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STUDIES ON STRAINS OF *AEROBACTER CLOACAE* RESPONSIBLE FOR ACUTE ILLNESS AMONG WORKERS USING LOW-GRADE STAINED COTTON [1]

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Two previous papers by Neal, Schneiter, and Caminita (*17*, *20*) have presented conclusive evidence that outbreaks of acute illness among workers using low-grade stained cotton were due to an endotoxic substance produced by a Gram-negative, rod-shaped microorganism present in large numbers in and on the cotton fibers. The organism was tentatively classed in the genus *Aerobacter* and, for convenience, it was referred to as the "cotton bacterium." The clinical syndrome of the acute illness described closely resembled that of mill fever reported among cotton mill operatives and hemp fever and grain fever reported among hemp plant workers and grain handlers, respectively. Therefore, samples of cotton mill dust, hemp mill dust, and grain elevator dust were examined and cultures of microorganisms identical with those isolated from the stained cotton were obtained. This paper presents the results of taxonomic studies on cultures of the "cotton bacterium" isolated from various sources.

SOURCE OF CULTURES

One hundred twenty-six samples of materials were received for analysis. These included 103 samples of various grades of raw cotton, 2 samples of cotton seed, 7 of cotton mill dust, 2 of soil from cotton-growing regions, 4 of grain elevator screenings, 2 of hemp mill dust, and 3 of whole hemp plants. A quantitative bacteriological examination was made of each sample as follows: A 1:100 dilution was prepared by aseptically weighing 1 gm. of material into 100 cc. of sterile

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

physiological saline in wide-mouth, screw cap bottles which were then shaken mechanically for 20 minutes. Serial dilutions from 1:1,000 to 1:100,000,000 were made from the resulting suspensions. In the early investigations aliquot portions of each serial dilution were plated on standard beef infusion agar for the detection of bacteria and on potato carrot dextrose agar [2] for the detection of fungi. The beef infusion agar plates were incubated for 24 hours at 37° C. while the potato carrot dextrose agar plates were incubated at room temperature.

It was immediately apparent that the stained cotton samples were heavily contaminated with one type of micro-organism which occurred to the exclusion of significant numbers of other types of bacteria. This organism formed characteristic, large, colorless to pale yellow, mucoid colonies on potato carrot dextrose agar. This medium was therefore adopted for routine analyses.

Very high plate counts (3,000,000 to more than 10,000,000,000 per gram) of mucoid bacteria were obtained on low-grade stained cotton, cotton dust, cotton seed from stained or bolly cotton, bolly cotton in the boll, low grades of tinged cotton, and grain elevator dust. Plate counts of about 500,000 mucoid organisms per gram were obtained on 2 samples of hemp dust and 1 sample of retted hemp plants. No mucoid organisms were found in soil in which cotton had grown, nor in most samples of white cotton and high-grade tinged cotton, nor in 3 samples of unretted hemp plants.

Two hundred and fifty-eight cultures were isolated from the various samples examined and from nose and throat swabs taken during or immediately after illness due to inhaling dust from low-grade cotton. After preliminary biochemical studies had indicated that the majority of these cultures were very similar, one representative culture from each source was selected and purified by repeated plating. One hundred and seven such cultures were subjected to intensive study.

On the basis of studies detailed below the characteristic mucoid organisms were classified as *Aerobacter cloacae*. While numerous workers have studied *A. cloacae* in connection with other members of the coliform group, no reference was found in the literature to a taxonomic study of this organism since the work of Jordan in 1890. Jordan's description is incomplete by present-day standards. Although *A. cloacae* is usually reported as a white organism, Rogers, Clark, and Evans (*18*) isolated yellow strains from grain, and MacConkey (*13*) isolated from horse feces, pond water, roof washings, oats, beans, malt, and corn a yellow organism which seems to be biochemically identical with the type 1 cultures described herein. While he did not name this yellow organism, he did designate as *B.*

[2] Formula: Potatoes, 2,000 gm.; carrots, 500 gm.; dextrose, 200 gm.; magnesium sulfate, 3.0 gm.; calcium carbonate, 2.0 gm.; agar, 150 gm.; and water, 10 liters. pH adjusted to 6.8.

cloacae a white organism which was biochemically identical with the type 2 cultures described below.

Because of the paucity of complete taxonomic studies on *A. cloacae* and because of the potential occupational hazard from organisms of this type if present in large numbers in dust from organic materials such as grain and vegetable fibers during their initial processing, it seems desirable to describe in detail the strains isolated from the low-grade cotton.

Four different types of *A. cloacae* were arbitrarily distinguished on the basis of certain carbohydrate fermentations. Type 1, a yellow organism, did not actively ferment adonitol, inositol, or inulin. It did ferment dulcitol and sorbitol. It was the predominant organism in 75 samples of stained or tinged cotton, 5 samples of cotton mill dust, 2 samples of cotton in the boll, 1 sample of cotton seed, 2 samples of hemp dust, a hemp plant after retting, and 2 samples of grain dust.

Type 2, a white organism, did not actively ferment adonitol, inositol, inulin, or dulcitol. It did ferment sorbitol. It was the predominant organism in 1 sample of stained cotton, 2 samples of cotton mill dust, and 2 samples of grain dust.

Type 3, represented by a small number of closely related cultures, was a white or a yellow organism, which did not actively ferment adonitol and inulin; it produced acid and sometimes gas in inositol. These cultures differed from each other only in their reactions on dulcitol and sorbitol. Type 3 organisms predominated in 4 samples of stained cotton, 2 samples of white cotton, and 1 sample of cotton seed.

Type 4 cultures varied widely in their fermentation reactions on the various carbohydrates. This type of organism predominated in 4 samples of stained cotton and 1 sample of hurds from a hemp breaker.

Table 1 summarizes the biochemical reactions by which the four types were arbitrarily differentiated.

TABLE 1.—*Differentiation characteristics of four types of* A. cloacae

	Fermentation of—			
	Dulcitol	Sorbitol	Inositol	Saccharides [1]
Type 1	+	+	−	+
Type 2	−	+	−	+
Type 3	±	±	+	+
Type 4	±	±	±	±

[1] See table 3.

Table 2 shows the number and kind of samples examined and incidence of the various types differentiated above. It should be noted that type 1 organisms predominated in 65 of 74 samples of

cotton and in 5 of 7 samples of cotton mill dust, all of which samples were reported or suspected to have caused acute illness. Type 1 is the organism which, on the basis of previous experimental work, is known to produce an endotoxic substance capable of causing acute illness (Neal, Schneiter, and Caminita (*17*)). Type 1 was not isolated, even on repeated examination, from the other 9 samples of cotton (of the total 74 samples) and 2 samples of cotton dust which had high plate counts of mucoid bacteria. One sample of such cotton and the two samples of cotton dust contained type 2 organisms; four samples of cotton contained type 3 organisms and three samples contained type 4. One cotton sample, reported to have been treated with ultraviolet light, contained no mucoid organisms. It will be noted, also, that type 1 organisms occurred in grain dust and, in comparatively low numbers, in hemp dust. This hemp dust was not reported to have caused illness.

TABLE 2.—*Number and types of* Aerobacter cloacae *isolated from various materials examined*

Type of material	Total number samples	Number samples containing type 1	Number samples containing type 2	Number samples containing type 3	Number samples containing type 4	Number samples containing no mucoid organisms other than spore formers	Average plate count mucoid organisms/gm. (millions)
Cotton, stained or tinged, reported or suspected to have caused illness	74	65	1	4	3	[1]1	678.0
Cotton mill dust, reported or suspected to have caused illness	7	5	2				2,480.0
Cotton, stained, reported *not* to have caused illness	5	2			1	2	399.0
Cotton, stained, history unknown	3	1				2	155.0
Cotton, stained or tinged, obtained from U. S. Dept. of Agriculture for comparison	10	7				3	114.5
Cotton, white, obtained from U. S. Dept. of Agriculture, for comparison	9			2		7	([2])
Cotton in boll (bolly cotton)	2	2					48.0
Cotton seed	2	1		1			39.0
Cotton plant debris	1					1	.0
Soil in which bolly cotton had grown	2					2	.0
Hemp dust	2	2					.6
Hemp plant after retting	1	1					.5
Hurds from hemp breaker	1				1		<.1
Hemp plant, unretted	3					3	.0
Grain dust (elevator screenings)	4	2	2				18.0

[1] This sample was reported to have been treated with ultraviolet light.

[2] These two samples had plate counts of 12,000 and 13,000 mucoid organisms per gram, respectively. It is felt that these samples were accidentally contaminated by contact with other samples since the counts were low and no other white cotton samples contained mucoid organisms.

DESCRIPTION OF THE ORGANISM

MORPHOLOGY

Form.—Short, thick rods with round ends. Cultures incubated at 20° C. show organisms uniform in size and shape while those incubated at 37° C. show occasional filaments and many coccoid forms (fig. 1). Broth cultures and old agar slant cultures commonly show poorly stained, granular, or bipolarly stained forms ranging from filamentous to coccoid in shape.

Size.—On potato carrot dextrose agar, incubated 24 hours at 37° C., the organisms average 2×0.7 microns. They are slightly larger when incubated at 20° C.

Arrangement.—Single. Occasionally paired.

Motility.—Actively motile in hanging drop preparations. Long peritrichous flagella were demonstrated with Maneval's (*15*) stain. In many preparations, however, organisms having one flagellum or several polar flagella were seen (fig. 2).

Staining reaction.—The organisms are Gram-negative when stained with Hucker's modification of the Gram stain (*16*, Leaflet IV), and nonacid fast. Cultures incubated at 37° C. often showed bipolar staining and granular forms.

Spore formation.—No spores were demonstrated in 7-day agar slant cultures with Schaeffer and Fulton's modification of the Wirtz method (*16*, Leaflet IV). The low thermal death time of the organisms (see below) also precludes spore formation.

Capsule formation.—A capsule is easily demonstrated with Anthony's (*16*, Leaflet IV) stain and is often visible with Gram's stain. Cultures grown on potato carrot dextrose agar for 24 hours at 37° C. are always encapsulated. Those on horse meat infusion agar for 24 hours at 37° C. may show a thin capsule. The heavy capsules are unevenly distributed around the organism, being concentrated at one or both ends (fig. 1). In this respect these organisms resemble *A. transcapsulatus* (*22*). The bipolar staining reaction noted above appeared to be due to two organisms being joined by their encapsulated ends.

Pigment production.—All type 1 and some type 3 and type 4 cultures produced pigment after a week's incubation on horse meat infusion agar, potato carrot dextrose agar, and in tryptose phosphate broth. Pigment is produced more rapidly at 20° C. or below than at 37° C.; it is completely soluble in absolute methanol and slightly soluble in weak alkali.

CULTURAL CHARACTERISTICS

Agar slant.—Potato carrot dextrose agar or beef infusion dextrose agar incubated 24 hours at 37° C.—Growth is smooth, shining, spreading, greyish, or yellowish. It is markedly mucoid, forming a deep pocket at the foot of the slant. All growth may flow down the slant into this pocket (fig. 3).

