yellow.

pure culture sludge developed by one species of zoogleal bacteria, such as culture 53, and a normal activated sludge. The effect of dispersion of the bacterial flocs in a pure culture sludge developed by this culture has been reported by Butterfield and the author (*5*).

Culture 55 was isolated from a sample of activated sludge received from the North plant at Lancaster, Pa., during the winter months. A pure culture sludge developed by this bacterium removed 69.1 percent of the oxidizable material from sterile synthetic sewage and 84.7 percent from sterilized domestic sewage during a 5-hour aeration period. The North plant sludge appeared to be in better condition than the sludge from the South plant at the various times examinations were made.

Zooglea culture 58 was isolated from the sludge which was the source of culture 55. This culture was originally culture Z–9, reported by Butterfield et al. (*4*). With pure culture sludges of about 1,905 p. p. m. suspended solids developed by this culture, 78.2 percent of the 5-day biochemical oxygen demand was oxidized during a 5-hour aeration period, using sterile synthetic sewage as a medium, while 88 percent of the 5-day biochemical oxygen demand was oxidized from sterilized domestic sewage, using sludges of about 2,138 p. p. m. suspended solids.

The above zooglea-forming cultures had been isolated from samples of sludge taken during the winter months.

Culture 60 was isolated from sludge collected from the Calumet Sewage Treatment Plant during the summer months. This culture when inoculated into sterile synthetic and sterilized domestic sewage developed-sludge like masses, but the degree of purification has not been determined. As it required several weeks to develop a pure culture sludge suitable for use in purification studies in sterile synthetic sewage and sterilized sewage, such rates have not been determined for cultures 60, 62, 82, 88, 100, 104, 105, 113, and 85.

Culture 62 was isolated from the experimental sludge plant at this laboratory after the sludge had been treated with toluene, in an attempt to get rid of fungus and other filamentous forms. The percentage of purification accomplished by this culture was not determined.

Culture 82 was isolated from a *Sphaerotilus* culture which had been growing in a medium containing 1,000 p. p. m. of dextrose. No experimental work has been done using a pure culture sludge developed by this organism.

Culture 83 was isolated from sludge developed at this laboratory by aerating settled domestic sewage at room temperature, in an 8-liter serum bottle, fed daily, by the fill-and-draw method, with settled domestic sewage. This culture was the only zooglea-forming

bacterium which produced a brownish color when grown on sterile potato. Pure culture sludge was developed by this culture in a sterile synthetic sewage by the fill-and-draw method. This sludge thus developed was used and reported by Ruchhoft (*12*) in the studies of glucose removal from substrates by activated sludge.

Culture 85 was isolated from the experimental activated sludge plant at this laboratory. This culture developed typical zoogleal flocs in sterile synthetic sewage under aeration, but no purification rates were determined.

Culture 86 was isolated from sludge developed at 20° C. after inoculating 8 liters of sterile synthetic sewage with 5 ml. of domestic sewage. Pure culture sludge of about 1,000 p. p. m. developed under aeration by this culture in sterile synethetic sewage oxidized an average of 64.0 percent of the 5-day biochemical oxygen demand in 5 hours. The percent purification accomplished by the organism in sterile domestic sewage has not been determined. The culture was the only zooglea-forming bacterium isolated from sludge that produced an orange color when grown on potato or in synthetic media. This culture was used by Butterfield and the writer (*19*) in a comparative study of the growth and purification of a zooglea-forming bacterium isolated from activated sludge when grown and operated as a trickling filter and as an activated sludge.

Culture 88 was not isolated at this laboratory. It was isolated from activated sludge and classified as *Zooglea ramigera* by Soriano (*14*). Sludge-like flocs developed in sterile synthetic sewage and sterilized settled sewage under aeration. No purification rates have been determined. This culture is similar to culture 85 in cultural characteristics.

Cultures 104 and 105 were isolated from flocs picked from a sludge developed by aerating domestic sewage at room temperature. The sludge was fed daily with 1,000 p. p. m. dextrose for a period of 10 to 12 weeks before these isolations were made. Microscopic examinations showed the sludge had the appearance of a good activated sludge at the time the isolations were made. The oxidation rates of these cultures have not been determined.

Culture 113 was picked from flocs formed in aerated domestic sewage kept at room temperature and fed daily with raw sewage. No purification studies have been completed with pure culture sludge developed by this organism in sterile synthetic sewage or sterilized domestic sewage.

CULTURES ISOLATED FROM TRICKLING FILTERS

All of the zooglea-forming organisms from trickling filters that have been studied have been isolated from the experimental trickling filter at the Stream Pollution Station, Cincinnati, Ohio. Other isolations

have been made from full-scale filter units at Osgood, Ind., and at Dayton, Ohio. These cultures all produced, under aeration, sludge-like flocs when inoculated into sterile synthetic sewage or sterilized domestic sewage.

Culture 87 is the only zoogleal culture isolated that has a distinct yellow color. It was isolated from the film covering the stones taken from the top section of the experimental trickling filter during the early spring. The percent of over-all purification accomplished by pure culture sludges of about 1,180 p. p. m. suspended solids developed by culture 87 in sterile synthetic sewage has been found to be 73.6 percent during a 5-hour aeration period. The percentage purification, when the sludge was developed and fed sterilized sewage, has not been determined.

Culture 100 was isolated from the top section of stones. The film was washed from the stone and a typical finger-like floc was picked, washed, and transferred to lactose broth. After flocs had developed, purification was done using the same technique as previously explained. Sludge-like flocs developed under aeration in sterile synthetic sewage or sterilized domestic sewage. No purification studies have been completed with this culture.

Culture 102 was isolated in a manner similar to culture 100. The floc was picked from film adhering to the stones of the top layer. No purification studies have been carried out using sludge developed by this organism.

Culture 103 was isolated from the film removed from the top layer of stones following the procedure used in the isolation of culture 87. The over-all purification during the 5-hour aeration period was found to be 82.2 percent of the 5-day biochemical oxygen demand of the sterile synthetic sewage. The purification properties of this organism have not been determined when developed on and fed sterilized domestic sewage.

Cultures 100, 102, and 103 were isolated from the trickling filter during the summer months. Sludges developed under aeration by inoculating sterile media, either synthetic or domestic sewage, are very similar to a sludge developed by a zooglea-forming organism isolated from the filter during colder months. The pure culture sludges developed by all of the trickling filter zoogleal organisms isolated resemble very closely in appearance pure culture sludges developed, under similar conditions and in similar media, by zooglea-forming organisms isolated from activated sludge. The rate of purification in the sterile synthetic sewage and sterilized domestic sewage is practically the same.

The effects of carbohydrates were determined by two methods. Dilute agar was prepared by dissolving in 1 liter of distilled water 5.0 gm. peptone, 7.25 gm. Na_2HPO_4, 0.7 gm. KH_2PO_4, 1 gm. agar and

5.0 gm. of the carbohydrate being tested. The agar was sterilized by autoclaving at 15 pounds for 15 minutes except when the sugar under consideration would break down by this method; intermittent sterilization was then used. Broth containing the same percentage of the various carbohydrates with the addition of brom cresol blue was also used. All tubes were read after incubating 14 and 31 days at 20° C.

The change of the pH value in the various sugar broths seems to indicate slight use of sugar. No gas production was observed. The results presented by Ruchhoft, Kachmar, and Moore (*12*) show that a pure culture sludge of a concentration of about 1,395 p. p. m. developed by culture 83, in sterile synthetic sewage with an average of 840 p. p. m. glucose added, removed 90.5 percent of the glucose in 23 hours. A pure culture sludge, developed in a similar manner by culture 86, with initial suspended solids of 686 p. p. m. in sterile synthetic sewage containing 505 p. p. m. glucose, utilized 84.7 percent in 24 hours. The pure culture sludges in the above experiments were kept under sufficient aeration to maintain aerobic conditions and the amount of sludge used was much greater; whereas in our tests using the pH range as an index of the utilization of sugar, very small amounts of inoculum were used and the tubes remained quiescent. In the experiments by Ruchhoft et al. (*12*) the glucose was utilized in the production of growth, and appears as bacterial protoplasm without the accumulation of intermediate acid end products. The hydrogen-ion concentration of the substrate as shown by Ruchhoft et al. (*12*) influences the rate of glucose removal as well as growth and floc formation which will be discussed later in this paper.

The presence of spores was determined by two methods: (1) the heat test—tubes being held after heating 20 minutes at 80° C. for 14 days at 20° C., and (2) staining with the Schaeffer and Fulton (*13*) modification technique. Liefson's (*10*) flagella stain was used. The semisolid KNO_3 medium of Zobell and Meyer (*18*) was used to determine nitrate reduction. If, during incubation, gas was produced with or without the reduction of nitrate, bubbles would be held in the media. No gas was produced in any instance.

From the cultural characteristics it is noted that all zoogleal cultures isolated are identical in nine characteristics: form, Gram stain, capsule, nonchain forming, photic, hydrogen sulfide, Voges-Proskauer, and the reaction with arabinose and raffinose. This would indicate that the zooglea-forming organisms isolated from activated sludge and trickling filters were closely related. Considering in addition the results of the following determinations—spores, production of indol, gelatin liquefaction, methyl red, citrate, and the reaction with cellobiose, dextrin, and salicin—the zoogleal cultures studied may be divided into nine groups as follows:

1. Cultures 53, 55, 62, 82, 88, and 113 isolated from activated sludge.
2. Cultures 58, 60, and 104 isolated from activated sludge.
3. Cultures 102 and 103 isolated from trickling filters.
4. Cultures 87 and 105 isolated from activated sludge and trickling filters.
5. Culture 100 isolated from trickling filter.
6. Culture 50 isolated from activated sludge.
7. Culture 83 isolated from activated sludge.
8. Culture 85 isolated from activated sludge.
9. Culture 86 isolated from activated sludge.

By adding the appearance of growth on agar slant, the relation to oxygen and the ability to reduce nitrates, groups 2 and 4 would be subdivided.

The close but not all-inclusive similarity brought out by the cultural characteristics is further evidence for the above-mentioned close relationship, implying that we are dealing with several varieties of a single group.

In the first four groups, each including more than one culture, similar reactions were obtained in very few of the additional cultural characteristics studied. The organisms within group 1 reacted similarly in regard to chromogenesis and their relation to oxygen. However, varying results were obtained from flagella stain, reduction of nitrates, peptonization of milk, and their ability to utilize xylose, glucose, galactose, mannose, lactose, melizitose, and mannitol.

Within group 2 similar results were obtained from growth in milk, chromogenesis, and the organisms' ability to utilize xylose. Differing results were obtained from flagella stain, their relation to oxygen, the formation of nitrites from nitrates, and their ability to utilize glucose, galactose, mannose, sucrose, lactose, melizitose, and mannitol. Both organisms listed in group 3 gave similar results with all of the tests used. The organisms listed in group 4 reacted in the same manner in regard to their utilization of xylose, lactose, growth in milk, their relation to oxygen, and flagella stain. Dissimilar results were obtained by the organisms in group 4 in the following: reduction of nitrates to nitrites, chromogenesis, and their utilization of glucose, mannose, sucrose, melizitose, and mannitol.

The results of the reactions used for group classification are presented in table 2, and it is shown clearly that the organisms being discussed are closely related. From the results listed, groups 1 and 9 differ only in utilization of carbohydrates. The same difference is observed for groups 4 and 8. Groups 6, 2, 7, and 5 differ in utilization of carbohydrates and growth in sodium citrate broth. Greater differences are observed in the results of the reactions of group 3 and the results of the other groups.

TABLE 2.—*Results of the reactions used for group classification*

Group	Spores	Indol	Gelatin liquefaction	Methyl red	Citrate	Carbohydrates 0.5 percent		
						Cellobiose	Dextrin	Salicin
3	−	+	+	+	+	A±	0	A±
1	+	−	+	−	−	A+		
9	+	−	+	−	−	A±	0	0
4	−	−	+	−	−	A+	A±	0
8	−	−	+	−	−	A±	0	A±
6	−	−	−	−	±			
2	−	−	−	−	−	A+	A±	A±
7	−	−	−	−	−	A+	A±	0
5	−	−	−	−	−	A±	0	0

EXPERIMENTAL WORK

The primary characteristics of these zoogleal bacteria were that they must grow in pure culture in flocs or colony formation in liquid media under aeration sufficient to maintain aerobic conditions. Experiments were instituted to determine (1) the minimum food requirement, (2) the effect of hydrogen-ion concentration on growth and floc formation, and (3) the effect of various substances commonly found in sewage in regard to growth and sludge production.

MINIMUM FOOD REQUIREMENT

Culture 86 was used in the tests to determine the minimum food requirement for floc production. Sterile synthetic sewage, of varying concentration, was inoculated with culture 86, incubated at 20° C. under aeration. The results given in table 3 indicate that synthetic sewage of less than 10 percent concentration would not support growth. A concentration of 17.5 percent to 37.5 percent produced growth but no floc formation. In a 50-percent concentration both growth and floc formation were observed after 4 days, but floc formation occurred sooner in full strength media.