Horse meat infusion agar and potato carrot agar without dextrose incubated 24 hours at 37° C.—Growth is smooth, shining, spreading, nonmucoid, butyrous, greyish on slant but cream colored when scraped up on a needle. Growth sometimes consists of many small separate colonies ("nailhead" appearance).

Agar plate.—Potato carrot dextrose agar.—Large mucoid, convex, spreading colonies, sometimes colorless and at other times yellowish, streaked with white (fig. 4). Subsurface colonies are lens-shaped or cuneiform and often crack the medium whence they grow typically on the surface. Occasionally a flat grey spreading colony is observed.

Horse meat infusion agar.—Colonies are small to medium, convex, smooth, and shining. They are colorless or off-white.

Broth.—Tryptose phosphate broth incubated 24 hours at either 37° C. or 20° C.—Growth is very luxuriant. The medium is turbid; a delicate pellicle forms which falls to the bottom of the tubes where heavy sediment collects. On prolonged incubation a thick ring forms around the tube at the surface of the medium. In cultures incubated at 20° C. this ring is yellow and sometimes viscous.

Blood agar plates.—The organisms are nonhemolytic.

BIOCHEMICAL FEATURES

Shortly after isolation each culture was tested on the differential chemical compounds listed below. After 2 years' cultivation on potato carrot dextrose agar, all available type 1, 2, and 3 cultures, were transplanted several times to horse meat infusion agar and a final series of tests was made on those compounds which seemed to have some differential value. Unless otherwise noted, incubation temperature was 37° C. Incubation time was 7 days, sometimes extending to 3 weeks. Readings were usually recorded at 24, 48, 72 hours, 1 week, and 3 weeks. Since type 4 cultures varied widely in their ability to ferment carbohydrates, it does not seem advisable to include a detailed report of their reactions. The following reactions apply to types 1, 2, and 3 only.

Lactose.—Acid is produced in lactose in 24 hours. Gas production and reversion to an alkaline reaction begin usually in 4 to 7 days. The amount of gas produced in 7 days ranges from 10 to 50 percent of the capacity of the Durham tubes. Type 2 and type 3 cultures

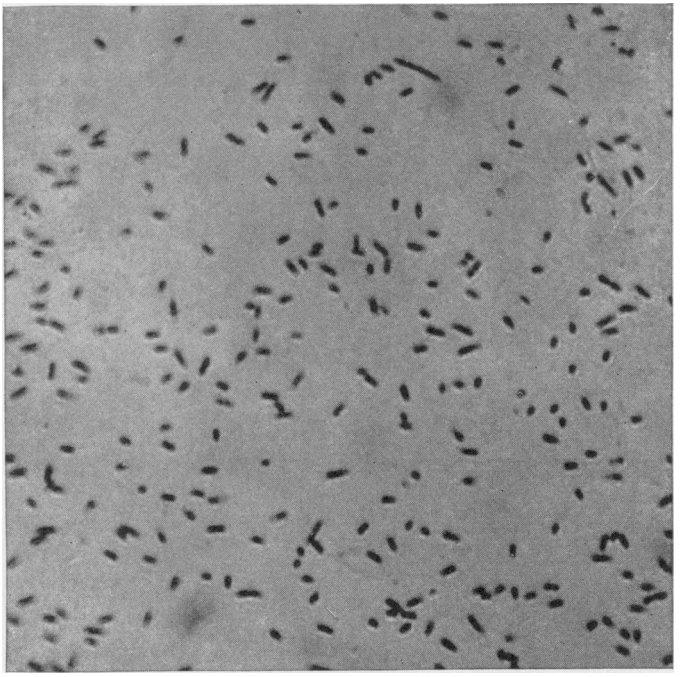

FIGURE 1.—Potato carrot dextrose agar slant culture, 24 hours at 37° C., Gram's stain. A few capsules are visible.

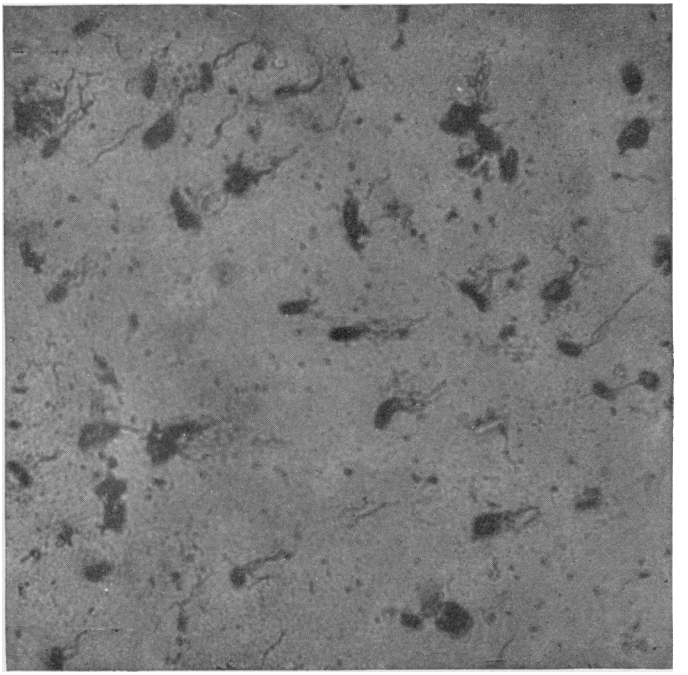

FIGURE 2.—Peritrichous flagella, Maneval's stain.

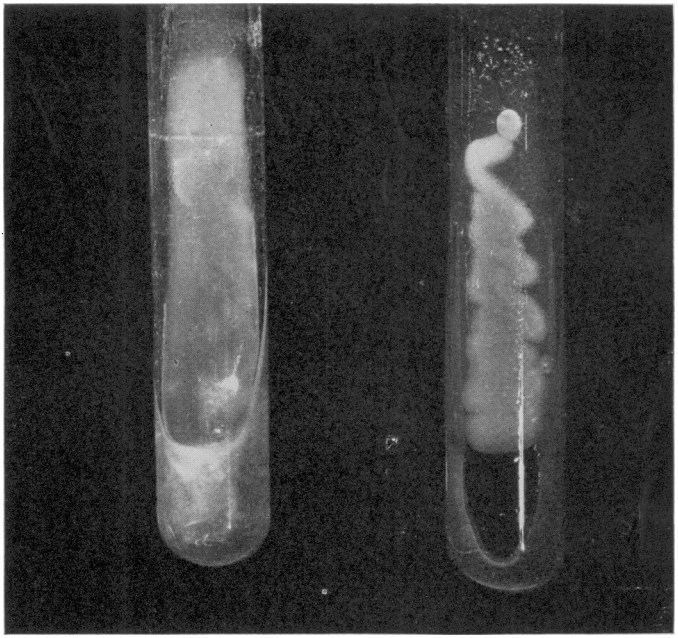

FIGURE 3.—Mucoid growth on potato carrot dextrose agar slant (left); butyrous growth on horse meat infusion agar slant (right).

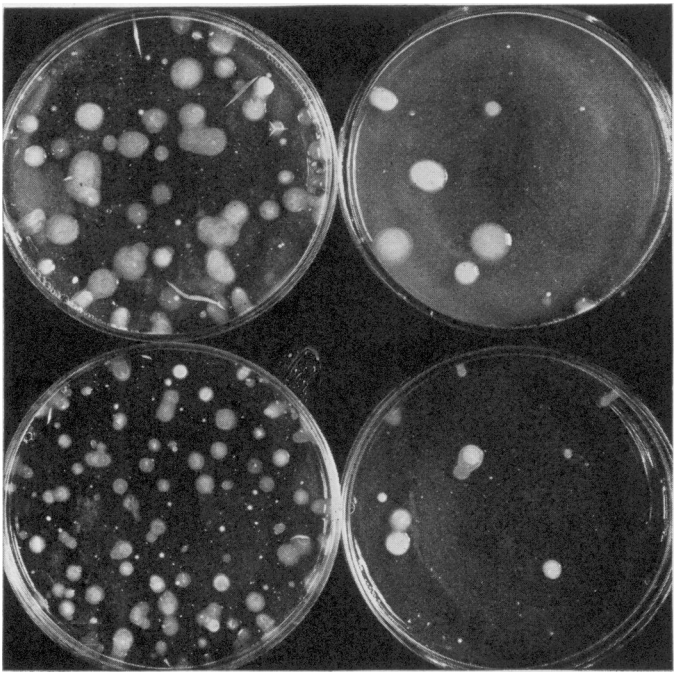

FIGURE 4.—Mucoid colonies on potato carrot dextrose agar.

generally produced 50 percent gas. Cultures varied in the speed of the fermentation. Some cultures, immediately after isolation, produced acid and gas in lactose in 24 hours. Other cultures failed to produce gas in 7 days. However, at some time during this study every type 1, 2, and 3 culture did produce acid and gas in lactose within 3 weeks. In the final tests 80 available cultures produced acid and gas in lactose, gas production beginning at 4 days' incubation. Thirteen of these cultures required 2 weeks to produce a significant amount of gas. Reduction of the indicator sometimes takes place but is not a constant characteristic of any one culture. The fermentation of lactose at 20° C. does not differ materially from that at 37° C.

Indol production.—Indol is not produced by any of the cultures. The medium consisted of Bacto tryptone (Difco), 10 gm., and distilled water, 1,000 cc., sterilized by autoclaving at 15 lbs., 121.6° C., for 15 minutes. The final pH was 7.0 to 7.2. Cultures were tested for indol production by the methods of Ruchhoft et al. (*19*) after an 18- to 24-hour incubation. Representative cultures incubated as long as 7 days did not produce indol.

Acetylmethylcarbinol production.—The Voges-Proskauer reaction is strongly positive, although after 18 months' maintenance on artificial culture media 4 of 80 available cultures gave a weak reaction. The medium consisted of Difco proteose peptone, 5 gm.; dextrose, 5 gm.; dipotassium phosphate (K_2HPO_4), 5 gm.; distilled water, 1,000 cc. The mixture was steamed 20 minutes, reaction corrected to pH 7.0, and filtered before autoclaving at 15 lbs., 121.6° C., for 15 minutes. Final pH was 6.8 to 7.0. Barritt's (*2*) reagent was used for testing for acetylmethylcarbinol after 24 to 48 hours' incubation at 37° C.

Methyl red reaction.—The methyl red reaction is negative. However, after 18 months' cultivation two typical cultures had a positive methyl red reaction and 35 others out of 80 retested had a weakly positive reaction. Cultures were incubated 4 days before testing.

Sodium citrate utilization.—All cultures showed a heavy cloudy growth after 24 hours' incubation at 37° C. in Koser's sodium citrate medium (*21*).

Uric acid utilization.—This medium (*21*) was used in the final tests. The 80 cultures used showed a fine hazy growth after 24 hours at 37° C. After a week's incubation a fine granular sediment was present in small amounts without an increase in turbidity.

Sodium hippurate hydrolysis.—Hajna and Damon (*7*), using 30 strains of *A. cloacae*, reported that these organisms failed to hydrolyze sodium hippurate. In these studies 81 cultures were inoculated into their medium. Nine cultures, including types 1, 2, and 3, gave a strongly positive test for hydrolysis after 3 days' incubation; a slight

degree of hydrolysis was produced by 13 other cultures, including types 1, 2, and 4.

Monosaccharide fermentation.—Arabinose, galactose, glucose, levulose, mannose, rhamnose, and xylose are fermented with production of acid and gas in 24 hours. The reaction of the medium is reversed to the alkaline side within 7 days. Reduction of the indicator was occasionally observed.

Disaccharide fermentation.—Cellobiose, maltose, sucrose, and trehalose are fermented with acid and gas production in 24 hours. The reaction of the medium is reversed within 7 days. Reduction of the indicator is often noted in trehalose. The fermentation of lactose has been discussed above.

Trisaccharide fermentation.—Raffinose is attacked with acid and gas production in 24 hours. The reaction of the medium is reversed to the alkaline side within 7 days.

Polysaccharide fermentation.—Dextrin is attacked with acid and gas production in 24 hours. The reaction of the medium is reversed within a week. Starch is fermented with acid and a small amount of gas in 24 hours. Gas, ranging in amount from a bubble to 25 percent of the capacity of the Durham tube, is produced within 7 days, and the reaction of the medium is reversed. Inulin is not actively fermented, but after 24 hours an acid reaction is noted in the Durham tube. Glass electrode pH determinations show a slight but significant decrease of pH in the medium. The alkalinity of the medium then increases within 7 days to about pH 8.5 to 9.0. A bubble of gas is sometimes present. No further change occurs after 3 weeks' incubation.

Alcohol fermentation.—Mannitol is fermented with acid and gas production in 24 hours and reversion of the reaction of the medium occurs within 7 days. Glycerol is attacked with acid production in 24 hours. After 4 or 5 days' incubation a bubble of gas sometimes appears. The reaction of the medium is reversed after 7 days' incubation and there is no further change after 3 weeks' incubation. Adonitol is not actively fermented. An acid reaction is present in the Durham tube within 24 hours, but the medium, examined by glass electrode, becomes progressively alkaline. This type of reaction was interpreted as negative.

Dulcitol is fermented by type 1 cultures with production of acid and gas in 24 hours. It is not actively fermented by type 2, the reaction being similar to that described above for adonitol. Type 3 cultures were variable in the production of acid and gas from dulcitol.

Inositol, like adonitol, is not actively fermented by types 1 and 2. Acid and gas is usually produced by type 3.

Sorbitol is attacked by types 1 and 2 with acid and gas production in 24 hours. It is not consistently fermented by type 3.

Glucoside fermentation.—Salicin is fermented with acid and gas production in 24 hours. There is no other change in the reaction of the medium after 7 days' incubation.

The base for the sugar differential media was made as follows: Meat extract, 3 gm.; Bacto proteose peptone No. 3, 10 gm.; sodium chloride, c. p., 5 gm.; phenol red (phenolsulfonephthalein), 0.02 gm.; and distilled water, 900 cc. The meat extract, peptone, and NaCl were dissolved in distilled water, steamed for 20 minutes, and the reaction was corrected to pH 7.6. The mixture was then reheated and the reaction recorrected if necessary. The phenol red indicator was added, the medium was filtered through paper and sterilized at 15 pounds pressure, 121.6° C., for 15 minutes. Five grams of the desired fermentable substance were dissolved in 100 cc. distilled water, sterilized by filtration through a Berkefeld or Seitz filter, and added to 900 cc. of the phenol red broth base. The medium was then dispensed into tubes which were steamed 20 minutes to drive air from the Durham tubes. The tubes were then incubated 24 hours at 37° C. to confirm sterility. Final pH value was 7.4 to 7.6.

Starch broth prepared by the above method failed to give a positive test for starch with iodine water. Therefore, Bacto nutrient broth was used, Difco soluble starch to make a 1-percent solution was added, and the mixture was steamed just long enough to dissolve the starch. The reaction was adjusted to pH 7.2 to 7.4 and the phenol red indicator was added, the medium was dispensed into tubes and autoclaved 10 minutes at 10 pounds, 115° C., for 10 minutes. The tubes were incubated 48 hours to confirm sterility. Final pH was 7.0. Control tubes made without indicator gave a positive test for starch with iodine water and a negative test for reducing sugars with Benedict's solution.

Litmus milk.—The medium is slightly acid after 24 hours' incubation. Coagulation and reduction of the indicator begin in 120 hours and are generally complete after 7 days' incubation. After 3 weeks' incubation gas may be present and some digestion may occur. The medium consisted of sterile skimmed milk heated to 80° C., with the reaction corrected to pH 7.6, and sterile saturated litmus solution added. The medium was dispensed into sterile tubes and incubated 24 hours at 37° C. to confirm sterility. Final pH was 7.4 to 7.6.

Gelatin liquefaction.—Gelatin is usually completely liquefied in 4 days. Some cultures liquefied gelatin in 48 hours and others took as

long as 11 days. Three cultures failed to liquefy gelatin in the final test although they had done so in previous tests. All cultures were incubated 3 weeks at 37° C. They were tested daily for liquefaction by placing inoculated and control tubes in the refrigerator until the control tubes had hardened, when readings were made on the inoculated tubes. The medium consisted of horse meat infusion broth, 1,000 cc., and Bacto gelatin, 125 gm. The gelatin was added to the broth and allowed to soak for 30 minutes. The mixture was then steamed until the gelatin dissolved. The reaction was corrected to pH 7.6 and the medium was dispensed into tubes and sterilized for 15 minutes at 15 lbs., 121.6° C. The final pH was 7.2 to 7.4.

Hydrogen sulfide production.—Production of hydrogen sulfide is doubtful. About half the cultures tested showed a light brown discoloration along the line of the stab after a week's incubation. The other cultures grew without discoloring the medium. The medium consisted of Bacto proteose peptone No. 3, 20 gm.; agar, 15 gm.; lead acetate, 0.5 gm.; dextrose, 1 gm.; and distilled water, 1,000 cc. The medium was tubed and sterilized at 15 lbs. pressure, 121.6° C., for 15 minutes. Final pH value was 6.6 to 6.8.

Eosin-methylene blue medium.—Levine's (*10*) e. m. b. medium streaked from 24-hour lactose broth cultures showed atypical, small, smooth, rose-colored colonies. Occasionally mucoid colonies were noted. However, none of the cultures developed colonies typical for *Aerobacter* on this medium.

Nitrate reduction.—Nitrates were reduced to nitrites in 24 hours. The medium consisted of Bacto peptone, 1 gm.; potassium nitrate (free of nitrite), 0.2 gm.; and distilled water, 1,000 cc. The medium was sterilized at 15 pounds, 121.6° C., for 15 minutes. The final pH value was 6.6 to 6.8. The test reagents recommended in the Manual of Methods for Pure Culture Study (*16*) were used.

According to Bergey's Manual of Determinative Bacteriology (*3*), the Imvic [3] reaction and fermentation of dextrose and lactose place the organisms in the genus *Aerobacter*, while the properties of gelatin liquefaction and incomplete glycerol fermentation distinguish *Aerobacter cloacae* from *A. aerogenes*. The organisms were therefore classified as *A. cloacae*.

The biochemical reactions for each of the 4 types of cultures studied are recorded in table 3.

[3] Indol production, methyl red and Voges-Proskauer reactions, and sodium citrate utilization.

TABLE 3.—*Biochemical reactions of* A. cloacae

	Cultures under investigation				American Type Culture Collection			
	Type 1	Type 2	Type 3	Type 4	222	529	961	962
Lactose[1]	AG	AG	AG	AG	AG	A	AB	AB
Indol	−	−	−	−	−	−	−	−
Methyl red	−	−	−	−	−	−	−	−
Voges-Proskauer	+	+	+	+	+	+	±	±
Sodium citrate	+	+	+	+	+	+	+	+
Adonitol[2]	−	−	−	−	−	−	−	−
Arabinose	AG	AG	AG	Variable	AG	AB	AG	AG
Cellobiose	AG	AG	AG	A	AG	A	AB	AB
Dextrin	AG	AG	AG	Variable	AG	AB	AB	AB
Dextrose	AG	AG	AG	Variable	AG	AB	A	A
Dulcitol[2]	AG	−	Variable	Variable	−	−	−	−
Galactose	AG	AG	AG	Variable	AG	AB	AG	AG
Glycerol	A	A	A	A	A	A	A	A
Inositol[2]	−	−	AG	Variable	−	−	−	−
Inulin[2]	−	−	−	−	−	−	−	−
Levulose	AG	AG	AG	Variable	AG	AB	AG	AG
Maltose	AG	AG	AG	Variable	AG	AB	AG	AG
Mannitol	AG	AG	AG	Variable	AG	A	A	A
Mannose	AG	AG	AG	Variable	AG	A	AB	AB
Raffinose	AG	AG	AG	−	AG	A	AG	AG
Rhamnose	AG	AG	AG	Variable	AG	AB	AG	AG
Salicin	AG	AG	AG		AG	A	A	AG
Sucrose	AG	AG	AG	Variable	AG	AG	AB	AG
Starch	AG	AG	AG		AB	A	A	A
Sorbitol	AG	AG	Variable	Variable	AG	A	A	A
Trehalose	AG	AG	AG	Variable	AG	A	AB	AB
Xylose	AG	AG	AG	Variable	AG	A	AG	AG
Uric acid	+	+	+		+	+	+	+
Nitrate reduction	+	+	+	+	+	+	+	+
Litmus milk	ACR	ACR	ACR	ACR	ACR	ACR	ACR	ACR
H_2S production	±	±	±	±	±	±	±	±
Gelatin liquefaction	+	+	+	+	+	+	+	+

[1] Slow fermentation. Acid in 24 hours; gas after 4 days.
[2] These reactions are recorded as negative although acid was always present in the fermentation tube. In the case of inulin a small amount of acid was produced in the medium in 24 hours, as determined by glass electrode.
A = acid.
G = gas.
B = bubble.
ACR = acid coagulation and reduction.

PHYSIOLOGICAL CHARACTERISTICS

Temperature relations.—Studies were made on 12 representative cultures of types 1, 2, and 3. The thermal death time as determined by a slightly modified Magoon's (*14*) method was 56°–57° C. for 10 minutes. One type 3 culture survived temperatures up to 60° C. for 10 minutes. Eighteen-hour cultures on potato carrot dextrose agar (encapsulated) and on horse meat infusion agar (unencapsulated) were tested.

Potato carrot dextrose agar slants, horse meat infusion agar slants, and tryptose phosphate broth tubes were used to determine optimum, minimum, and maximum growth temperatures. The optimum ranges between 25° and 37° C. The minimum is between 5° and 10° C. In this range growth is very slow. The maximum growth temperature is between 42° and 45° C. in tryptose phosphate broth or on horse meat infusion agar slants. The organism grows very poorly on potato carrot dextrose agar in this temperature range.

Relation to reaction (pH) of medium.—The optimum pH is between 6.0 and 9.5. The test medium used was Bacto standard nutrient broth adjusted to the desired pH with N/1 sodium hydroxide. The organism incubated at 37° C. does not grow in this medium at pH 4.0 or at pH 10.0. All pH determinations were made with a glass electrode.