TABLE 3.—*Growth of pure culture zooglea in synthetic media of varying food concentration*

Culture	Percent food concentration [1]	Peptone p. p. m.	Results
86	1	3	No growth, aerated 7 days.
86	10	30	No growth, aerated 7 days.
86	17.5	52.5	Turbid, no floc, aerated 10 days.
86	25	75	Turbid, no floc, aerated 4 days.
86	37.5	112.5	Turbid, no floc, aerated 10 days.
86	50	150	Turbid and floc in bottom, aerated 4 days.
86	100	300	Turbid and floc throughout, aerated 2 days.

[1] Percent of standard synthetic sewage added.

pH EFFECTS

Observations on the influence of hydrogen-ion concentration on growth were made as follows: Flasks containing 100 ml. of synthetic sewage were sterilized and the pH was adjusted using sterile 10 percent H_3PO_4 or sterilized 10 percent NaOH to cover the range from pH 3.5 to pH 10.0. After such adjustment the flasks were inoculated with a suspension of culture 86, previously shaken with sterile glass beads, and examinations to determine the initial total count per ml. and the pH were made. The flasks were incubated at 20° C. for 24 hours and the pH value and total counts were again determined. From the results presented in table 4 it will be observed that there was a tendency for the lower pH to rise during the incubation period and for the pH values in the higher range to drop.

TABLE 4.—*Pure culture 86 in standard synthetic sewage of varying pH readings. Flasks and plates incubated at 20° C.*

pH readings		Series 1, bacteria per ml.		Series 2, bacteria per ml.		Series 3, bacteria per ml.	
0 hour	24 hour	0 hour	24 hour	0 hour	24 hour	0 hour	24 hour
3.5	3.8		No growth [1]				
4.0	4.6		do [1]				
4.5	4.7		do [1]				
5.0	5.5	97	Less than 1	156	Less than 10,000		
5.5	5.9	97	do	156	do	13	Less than 10.
6.0	6.4	97	195	156	400,000	13	100.
6.5	6.8	97	21,500	156	650,000	13	200.
7.0	7.1	97	3,020,000	156	1,560,000	13	56,900.
7.5	7.2	97	267,000,000	156	370,000	13	82,500.
8.0	7.4	97	68,000,000	156	35,000	13	120,000.
8.5	7.6	97	69,000,000	156	890,000	13	186,000.
9.0	8.7					13	57,800.
9.5	8.6		No growth [1]				
10.0	9.8		do [1]				

[1] Observations based on turbidity readings.

The number of organisms per ml. increased most rapidly at a pH of 7.0 to 8.0. Incubating the flasks for an additional 24 hours, for a total of 48 hours at 20° C, produced very little change in the flasks of the upper range. Microscopic examination of the flasks at the end of the 24-hour period showed few flocs present in the flasks of pH 6.5 and no flocs at lower pH values. Floc formation increased up to a hydrogen-ion concentration of pH 7.5 and decreased with further increase in pH. In the development of pure culture sludges, this principle has been followed by adjusting the pH value after feeding to pH 6.5. During a 24-hour aeration period the hydrogen-ion concentration increases to pH 7.6. Therefore, judging by the bacterial counts obtained and from the microscopic appearance of the growth, the optimum pH for growth and floc formation appears to be pH 6.8 to 7.5.

EFFECT OF VARIOUS SUBSTANCES COMMONLY FOUND IN SEWAGE

Various substances commonly found in sewage, such as soap, sodium ricinoleate, sodium formate, sodium oleate, creatine, pectin, glucose, glycerine, and sucrose, were added to synthetic sewage in an effort to stimulate growth and floc formation. Glucose was the only substance used which when added increased floc formation materially, as is also shown by the studies of Ruchhoft et al. (*12*). No increase was observed when glucose was added to synthetic sewage if beef extract or urea was omitted. Cultures 53, 60, 83, 87, 104, and 113 were used for these experiments.

TABLE 5.—*Effect of various substances on growth of pure culture zooglea organisms*

Basic medium	Substance added	Amount per liter added	Culture	Results
Synthetic sewage	Soap [1]	0.1 gm	53	Floc developed; few free organisms.
Do	Na. ricinoleate	0.1 gm	53	No floc developed; few free organisms.
Do	do	0.05 gm	53	Little floc developed; supernatant turbid pink color.
Do	do	0.025 gm	53	Floc developed.
Do	do	0.05 gm	53	Small amount floc developed; pink color.
Do	do	1.5 gm	53	Do.
Do	Na. formate	0.5 gm	53	Normal amount floc developed.
Do	No. oleate	0.05 gm	53	Do.
Do	Creatine	0.05 gm	53	Very little floc developed.
Do	Certo	5 ml	113	Floc developed.
Do	do	2 ml	53	Supernatant turbid; small amount floc.
Do	do	1 ml	113	Growth, but no floc.
Do	Pectin [2]	2 ml	53	Floc developed; no increase in amount.
Do	do [2]	1 ml	53	Small flocs; no increase in amount.
Do	do [2]	0.5 ml	53	Small loose flocs.
Do.[3]	Glucose	0.1 gm	53	Supernatant turbid; no flocs.
Do.[3]	do	0.1 gm	53	Do.
Do.[3]	do	0.1 gm	113	Flocs; no increase in amount sludge.
Do.[4]	Glycerine	0.05 gm	53	Sludge developed.
Do.[4]	do	0.05 gm	113	Do.
Do.[4]	do	0.05 gm	53	Do.
Do.[4]	do	0.05 gm	113	Do.
Do.[4]	Sucrose	0.05 gm	53	No flocs; supernatant turbid.
Do.[4]	Glycerine	0.05 gm	53	Floc developed slowly.
Do.[4]	Glucose	10.0 gm	53	No growth.
Do	Glycerine	0.5 gm	53	Sludge developed.
Do	do	0.5 gm	60	Sludge developed; no increase in amount.
Do.[4]	Glucose	0.05 gm	53	No increase in amount; floc developed.
Do.[4]	Glycerine	0.05 gm	53	Do.
Do.[4]	do	0.05 gm	83	Do.
Do	Glucose	0.5 gm	87	Amount of floc increased.
Do	do	0.5 gm	104	Slight increase in floc.
Do	do	0.5 gm	104	Do.
Do	do	0.5 gm	83	Amount of floc increased.

[1] Bell's Castile hand soap.
[2] Pectin extracted from grapefruit rind.
[3] Beef extract omitted from synthetic.
[4] Urea omitted from synthetic.

SUMMARY

The predominant bacteria of activated sludge and of trickling filters have been isolated in pure culture. It would appear that these zoogleal bacteria might be considered in one group. All cultures studied, isolated from activated sludge and trickling filters, were short Gram-negative rods, failed to produce H_2S or acetyl methyl carbinol, produced acid in broth containing arabinose, produced no change in

broth containing raffinose, and produced capsules which bound the cells together in a capsular matrix tenaciously enough to remain intact under agitation sufficiently violent to keep the flocs suspended and to maintain aerobic conditions. Such sludge flocs of about 1,500 p. p. m. suspended solids composed entirely of masses of bacterial cells in pure culture will remove in 3 hours 36.3 to 84.2 percent, and in 5 hours 55.6 to 91.6 percent of the 5-day biochemical oxygen demands of polluted water or sewage.

The cultures isolated may be divided into nine related groups determined by the following characteristics: formation of spores, indol reaction, gelatin liquefaction, methyl red test, growth in citrate media, motility, and the pH reading in broth containing cellobiose, dextrin, and salicin.

The floc-forming organisms isolated from trickling filters will develop, under aeration, an activated sludge and will function similarly to a sludge developed by the floc-forming organisms isolated from activated sludge. This indicates that the zoogleal organisms found in trickling filters and in activated sludge flocs are closely related.

REFERENCES

(1) Bergey, D. H.: Manual of Determinative Bacteriology. 5th ed. Williams & Wilkins, Baltimore, Md., 1939.

(2) Buswell, A. M., and Suter, Max: A study of floc-forming organisms in chlorinated water supplies. J. Bact., **39**: 583–587 (1940).

(3) Butterfield, C. T.: Studies of sewage purification. II. A zoogleal-forming bacterium isolated from activated sludge. Pub. Health Rep., **50**: 671–684 (1935).

(4) Butterfield, C. T., Ruchhoft, C. C., and McNamee, P. D.: Studies of sewage purification. VI. Biochemical oxidation by sludges developed by pure cultures of bacteria isolated from activated sludge. Pub. Health Rep., **52**: 387–412 (1937).

(5) Butterfield, C. T., and Wattie, Elsie: Studies of sewage purification. VIII. Observations on the effect of variations in the initial numbers of bacteria and of the dispersion of sludge flocs in the course of oxidation of organic material by bacteria in pure culture. Pub. Health Rep., **53**: 1912–1934 (1938).

(6) Dienert, F.: Study of the mechanism of purification of sewage waters by biological processes. Revue d'Hygiene, **59**: 5–16 (1937). (Translated by J. B. Lackey.)

(7) Gilcreas, F. W.: Laboratory control of sewage treatment. IX. The biology of sewage. Municipal Sanitation, **8**: 225 (1937).

(8) Heukelekian, H., and Littman, M. L.: Carbon and nitrogen transformation in the purification of sewage by the activated sludge process. II. Morphological and biochemical studies of zoogleal organisms. Sewage Works J., **9**: 752 (1939).

(9) Heukelekian, H., and Schulhoff, H. B.: Studies on the clarifying organisms in activated sludge. Sewage Works J., **10**: 43 (1938).

(10) Liefson, E.: A method of staining bacterial flagella and capsules together with a study of the origin of flagella. J. Bact., **20**: 203–211 (1930).

(11) Ruchhoft, C. C., Butterfield, C. T., McNamee, P. D., and Wattie, E.: Studies of sewage purification. IX. Total purification, oxidation, absorption and synthesis of nutrient substrates by activated sludge. Pub. Health Rep., **54**: 468–496 (1939). Sewage Works J., **11**: 195 (1939).

(12) Ruchhoft, C. C., Kachmar, J. F., and Moore, W. A.: Studies of sewage purification. XI. The removal of glucose from substrates by activated sludge. Pub. Health Rep., **55**: 393–423 (1940).

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(13) Schaeffer, A. B., and Fulton, M. D.: Manual of Methods for Pure Culture Study of Bacteria. Leaflet IV. 7th ed. Staining Procedures. 1939. Edited by Committee on Bacteriological Technic of the Society of American Bacteriologists.
(14) Soriano, S.: Personal communication—culture isolated while on sabbatical leave and working at the Univeristy of Wisconsin, Madison, Wisconsin. Prof. of Microbiology, Faculty of Agronomy. University of Buenos Aires. Rep. Argentina.
(15) Theriault, E. J., McNamee, P. D., and Butterfield, C. T.: Experimental studies of natural purification in polluted waters. V. The selection of dilution waters for use in oxygen demand tests. Pub. Health Rep., 46: 1084–1116 (1931).
(16) Whitehead, H. C., and O'Shaughnessy, F. I. C.: Improving the efficiency of activated sludge. Surveyor and Municipal and County Engineer, 89: 407–410 (1936).
(17) Winogradsky, H.: Sur la microflore nitrificatrice des boues activies de Paris. Compt. Rend. Acad. Sci. (Paris), 200: 1886–1888 (1935).
(18) Zobell, C. E., and Meyer, K. F.: Metabolism studies on the Brucella group. VI. Nitrate to nitrite reduction. J. Infect. Dis., 51: 99–108 (1932).
(19) Butterfield, C. T., and Wattie, Elsie: Studies of sewage purification. XV. Effective bacteria in purification by trickling filters. Pub. Health Rep., 56: 2445–2464 (1941).

THE CHEMOTHERAPEUTIC ACTION OF A N-PHOSPHORYL DERIVATIVE OF 4-4'-DIAMINODIPHENYLSULFONE[1]

By M. I. Smith, *Chief Pharmacologist*, S. M. Rosenthal, *Senior Pharmacologist*, and E. L. Jackson, *Chemist, United States Public Health Service*

It has been shown (*1*) that of a series of sulfonamides, sulfones, and certain phosphorus analogues 4–4'–diaminodiphenylsulfone proved the most effective bacteriostatic agent against the tubercle bacillus and the most effective in retarding the progress of experimental tuberculous infection in guinea pigs. The excessive toxicity of this compound made it desirable to develop derivatives of lower toxicity without sacrificing specificity. The present report concerns the pharmacologic and chemotherapeutic properties of a N–phosphoryl derivative of 4–4'–diaminodiphenylsulfone.

Other attempts have been made to reduce the toxicity of 4–4'–diaminodiphenylsulfone by substitutions following Buttle's (*2*) demonstration of its high antibacterial action. The acetylated derivatives introduced by Fourneau (*3*) proved of little superiority over the parent substance. The formaldehyde sulfoxylate and bisulfite derivatives of Bauer and Rosenthal (*4*), though much less toxic than 4–4'–diaminodiphenylsulfone, were also less active. The substituted derivative 4–amino–4'–hydroxy–diphenylsulfone and others prepared by Raiziss (*5*) similarly had less antistreptococcic action than 4–4'–diaminodiphenylsulfone and but little antipneumococcic action. The sulfonated glucose derivative (promin) likewise has but little advantage over sulfanilamide in streptococcus infection and sulfapyridine in pneumococcus infection (*6,7*). Roblin and associates (*8*) reported an active

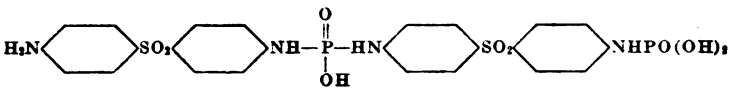
[1] From the Division of Chemotherapy, National Institute of Health.

sulfanyl derivative, but gave no details. Dewing and associates (*9*) prepared a series of derivatives, but these were for the most part inactive. A nicotinic acid derivative[3] currently prepared in this laboratory by Hugo Bauer has little antistreptococcic or antipneumococcic activity. Its action against the tubercle bacillus is under investigation.

The phosphoryl derivative of 4–4′–diaminodiphenylsulfone which forms the subject of the present report has been prepared and is being studied in connection with investigations on the chemotherapy of tuberculosis to be reported later. Observations with this compound in other experimental bacterial infections, however, appear sufficiently interesting to warrant the publication of a preliminary note at this time.

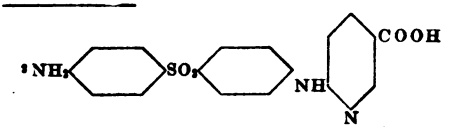
CHEMICAL AND PHYSICAL PROPERTIES

The N–phosphoryl derivative was obtained by the reaction of phosphorus oxychloride with 4–4′–diaminodiphenylsulfone and subsequent hydrolysis of the resulting chlorophosphoryl derivative. The results of analyses of the substance for carbon, hydrogen, nitrogen, phosphorus, and sulfur correspond to the empirical formula $C_{24}H_{24}O_9N_4P_2S_2$. The compound probably has the following structure:



It is an amorphous powder, which is only slightly soluble in water and the usual organic solvents at room temperature. Its sodium salt is freely soluble in water. Aqueous solutions having a pH of 6 to 7 may be prepared by treating the substance with N/1 NaOH solution in the proportion of 1.8 to 2.2 cc. per gm. of compound. In agreement with the above formula the diazotization value of the compound by the Bratton and Marshall (*10*) procedure is slightly over 20 percent of that found for 4–4′–diaminodiphenylsulfone. An article on the synthesis of this N–phosphoryl derivative will be published elsewhere by one of us. (E. L. J.)

The usual procedure for the estimation of the sulfonamide drugs in the body fluids is applicable to the determination of the N–phosphoryl derivative in the urine, but does not yield quantitative results in blood. When added to urine and measured by the Bratton and Marshall technique, the compound is recoverable 100 percent; after hydrolysis, it is recoverable to the extent of 250 percent. The latter figure is



consistent with results obtained by hydrolysis of the compound for 1 hour at 100° C. with N/10 to N/1 HCl; the diazotization value is increased by this hydrolysis reaction to a maximum of about 50 percent of the value shown by 4–4′–diaminodiphenylsulfone. When added to blood or tissues, the substance is recoverable only to the extent of about 40 to 60 percent whether determined as "free," or as "total" after hydrolysis. Added to plasma it is nearly completely recoverable, but when added to erythrocytes, 30 to 40 percent is recoverable. The reason for this is not clear, though this obviously complicates the problem of the fate of the substance in the body.

TOXICITY

Given to mice the compound is tolerated in doses of 0.5 gm. per kg. whether given orally or subcutaneously. A dose of 1.0 gm. per kg. gave 80 percent mortality in mice when injected subcutaneously and 60 percent mortality when given orally. Rats receiving 0.5 gm. per kg. injected intravenously uniformly survived. Guinea pigs tolerate 1.5 gm. per kg. when given orally and 0.5 gm. per kg. when given subcutaneously. Four out of 10 animals survived a single dose of 0.75 gm. per kg. injected subcutaneously.[3] These data are shown in table 1. Since the same mortality rate was obtained from a single oral or subcutaneous injection of 0.3 gm. per kg. 4–4′–diaminodiphenylsulfone (the latter as 7.5 percent solution in propylene glycol), it would appear that this compound is at least half and possibly one-fifth as toxic as 4–4′–diaminodiphenylsulfone. Guinea pigs and mice survived repeated daily subcutaneous injections of from 0.2 to 0.3 gm. per kg. for a period of from 2 to 4 weeks, and the daily oral administration of 0.5 gm. per kg. in guinea pigs over a period of 3 weeks had little effect on the growth curve.

TABLE 1.—*Acute toxicity*

Animal species	Dose (Gm. per kg.)	Route	Number	Survival (Percent)
Mice	0.5	Oral	10	90
	1.0	do	10	40
	0.5	Subcutaneous	10	80
	1.0	do	10	20
Rats	0.5	Intravenous	10	100
	0.75	do	7	0
Guinea pigs	0.5	Subcutaneous	10	80
	0.75	do	10	40
	1.0	do	5	0
	1.5	Oral	10	100

ABSORPTION AND EXCRETION

These were studied in guinea pigs and rabbits. As stated previously, the failure to recover the drug quantitatively when added to blood makes blood level determinations a procedure of uncertain value.

[3] Large doses injected subcutaneously often produce local irritant action.

However, it was deemed necessary to do some of this work even if the information so obtained has only a partial value. The absorption of the drug in the guinea pig appears to be equally as satisfactory whether given orally or subcutaneously. The relative blood levels are shown in table 2, and the percent excretion is shown in table 3. In rabbits similar observations were made with the drug administered orally, subcutaneously, or intravenously in doses of from 50 to 500 mg. per kg. with the results as shown in table 4.

TABLE 2.—*Blood levels in guinea pigs after 0.2 gm. per kg.*

Time (hours)	Mgs. percent	
	Oral	Subcutaneous
½	----	6.5
1	8.0	----
2	----	5.5
2¼	6.0	----
22	0.2	0.2

TABLE 3.—*Urinary excretion in guinea pigs, percent of dose administered*

No.	Subcutaneous injection 0.5 gm. per kg.					Oral administration 1.5 gm. per kg.					
	First day	Second day	Third day	Fourth day	Total	First day	Second day	Third day	Fourth day	Fifth and sixth days	Total
1	49	9	2	1	61	13	14	11	7	6	51
2	30	12	5	2	49	18	14	11	5	7	55
3	35	16	9	1	61	18	8	9	3	2	40
4	27	15	8	3	53	14	11	12	5	3	45

TABLE 4.—*Blood levels and urinary excretion in rabbits*

No.	Dose (mg. per kg.)	Route	Blood level (mg. percent)		Urinary excretion (percent of dose)	
			Peak within 6 hours	At 24 hours	At 24 hours	At 48 hours
1	50	Intravenous	4.2	0	60	----
2	200	do	12.0	0.2	65	73
3	200	Subcutaneous	5.1	1.0	57	63
4	200	Oral	2.9	1.6	62	71
5	500	do	4.4	0.6	43	47
6	500	do	6.8	1.2	55	63

BACTERIOSTATIC TESTS

The bacteriostatic action of the phosphoryl derivative was compared with diaminodiphenylsulfone and sulfadiazine. Because addition of this compound to media containing peptone caused precipitation, tests were run in peptone-free beef infusion broth containing 0.2-percent glucose (pH 7.0). It has been previously found that in such a medium relatively high bacteriostatic values are obtained with the sulfonamide

drugs. Approximately 10,000 organisms per 10 cc. from an 18-hour culture in rabbit-blood broth were employed. The following values represent the highest dilutions which inhibited growth for 24 hours:

	Pneumococcus I	*Streptococcus No. 1885* [1]
Phosphoryl derivative	50, 000	25, 000
Diaminodiphenylsulfone	200, 000	100, 000
Sulfadiazine	50, 000	25, 000

[1] Hemolytic, Group A.

In vitro diaminodiphenylsulfone was the most active of the three compounds.

THERAPEUTIC TESTS

These were carried out with the same strains of organisms employed in the bacteriostatic experiments. They have been employed in this laboratory for several years and maintained at a virulence such that 0.5 cc. of an 18-hour broth culture intraperitoneally is lethal to mice in dilutions up to 10^{-9}.

Upon hemolytic streptococcal infections in mice, it is seen (table 5) that the phosphorylated sulfone was of the same order of activity as the parent sulfone when both are given subcutaneously. On oral administration the sulfone increases in activity while the phosphorus compound gave evidence of being less active. Sulfadiazine orally was approximately one-sixteenth as active as the diaminosulfone, and less than one-eighth as active as the phosphoryl derivative.

TABLE 5.—*Comparative effects upon hemolytic streptococcus infections in mice*

Infective dose	Therapy	Route	Number of mice	Death in days								Mortality (percent)
				1	2	3	4	5	6	7	8–14	
0.5 cc., intraperitoneal injection, 10^{-6} dilution.	Diaminodiphenylsulfone:											
	0.125 mg. B D × 4 days	Oral	30		1			1	4	2	3	36.6
	0.25 mg. B D × 4 days	do	20		1				1	1		15
	0.125 mg. B D × 4 days	Subcutaneous.	16	1	6	1			1		1	62.5
	Phosphoryl derivative:											
	0.125 mg. B D × 4 days	do	35	1	5	1	2	5	4	1	2	60
	0.25 mg. B D × 4 days	do	40	2	3		1	2	1		5	35
	0.125 mg. B D × 4 days	Oral	15	2	7	1		3				86.6
	0.25 mg. B D × 4 days	do	19				2	1	2			26.3
	Sulfadiazine:											
	2.0 mg. B D × 4 days	do	15		1			1	2	1	1	40
	Controls		49	45	3							98

Upon type I pneumococcus infections diaminodiphenylsulfone orally in the maximum tolerated dose (2 mg. daily) caused prolongation of life, but only 2 of 52 mice survived the infection (table 6). The phosphoryl derivative subcutaneously gave more favorable results, and with 3 to 5 mg. daily for 4 days approximately half of the animals survived. Sodium sulfadiazine subcutaneously in equivalent doses was

less active while sulfadiazine orally was approximately one-tenth as active as the phosphoryl derivative.

TABLE 6.—*Comparative effects upon pneumococcus type I infections in mice*

Infective dose	Therapy	Route	Number of mice	Death in days								Mortality (per cent)
				1	2	3	4	5	6	7	8–14	
0.5 cc., intraperitoneal injection, 10^{-6} dilution.	Diaminodiphenylsulfone:											
	1.0 mg. B D × 4 days	Oral	52	2	6	2	1	8	14	8	9	96.0
	Phosphoryl derivative:											
	1.0 mg. B D × 4 days	Subcutaneous.	45	2	8	6	4	2	3	6	8	86.6
	1.5 mg. B D × 4 days	do	15					1	3	2	2	53.3
	2.5 mg. B D × 4 days	do	42		3	1		6	3	6	5	57.0
	1.0 mg. B D × 4 days	Oral	15		8	2			1	3	1	100.0
	Sulfadiazine:											
	10.0 mg. B D × 4 days	do	15			2	5		2	1		66.6
	15.0 mg. B D × 4 days	do	15		1	1	1		1	2	4	66.6
	25.0 mg. B D × 4 days	do	13					1	1	1	3	46.0
	Sodium sulfadiazine:											
	1.0 mg. B D × 4 days	Subcutaneous.	13		2			3	3	2	2	92.3
	2.5 mg. B D × 4 days	do	30	3		1		2	7	5	3	70.0
	5.0 mg. B D × 4 days [1]	do	9							2	1	33.3
	Phosphoryl derivative:											
	0.1 percent in diet for 7 days.		20		8	8				2		90.0
	0.2 percent in diet for 7 days.		20		4	5	1	1	1	1	2	75.0
	Sulfadiazine:											
	0.1 percent in diet for 7 days.		19		6	5	1		1	3	2	94.7
	0.2 percent in diet for 7 days.		20		4	2	1			1	3	55.0
	Controls		75	63	12							100.0
0.5 cc., intraperitoneal injection, 10^{-6} dilution.	Phosphoryl derivative:											
	1.0 mg. B D × 5 days	Subcutaneous.	15		6		1	1	1	2	2	86.6
	2.5 mg. B D × 5 days	do	15	1	3			2	1	4	1	80.0
	Sodium sulfadiazine:											
	1.0 mg. B D × 5 days	do	15	3	5	1	3	1		2		100.0
	2.5 mg. B D × 5 days	do	15		1	1	2	1	1	3	1	66.6
	Controls		9	9								100.0

[1] Toxic dose. 6 additional mice died of drug toxicity.

The decreased activity of the phosphoryl derivative when administered orally was further shown in a comparison with sulfadiazine upon pneumococcal infection when the drugs were administered in the diets for 1 week after inoculation. The drug diets were begun 2 days prior to inoculation. Upon diets containing 0.1 percent of the drugs, survivors for 2 or more days were insufficient for quantitative comparison of drug intake. With 0.2 percent concentrations in the diet, the average daily food consumption was 3.1 gm. for the phosphorus compound and 2.8 gm. for sulfadiazine. Sulfadiazine with an average daily drug intake of 5.7 mg. resulted in mortality of 55 percent while the phosphorus compound with an average daily intake of 6.3 mg. gave a mortality of 75 percent.

No close correlation between *in vitro* and *in vivo* effects was observed. The bacteriostatic action of the phosphoryl derivative and sulfadiazine were of the same order, while the sulfone was approximately 4 times as active.