Oxygen relationships.—The organism is a facultative anaerobe. Tubes of chopped meat medium from which air had been removed by steaming and quick cooling were employed for these studies.

Dextrose dissimilation.—Carbon dioxide and hydrogen were produced from dextrose in a medium containing 1.0-percent Witte peptone, 0.5-percent anhydrous K_2HPO_4, and 1.0-percent dextrose. The ratio of CO_2 to H_2 was not determined.

SEROLOGICAL CHARACTERISTICS

Antigenicity.—Previous serological studies (*20*) showed that identical antibodies could be produced in rabbit blood by intravenous injections of saline suspensions of viable and killed organisms, and by Berkefeld filtrates of 24- and 48-hour and 7-day tryptose broth cultures. No such antibodies were present in the blood of normal nonimmunized rabbits. The agglutinin titer of the serum tended to decrease with increasing length of time after the last injection of the antigen.

In order to determine whether the various cultures could be separated into distinct serological groups, serums were prepared against 4 type 1 cultures and 1 type 3 culture. Saline suspensions prepared from 24-hour potato carrot dextrose agar slant cultures and containing about one billion killed organisms per cc. were used. The animals were immunized by injecting gradually increasing doses of antigen, usually on alternate days throughout a 2-week period. Blood was drawn by cardiac puncture 3 or 4 days after the last injection. The serum was tested for agglutinins by dilution in the usual manner with sterile physiological saline throughout a range from 1:10 to 1:5,120. Five-tenths cc. of antigen, consisting of filtered, standardized, uniform suspensions of 18- to 24-hour potato carrot dextrose agar slant cultures of the organisms to be tested, was added to each dilution. After thorough mixing, all agglutination tubes were incubated in a constant temperature bath at 37° C. for 2 hours, followed by refrigeration at 5° C. overnight. The highest titer obtained was 1 plus at a serum dilution of 1:5,120.

Approximately 80 cultures were tested against the 5 serums. Those cultures which agglutinated to a titer of 1 plus in the 1:5,120 dilution were considered to be homologous. The results listed below indicate the high degree of serological heterology prevailing among the cultures regardless of biochemical type.

Sixteen type 1 cultures were found to be homologous with the first type 1 serum; 18 type 1 cultures were homologous with the second type 1 serum; and 3 type 1 cultures with the third. Five type 1 and 2 type 2 cultures were homologous with the fourth type 1 serum. Two type 3, 1 type 2, and 1 type 1 culture were homologous with the type 3 serum. One type 2 culture was homologous with both a type 1 serum and a type 3 serum. Many of the other cultures, although not homologous, showed marked agglutination with all the serums in the low dilutions, sometimes to a titer as high as 1 plus in the 1:1,280 dilution. In view of the tendency toward cross agglutination, the attempt to separate the cultures into distinct serological groups was abandoned. Agglutinin absorption tests were not performed. It has been shown that encapsulated strains of *B. aerogenes* differ serologically but become antigenically the same when decapsulated (*9*). Further work may prove this to be true for strains of *A. cloacae* also.

Toxin production.—Shwartzman tests and Dolman and Hammon tests (*20*) showed that a heat-stable, endotoxic substance is liberated by type 1 organisms. This endotoxic substance can be neutralized by homologous immune serum.

PATHOGENICITY AND TOXICITY

The type 1 organism has a very low pathogenicity for experimental animals. Kittens, hamsters, guinea pigs, monkeys, rabbits, and chickens exposed to dust from low-grade stained cotton (plate count 100,000,000 per gram) for one or more 7-hour periods showed no symptoms. Intranasal application of growth from 24-hour potato carrot dextrose agar slants into several species of animals did not produce ill effects. Subcutaneous injection of viable cultures into rabbits caused abscess formation from which the organisms could be recovered. Massive doses of viable organisms were required to kill mice and guinea pigs when injected intraperitoneally. Toxic filtrates (Berkefeld filtrates of 7-day tryptose phosphate broth cultures), viable cultures, and heat-killed cultures, respectively, were administered in amounts ranging from 0.25 to 1.0 cc. to 14-day-old chicks intradermally, intraperitoneally, intravenously, and by gavage without producing any noticeable symptoms. Gross autopsy findings were normal on birds that were sacrificed and examined.

Rabbits were killed by intravenous injections of sterile filtrates of 7-day tryptose broth cultures. The lethal dosage varied considerably, ranging from 0.02 to 0.5 cc. per kg. of body weight.

Human beings were made acutely ill by inhaling for 10 minutes dust from cotton of the same lot as that to which animals were exposed. The typical organisms were recovered from the upper respiratory tract by swabs streaked on potato carrot dextrose agar immediately after exposure to the cotton dust; however, the organisms could not

be recovered by this method 24 to 48 hours later. The organisms were not isolated from the blood stream of approximately 40 individuals who had had the acute illness. The same type of illness could also be caused in human beings by inhalation of sterile filtrates of 7-day tryptose broth cultures. One-tenth cc. of such filtrates injected intradermally into human beings caused severe cutaneous reactions and systemic symptoms within 3 hours.

As reported previously (*20*), the type 1 organisms did not infect cotton seedlings. Further work to determine whether this type would attack cotton bolls was carried out. Bolls averaging an inch in diameter were inoculated according to the method of Hopkins (*8*) with saline suspensions of 18- to 24-hour potato carrot dextrose agar slant cultures of *A. cloacae*, type 1, of *A. aerogenes*, and with sterile distilled water, respectively. Inoculated bolls continued to grow, and none fell from the plants. Bolls were harvested 1 and 2 weeks after inoculation and after the plants had been killed by frost, and examined as follows: Each boll was dipped into 0.1-percent mercuric chloride and then divided crosswise with a sterile scalpel. One gram of individual sections of seed and fiber were analyzed quantitatively for bacterial content and representative colonies were picked from plates and examined biochemically.

Externally, the inoculated bolls had the same appearance as the uninoculated bolls. Internally, at the site of inoculation, the boll wall was yellow or darkened, the immature fibers were yellow, and the seed coats brown. Usually only the section of the boll directly inoculated and one or two adjacent sections were attacked. The typical organisms injected in each case could be recovered in enormous numbers from the infected fibers. Microscopic examination showed the organisms growing in and on the fibers. Uninoculated bolls were sterile; those inoculated with sterile distilled water usually showed a mixed bacterial flora. On the basis of this work it would seem that organisms of the genus *Aerobacter* are saprophytic in cotton bolls, which offer a favorable medium for their development.

It was concluded that these strains of *A. cloacae* are mildly pathogenic for experimental animals, toxic to rabbits and human beings, and not pathogenic for cotton plants.

COMPARISON WITH CULTURES OF *A. CLOACAE* FROM AMERICAN TYPE CULTURE COLLECTION

Four cultures of *A. cloacae*, Nos. 222, 529, 961, and 962, were obtained from the American Type Culture Collection and tested biochemically in the media described above. Culture 222, submitted by Jordan who originally described the species, was identical biochemically with the type 2 strain. Culture 529 was nonmotile, the

individual organisms were rather long and in chains. Its Imvic was −−++; it fermented glycerol with acid production and liquefied gelatin. It was, however, rather inactive on most of the carbohydrate media and it did not reduce nitrates to nitrites. Cultures 961 and 962 were almost identical biochemically. Both, however, present different biochemical reactions now than those described by Levine (*11*). According to the scheme of classification presented herein they would be grouped as type 4. None of the four cultures was mucoid, even after repeated transfers on potato carrot dextrose agar, and none produced yellow pigment. All four cultures tested against two type 1 serums agglutinated in low titers.

DISCUSSION

The strains of *A. cloacae*, types 1, 2, and 3 described, differ from each other biochemically only in their ability to ferment dulcitol, inositol, and sorbitol. Type 4 cultures differ from each other in their ability to ferment a number of the common carbohydrate test substances. Over a period of 2 years during which 107 cultures, representing all 4 types, have been studied in the laboratory, the biochemical reactions of each culture, except type 4 organisms, have remained constant. Pigment production by type 1 cultures at temperatures ranging from 5° to 37° C. and the property of mucoid growth by all types on solid media containing glucose have also remained constant. A comparison of these cultures with 4 type cultures of *A. cloacae* (Nos. 222, 961, 962, and 529) obtained from the American Type Culture Collection showed that types 1, 2, and 3 closely resembled type culture No. 222 except for the following characteristics: Type 1 cultures fermented dulcitol, produced pigment, and showed mucoid growth on dextrose agar; type 2 cultures were identical with No. 222 except for mucoid growth on dextrose agar; type 3 cultures differed from each other in their reactions on dulcitol and sorbitol and in pigment production; they were always mucoid on dextrose agar. Type 4 cultures were similar to type cultures No. 961 and No. 962 except for mucoid growth and pigment production. It would seem, therefore, that the characteristics of pigment production and/or mucoid growth differentiate the strains under study from strains previously described.

While other investigators have observed pigment production and mucoid growth in organisms of this group, no reference was found in the literature to studies of factors governing pigment production and mucoid growth in *A. cloacae*. In view of the known instability of these characteristics in the case of other organisms, however, it does not seem justifiable to distinguish a separate variety of *A. cloacae* on this basis without further study.

A few type 1 cultures only were tested specifically for toxin production. However, it is logical to assume that types 2, 3, and 4 also produce toxin because these types only could be isolated from a few samples of cotton known to have caused illness. None of the four strains from the American Type Culture Collection evolved a toxic substance that would sensitize rabbits to the toxin produced by the type 1 cultures being studied. In precipitin tests for toxin with immune serum prepared against a type 1 culture, precipitinogens prepared from the four American Type Culture strains did not show a high titer. Since these four strains never developed a mucoid appearance and a toxic substance could not be demonstrated by available test, it is suggested that toxin production by *A. cloacae* may be correlated with the property of mucoid growth.

While slowly decreasing in numbers, the organisms are known to remain viable in baled cotton for at least 3 years. This type of nonsporulating organism does not usually survive such a long time under unfavorable conditions for growth. It has been suggested that the mucilaginous substance microscopically visible around the organisms attached to cotton fibers serves as a protective agent. The relationship, if any, between this mucilaginous substance and toxin production is unknown.

While outbreaks of food poisoning have been attributed to the ingestion of toxic substances contained in products contaminated with *A. cloacae* (*4*, *6*), the illness among workers handling low-grade cotton appears to be the first reported instance of respiratory disease due to the inhalation of such toxic products. Since *A. cloacae* is commonly known to be widely distributed in nature, it might be expected as a major contaminant of organic plant materials offering suitable conditions for its growth such as the fermented fibers of hemp, flax, and jute. During these studies type 1 organisms were isolated from hemp mill dust and from retted hemp plants. Illness similar to that observed in workers handling stained cotton has been reported in hemp, flax, and jute workers (*1*). *A. cloacae* occurs naturally on grains, and workers exposed for several hours to heavy concentrations of grain dust are subject to "grain fever" or "thresher's fever" (*12*), a clinical entity very similar to the acute illness caused by the inhalation of low-grade cotton dust. The type 1 and type 2 strains of *A. cloacae* were found to occur in large numbers in grain elevator screenings. Recently a syndrome has been described in workers exposed to dust from bagasse (sugar cane fiber) for which no cause has been directly ascribed (*5*). It is thought that bagasse with a suitable sugar content would offer a favorable medium for this group of organisms.