COMMENTS

Litchfield, White, and Marshall (*11*) using the drug-diet technique found that diaminodiphenylsulfone possesses an activity ratio against a pneumococcal infection nearly 7 times that of sulfapyridine and sulfathiazole and 16 times that of sulfanilamide. The average daily intake of the sulfone for a 50 percent survival in their experiments was 1.6 mg. The toxicity of the sulfone is high, and numerous attempts have been made to prepare active derivatives with lowered toxicity. Against pneumococcal infections none of these derivatives has shown particular promise.

The present experiments indicate that the phosphoryl derivative of 4–4′–diaminodiphenylsulfone appears different from the parent substance or its other known derivatives. Many of the sulfone derivatives studied heretofore, though less toxic than the parent substance are also less active, and what activity they possess appears to be due to the sulfone set free in the body. The phosphoryl derivative is considerably less toxic, and while its activity against streptococcus infection is somewhat less weight for weight than that of the sulfone, its activity against pneumococcus infection in relation to the tolerated dose is much better than that of the sulfone. Thus the phosphoryl derivative protected about 50 percent of mice against pneumococcus infection with a daily dosage of about one-half to one-third of the tolerated dose while the sulfone failed to protect under the same experimental conditions with maximum tolerated doses. Whether this is due to greater antibacterial specificity or to lower toxicity of the phosphoryl derivative as compared with the sulfone, it is not possible to state definitely at present. However, the increased activity towards pneumococcus as compared with streptococcus infections lends some support to specificity of action.

Accurate estimations of the phosphoryl derivative in the blood cannot be made at present; consequently, comparisons of antibacterial activity on the basis of blood concentrations cannot be made. However, the following comparison of the phosphoryl derivative with the sulfone and sulfadiazine giving dosage levels which, when administered twice daily, will produce approximately the same degree of protection in mice may be helpful.

	Streptococcus	*Pneumococcus*
4–4′–Diaminodiphenylsulfone (oral)	0. 125 mg.	>1. 0 mg.
Phosphoryl derivative (s. c.)	0. 25 mg.	1. 5–2. 5 mg.
Sulfadiazine (oral)	2. 0 mg.	15. 0–25. 0 mg.

Since the tolerated dose of the sulfone in mice is about 150 mg. per kg., while that of the phosphorylated sulfone is about 500 mg. per kg., it would appear that the phosphoryl derivative has a therapeutic index of 20 as compared with 13 for the sulfone against streptococcus

infection. The relative therapeutic index against pneumococcus infection is even more favorable. The therapeutic effectiveness of the phosphoryl derivative in experimental pneumococcus infection in mice also appears to surpass that of sulfadiazine.

The more important chemotherapeutic properties of the phosphoryl derivative of 4–4′–diaminodiphenylsulfone may be expressed numerically, in comparison with the parent sulfone, as follows (the corresponding value for the latter being in each case 1):

1. Diazotization value	
a. Before hydrolysis	0. 2
b. After hydrolysis	0. 5
2. Acute toxicity	
a. Oral	0. 2
b. Subcutaneous	0. 4
3. Antistreptococcic action	
a. In vitro	0. 25
b. In vivo	0. 5
4. Antipneumococcic action	
a. In vitro	0. 25
b. In vivo	greater than 1. 0

Earlier investigations of phosphorylated derivatives of phenols (*12*) and alcohols (*13*) showed that such compounds often acquired new pharmacologic properties though the direction of change could not always be predicted. The present experiments with the phosphoryl derivative of 4–4′–diaminodiphenylsulfone appears to be another example illustrating the same principle.

SUMMARY

The pharmacologic action and chemotherapeutic activity of a N–phosphoryl derivative of 4–4′–diaminodiphenylsulfone have been studied. Its toxicity is from one-half to one-fifth that of its parent substance. Its chemotherapeutic activity against streptococci is of about the same order as that of its parent substance.

When administered parenterally under the conditions of our experiments, this compound exhibited a curative action in experimental pneumococcus infections in mice that could not be demonstrated with the parent sulfone; the results obtained compare favorably with those from sulfadiazine and sodium sulfadiazine.

REFERENCES

(*1*) Smith, M. I., Emmart, E. W., and Westfall, B. B.: The action of certain sulfonamides, sulfones and related phosphorus compounds in experimental tuberculosis. J. Pharmacol. & Exp. Ther., **74**: 163 (1942).

(*2*) a. Buttle, G. A. H., Camb, M. A., Stephenson, D., Smith, S., Dewing, T., and Foster, G. E.: Treatment of streptococcal infection in mice with 4:4′–diaminodiphenylsulfone. Lancet, **1**: 1331 (1937).

b. Buttle, G. A. H., Dewing, T., Foster, G. E., Gray, W. H., Smith, S., and Stephenson, D.: Action of substances allied to 4:4′-diaminodiphenylsulfone in streptococcal and other infections in mice. Biochem. J., **32**: 1101 (1938).

(*3*) Fourneau, E., Trefouel, J., Trefouel, Mme., Nitti F., and Bovet, D.: Action antistreptococcique des derives sulfures organiques. C. R. Acad. Sci. Paris, **205**: 299 (1937).

(*4*) Bauer, H., and Rosenthal, S. M.: Studies in chemotherapy. VII. Some new sulphur compounds active against bacterial infections. Pub. Health Rep., **53**: 40 (1938).

(*5*) a. Raiziss, G. W., Clemence, L. W., Severac, M., and Moetsch, J. C.: Chemistry and chemotherapy of 4:4′-diaminodiphenylsulfone, 4-amino-4′-hydroxydiphenylsulfone and related compounds. J. Am. Chem. Soc., **61**: 2763 (1939).

b. Raiziss, George W., Kolmer, John A., and Rule, Anna M.: Sulfone compounds in the treatment of experimental pneumococcal infections. J. Inf. Dis. **66**: 138 (1940).

(*6*) Greey, P. H., MacLaren, D. B., and Lucas, C. C.: Comparative chemotherapy in experimental pneumococcal infections. Can. Med. Assoc. J., **40**: 319 (1939).

(*7*) Toomey, J. A., and Roach, F. E.: Promin in treatment of some acute infections. J. Pediat., **18**: 1 (1941).

(*8*) Roblin, R. O., Williams, J. H., and Anderson, G. W.: Studies in chemotherapy. III. Sulfones. J. Am. Chem. Soc., **63**: 1930 (1941).

(*9*) Dewing, T., Gray, W. H., Platt, B. C., and Stephenson, D.: Antibacterial substances allied to sulfanilamide. J. Chem. Soc., 239 (April 1942).

(*10*) Bratton, A. C., and Marshall, E. K.: New coupling component for sulfanilamide determination. J. Biol. Chem., **128**: 537 (1939).

(*11*) Litchfield, J. T., White, H. J., and Marshall, E. K., Jr.: The comparative therapeutic activity of sulfanilamide, sulfapyridine, sulfathiazole, and diaminodiphenylsulfone in type I pneumococcus infections in mice. J. Pharmacol. & Exp. Ther., **69**: 166 (1940).

(*12*) Smith, M. I.: A comparative pharmacologic study of the three phosphoric esters of orthocresol. J. Pharmacol. & Exp. Ther., **51**: 217 (1934).

(*13*) Smith, M. I.: The pharmacologic action of some alcoholic phosphoric esters. Natl. Inst. Health Bull. No. 165, No. II, Government Printing Office (1936).

PROVISIONAL MORTALITY RATES FOR THE FIRST HALF OF 1942

The mortality rates in this report are based upon preliminary data from 26 States, the District of Columbia, Alaska, and Hawaii for the first 6 months of 1942. Comparative data for the first 6 months of 1940 and 1941 are also presented for 24 States.

This report is made possible through a cooperative arrangement with the respective States which voluntarily furnish provisional monthly tabulations of current birth and death statistics to the United States Public Health Service which analyzes and publishes the data. Because of lack of uniformity in the method of classifying deaths according to cause, as well as some delay in filing certificates, these data are preliminary and may differ in some instances from the final figures subsequently published by the Bureau of the Census.

In the past, however, these preliminary reports have accurately reflected the trend in mortality rates for the country as a whole. Some deviation from the final figures, especially those for specific causes of death, for individual States may be expected because of the

provisional nature of the information. Nevertheless, it is believed that the trend in mortality within each State is correctly represented. Comparisons of specific causes of death for different States are subject to error because of variations in tabulation procedure and promptness of filing the original certificates. Such comparisons should be based upon the final figures published by the Bureau of the Census.

The mortality record of the first half of 1942 has been one of the most favorable in recent years. The death rate from all causes, 10.5 per 1,000 population, was about 5 percent less than the corresponding rate for 1941, and 8 percent less than the rate for 1940. All except five of the States for which information is available reported a decrease in the death rate.

The decrease in the mortality rate from all causes resulted from a decrease in the death rate from each of the important causes of death except cancer. For the latter cause, the death rate for the first half of 1942, 119 per 100,000 population, represented a slight increase over the rate for the first half of 1941, 118 per 100,000 population. The only other cause of death with a higher rate in 1942 than in 1941 was meningococcus meningitis, a relatively minor cause of death.

Each of the remaining important causes of death, tuberculosis, influenza, pneumonia, diabetes, cerebral hemorrhage, heart disease, nephritis, and accidents, was less prevalent during the first 6 months of 1942 than during the corresponding period of 1941. Most of the decline in the death rate from accidental causes resulted from a decrease in the rate from automobile accidents which was 14 percent less than the rate for 1941, although it was still slightly higher than the corresponding rate for 1940. All except three of the States for which data are available reported a decrease in the death rate from automobile accidents.

Both the infant and maternal mortality rates continued to decline; the former was about 10 percent and the latter nearly 20 percent less than the rate for 1941. The birth rate increased about 6 percent.

Provisional mortality from certain causes in the first 6 months of 1942, with comparative provisional data for the corresponding period in preceding years

State and period	All causes, rate per 1,000 population (annual basis)	Births (exclusive of stillbirths) per 1,000 population (annual basis)	Rate per 1,000 live births		Death rate per 100,000 population (annual basis)																		
			Total infant mortality	Maternal mortality	Typhoid fever (1–2)	Dysentery (27)	Diarrhea and enteritis under 2 years (119)	Scarlet fever (8)	Diphtheria (10)	Whooping cough (9)	Measles (35)	Cerebrospinal (meningococcus) meningitis (6)	Acute poliomyelitis and polioencephalitis (36)	Acute infectious encephalitis (lethargic) (37)	Tuberculosis, all forms (13–22)	Syphilis (30)	Influenza (grippe) (33)	Pneumonia, all forms (107–109)	Cancer, all forms (45–55)	Diabetes mellitus (61)	Cerebral hemorrhage, embolism, and thrombosis (83 a, b)	Diseases of the heart (90–93)	Nephritis, all forms (130–132)	All accidents, including automobile accidents (169–195)	Automobile accidents (170 a, b, c)
24 States[1]																									
January–June:																									
1942	10.5	18.7	44	2.6	0.4	0.6	4.3	0.4	0.5	2.1	1.3	0.8	0.2	0.4	45.8	12.3	11.2	54	119	26.6	92	302	76	65	20.2
1941	11.1	17.7	49	3.2	.6	(2)	4.4	.4	.6	3.0	2.9	.6	.3	.6	48.9	(2)	27.5	61	118	28.7	93	316	81	69	23.4
1940	11.4	16.8	50	4.2	.6	(2)	4.2	.7	.9	2.2	.5	.7	.2	.6	49.4	(2)	23.8	70	117	28.7	98	319	87	66	19.8
January–March:																									
1942	11.0	18.5	49	2.8	.3	.4	3.6	.4	.8	2.2	1.4	.7	.2	.4	45.1	12.6	15.4	68	119	28.4	96	323	79	65	22.1
1941	11.9	17.4	53	3.0	.5	(2)	3.6	.4	.8	3.1	1.6	.7	.3	.5	48.8	(2)	46.2	81	117	31.3	98	342	86	68	23.0
1940	12.3	16.5	54	4.2	.5	(2)	3.5	.8	1.2	2.2	.5	.7	.3	.6	49.1	(2)	36.4	91	119	31.4	105	344	93	67	19.1
April–June:																									
1942	10.0	19.0	41	2.5	.4	.9	5.0	.3	.2	2.0	1.1	.9	.2	.4	46.5	12.0	6.9	40	118	24.8	87	281	72	64	18.2
1941	10.4	17.9	45	3.4	.6	(2)	5.3	.4	.4	3.0	4.2	.5	.3	.6	49.1	(2)	9.0	41	118	26.2	89	290	76	69	23.8
1940	10.6	17.2	46	4.1	.7	(2)	5.0	.5	.5	2.2	.5	.6	.2	.7	49.6	(2)	11.2	49	116	26.0	91	295	81	65	20.5
Industrial policyholders:[3]																									
1942	7.7	----	----	----	.3	----	[4] 3.3	.5	.5	1.0	.7	----	----	----	43.0	10.8	5.5	36	103	29.6	63	[5] 166	[6] 52	49	18.1
1941	8.0	----	----	----	.5	----	[4] 2.6	.6	.6	1.4	1.3	----	----	----	44.7	11.9	12.7	41	105	30.3	64	[5] 170	[6] 56	47	19.3
1940	8.1	----	----	----	.4	----	[4] 3.7	.7	.9	1.3	.4	----	----	----	46.4	12.3	11.8	47	102	31.3	64	[5] 172	[6] 61	42	15.3
Alaska:																									
1942	17.5	25.9	121	1.0	(7)	(7)	7.9	(7)	2.6	15.9	5.3	5.3	(7)	(7)	341.8	5.3	18.5	135	80	7.9	66	217	40	254	10.6
1941	20.8	28.4	121	5.9	5.4	(2)	(7)	(7)	(7)	10.8	86.3	2.7	5.4	(7)	498.8	(2)	102.4	154	84	8.1	92	186	38	170	(7)
1940	20.2	26.1	156	2.1	(7)	(2)	5.5	(7)	8.2	43.8	246.2	(7)	(7)	(7)	440.5	(2)	8.2	175	79	(7)	93	233	16	137	(7)
Colorado:																									
1942	11.2	19.3	52	2.2	.5	.5	1.4	1.1	1.2	4.1	2.6	.5	.4	.2	55.7	10.6	11.6	92	120	18.3	97	264	77	87	26.6
1941	10.9	18.4	52	3.1	.9	(2)	4.8	1.1	1.6	5.7	2.1	.7	.7	.7	52.9	(2)	30.6	71	111	14.4	82	287	82	68	23.6
1940	11.2	19.2	51	3.4	.7	(2)	2.1	.5	1.6	2.7	2.0	.5	1.1	.4	51.8	(2)	16.1	87	121	17.9	89	269	77	78	23.6