Finally, an investigation into conditions governing the production of toxin by this group of organisms in materials with which the worker is in close contact during processing is indicated.

SUMMARY AND CONCLUSIONS

A Gram-negative, motile, mucoid, nonsporulating, rod-shaped micro-organism was found to occur in large numbers in low-grade, stained cotton, which caused illness among workers; the same organism was also found in large numbers in dust from cotton mills, hemp mills, and grain elevators. Workers exposed to hemp dust or grain dust are known to suffer from an illness similar to that described in workers in low-grade cotton.

An intensive study was made of 107 cultures isolated from samples of raw cotton, cotton seed, cotton mill dust, elevator dust screenings, hemp mill dust, and retted hemp plants, and from patients in cases of illness.

The organisms were slow lactose fermenters, their Imvic reaction was $--++$; most of the cultures isolated actively attacked the usual carbohydrate test substances except adonitol, inositol, and inulin; they produced only acid in glycerol and liquefied gelatin; they gave a characteristic heavy mucoid growth on dextrose agar; and under suitable conditions most of them produced yellow pigment. According to Bergey's Manual, fifth edition, the Imvic reaction, gelatin liquefaction, and lack of active glycerol fermentation are characteristic of *Aerobacter cloacae* as distinguished from *A. aerogenes*. The organisms referred to in previous papers as the "cotton bacterium" were therefore classified as *A. cloacae*. There did not appear to be sufficient difference between the strains studied and cultures of *A. cloacae* obtained for comparison from the American Type Culture Collection to justify classifying these strains as a new variety of *A. cloacae*.

Four types were arbitrarily differentiated on the basis of biochemical tests: Type 1 cultures produced yellow pigment and fermented dulcitol and sorbitol but not inositol; type 2 cultures produced white growth and fermented sorbitol but not inositol or dulcitol; type 3 cultures were white or yellow, fermented inositol but usually not dulcitol and sorbitol; type 4 cultures varied widely in fermentation reactions on the various carbohydrates. Type 1, 2, and 3 cultures fermented saccharides rapidly with reversion of the reaction of the medium.

Type 1 cultures produced an endotoxin capable of causing illness in human beings when inhaled, although the bacteria themselves did not appear to survive longer than 48 hours in the human respiratory tract. They had a low pathogenicity for laboratory animals. Small

doses of the endotoxin injected intravenously killed rabbits but not mice or chickens.

Antibodies could be produced in rabbits by intravenous injections of either the endotoxic substance or killed cultures. The cultures tested against immune rabbit serums appeared to be heterologous although a considerable degree of cross agglutination appeared in low dilutions.

The organisms were not pathogenic when inoculated into cotton seedlings but were saprophytic in immature cotton bolls into which they were introduced.

ACKNOWLEDGMENTS

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REFERENCES

(1) Arlidge, J. T.: The Hygiene Diseases and Mortality of Occupations. Percival & Co., London, 1892.
(2) Barritt, M. M.: The intensification of the Voges-Proskauer reaction by the addition of α-naphthol. J. Path. and Bact., 42: 441–454 (1936).
(3) Bergey's Manual of Determinative Bacteriology. Fifth ed. The Williams & Wilkins Co., Baltimore, 1939.
(4) Buchanan, E. B., and Megrail, E.: Two outbreaks of food poisoning probably due to *B. cloacae*. J. Infec. Dis., 44: 235–242 (1929).
(5) Castleden, L. I. M., and Hamilton-Paterson, J. L.: Bagassosis; an industrial lung disease. Brit. Med. J., 2: 478–480 (1942).
(6) Gilbert, R., et al.: Food poisoning due to toxic substances formed by strains of the Cloacae-Aerogenes group. Am. J. Pub. Health, 22: 721–726 (1932).
(7) Hajna, A. A., and Damon, S. R.: Differentiation of *A. aerogenes* and *A. cloacae* on the basis of the hydrolysis of sodium hippurate. Am. J. Hyg., 19: 545–548 (1934).
(8) Hopkins, J. C.: An introductory note on two bacteria causing an internal rot of cotton bolls. Ann. Appl. Biol., 13: 260–265 (1926).
(9) Julianelle, L. A.: Immunological specificity of *Bacterium aerogenes* and its antigenic relation to pneumococcus, Type II, and Friedländer's bacillus, Type B. J. Immunol., 32: 21–33 (1937).
(10) Levine, M.: Bacteria fermenting lactose and their significance in water analysis. Iowa State Coll. of Agr. and Mech. Arts, Bull. 62, 1921.
(11) Levine, M.: Bacteria in creamery wastes. Iowa Engin. Exper. Sta. Bull. 77, 1926.
(12) Light, A. B.: "Thresher's fever." Ther. Gaz., 3rd ser., 34: 615–619 (1918).
(13) MacConkey, A.: Further observations on the differentiation of lactose-fermenting bacilli, with special reference to those of intestinal origin. J. Hyg., 9: 86–103 (1909).
(14) Magoon, C. A.: Studies upon bacterial spores. I. Thermal resistance as affected by age and environment. J. Bact., 11: 253–283 (1926).
(15) Maneval, W. E.: The staining of flagella of bacteria, with special reference to mordants. J. Bact., 21: 313–321 (1931).
(16) Society of American Bacteriologists, Committee on Bacteriological Technic: Manual of Methods for Pure Culture Study of Bacteria. Leaflet IV and Leaflet V. The Society, Geneva, N. Y., 1939.
(17) Neal, P. A., Schneiter, R., and Caminita, B. H.: Report on acute illness among rural mattress-makers using low-grade stained cotton. J. Am. Med. Assoc., 119: 1074–1082 (1942).
(18) Rogers, L. A., Clark, W. M., and Evans, A. C.: The characteristics of bacteria of the colon type occurring on grains. J. Infec. Dis., 17: 137–159 (1915).

(19) Ruchhoft, C. C., et al.: Coli-aerogenes differentiation in water analysis. II. The biochemical differential tests and their interpretation. J. Bact., 22: 125–181 (1931).
(20) Schneiter, R., Neal, P. A., and Caminita, B. H.: Etiology of acute illness among workers using low-grade stained cotton. Am. J. Pub. Health, 32: 1345–1359 (1942).
(21) Standard Methods for the Examination of Water and Sewage. Eighth ed., Am. Pub. Health Assoc., New York, 1936.
(22) Thompson, R.: An organism with a transverse capsule. J. Bact., 28: 41–42 (1934).

A SOAP WHICH INDICATES THE PRESENCE OF MERCURY FULMINATE [1]

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To help reduce the incidence of mercury fulminate dermatitis in the explosives industry (*1*) a liquid soap has been developed which indicates, by a change in color, the presence of traces of mercury fulminate upon the skin. This reagent soap has the following composition:

Diphenylthiocarbazone	0.18 gm.
Triethanolamine (technical)	250 cc.
Liquid soap	750 cc.
Hydroquinone	0.015 gm.

The soap is orange in color. In the presence of traces of mercury salts it changes rapidly to a deep, easily recognizable purple.

EXPERIMENTAL

Preparation.—Diphenylthiocarbazone, which is obtainable from most chemical supply houses as "dithizone" reagent, is added to the triethanolamine and the mixture rotated or gently shaken without warming until the solution is complete. The technical triethanolamine used in these experiments was colorless and had the following composition:

Triethanolamine, not less than 80 percent.
Diethanolamine, not more than 15 percent.
Ethanolamine, not more than 2.5 percent.

To this mixture is added the hydroquinone dissolved in the commercial liquid soap preparation, which, in our experiments, met the Government purchase specifications P–S–618 [2] and contained no extraneous substances reacting with diphenylthiocarbazone in triethanolamine solution.

Mode of action.—Fundamental to the success of any reagent soap are rapidity of reaction, sensitivity, stability, detergency, clarity of color changes, and innocuous nature. In the experiments leading to the development of the triethanolamine-diphenylthiocarbazone re-

[1] From the Dermatoses Investigations Section, Division of Industrial Hygiene, National Institute of Health.
[2] Federal Standard Stock Catalogue, section IV, part 5.

agent a number of other reagents for mercury fulminate were tested. These failed because, from the point of view of utility in a reagent soap, the reactions were much too slow (e. g., with potassium ferricyanide), because they required toxic chemicals (e. g., phenylhydrazine), or because a positive test resulted in changes which could be observed only by the technically trained eye.

A means of bringing mercury fulminate into solution was first sought as a general method of speeding test reactions. Technical triethanolamine was found satisfactory for this purpose; its inclusion in the formula of the reagent soap is based upon the work of Majrich (*2*) in which the ethanolamines are shown to be solvents for mercury fulminate. It was then found that diphenylthiocarbazone in triethanolamine solution changes color strikingly and sensitively in the presence of mercury ion, and this indicating system was accordingly incorporated into the soap.

Sensitivity.—Since under working conditions mercury fulminate is spread over or embedded in the skin rather than dissolved in it, it was necessary to determine the sensitivity of the reagent soap in terms of concentration of mercury ions per unit of skin area required to give the test, rather than the more common concentration per unit volume. This was accomplished by employing test papers on which a known and mechanically fixed area contained a known amount of mercury salt (mercuric chloride) (*3*). By this technique it was shown that one drop, or about 0.05 cc., of reagent soap solution will indicate the presence of 2γ (0.000002 gm.) of mercury ion per square centimeter. This extreme sensitivity is not applicable to skin surfaces because it requires comparison with controls; however, the results with 10 γ were unequivocal and applicable to the detection of mercury ions upon the skin.

It is evident that a practical industrial indicating soap must produce changes which require little judgment to evaluate on the part of the worker. The concentration of diphenylthiocarbazone used in the reagent soap was determined by testing graduated concentrations on the hands of workers on a fuse line where mercury fulminate was used in a primer mix, with antimony sulfide, potassium chlorate, and ground glass. The concentration of diphenylthiocarbazone recommended here produced color changes clearly perceptible to these workers without staining their hands.

Effect upon skin and hair.—The degree of staining of the skin in this case depends upon the concentrations of both diphenylthiocarbazone and mercury fulminate. With the concentration of reagent given even high concentrations of mercury fulminate upon the skin will produce no staining, although deeply embedded particles may result in a fugitive tattoo. Higher concentrations of diphenylthiocarbazone, e. g.,

0.25 gm. per liter of soap, regularly produced staining even with traces of mercury fulminate.

High concentrations of the mercury-diphenylthiocarbazone complex will perceptibly color only the lightest shades of hair; however, this may be interpreted as a contraindication to the regular use of the reagent soap as a shampoo.

The use of technical triethanolamine upon the skin in a number of dermatological preparations has been reported (*4*) and no skin hazard is to be anticipated from this source.

Stability.—The auto-oxidation of diphenylthiocarbazone in alkaline solution is an established phenomenon (*5*). We have observed, however, that in the reagent soap a decrease in the content of triethanolamine from 25 percent to 5 percent increases the rate of auto-oxidation about 10 times, i. e., 100 cc. samples in full, stoppered bottles, and not containing hydroquinone, lose their potency completely in 10 days and 1 day, respectively. A solution of diphenylthiocarbazone in 25 percent triethanolamine and 75 percent water is perfectly stable for several weeks under these conditions. It is a fair conclusion, therefore, that the soap is the cause of the rapid degradation of the mercury reagent. This is perfectly consistent with the strong tendency of unsaturated fatty acids (present in liquid soaps as their sodium or potassium salts) to form peroxides, which in this case catalyse the oxidation of diphenylthiocarbazone. To overcome this action, hydroquinone was added as an antioxidant (*6*). In this manner the stability of the reagent soap was extended to six weeks. Its reactivity may then be renewed by the addition of fresh diphenylthiocarbazone.

Precautions and limitations.—It has been shown that diphenylthiocarbazone gives characteristic colorations with a number of ions which may be divided into groups according to whether the test is carried out under basic or acidic conditions (*7*). The accompanying list (table 1) reviews those ions which give positive tests under basic conditions.

TABLE 1.—*The colors of the metal-diphenylthiocarbazone compounds in CCl_4*

[One solvent is given for the sake of consistency; in the several cases in which the colors are known both in CCl_4 and water (Hg^{++}, Ag^{+}, Cu^{++}, Ni^{++}, Co^{++}) they are the same and it is probable that this similarity obtains throughout the list.]

Ion	pH	Color in CCl_4	Ion	pH	Color in CCl_4
Cu^{++}	Alkaline	Green brown.	Ag^{+}	Alkaline	Violet.
Au^{+}	do	Red.	Zn^{++}	Weak alkaline	Red purple.
Hg^{+}	Weak alkaline	Violet.	Tl^{+}	9–12	Red.
Pb^{++}	8–10	Red.	Bi^{+++}	7–9	Orange.
Sn^{++}	6–8	Purple red.	Mn^{++}	11	Brown red.
Co^{++}	7–9	Violet.	Ni^{++}	Weak	Brown.
Cd^{++}	5 percent NaOH	Red.			

Any of these ions may be expected to interfere with the effectiveness of the soap, and their presence in either metal soap dispenser parts or primer parts which are constantly being handled may be

sufficient to discolor the soap. In using this reagent cleanser it is therefore suggested that, after it is certain that the local tap water does not contain interfering amounts of any of these metals, the workers be instructed to wash with it until it retains its original color. The skin will then be free of mercury and of any of the interfering ions.

SUMMARY

A soap solution which is a reagent for mercury fulminate is described. The active ingredients of this reagent soap are triethanolamine and diphenylthiocarbazone.

REFERENCES

(1) Schwartz, Louis: Occupational Dermatoses, pp. 185–186, *in* A Manual of Industrial Hygiene, Wm. Gafafer, editor. W. B. Saunders Co., Philadelphia, 1943.
(2) Majrich, A.: Z. ges. Schiess-Sprengstoffw., **31**: 147–148 (1936).
(3) Yagoda, H.: Applications of confined spot tests in analytical chemistry. Ind. and Eng. Chem., anal. ed., **9**: 79 (1937).
(4) Goodman, H.: Cosmetic dermatology. I. Triethanolamine. Arch. Derm. and Syph., **36**: 116–118 (1937).
(5) Clifford, P. A.: J. Assoc. Off. Ag. Chem., **26**: 26 (1943).
(6) Olcott, H. S.: J. Am. Chem. Soc., **56**: 2392 (1934).
(7) Fischer, H., and Leopoldi, G.: Chem.-Ztg., **64**: 231–233 (1940).

DEATHS DURING WEEK ENDED JULY 17, 1943

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

	Week ended July 17, 1943	Corresponding week, 1942
Data for 88 large cities of the United States:		
Total deaths	7,782	7,690
Average for 3 prior years	7,342	
Total deaths, first 28 weeks of year	259,350	235,281
Deaths under 1 year of age	583	539
Average for 3 prior years	490	
Deaths under 1 year of age, first 28 weeks of year	17,958	15,168
Data from industrial insurance companies:		
Policies in force	65,631,999	64,948,767
Number of death claims	12,255	10,029
Death claims per 1,000 policies in force, annual rate	9.7	8.1
Death claims per 1,000 policies, first 28 weeks of year, annual rate	10.2	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 JULY 24, 1943

Summary

An increase was again recorded in the incidence of poliomyelitis. A total of 329 cases was reported, as compared with 297 last week and a 5-year (1938–42) median of 124. Of the current total, 249 cases, or 76 percent, were reported in three States, as follows (last week's figures in parentheses): California, 111 (90); Texas, 96 (102); Oklahoma, 42 (39). The combined reports of these States have constituted, for the past 3 weeks, 84, 85, and 78 percent of the respective weekly totals, and for the first 29 weeks of the year 67 percent (1,312 cases) of the total of 1,955 cases in the country as a whole.

Meningococcus meningitis incidence decreased from 264 cases for the preceding week to 237 for the current week, notwithstanding increases of from 6 to 10 cases each in Illinois, Michigan, and California and minor increases in some other States. The 5-year median for the current week is 34 cases.

Current weekly totals reported for diphtheria, influenza, measles, and whooping cough were above the corresponding 5-year medians, while those for scarlet fever, smallpox, and typhoid fever were below.

Cumulative figures for the diseases included in the table for the first 29 weeks of the year (figures for the corresponding period of 1942 in parentheses) are as follows: Anthrax, 37 (51); diphtheria, 6,615 (6,765); dysentery, all forms, 12,371 (7,884); infectious encephalitis, 336 (258); influenza, 79,477 (79,322); leprosy, 17 (32); measles, 528,294 (461,421); meningococcus meningitis, 12,779 (2,188); poliomyelitis, 1,955, (875); Rocky Mountain spotted fever, 258 (277); scarlet fever, 94,785, (86,642); smallpox, 596 (596); tularemia, 534 (574); typhoid and paratyphoid fever 2,424 (3,116); endemic typhus fever, 1,638 (1,271); whooping cough, 118,067 (109,174).

Deaths in 88 large cities of the United States totaled 7,532 for the current week as compared with 7,416 for the preceding week and a 3-year (1940–42) average of 7,568. The cumulative total for the first 29 weeks of the year is 253,067, as compared with 229,645 for the same period of 1942.

Telegraphic morbidity reports from State health officers for the week ended July 24, 1943, and comparison with corresponding week of 1942 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 1938–42	Week ended—		Median 1938–42	Week ended—		Median 1938–42	Week ended—		Median 1938–42
	July 24, 1943	July 25, 1942		July 24, 1943	July 25, 1942		July 24, 1943	July 25, 1942		July 24, 1943	July 25, 1942	
NEW ENGLAND												
Maine	0	0	1	------	------	------	43	28	26	0	2	0
New Hampshire	0	0	0	------	------	------	5	0	2	1	0	0
Vermont	0	0	0	------	------	------	65	57	25	1	1	0
Massachusetts	6	2	2	------	------	------	222	185	207	11	2	0
Rhode Island	0	0	0	1	------	------	73	38	29	5	0	0
Connecticut	0	0	1	------	1	1	69	83	49	7	0	0
MIDDLE ATLANTIC												
New York	9	4	10	[1] 2	[1] 4	[1] 2	668	184	491	26	9	4
New Jersey	0	2	3	3	2	2	513	122	122	11	0	1
Pennsylvania	6	9	9	------	------	------	92	98	201	22	3	3
EAST NORTH CENTRAL												
Ohio	7	2	7	5	8	4	156	73	58	7	0	0
Indiana	2	4	4	3	------	3	48	14	14	0	1	1
Illinois	10	9	15	12	4	4	232	44	58	16	1	1
Michigan [2]	2	2	3	2	1	------	793	105	241	15	0	0
Wisconsin	1	3	2	7	12	9	468	280	373	4	0	0
WEST NORTH CENTRAL												
Minnesota	5	1	1	------	2	1	105	40	23	0	0	0
Iowa	1	0	1	------	------	------	24	57	53	0	1	0
Missouri	4	2	4	------	1	------	20	8	8	11	0	0
North Dakota	0	4	2	------	2	------	40	10	8	0	0	0
South Dakota	0	1	1	------	------	------	7	10	3	1	0	0
Nebraska	4	1	0	1	4	------	10	6	6	0	1	0
Kansas	0	2	2	1	1	1	52	23	23	2	0	0
SOUTH ATLANTIC												
Delaware	0	0	0	------	------	------	2	1	4	0	0	0
Maryland [2]	0	6	1	2	1	1	58	15	15	7	4	2
District of Columbia	0	0	1	------	------	------	34	8	8	1	0	0
Virginia	7	11	10	39	24	20	46	13	47	10	7	1
West Virginia	4	1	3	2	------	1	88	5	6	0	0	0
North Carolina	4	1	7	------	------	------	37	19	51	7	0	0
South Carolina	12	0	3	133	92	78	14	16	13	4	0	0
Georgia	4	1	3	10	8	9	10	7	7	2	2	1
Florida	4	3	3	9	4	4	10	11	10	1	0	0
EAST SOUTH CENTRAL												
Kentucky	1	1	3	1	------	------	19	3	24	0	1	2
Tennessee	4	3	2	3	8	12	16	27	27	6	0	0
Alabama	1	4	5	31	11	11	27	9	26	5	3	3
Mississippi [2]	2	8	7	------	------	------	------	------	------	3	0	0
WEST SOUTH CENTRAL												
Arkansas	4	3	3	------	5	10	11	31	22	4	0	0
Louisiana	12	1	5	6	1	4	5	8	4	3	0	0
Oklahoma	3	2	2	2	4	4	9	6	6	3	0	0
Texas	23	27	22	231	79	74	101	94	94	4	3	1
MOUNTAIN												
Montana	0	0	0	------	9	1	65	25	25	0	0	0
Idaho	0	0	0	2	------	------	4	34	3	0	0	0
Wyoming	0	0	0	------	15	------	8	13	3	0	0	0
Colorado	3	2	10	2	13	5	9	39	24	0	1	0
New Mexico	0	1	1	1	2	------	8	0	8	0	0	0
Arizona	0	5	3	31	1	13	12	7	14	1	0	0
Utah [2]	0	0	0	------	------	------	33	102	37	1	0	0
Nevada	0	0	------	------	------	------	5	9	------	1	0	------
PACIFIC												
Washington	6	1	1	------	------	------	36	177	22	4	0	0
Oregon	7	1	1	5	5	5	32	47	36	7	1	0
California	11	7	15	37	3	11	288	550	277	23	2	1
Total	169	137	148	584	327	318	4,701	2,739	3,126	237	45	34
29 weeks	6,615	6,765	8,192	79,477	79,322	150,548	528,294	461,421	461,421	12,779	2,188	1,303

See footnotes at end of table.

Telegraphic morbidity reports from State health officers for the week ended July 24, 1943, and comparison with corresponding week of 1942 and 5-year median—Con.