Connecticut:																									
1942	9.3	15.2	30	2.1	(7)	(7)	2.6	(7)	(7)	.2	.3	.5	(7)	.1	32.1	6.7	2.3	29	129	31.6	85	307	64	53	14.3
1941	9.6	12.3	36	3.2	.2	(2)	1.8	.2	(7)	.4	.1	.2	(7)	.1	34.5	(2)	8.1	37	152	35.0	84	333	69	57	17.3
1940	10.7	12.4	42	3.0	.1	(2)	1.8	(7)	.2	.4	.2	(7)	.2	.5	34.6	(2)	6.6	58	149	22.1	119	331	66	56	16.4
Delaware:																									
1942	12.1	18.4	36	.8	(7)	(7)	3.7	2.9	(7)	(7)	(7)	(7)	(7)	.7	58.2	12.5	8.1	62	131	30.2	111	400	131	72	22.1
1941	12.6	18.2	46	(7)	(7)	(2)	5.2	1.5	(7)	3.7	(7)	(7)	(7)	(7)	55.9	(2)	20.9	69	122	27.6	107	392	145	63	23.9
1940	12.7	16.3	56	5.5	(7)	(2)	4.5	2.3	(7)	3.0	(7)	.8	(7)	(7)	50.4	(2)	18.1	72	143	33.1	110	378	137	56	21.1
District of Columbia:																									
1942	10.8	23.5	42	2.5	.2	(7)	12.7	.2	.2	2.9	(7)	2.6	.2	(7)	63.0	20.3	3.6	67	133	28.0	73	280	94	64	16.5
1941	12.3	23.5	48	2.6	.3	(2)	7.3	(7)	.5	2.2	.5	1.1	.3	(7)	62.0	(2)	6.8	84	151	26.7	91	318	103	70	20.7
1940	13.4	20.9	48	2.0	.6	(2)	7.4	.3	.3	1.8	(7)	.9	.6	(7)	66.2	(2)	11.6	97	140	33.3	99	357	132	63	19.6
Florida:																									
1942	11.6	17.1	59	4.0	2.0	2.5	8.9	.1	1.6	2.5	4.5	1.0	.1	.1	48.4	17.7	20.0	56	97	20.6	114	281	76	95	29.9
1941	13.9	17.0	59	7.0	1.3	(2)	8.3	.1	.9	1.8	1.5	.8	1.8	.5	48.4	(2)	46.0	60	107	22.9	126	321	95	122	41.0
1940	13.2	15.3	60	7.8	1.1	(2)	6.9	.1	.9	2.0	.6	.4	.2	.4	54.8	(2)	44.7	69	99	20.8	125	335	96	99	38.8
Georgia:																									
1942	8.6	19.6	57	4.2	.4	1.8	5.6	.1	1.0	2.9	3.4	.7	.3	.1	37.9	13.0	20.4	60	60	11.8	85	160	97	58	19.6
1941	10.2	19.8	65	4.0	.5	(2)	9.0	.3	1.0	4.5	7.5	.5	.5	.1	44.1	(2)	55.4	67	61	13.2	98	191	104	66	26.2
1940	10.3	18.9	61	5.4	.7	(2)	6.2	.4	1.5	2.8	1.0	.4	.3	.1	49.7	(2)	47.9	84	59	11.3	98	199	103	59	20.0
Hawaii:																									
1942	7.6	23.2	43	2.2	2.3	(7)	6.0	(7)	(7)	5.1	.5	.5	(7)	(7)	56.8	13.4	3.7	51	67	15.7	43	140	62	81	18.0
1941	7.2	22.2	47	1.9	1.4	(2)	12.7	(7)	.9	1.9	2.3	(7)	.5	.5	80.7	(2)	2.8	45	81	16.9	54	127	54	70	19.2
1940	7.4	22.3	49	2.3	2.9	(2)	6.7	(7)	2.4	2.4	(7)	.5	1.9	(7)	64.1	(2)	6.2	49	68	15.7	44	117	62	52	11.9
Idaho:																									
1942	8.6	20.7	37	2.0	.4	(7)	(7)	(7)	.7	.7	.7	.4	(7)	.4	14.5	2.6	12.2	55	74	14.8	85	244	55	74	15.9
1941	8.5	22.5	39	2.0	1.1	(2)	1.5	.8	(7)	5.3	.4	.8	(7)	1.1	14.3	(2)	17.7	37	84	20.7	77	214	58	78	23.7
1940	9.2	22.1	36	4.0	.4	(2)	3.8	1.1	.4	1.5	1.5	1.5	(7)	1.5	21.0	(2)	17.2	39	89	16.8	63	242	58	87	27.5
Indiana:																									
1942	11.0	17.7	39	3.2	.2	.2	3.0	.5	.3	1.6	.4	.1	.2	.4	38.6	10.1	20.1	58	121	12.8	136	255	81	74	29.5
1941	11.7	16.3	43	2.9	.5	(2)	2.0	.9	1.0	1.8	2.7	.7	.1	.1	39.6	(2)	32.5	61	121	15.6	144	295	68	86	36.4
1940	12.2	16.1	46	3.5	.4	(2)	2.8	1.5	.8	2.8	.1	.2	.1	.5	40.0	(2)	34.4	76	123	17.4	152	340	86	74	28.8
Iowa:																									
1942	9.6	17.3	37	2.2	(7)	.2	2.1	.2	.2	1.6	1.0	.1	.1	.4	14.9	6.3	10.6	41	134	24.3	107	286	61	63	16.7
1941	10.0	17.0	39	2.7	.2	(2)	1.3	.6	.3	1.7	.9	.3	.1	.3	13.9	(2)	21.1	53	129	26.5	105	297	63	57	18.4
1940	10.6	15.5	42	4.1	.2	(2)	1.6	.9	.9	.6	.7	.7	.5	1.1	17.4	(2)	26.6	59	135	29.0	116	312	72	61	16.8
Kansas:																									
1942	10.6	17.2	40	2.5	(7)	.3	3.2	.4	(7)	1.2	2.3	.3	.1	1.0	24.1	10.6	21.1	40	125	28.0	116	310	90	75	20.4
1941	11.0	16.2	43	2.3	.1	(2)	2.0	.3	.3	4.3	3.3	.8	.1	1.0	25.8	(2)	35.3	47	120	28.1	109	303	103	72	25.9
1940	10.7	14.8	41	3.9	(7)	(2)	2.3	.4	.8	1.8	1.7	.7	.1	1.0	24.7	(2)	29.4	44	125	27.7	107	292	105	71	21.8
Kentucky:																									
1942	9.8	(2)	(2)	(2)	1.0	2.1	5.1	1.0	1.4	4.2	1.5	1.0	.6	.2	65.6	9.4	22.2	71	83	15.1	91	236	80	69	17.6
1941	10.9	20.0	60	4.9	1.4	(2)	6.2	1.1	1.0	8.3	8.4	1.1	.9	.1	76.0	(2)	65.7	74	81	17.1	103	231	80	71	25.0
1940	10.7	20.4	55	4.8	1.8	(2)	5.4	1.1	1.5	4.5	.8	1.3	1.0	.4	69.9	(2)	42.7	74	81	15.1	111	227	74	74	20.0
Louisiana:																									
1942	9.7	19.6	58	3.4	1.5	1.7	8.3	.1	1.6	2.5	2.0	.8	.5	.2	53.2	23.9	17.3	57	88	17.8	69	241	72	55	17.5
1941	10.4	20.3	65	4.6	2.4	(2)	9.6	.3	1.1	2.1	.2	.8	.3	.1	59.0	(2)	45.1	67	85	16.4	68	264	88	65	21.5
1940	11.8	18.9	69	6.3	2.5	(2)	11.3	.1	1.6	7.0	1.0	.6	.4	.2	63.1	(2)	49.3	98	89	20.0	71	273	92	70	19.9

See footnotes at end of table.

Provisional mortality from certain causes in the first 6 months of 1942, with comparative provisional data for the corresponding period in preceding years—Continued

State and period	All causes, rate per 1,000 population (annual basis)	Births (exclusive of stillbirths) per 1,000 population (annual basis)	Rate per 1,000 live births		Death rate per 100,000 population (annual basis)																		
			Total infant mortality	Maternal mortality	Typhoid fever (1–2)	Dysentery (27)	Diarrhea and enteritis under 2 years (119)	Scarlet fever (8)	Diphtheria (10)	Whooping cough (9)	Measles (35)	Cerebrospinal (meningococcus) meningitis (6)	Acute poliomyelitis and polioencephalitis (36)	Acute infectious encephalitis (lethargic) (37)	Tuberculosis, all forms (13–22)	Syphilis (30)	Influenza (grippe) (33)	Pneumonia, all forms (107–109)	Cancer, all forms (45–55)	Diabetes mellitus (61)	Cerebral hemorrhage, embolism, and thrombosis (83, a, b)	Diseases of the heart (90–95)	Nephritis, all forms (130–132)	All accidents, including automobile accidents (169–195)	Automobile accidents (170 a, b, c)
24 States—Continued																									
Maine:																									
1942	12.6	19.9	44	2.0	.2	(?)	4.9	.7	.2	1.2	2.1	2.4	(?)	.9	31.3	7.5	13.6	64	145	32.7	129	367	84	80	19.3
1941	13.4	17.6	56	2.8	.2	(?)	5.0	.2	.5	2.6	1.4	1.2	.2	.5	33.8	(?)	33.5	74	154	32.1	132	400	99	71	18.9
1940	12.3	17.2	57	4.4	.5	(?)	5.2	1.2	.9	1.7	1.7	1.7	(?)	.2	28.7	(?)	16.1	61	145	31.8	133	370	95	65	17.5
Maryland:																									
1942	12.1	19.5	45	2.4	.2	.2	5.8	.2	.2	1.1	1.3	2.9	(?)	.4	77.1	20.2	6.7	67	134	31.3	97	361	121	77	24.2
1941	12.7	17.6	58	2.2	.4	(?)	4.6	.1	(?)	4.7	1.1	1.2	.1	.3	79.4	(?)	16.8	77	147	33.7	97	369	127	78	28.0
1940	13.1	16.3	52	3.1	.4	(?)	3.2	.4	.4	2.8	.1	.6	.2	.4	84.9	(?)	13.8	86	137	34.6	105	377	145	72	22.1
Montana:																									
1942	9.8	19.6	41	2.2	(?)	(?)	2.1	1.1	1.4	1.1	(?)	1.1	(?)	.7	35.7	12.1	7.5	53	107	12.5	97	260	57	73	19.6
1941	10.2	20.4	40	1.1	.4	(?)	3.2	2.2	2.5	1.1	.4	.4	1.4	.7	39.4	(?)	28.3	57	103	15.8	96	252	58	82	26.9
1940	10.3	20.0	41	3.9	.7	(?)	2.2	3.2	.7	.7	1.4	1.1	(?)	1.8	43.8	(?)	16.9	58	116	11.1	101	241	55	84	22.6
Nevada:																									
1942	15.3	20.5	55	(?)	1.8	3.5	10.5	1.8	(?)	12.3	1.8	3.5	1.8	3.5	73.8	12.3	7.0	77	128	15.8	114	394	58	271	87.9
1941	11.7	17.6	42	3.0	5.4	(?)	3.6	(?)	(?)	1.8	(?)	(?)	(?)	1.8	64.4	(?)	10.7	50	127	16.1	70	315	57	190	84.1
1940	12.3	18.5	44	5.9	(?)	(?)	5.4	(?)	1.8	(?)	(?)	1.8	1.8	(?)	63.6	(?)	9.1	78	114	21.8	69	303	60	149	61.8
New Mexico:																									
1942	9.7	25.2	92	2.6	.7	2.9	13.0	(?)	2.2	9.8	10.9	.4	.7	1.1	58.7	11.6	19.6	80	50	6.5	45	133	49	71	25.3
1941	10.3	27.1	85	3.7	1.8	(?)	17.4	.4	2.6	11.1	15.1	(?)	.4	.7	71.7	(?)	24.4	67	54	11.8	43	120	52	85	38.4
1940	10.4	33.1	71	4.6	1.1	(?)	14.7	(?)	1.9	10.2	.4	(?)	(?)	(?)	80.1	(?)	16.6	62	52	10.5	45	114	56	82	34.6
New York:																									
1942	11.1	16.5	33	2.0	.1	.2	2.7	.2	.1	.9	.1	.9	.1	.8	47.8	14.6	2.1	44	165	39.6	74	403	58	58	15.2
1941	11.6	15.1	35	2.3	.2	(?)	2.7	.3	(?)	.8	.6	.6	(?)	.9	48.8	(?)	6.6	55	158	43.5	76	418	64	58	16.1
1940	11.8	14.3	39	3.5	.1	(?)	3.1	.5	.1	1.2	.4	.1	(?)	1.0	49.1	(?)	4.5	57	157	43.1	77	415	70	59	14.8

North Carolina:																							
1942	8.3	23.1	54	3.8	.3	1.2	7.7	.3	.7	4.0	2.9	.5	.1	.2	46.1	7.7	11.9	60	57	13.4	86	167	81	66	25.1
1941	9.5	23.3	64	4.8	.6	(2)	7.7	.3	1.6	6.3	4.3	.5	.3	.4	50.2	(2)	46.1	76	57	14.3	84	167	89	70	32.5
1940	9.6	22.4	62	6.0	.5	(2)	7.2	.4	2.9	2.3	.5	.5	.2	.3	52.4	(2)	37.9	79	59	15.4	91	159	107	60	22.7
North Dakota:																							
1942	6.8	17.9	42	3.2	.3	.6	7.3	.3	(7)	1.9	2.2	.3	(7)	1.9	22.5	1.9	6.0	34	82	17.4	73	176	35	39	9.8
1941	8.5	21.8	45	1.9	(7)	(2)	3.8	.6	1.6	2.2	.6	(7)	.3	1.3	19.9	(2)	18.0	47	88	20.6	74	215	45	65	13.0
1940	8.0	21.2	41	1.5	.9	(2)	5.0	1.6	1.6	2.2	.3	(7)	(7)	1.6	18.2	(2)	13.8	43	97	24.5	73	206	43	49	13.8
Ohio:																							
1942	11.2	18.0	40	2.1	.3	.2	3.2	.5	.3	2.2	.8	.2	.1	.1	41.5	11.7	11.7	55	134	32.5	109	334	79	80	27.7
1941	11.6	15.6	43	2.8	.3	(2)	3.5	.3	.2	2.8	2.8	.2	.3	.7	43.8	(2)	23.6	55	136	31.8	108	331	79	83	29.9
1940	11.9	15.7	40	3.8	.8	(2)	3.3	.8	.5	1.7	(7)	.6	(6)	.5	42.6	(2)	20.5	67	136	31.8	117	334	83	83	24.6
Oklahoma:																							
1942	10.1	22.3	42	3.5	1.1	1.6	1.7	.3	2.1	2.3	8.3	.5	.7	.5	52.9	7.3	18.4	63	97	18.6	93	214	60	80	19.4
1941	9.3	19.3	56	3.1	.9	(2)	2.2	.3	2.4	7.3	1.1	.4	.5	.4	49.2	(2)	40.8	70	84	16.9	82	199	60	58	20.1
1940	9.1	17.8	48	3.6	1.5	(2)	3.4	.4	2.8	1.6	.3	1.4	1.1	.8	49.4	(2)	33.8	73	83	16.1	81	169	64	56	17.4
Pennsylvania:																							
1942	11.2	19.4	39	1.9	.3	.1	3.7	.4	.1	1.5	.8	.8	.1	.3	39.5	10.9	8.0	47	126	34.5	90	355	89	50	16.4
1941	11.5	17.7	41	2.4	.4	(2)	3.3	.4	.3	1.6	1.8	.3	.2	.8	42.4	(2)	18.2	54	124	38.3	90	367	94	55	16.5
1940	11.8	15.4	48	2.9	.5	(2)	3.7	.6	.5	1.3	.1	1.0	.2	.6	42.7	(2)	17.1	63	123	38.5	89	365	105	54	14.3
Tennessee:																							
1942	9.2	18.5	55	3.1	.3	1.0	3.5	.7	.5	2.3	1.9	.5	.5	.5	72.6	11.9	24.1	68	75	13.2	85	186	61	61	17.5
1941	10.1	17.6	60	4.0	.8	(2)	5.3	.4	.8	6.5	7.4	1.2	.3	.8	84.1	(2)	55.8	81	79	14.3	81	183	66	57	19.1
1940	10.6	16.4	59	5.6	.8	(2)	3.6	.7	1.2	3.0	1.1	.7	.1	.4	78.8	(2)	50.1	95	71	16.5	89	212	64	61	14.9
Utah:																							
1942	8.9	26.7	35	1.6	.4	.7	2.5	1.1	(7)	.4	1.8	(7)	(7)	.4	12.9	5.4	11.2	38	96	14.4	61	269	58	79	26.3
1941	8.3	24.0	31	1.1	.4	(2)	3.6	(7)	(7)	2.2	(7)	.4	(7)	.4	12.3	(2)	15.2	31	78	22.5	60	262	53	72	28.7
1940	8.9	24.5	39	2.4	.4	(2)	2.2	2.2	(7)	2.9	1.5	.7	.4	.4	17.1	(2)	18.6	42	92	17.1	55	257	55	75	27.7
Vermont:																							
1942	11.5	18.2	47	2.2	.6	.6	4.5	.6	.6	3.4	.6	.6	(7)	(7)	32.0	4.5	9.5	55	138	29.2	121	371	80	33	11.2
1941	12.0	18.2	53	2.8	.6	(2)	5.6	.6	(7)	(7)	1.2	1.7	(7)	(7)	39.8	(2)	25.8	60	140	29.2	122	399	91	51	14.6
1940	11.8	18.5	35	4.5	1.1	(2)	4.5	(7)	(7)	2.8	(7)	.6	(7)	(7)	39.7	(2)	16.8	82	127	22.4	129	329	80	44	10.6
Virginia:																							
1942	10.7	21.0	59	3.2	.5	1.8	4.9	.4	1.1	4.3	1.0	1.9	.2	.6	59.6	16.5	16.4	71	85	19.3	109	254	87	77	25.0
1941	12.0	19.9	73	4.4	.4	(2)	5.0	.3	1.2	8.9	10.6	1.3	.3	.1	66.2	(2)	49.9	80	82	21.1	109	276	106	86	32.0
1940	11.8	19.1	64	4.9	.4	(2)	4.5	.4	2.0	4.3	1.2	1.6	.3	.8	62.0	(2)	41.7	93	78	22.0	109	273	116	75	25.2

[1] The District of Columbia is included as a State. Estimated population July 1, 1942, 66,402,500. Includes all of the States listed below except Delaware, Kansas, and Oklahoma.

[2] Data not available.

[3] These data are taken from the July 1942 Statistical Bulletin published by the Metropolitan Life Insurance Co. The rates for 1941 and 1942 are subject to correction as they are based on provisional estimates of lives exposed to risk. Data do not include all diseases reported to the Public Health Service.

[4] Classified as diarrhea and enteritis, age not specified.

[5] International List (1940) titles 92, 93 c, d, e, and 95 only.

[6] Chronic nephritis only.

[7] No deaths reported.

[8] Less than 1/10 of 1 per 100,000 inhabitants.

DEATHS DURING WEEK ENDED SEPTEMBER 26, 1942

[From the Weekly Mortality Index, issued by the Bureau of the Census, Department of Commerce]

	Week ended Sept. 26, 1942	Corresponding week 1941
Data from 88 large cities of the United States:		
Total deaths	7,660	7,380
Average for 3 prior years	7,549	
Total deaths, first 38 weeks of year	316,488	319,431
Deaths per 1,000 population, first 38 weeks of year, annual rate	11.6	11.7
Deaths under 1 year of age	592	513
Average for 3 prior years	509	
Deaths under 1 year of age, first 38 weeks of year	21,564	19,872
Data from industrial insurance companies:		
Policies in force	65,043,991	64,486,432
Number of death claims	10,068	10,571
Death claims per 1,000 policies in force, annual rate	8.1	8.5
Death claims per 1,000 policies, first 38 weeks of year, annual rate	9.2	9.6

PREVALENCE OF DISEASE

No health department, State or local, can effectively prevent or control disease without knowledge of when, where, and under what conditions cases are occurring

UNITED STATES

REPORTS FROM STATES FOR WEEK ENDED OCTOBER 3, 1942

Summary

The number of reported cases of meningococcus meningitis increased during the week from 39 to 48. More than one-half of the cases occurred in the Middle Atlantic and South Atlantic States, while one-fourth occurred in the Pacific States (6 in California—the largest number reported by any one State).

A slight decrease was reported in the incidence of poliomyelitis—217 cases as compared with 220 for the preceding week. The 5-year (1937–41) median for the week is 469 cases. The highest incidence is in the East North Central (64 cases) and the Middle Atlantic States (34). The number of cases reported to date this year (2,835 cases) is below that for the corresponding period of any other year since 1938.

Expected seasonal increases were recorded for diphtheria, influenza, measles, and scarlet fever. The accumulated totals to date this year for diphtheria, smallpox, and typhoid fever are below the figures for the corresponding period of any previous year for which comparable records are available. Of 959 cases of influenza (5-year median, 599), 379 occurred in Texas and 171 in South Carolina.

The number of reported cases of endemic typhus fever declined from 145 to 76. The latter is the lowest weekly figure since the week ended July 18. The indications are, however, that the total number of cases which will be reported this year will exceed that for any of the preceding five years.

Other reports received during the week include 3 cases of undulant fever in Maryland and 1 case in Pennsylvania, 29 cases of amebic dysentery, 279 cases of bacillary (118 in Texas and 73 in New York), 200 cases of unspecified dysentery (142 in Virginia and 33 in Arizona), 9 cases of infectious encephalitis, 1 case of leprosy (in California), 7 cases of Rocky Mountain spotted fever (all in the eastern States), 1 case of smallpox (in Wisconsin), and 12 cases of tularemia.

The death rate for 88 large cities in the United States increased rather sharply during the current week—from 10.7 per 1,000 population last week to 11.5 (same week last year, 10.7; 3-year average, 10.6). The cumulative rate to date is 11.6, as compared with 11.7 for the same period of 1941.

Telegraphic morbidity reports from State health officers for the week ended October 3, 1942, and comparison with corresponding week of 1941 and 5-year median

In these tables a zero indicates a definite report, while leaders imply that, although none were reported, cases may have occurred.

Division and State	Diphtheria			Influenza			Measles			Meningitis, meningococcus		
	Week ended—		Median 1937–41	Week ended—		Median 1937–41	Week ended—		Median 1937–41	Week ended—		Median 1937–41
	Oct. 3, 1942	Oct. 4, 1941		Oct. 3, 1942	Oct. 4, 1941		Oct. 3, 1942	Oct. 4, 1941		Oct. 3, 1942	Oct. 4, 1941	
NEW ENG.												
Maine	0	0	1	------	------	1	0	28	8	1	0	0
New Hampshire	0	0	0	------	------	------	0	7	0	0	0	0
Vermont	0	0	0	------	------	------	17	0	0	0	0	0
Massachusetts	3	4	3	------	------	------	40	53	52	3	3	0
Rhode Island	4	0	0	------	------	------	4	2	1	0	0	0
Connecticut	8	0	1	1	1	1	2	4	5	1	1	0
MID. ATL.												
New York	9	8	13	[1] 6	------	[1] 6	42	48	60	5	3	2
New Jersey	1	2	6	5	3	5	25	24	24	3	0	0
Pennsylvania	6	8	14	------	------	------	51	90	90	4	2	2
E. NO. CEN.												
Ohio	6	30	30	5	4	14	22		18	0	0	1
Indiana	4	20	20	14	23	14	7		3	0	0	0
Illinois	11	17	17	6	6	6	13		22	0	0	1
Michigan [2]	5	7	11	3	2	2	21		30	0	1	1
Wisconsin	1	0	0	34	25	25	48		39	0	0	0
W. NO. CEN.												
Minnesota	1	1	3	1	------	2	1	23	6	0	0	0
Iowa	7	4	5	------	5	5	12	6	6	0	0	0
Missouri	3	2	6	------	1	1	3	12	4	1	0	0
North Dakota	0	3	3	3	3	5	4	6	2	0	0	0
South Dakota	0	7	1	------	------	------	0	2	3	0	0	0
Nebraska	5	1	1	3	------	------	22	8	4	1	0	0
Kansas	2	2	5	9	8	2	5	4	4	0	1	1
SO. ATL.												
Delaware	1	1	0	------	------	------	0	0	0	0	0	0
Maryland [2]	5	2	6	2	------	3	8	14	5	5	1	1
Dist. of Col	1	5	2	------	1	1	0	3	3	0	0	0
Virginia	20	36	39	111	183	32	7	24	16	4	0	0
West Virginia	8	5	10	3	7	7	2	53	5	2	0	1
North Carolina	76	120	115	------	------	2	5	35	21	0	1	0
South Carolina	31	26	26	171	110	139	2	18	3	3	0	0
Georgia	25	44	38	28	22	15	3	5	2	0	0	0
Florida	7	15	12	------	3	3	2	4	2	0	0	0
E. SO. CEN.												
Kentucky	12	14	24	2	------	3	0	9	12	0	0	1
Tennessee	24	10	26	19	5	14	6	38	38	0	0	0
Alabama	18	34	34	19	------	10	1	4	5	0	1	1
Mississippi [2]	10	24	19	------	------	------	------	------	------	0	0	0
W. SO. CEN.												
Arkansas	17	32	21	29	20	14	3	32	3	0	0	0
Louisiana	6	9	13	5	39	3	2	0	1	0	1	1
Oklahoma	9	17	12	10	28	28	1	2	2	1	0	0
Texas	49	53	43	379	357	135	4	10	13	1	0	2
MOUNTAIN												
Montana	4	16	0	1	2	2	1	3	16	0	0	0
Idaho	1	0	0	------	5	3	23	2	2	0	0	0
Wyoming	1	2	2	16	------	------	12	2	3	0	0	0
Colorado	17	2	5	19	23	7	8	16	8	0	0	0
New Mexico	5	0	3	1	------	------	0	4	4	0	0	0
Arizona	2	3	2	31	39	39	3	17	3	0	0	0
Utah [2]	0	0	0	------	4	2	54	5	2	1	0	0
Nevada	1	0	------	------	------	------	2	0	------	0	0	------
PACIFIC												
Washington	2	1	3	1	------	------	87	5	6	3	0	0
Oregon	3	1	3	5	6	7	30	14	8	3	0	0
California	17	11	12	17	39	16	42	77	72	6	1	1
Total	448	599	609	959	974	599	647	824	824	48	16	23
39 weeks	9,374	9,879	14,808	84,770	494,500	162,982	470,048	828,494	351,182	2,671	1,587	1,587