Division and State	Poliomyelitis			Scarlet fever			Smallpox			Typhoid and paratyphoid fever [3]		
	Week ended—		Median 1938–42	Week ended—		Median 1938–42	Week ended—		Median 1938–42	Week ended—		Median 1938–42
	July 24, 1943	July 25, 1942		July 24, 1943	July 25, 1942		July 24, 1943	July 25, 1942		July 24, 1943	July 25, 1942	
NEW ENGLAND												
Maine	0	1	0	16	1	5	0	0	0	0	0	1
New Hampshire	0	0	0	2	0	1	0	0	0	0	0	0
Vermont	0	2	0	2	0	2	0	0	0	0	1	0
Massachusetts	0	3	1	93	64	37	0	0	0	8	1	1
Rhode Island	1	0	0	10	0	3	0	0	0	0	0	0
Connecticut	2	0	1	18	4	12	0	0	0	0	0	1
MIDDLE ATLANTIC												
New York	10	2	4	79	74	84	0	0	0	8	9	12
New Jersey	0	4	1	19	16	24	0	0	0	3	2	5
Pennsylvania	2	3	3	41	42	77	0	0	0	6	10	15
EAST NORTH CENTRAL												
Ohio	2	1	1	47	79	51	0	0	0	39	13	8
Indiana	1	4	1	10	5	14	0	0	2	3	2	7
Illinois	7	12	5	37	43	63	2	0	1	5	3	11
Michigan [2]	1	7	7	26	39	76	2	1	1	45	1	3
Wisconsin	1	0	0	49	34	34	0	1	2	0	0	0
WEST NORTH CENTRAL												
Minnesota	0	0	0	10	42	27	0	0	0	0	1	0
Iowa	0	1	1	8	7	12	1	0	3	0	0	4
Missouri	4	2	1	10	15	12	0	0	3	5	2	12
North Dakota	0	1	1	0	2	3	0	0	3	0	0	0
South Dakota	0	0	0	5	11	6	0	0	1	0	1	0
Nebraska	1	0	1	4	1	3	0	1	1	0	1	0
Kansas	7	2	0	13	10	18	0	0	0	1	5	5
SOUTH ATLANTIC												
Delaware	0	0	0	1	3	1	0	0	0	0	0	0
Maryland [2]	1	0	0	21	13	10	0	0	0	2	3	6
District of Columbia	0	0	0	3	7	1	0	0	0	0	0	0
Virginia	2	3	2	3	4	11	0	0	0	2	10	10
West Virginia	0	2	2	13	21	13	0	0	0	8	14	12
North Carolina	3	2	2	6	10	10	0	0	0	3	12	12
South Carolina	2	3	3	5	1	2	0	0	0	8	5	15
Georgia	1	4	4	11	7	10	0	0	0	14	26	24
Florida	0	1	1	1	0	3	0	0	0	3	1	4
EAST SOUTH CENTRAL												
Kentucky	0	20	4	7	20	15	1	0	0	9	17	17
Tennessee	0	11	2	18	14	11	0	0	0	6	23	23
Alabama	0	3	3	10	5	6	0	0	0	12	8	8
Mississippi [2]	0	5	3	2	3	5	0	0	0	14	5	7
WEST SOUTH CENTRAL												
Arkansas	6	15	1	9	1	2	0	0	0	9	19	26
Louisiana	10	3	3	2	3	5	0	0	0	7	14	17
Oklahoma	42	0	0	6	9	9	0	0	0	3	12	12
Texas	96	2	3	18	17	17	2	0	0	25	28	43
MOUNTAIN												
Montana	0	0	0	4	2	6	0	0	0	1	2	0
Idaho	0	0	0	0	4	2	0	0	0	0	1	1
Wyoming	0	0	0	7	0	1	0	0	0	1	0	0
Colorado	5	0	0	23	9	9	0	0	1	1	3	4
New Mexico	2	1	1	0	1	3	1	0	0	5	3	3
Arizona	4	0	0	8	1	3	3	0	1	3	1	2
Utah [2]	0	0	0	7	4	6	0	0	0	0	3	2
Nevada	0	0	------	0	0	------	0	0	------	0	1	------
PACIFIC												
Washington	2	0	0	13	4	11	0	0	0	0	3	2
Oregon	3	3	0	6	1	4	0	0	1	1	0	2
California	111	1	8	99	34	42	0	0	1	4	3	6
Total	329	124	124	807	692	814	12	3	29	264	269	345
29 weeks	1,955	875	1,067	94,785	86,642	113,489	596	596	1,872	2,424	3,116	3,444

See footnotes at end of table.

Telegraphic morbidity reports from State health officers for the week ended July 24, 1943, and comparison with corresponding week of 1942 and 5-year median—Con.

Division and State	Whooping cough			Week ended July 24, 1943								
	Week ended—		Median 1938–42	Anthrax	Dysentery			Encephalitis, infectious	Leprosy	Rocky Mt. spotted fever	Tularemia	Typhus fever
	July 24, 1943	July 25, 1942			Amebic	Bacillary	Unspecified					
NEW ENGLAND												
Maine	64	22	28	0	0	0	0	0	0	0	0	0
New Hampshire	0	4	4	0	0	0	0	0	0	0	0	0
Vermont	10	50	20	0	0	0	0	0	0	0	0	0
Massachusetts	66	141	132	0	0	0	0	2	0	0	0	0
Rhode Island	43	22	15	0	0	0	0	0	0	0	0	0
Connecticut	27	67	51	0	0	0	0	0	0	0	0	0
MIDDLE ATLANTIC												
New York	269	341	341	0	1	11	0	0	0	0	0	0
New Jersey	184	254	254	0	0	0	0	0	0	2	0	0
Pennsylvania	255	274	336	0	0	0	0	0	0	1	0	0
EAST NORTH CENTRAL												
Ohio	193	183	183	0	0	1	0	0	0	1	0	0
Indiana	61	49	30	0	0	0	0	0	0	1	0	0
Illinois	223	415	363	0	2	1	0	3	0	2	1	0
Michigan [2]	354	170	269	0	1	4	0	0	0	0	0	0
Wisconsin	304	243	243	0	0	0	0	0	0	0	0	0
WEST NORTH CENTRAL												
Minnesota	85	39	39	0	1	0	0	0	0	0	1	0
Iowa	47	30	30	0	0	0	0	0	0	0	0	0
Missouri	36	38	49	0	0	0	0	0	0	1	0	0
North Dakota	35	4	10	0	0	0	0	0	0	1	0	0
South Dakota	4	0	10	0	1	0	0	0	0	0	0	0
Nebraska	9	8	13	0	0	0	0	0	0	0	0	0
Kansas	58	47	53	0	0	0	0	0	0	0	2	0
SOUTH ATLANTIC												
Delaware	0	2	5	0	0	0	0	0	0	5	0	0
Maryland [2]	112	46	57	0	0	0	2	1	0	3	1	0
District of Columbia	54	21	13	0	0	0	0	0	0	0	0	0
Virginia	103	46	76	0	0	0	439	0	0	4	0	0
West Virginia	71	20	26	0	0	0	0	0	0	0	0	1
North Carolina	268	146	239	0	0	12	0	0	0	5	0	2
South Carolina	131	49	49	0	0	46	0	0	0	0	2	10
Georgia	28	28	46	0	1	22	0	0	0	0	0	35
Florida	12	19	19	0	3	1	0	0	0	0	0	9
EAST SOUTH CENTRAL												
Kentucky	57	84	49	0	0	25	0	0	0	0	0	0
Tennessee	66	34	48	0	1	0	17	0	0	3	1	1
Alabama	54	27	27	0	0	0	0	0	0	1	0	17
Mississippi [2]	------	------	------	0	0	0	0	0	0	0	0	2
WEST SOUTH CENTRAL												
Arkansas	25	32	23	0	4	36	0	0	0	0	2	1
Louisiana	7	11	26	0	1	28	0	0	1	0	0	2
Oklahoma	18	4	19	0	0	0	0	0	0	0	0	0
Texas	336	164	164	0	89	409	0	0	0	0	3	50
MOUNTAIN												
Montana	36	27	6	0	0	0	0	0	0	0	0	0
Idaho	5	6	6	0	0	0	0	0	0	1	0	0
Wyoming	4	6	7	0	0	0	0	1	0	3	3	0
Colorado	9	15	28	0	0	1	0	1	0	0	0	0
New Mexico	4	13	19	0	0	1	1	0	0	0	0	0
Arizona	30	3	13	0	0	0	28	0	0	0	0	0
Utah [2]	66	19	50	0	0	0	0	0	0	0	0	0
Nevada	0	4	------	0	0	0	0	0	0	0	0	0
PACIFIC												
Washington	70	49	49	0	0	0	0	0	0	0	0	0
Oregon	56	17	23	0	0	0	0	0	0	0	0	0
California	242	146	240	0	1	21	0	5	0	1	0	1
Total	4, 191	3, 439	4, 061	0	106	619	487	13	1	35	16	131
29 weeks	118, 067	109, 174	113, 405	37	1, 154	8, 128	3, 089	336	17	258	534	1, 638
29 weeks, 1942	------	------	------	51	581	4, 380	2, 923	258	32	277	574	1, 271

[1] New York City only.
[2] Period ended earlier than Saturday.
[3] Including paratyphoid fever cases reported separately as follows: Massachusetts, 7; New York, 1; Illinois, 2; Michigan, 40; Florida, 1; Texas, 2; California, 2.