See footnotes at end of table.

Telegraphic morbidity reports from State health officers for the week ended October 3, 1942, and comparison with corresponding week of 1941 and 5-year median—Continued

Division and State	Poliomyelitis			Scarlet fever			Smallpox			Typhoid and paratyphoid fever		
	Week ended—		Median 1937–41	Week ended—		Median 1937–41	Week ended—		Median 1937–41	Week ended—		Median 1937–41
	Oct. 3, 1942	Oct. 4, 1941		Oct. 3, 1942	Oct. 4, 1941		Oct. 3, 1942	Oct. 4, 1941		Oct. 3, 1942	Oct. 4, 1941	
NEW ENG.												
Maine	1	7	0	4	4	4	0	0	0	3	3	2
New Hampshire	0	2	0	6	3	3	0	0	0	0	0	0
Vermont	3	1	1	2	0	4	0	0	0	1	0	0
Massachusetts	1	10	4	94	81	40	0	0	0	11	6	2
Rhode Island	0	1	0	3	0	1	0	0	0	0	0	0
Connecticut	3	12	5	25	10	10	0	0	0	1	1	2
MID. ATL.												
New York	20	87	45	112	92	101	0	0	0	12	11	18
New Jersey	9	22	12	32	38	38	0	0	0	2	3	5
Pennsylvania	5	51	31	80	65	85	0	0	0	17	18	18
E. NO. CEN.												
Ohio	7	32	32	77	118	121	0	0	0	5	18	18
Indiana	3	1	4	35	36	68	0	0	1	1	4	4
Illinois	37	18	18	76	75	138	0	0	0	14	7	29
Michigan [2]	16	19	44	51	71	100	0	0	0	6	11	4
Wisconsin	1	2	8	57	66	66	1	0	0	1	1	2
W. NO. CEN.												
Minnesota	5	15	23	28	23	28	0	0	2	0	0	2
Iowa	5	2	16	37	34	34	0	0	1	1	3	3
Missouri	3	0	2	32	22	25	0	1	0	7	6	13
North Dakota	1	1	1	3	5	11	0	0	1	0	1	1
South Dakota	0	1	1	5	8	8	0	0	0	0	1	2
Nebraska	15	0	1	14	10	12	0	0	0	0	2	1
Kansas	9	0	4	29	41	56	0	0	0	2	2	5
SO. ATL.												
Delaware	2	0	0	3	4	3	0	0	0	1	0	0
Maryland [2]	1	18	2	17	18	18	0	0	0	7	5	6
Dist. of Col	0	12	2	14	17	6	0	0	0	0	2	1
Virginia	2	10	3	41	15	24	0	0	0	6	7	18
West Virginia	0	1	1	43	21	35	0	0	0	2	8	14
North Carolina	8	7	3	78	77	77	0	0	0	2	4	10
South Carolina	3	8	1	13	15	13	0	0	0	2	7	11
Georgia	2	11	*2	36	39	27	0	0	0	13	7	10
Florida	1	7	0	1	4	4	0	0	0	5	8	4
E. SO. CEN.												
Kentucky	3	6	6	29	32	52	0	0	0	5	11	11
Tennessee	7	27	2	75	54	49	0	0	0	14	18	11
Alabama	1	22	1	32	36	30	0	0	1	4	6	6
Mississippi [2]	1	7	1	18	13	13	0	1	0	2	7	7
W. SO. CEN.												
Arkansas	7	4	2	4	11	14	0	0	0	8	9	13
Louisiana	2	1	1	6	11	5	0	0	0	4	6	16
Oklahoma	2	3	3	10	16	14	0	0	1	7	6	6
Texas	4	4	7	32	13	24	0	1	1	22	17	31
MOUNTAIN												
Montana	0	1	1	10	8	9	0	0	0	1	0	0
Idaho	0	0	1	10	1	7	0	0	0	0	0	1
Wyoming	5	0	0	2	3	3	0	0	0	0	0	0
Colorado	2	1	1	10	15	18	0	1	1	2	12	12
New Mexico	1	1	1	1	8	4	0	0	0	9	1	7
Arizona	1	0	0	2	3	3	0	0	0	3	2	2
Utah [2]	1	1	2	7	6	6	0	0	0	0	0	0
Nevada	0	0	------	0	0	------	0	0	------	1	0	------
PACIFIC												
Washington	0	7	6	19	41	18	0	0	1	2	0	2
Oregon	0	8	3	3	10	10	0	0	1	0	0	1
California	17	5	8	67	74	95	0	0	1	7	9	9
Total	217	456	469	1, 385	1, 367	1, 487	1	4	19	213	250	387
39 weeks	2, 835	6, 845	6, 845	94, 716	95, 332	122, 665	640	1, 179	8, 284	5, 350	6, 655	10, 090

See footnotes at end of table.

Telegraphic morbidity reports from State health officers for the week ended October 3, 1942—Continued

Division and State	Whooping cough		Week ended Oct. 3, 1942								
	Week ended—		Anthrax	Dysentery			Encephalitis	Leprosy	Rocky Mountain spotted fever	Tularemia	Typhus fever
	Oct. 3, 1942	Oct. 4, 1941		Amebic	Bacillary	Unspecified					
NEW ENG.											
Maine	36	10	0	0	0	0	0	0	0	0	0
New Hampshire	2	14	0	0	0	0	0	0	0	0	0
Vermont	17	2	0	0	0	0	0	0	0	0	0
Massachusetts	113	138	0	0	3	0	0	0	0	0	0
Rhode Island	26	43	0	0	0	0	0	0	0	0	0
Connecticut	38	47	0	0	3	0	0	0	0	0	0
MID. ATL.											
New York	329	359	0	1	73	0	2	0	1	0	0
New Jersey	133	133	0	0	1	0	0	0	0	0	0
Pennsylvania	250	204	0	0	0	0	0	0	0	0	0
E. NO. CEN.											
Ohio	129	225	0	0	0	0	0	0	0	0	0
Indiana	30	10	0	0	0	0	0	0	0	0	0
Illinois	166	182	0	2	14	0	2	0	0	0	0
Michigan [2]	193	347	0	0	5	0	0	0	0	0	0
Wisconsin	187	206	0	0	0	0	0	0	0	0	0
W. NO. CEN.											
Minnesota	34	47	0	3	0	0	0	0	0	1	0
Iowa	23	5	0	0	0	0	0	0	0	0	0
Missouri	5	9	0	0	0	0	0	0	0	0	0
North Dakota	22	27	0	0	0	0	0	0	0	0	0
South Dakota	0	1	0	0	0	0	0	0	0	0	0
Nebraska	7	0	0	0	0	0	0	0	0	0	0
Kansas	22	41	0	0	0	0	1	0	0	2	0
SO. ATL.											
Delaware	0	3	0	0	0	0	0	0	0	0	0
Maryland [1]	64	57	0	0	0	20	0	0	2	0	0
Dist. of Col	6	15	0	0	0	0	0	0	0	0	1
Virginia	19	24	0	1	0	142	0	0	0	1	0
West Virginia	7	14	0	0	0	1	0	0	6	0	0
North Carolina	35	79	0	0	0	0	0	0	1	0	1
South Carolina	22	31	0	0	18	0	0	0	0	0	7
Georgia	10	55	0	0	2	0	0	0	0	0	22
Florida	3	16	0	0	0	0	0	0	0	0	3
E. SO. CEN.											
Kentucky	26	72	0	0	0	0	0	0	1	1	0
Tennessee	29	26	0	0	0	5	0	0	2	2	4
Alabama	8	15	0	0	0	0	0	0	0	0	7
Mississippi [2]	----	----	0	0	0	0	0	0	0	0	2
W. SO. CEN.											
Arkansas	3	27	0	3	12	0	0	0	0	2	5
Louisiana	3	2	0	4	1	0	0	0	0	0	3
Oklahoma	3	11	0	0	0	0	0	0	0	0	0
Texas	104	68	0	11	118	0	0	0	0	1	21
MOUNTAIN											
Montana	28	8	0	0	1	0	0	0	0	0	0
Idaho	1	0	0	0	0	0	0	0	0	0	0
Wyoming	11	7	0	0	0	0	0	0	0	0	0
Colorado	19	68	0	0	5	0	0	0	0	0	0
New Mexico	26	18	0	0	18	0	0	0	0	0	0
Arizona	3	4	0	0	0	33	2	0	0	0	0
Utah [2]	21	11	0	0	0	0	0	0	0	1	0
Nevada	0	4	0	0	0	0	0	0	0	0	0
PACIFIC											
Washington	18	55	0	0	0	0	0	0	0	0	0
Oregon	6	11	0	0	0	0	0	0	0	0	0
California	213	186	0	4	5	0	2	1	0	1	0
Total	2,450	2,937	0	29	279	201	9	1	7	12	76
39 weeks	139,386	165,432	----	----	----	----	----	----	----	----	----

[1] New York City only.
[2] Period ended earlier than Saturday.

WEEKLY REPORTS FROM CITIES

City reports for week ended September 19, 1942

This table lists the reports from 83 cities of more than 10,000 population distributed throughout the United States, and represents a cross section of the current urban incidence of the diseases included in the table.

	Diphtheria cases	Encephalitis, infectious, cases	Influenza		Measles cases	Meningitis, meningococcus, cases	Pneumonia deaths	Poliomyelitis cases	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
			Cases	Deaths								
Atlanta, Ga	1	0	9	0	0	0	2	0	3	0	0	0
Baltimore, Md	0	0	-----	0	3	1	6	0	9	0	1	35
Barre, Vt	0	0	-----	0	2	0	0	0	0	0	0	3
Billings, Mont	0	0	-----	0	0	0	1	0	0	0	0	0
Birmingham, Ala	0	0	1	1	0	0	3	0	3	0	1	1
Boise, Idaho	0	0	-----	0	0	0	0	0	0	0	0	0
Boston, Mass	0	0	-----	0	1	3	6	1	20	0	0	39
Bridgeport, Conn	0	0	-----	0	0	0	0	0	0	0	0	1
Brunswick, Ga	0	0	-----	0	0	0	0	0	0	0	0	0
Buffalo, N. Y	1	0	-----	0	0	0	5	0	0	0	0	14
Camden, N. J	3	0	-----	0	0	0	1	0	0	0	0	5
Charleston, S. C	0	0	1	0	0	0	0	1	0	0	0	2
Charleston, W. Va	0	0	-----	0	0	0	0	0	0	0	0	0
Chicago, Ill	11	0	1	0	7	0	18	22	19	0	3	180
Cincinnati, Ohio	1	0	-----	1	0	0	1	2	5	0	0	26
Cleveland, Ohio	1	0	3	0	1	2	5	3	17	0	0	46
Columbus, Ohio	0	0	1	1	1	0	2	0	11	0	0	2
Concord, N. H	0	0	-----	0	0	0	0	0	1	0	0	0
Cumberland, Md	0	0	-----	0	0	0	0	0	0	0	0	0
Dallas, Texas	2	0	-----	0	0	0	3	0	1	0	0	6
Denver, Colo	4	0	10	0	4	0	5	0	2	0	1	7
Detroit, Mich	1	1	-----	0	10	0	14	1	14	0	1	129
Duluth, Minn	0	0	-----	0	1	0	1	0	1	0	0	7
Fall River, Mass	2	0	-----	0	0	0	1	0	4	0	0	7
Fargo, N. Dak	0	0	-----	0	0	0	0	0	0	0	0	0
Flint, Mich	0	0	-----	0	0	0	0	0	0	0	0	6
Fort Wayne, Ind	0	0	-----	0	0	0	1	0	0	0	0	0
Frederick, Md	0	0	-----	0	0	0	0	0	0	0	0	0
Galveston, Texas	0	0	-----	0	0	0	2	0	0	0	0	1
Grand Rapids, Mich	0	0	-----	0	0	0	0	0	1	0	0	8
Great Falls, Mont	0	0	-----	0	0	0	0	0	0	0	0	0
Hartford, Conn	0	0	-----	0	1	0	1	5	2	0	1	17
Helena, Mont	0	0	-----	0	0	0	0	0	1	0	0	0
Houston, Tex	2	0	1	0	0	0	4	1	1	0	1	4
Indianapolis, Ind	2	0	-----	0	2	0	7	0	6	0	0	18
Kansas City, Mo	0	0	-----	0	2	0	2	1	7	0	0	0
Kenosha, Wis	0	0	-----	0	1	0	0	0	2	0	0	3
Los Angeles, Calif	4	0	6	0	9	1	12	3	6	0	0	23
Lynchburg, Va	0	0	-----	0	0	0	1	0	0	0	0	1
Memphis, Tenn	1	0	-----	1	2	0	1	1	1	0	0	13
Milwaukee, Wis	0	0	-----	0	7	0	7	0	7	0	0	38
Minneapolis, Minn	0	1	-----	1	1	0	2	3	7	0	0	3
Missoula, Mont	0	0	-----	0	0	0	0	0	0	0	0	2
Mobile, Ala	1	0	-----	0	0	0	2	1	1	0	0	0
Nashville, Tenn	0	0	1	0	0	0	2	0	1	0	0	4
Newark, N. J	0	0	1	0	1	0	0	1	4	0	0	15
New Haven, Conn	0	0	-----	0	0	0	0	0	3	0	0	13
New Orleans, La	0	0	-----	0	0	1	9	0	1	0	0	1
New York, N. Y	3	0	3	2	9	7	40	11	35	0	3	171
Omaha, Nebr	1	0	-----	0	1	0	2	1	1	0	0	0
Philadelphia, Pa	1	0	-----	0	3	1	16	1	19	0	3	113
Pittsburgh, Pa	6	0	2	1	1	0	7	0	3	0	2	9
Portland, Maine	0	0	-----	0	3	5	2	0	0	0	0	15
Providence, R. I	0	0	-----	0	1	0	1	0	1	0	1	17
Pueblo, Colo	1	0	-----	0	0	0	2	0	2	0	0	0

City reports for week ended September 19, 1942—Continued

	Diphtheria cases	Encephalitis, infectious, cases	Influenza		Measles cases	Meningitis, meningococcus, cases	Pneumonia deaths	Poliomyelitis cases	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
			Cases	Deaths								
Racine, Wis	0	0	------	0	1	0	1	0	5	0	0	3
Raleigh, N. C	0	0	------	0	0	0	1	0	0	0	0	6
Reading, Pa	0	0	------	0	0	0	1	0	0	0	0	6
Richmond, Va	1	0	1	1	0	0	4	0	2	0	0	0
Roanoke, Va	0	0	------	0	4	0	0	0	0	0	0	0
Rochester, N. Y	0	0	------	0	0	0	3	2	2	0	2	18
Sacramento, Calif	1	0	------	0	1	0	0	0	2	0	1	6
St. Joseph, Mo	0	0	------	0	0	0	1	0	1	0	0	0
Saint Louis, Mo	0	0	------	0	8	0	15	1	3	0	3	3
St. Paul, Minn	0	0	------	0	1	0	1	1	4	0	0	29
San Antonio, Tex	0	0	1	1	0	0	2	0	1	0	1	1
San Francisco, Calif	0	0	------	1	7	0	6	0	3	0	0	7
Savannah, Ga	0	0	3	1	0	0	1	0	0	0	0	1
Seattle, Wash	2	0	------	0	5	0	0	0	1	0	0	12
Shreveport, La	0	0	------	0	0	0	1	0	0	0	1	0
South Bend, Ind	0	0	------	0	0	0	0	0	0	0	0	0
Spokane, Wash	0	0	------	0	4	0	1	1	3	0	0	0
Springfield, Ill	0	0	------	0	1	0	1	0	0	0	0	9
Springfield, Mass	0	0	------	0	0	1	4	0	7	0	0	3
Superior, Wis	0	0	------	0	1	0	0	0	0	0	0	1
Syracuse, N. Y	0	0	------	0	1	0	1	1	1	0	0	16
Tacoma, Wash	0	0	------	0	6	0	0	0	1	0	0	0
Tampa, Fla	0	0	------	0	0	0	0	0	0	0	0	0
Terre Haute, Ind	1	0	------	0	0	0	1	0	0	0	0	1
Topeka, Kans	0	0	------	0	0	0	1	0	3	0	0	0
Trenton, N. J	0	0	------	0	1	0	0	0	0	0	0	6
Washington, D. C	1	0	------	0	1	1	5	0	8	0	1	23
Wheeling, W. Va	0	0	------	0	0	0	0	0	1	0	0	0
Wichita, Kans	0	0	------	0	0	0	2	0	3	0	0	2
Wilmington, Del	0	0	------	0	1	0	2	3	2	0	0	2
Wilmington, N. C	1	0	------	0	0	0	0	0	1	0	0	5
Winston-Salem, N. C	1	0	------	0	2	0	0	0	1	0	0	0
Worcester, Mass	0	0	------	0	0	1	6	1	11	0	0	53

Anthrax—Cases: Philadelphia, 1.
Dysentery, amebic—Cases: Cleveland, 4; Newark, 1; New York, 2; Philadelphia, 1; Rochester, 1.
Dysentery, bacillary—Cases: Baltimore, 2; Detroit, 12; Los Angeles, 11; Nashville, 6; New Haven, 1; New York, 20; Philadelphia, 2; Richmond, 2; Syracuse, 1.
Rocky Mountain spotted fever—Cases: Kansas City, 2.
Typhus fever—Cases: Atlanta, 2; Dallas, 1; Houston, 3; Mobile, 2; New Orleans, 2; Richmond, 1; Savannah, 4; Shreveport, 1; Winston-Salem, 2.

Rates (annual basis) per 100,000 population for the group of 88 cities in the preceding table (estimated population, 1942, 33,892,618)

Period	Diphtheria cases	Influenza		Measles cases	Pneumonia deaths	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
		Cases	Deaths						
Week ended Sept. 19, 1942	7.85	6.92	1.85	18.31	39.69	44.31	0.00	4.15	187.69
Average for week 1937–41	11.04	6.53	1.55	[1] 23.79	39.80	43.38	0.31	9.17	171.33

[1] Median.

FOREIGN REPORTS

CANADA

Provinces—Communicable diseases—Week ended September 5, 1942.—During the week ended September 5, 1942, cases of certain communicable diseases were reported by the Dominion Bureau of Statistics of Canada as follows:

Disease	Prince Edward Island	Nova Scotia	New Brunswick	Quebec	Ontario	Manitoba	Saskatchewan	Alberta	British Columbia	Total
Cerebrospinal meningitis		2		2	4				1	9
Chickenpox		2		34	27	12	3	1	24	103
Diphtheria		17		17	1	1	2		2	40
Dysentery				15					2	17
Encephalomyelitis						2	1			3
German measles				2	5			1	1	9
Influenza		15			7		1			23
Lethargic encephalitis							1			1
Measles		1		16	6	3	13		2	41
Mumps		14	1	7	81	9	27	6	42	187
Pneumonia		2			5				8	15
Poliomyelitis		14	8	8	6	2	2		7	47
Scarlet fever		8	4	26	50	3	11	27	13	142
Tuberculosis		3	11	164	25	75	9	60	9	356
Typhoid and paratyphoid fever		1	2	16	1	1	2		3	26
Undulant fever		1		1	1				1	4
Whooping cough		2		326	42	19	5	10	39	443
Other communicable diseases		4			256	25			2	287

CHILE

Santiago—Cerebrospinal meningitis.—Following is a table showing the number of cases of cerebrospinal meningitis and deaths from the same cause reported in Santiago, Chile, by 4-week periods from the beginning of the present epidemic:

4 weeks ended—	Cases	Deaths	4 weeks ended—	Cases	Deaths
1941—Oct. 4 [1]	45	11	1942—Continued.		
Nov. 1	65	19	Apr. 18	36	7
Nov. 29	73	15	May 16	45	12
Dec. 27	73	28	June 13	84	16
1942—Jan. 24	57	17	July 11	250	42
Feb. 21	33	12	Aug. 8	727	69
Mar. 21	25	8	Sept. 5	919	

[1] For the period Dec. 29, 1940, to Sept. 6, 1941, only 9 cases with 2 deaths were reported.

REPORTS OF CHOLERA, PLAGUE, SMALLPOX, TYPHUS FEVER, AND YELLOW FEVER RECEIVED DURING THE CURRENT WEEK

NOTE.—Except in cases of unusual prevalence, only those places are included which had not previously reported any of the above-mentioned diseases, except yellow fever, during the current year. All reports of yellow fever are published currently.

A cumulative table showing the reported prevalence of these diseases for the year to date is published in the PUBLIC HEALTH REPORTS for the last Friday in each month.

(Few reports are available from the invaded countries of Europe and other nations in war zones.)

Plague

Indochina—Laos.—During the period September 1–10, 1942, 1 fatal case of plague was reported in Laos, Indochina.

Typhus Fever

Algeria.—During the period August 11–20, 1942, 189 cases of typhus fever were reported in Algeria.

Iraq.—During the week ended August 15, 1942, 4 cases of typhus fever were reported in Iraq.

Morocco.—During the week ended September 5, 1942, 40 cases of typhus fever were reported in Morocco.

Yellow Fever

Ivory Coast.—On September 17, 1942, 1 suspected case of yellow fever was reported in Ivory Coast, no specific location being given.

COURT DECISION ON PUBLIC HEALTH

Hard clams—prohibition of digging in certain waters—action upheld.[1]—(New York Supreme Court; *Matter of De Roche;* decided 1942.) In March 1942 the New York State Conservation Commission by an order and the New York City Board of Health by a resolution prohibited the digging of hard clams in Raritan Bay. By statute authority was vested (*a*) in the conservation commission to certify those lands from which shellfish could be taken for use as food and (*b*) in the board of health to regulate all matters affecting health in the city. The petitioner sought to have the order and resolution rescinded and to have the commission and board (*a*) directed to reopen the bay for clam digging, (*b*) restrained from enforcing the order and resolution, and (*c*) directed to fix and determine on a scientific, fair, accurate, and reasonable basis a standard of purity and sanitary condition for hard clams to be taken or sold and for the waters overlying such hard clams. The power of the commission or the board to act was not challenged by the petitioner but he contended that their action treating a scientific subject had been unscientific and, as a result, was arbitrary, capricious, and unreasonable and, consequently, illegal.

The returns filed by the respondent commission and board showed that the waters were made available for removal of hard clams on January 1, 1940, and continued as an operating area for clammers until the above-mentioned order and resolution were made. When the waters were opened to the clam industry the two respondents made tests and came to certain conclusions based upon standards of safety accepted by the various authorities then vested with control. In making these tests the respondents had collaborated with a bacteriologist affiliated with those businessmen interested in the clam industry and at that time (autumn, 1939) the unanimous opinion was that "where tested waters showed a score of 70 coliforms per 100 milliliters that degree of pollution indicated an absence of pathogenic organisms or at least that condition could be assumed with safety." Later there was an unaccountable rise in the coliform scores in the bay and the authorities of the States of New York and New Jersey and of the city of New York requested the United States Public Health Service to make a comprehensive investigation of the waters of

[1] This is believed to be the first court decision recognizing the coliform scoring of waters as a criterion of shellfish safety.

Raritan Bay with relation to the harvesting for human consumption of hard clams found therein. A report was made by the Public Health Service on the public health aspects of clamming in Raritan Bay and the Supreme Court of New York said that an examination of the report "reveals a carefully prepared document and reflects most deliberate planned action wherein the utmost care was exercised in making the tests and performing experiments to the end that an accurate, fair, convincing result would be obtained. The report shows the bay waters to be dangerously polluted with sewage exposing the public to typhoid fever." This report, together with experiments, tests, and reports made by the respondents themselves, formed the basis for the prohibitory action attacked by the petitioner. The petitioner argued that the report failed to show a single contaminated clam taken from the bay but, in the court's view, that was an unimpressive criticism. The action of the respondents was upheld, the court stating in part as follows: "The presence of polluted waters is sufficient. Authorities should not wait until contamination becomes real. The only point before the court is whether respondents in adopting the resolution and making the order acted arbitrarily, capriciously or unreasonably, which is the charge of petitioner. Respondents have acted. The law authorized their acts. Their competence is not questioned. They have decided after investigation and careful consideration. In their returns they set forth the sources of information which prompted them to act. I find that these sources are unassailable. On the merits this court approves of the prohibitory action, but if it did not, on the showing herein presented, it would be unwarranted in substituting its judgment for that of the administrative authorities charged with the responsibility."

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