WEEKLY REPORTS FROM CITIES

City reports for week ended July 10, 1943

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								
NEW ENGLAND												
Maine:												
Portland	0	1	------	0	12	2	2	0	0	0	0	6
New Hampshire:												
Concord	0	0	------	0	0	0	0	0	0	0	0	0
Massachusetts:												
Boston	0	0	------	0	76	2	10	0	57	0	0	15
Fall River	0	0	------	0	14	1	0	0	1	0	1	5
Springfield	0	0	------	0	16	2	0	1	7	0	0	0
Worcester	0	0	------	0	2	0	3	0	2	0	0	8
Rhode Island:												
Providence	0	0	------	1	90	0	0	0	4	0	0	10
Connecticut:												
Bridgeport	0	0	------	0	0	0	0	0	0	0	0	1
Hartford	1	0	------	0	2	0	3	0	3	0	1	6
New Haven	0	0	------	0	15	0	0	0	0	0	0	0
MIDDLE ATLANTIC												
New York:												
Buffalo	0	0	------	0	2	1	4	0	4	0	0	4
New York	11	3	1	1	522	26	39	3	44	0	1	74
Rochester	0	0	------	0	14	0	2	0	2	0	0	9
Syracuse	0	0	------	0	15	0	1	0	2	0	0	15
New Jersey:												
Camden	0	0	------	0	0	0	0	0	1	0	0	0
Newark	0	0	------	0	49	0	2	0	3	0	0	38
Trenton	0	0	------	0	0	0	2	0	1	0	0	3
Pennsylvania:												
Philadelphia	0	0	------	2	81	8	19	0	14	0	1	83
Pittsburgh	2	1	------	0	4	3	5	0	5	0	0	20
Reading	0	0	------	0	3	0	0	0	0	0	0	9
EAST NORTH CENTRAL												
Ohio:												
Cincinnati	0	0	------	0	7	3	1	0	4	0	0	7
Cleveland	6	0	------	0	11	4	5	0	16	0	0	63
Columbus	0	0	------	0	12	1	0	0	4	0	1	2
Indiana:												
Fort Wayne	0	0	------	0	4	0	0	0	1	0	0	0
Indianapolis	0	0	7	0	0	0	2	0	5	0	0	6
South Bend	0	0	------	0	5	0	0	0	0	0	0	0
Terre Haute	0	0	------	0	0	0	0	0	0	0	0	0
Illinois:												
Chicago	5	0	1	0	166	5	12	2	20	0	1	67
Springfield	0	0	------	0	1	0	1	0	1	0	0	3
Michigan:												
Detroit	0	0	------	0	241	4	5	1	14	0	0	50
Flint	0	0	------	0	3	0	0	0	0	0	0	7
Grand Rapids	0	0	------	0	76	0	1	0	4	0	0	10
Wisconsin:												
Kenosha	0	0	------	0	1	0	0	0	5	0	0	2
Milwaukee	0	0	------	0	133	1	0	0	13	0	0	35
Racine	0	0	------	0	4	1	0	0	0	0	0	3
Superior	1	0	------	0	30	0	0	0	1	0	0	0
WEST NORTH CENTRAL												
Minnesota:												
Duluth	1	0	------	0	72	0	0	0	0	0	0	4
Minneapolis	0	0	------	0	5	1	1	1	7	0	0	1
St. Paul	0	0	------	0	11	0	5	0	6	0	0	42
Missouri:												
Kansas City	0	0	------	0	24	1	5	0	10	0	0	11
St. Joseph	0	0	------	0	0	0	0	0	1	0	0	0
St. Louis	1	0	------	0	12	3	8	0	4	0	0	22
Nebraska:												
Omaha	0	0	------	0	1	1	5	0	1	0	0	0
Kansas:												
Topeka	0	0	------	0	8	0	0	0	1	0	0	12
Wichita	0	0	------	0	7	0	2	3	0	0	0	14

City reports for week ended July 10, 1943—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								
SOUTH ATLANTIC												
Delaware:												
Wilmington	1	0	------	0	3	1	0	0	0	0	0	6
Maryland:												
Baltimore	0	0	1	0	66	4	8	0	8	0	0	83
Cumberland	0	0	------	0	0	0	0	0	0	0	0	0
Frederick	0	0	------	0	0	0	0	0	0	0	0	0
District of Columbia:												
Washington	0	0	------	0	39	5	9	0	9	0	0	38
Virginia:												
Lynchburg	0	0	------	0	9	0	0	0	0	0	0	19
Richmond	0	0	------	0	12	0	0	1	0	0	1	38
West Virginia:												
Charleston	0	0	------	0	0	0	0	0	2	0	1	2
Wheeling	0	0	------	0	0	0	3	0	0	0	0	18
North Carolina:												
Winston-Salem	0	0	------	0	0	0	2	0	0	0	0	24
South Carolina:												
Charleston	0	0	------	0	0	0	0	0	1	0	0	4
Georgia:												
Atlanta	0	0	1	0	2	1	1	0	5	0	0	4
Brunswick	0	0	------	0	1	0	0	0	0	0	0	0
Savannah	0	0	------	0	1	2	0	0	1	0	1	1
Florida:												
Tampa	0	0	------	0	0	0	2	0	0	0	0	0
EAST SOUTH CENTRAL												
Tennessee:												
Memphis	0	0	------	0	4	0	3	0	3	0	1	10
Nashville	0	0	------	0	4	0	4	0	1	0	0	10
Alabama:												
Birmingham	0	0	1	0	2	0	4	0	0	0	0	5
Mobile	0	0	------	0	0	0	1	0	0	0	0	0
WEST SOUTH CENTRAL												
Arkansas:												
Little Rock	0	0	------	0	0	0	1	0	0	0	0	2
Louisiana:												
New Orleans	2	0	7	1	2	0	7	0	1	0	0	5
Shreveport	0	0	------	0	0	1	2	7	0	0	0	0
Texas:												
Dallas	1	0	------	0	1	0	4	8	2	0	0	2
Galveston	0	0	------	0	0	0	3	1	1	0	0	1
Houston	0	0	------	0	0	1	8	9	1	0	0	3
San Antonio	4	0	------	1	0	0	5	0	1	0	0	6
MOUNTAIN												
Montana:												
Billings	0	0	------	0	6	0	2	0	1	0	0	0
Great Falls	0	0	------	0	9	0	0	0	0	0	0	1
Helena	0	0	------	0	0	0	0	0	0	0	0	0
Missoula	0	0	------	0	0	0	0	0	0	0	0	0
Colorado:												
Pueblo	0	0	------	0	0	0	0	0	4	0	0	5
Utah:												
Salt Lake City	0	0	------	0	11	0	0	0	6	0	0	38
PACIFIC												
Washington:												
Seattle	4	0	------	1	48	0	1	0	3	0	0	6
Spokane	1	0	------	0	8	1	0	0	1	0	0	2
Tacoma	0	0	------	0	0	0	0	0	0	0	0	0
California:												
Los Angeles	0	0	3	0	75	3	6	8	12	0	0	27
Sacramento	2	0	------	0	2	1	1	2	2	0	0	8
San Francisco	0	0	2	0	15	2	13	4	10	0	0	8
Total	43	5	24	7	2,071	92	230	51	343	0	10	1,043
Corresponding week, 1942	45	3	14	4	1,377	25	236	16	308	0	6	1,266
Average, 1938–42	57	------	26	[1] 10	[2] 1,668	------	[1] 231	------	425	2	33	1,251

[1] 3-year average, 1940–42.
[2] 5-year median.

Dysentery, amebic.—Cases: San Francisco, 1.
Dysentery, bacillary.—Cases: Buffalo, 5; Baltimore, 1; Charleston, S. C., 29; Nashville, 4; Los Angeles, 2.
Dysentery, unspecified.—Cases: San Antonio, 13.
Tularemia.—Cases: St. Louis, 1; Nashville, 1.
Typhus fever.—Cases: St. Louis, 1; Charleston, S. C., 1; Savannah, 3; New Orleans, 2; Dallas, 1; Houston, 1.

Rates (annual basis) per 100,000 population, by geographic groups, for the 83 cities in the preceding table (estimated population, 1942, 34,215,500)

	Diphtheria case rates	Encephalitis, infectious, case rates	Influenza		Measles case rates	Meningitis, meningococcus, case rates	Pneumonia death rates	Poliomyelitis case rates	Scarlet fever case rates	Smallpox case rates	Typhoid and paratyphoid fever case rates	Whooping cough case rates
			Case rates	Death rates								
NEW ENGLAND	2.5	2.5	0	2.5	567	17.5	44.9	2.5	184.8	0	5.0	127
MIDDLE ATLANTIC	5.8	1.8	.4	1.3	308	16.9	33.0	1.3	33.9	0	.9	114
EAST NORTH CENTRAL	7.0	0	4.7	0	405	11.1	15.8	1.8	51.4	0	1.2	149
WEST NORTH CENTRAL	4.0	0	0	0	277	11.9	51.4	7.9	59.3	0	0	210
SOUTH ATLANTIC	1.8	0	3.6	0	236	23.1	35.5	1.8	46.2	0	5.3	421
EAST SOUTH CENTRAL	0	0	5.9	0	59	0	71.3	0	23.8	0	5.9	148
WEST SOUTH CENTRAL	20.5	0	20.5	5.9	9	5.9	88.0	73.3	17.6	0	0	56
MOUNTAIN	0	0	0	0	463	0	35.6	0	195.9	0	0	784
PACIFIC	12.2	0	8.7	1.7	259	12.2	36.7	24.5	48.9	0	0	89
Total	6.6	.8	3.7	1.1	316	14.0	35.1	7.8	52.3	0	1.5	159

FOREIGN REPORTS

CANADA

Provinces—Communicable diseases—Week ended June 26, 1943.—During the week ended June 26, 1943, 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
Chickenpox		36	3	94	151	26	44	31	100	485
Diphtheria		1	2	13	2		1			19
Dysentery (bacillary)				1						1
Encephalitis (infectious)						1		5		6
German measles		2		8	76	4	6	38	7	141
Influenza		6	4		15	1	3		1	30
Measles	2	90	2	230	963	127	44	244	133	1,835
Meningitis, meningococcus		1		1	2					4
Mumps		89	8	17	238	50	12	50	39	503
Poliomyelitis		1				2				2
Scarlet fever	4	19	8	49	85	33	26	47	21	292
Tuberculosis (all forms)	4	4	1	118	67	30	3	12	34	273
Typhoid and paratyphoid fever		1	1	8	1					11
Undulant fever				1	3	1				5
Whooping cough		20	1	59	104	21	19	21	20	265

CUBA

Habana—Communicable diseases—4 weeks ended May 29, 1943.—During the 4 weeks ended May 29, 1943, certain communicable diseases were reported in Habana, Cuba, as follows:

Disease	Cases	Deaths	Disease	Cases	Deaths
Diphtheria	22		Paratyphoid fever	2	
Malaria	5		Tuberculosis	2	
Measles	17		Typhoid fever	22	4

JAMAICA

Notifiable diseases—4 weeks ended July 3, 1943.—During the 4 weeks ended July 3, 1943, cases of certain notifiable diseases were reported in Kingston, Jamaica, and in the island outside of Kingston, as follows:

Disease	Kingston	Other localities	Disease	Kingston	Other localities
Chickenpox	13	22	Scarlet fever		1
Dysentery	5	2	Tuberculosis	26	105
Erysipelas	1		Typhoid fever	3	36
Leprosy	1	5	Typhus fever	1	
Puerperal septicemia		1			

SWITZERLAND

Notifiable diseases—December 1942.—During the month of December 1942, cases of certain notifiable diseases were reported in Switzerland as follows:

Disease	Cases	Disease	Cases
Cerebrospinal meningitis	11	Mumps	220
Chickenpox	354	Paratyphoid fever	6
Diphtheria and croup	338	Poliomyelitis	24
Dysentery	22	Scarlet fever	309
German measles	16	Tuberculosis	376
Hepatitis, epidemic	360	Typhoid fever	16
Influenza	69	Undulant fever	2
Measles	248	Whooping cough	99

WORLD DISTRIBUTION OF CHOLERA, PLAGUE, SMALLPOX, TYPHUS FEVER, AND YELLOW FEVER

From medical officers of the Public Health Service, American consuls, International Office of Public Health, Pan American Sanitary Bureau, health section of the League of Nations, and other sources. The reports contained in the following tables must not be considered as complete or final as regards either the list of countries included or the figures for the particular countries for which reports are given.

CHOLERA

[C indicates cases]

NOTE.—Since many of the figures in the following tables are from weekly reports, the accumulated totals are for approximate dates.

Place		January–April 1943	May 1943	June 1943—week ended—			
				5	12	19	26
ASIA							
Ceylon	C	47	1	1			
India	C	83,023	5,561	1,854	1,051		
Calcutta	C	854	513	136	154	119	
Madras	C	964					
Vizagapatam	C	4					
India (French)	C	49					
Chandernagor	C	4					
Karikal	C	28					
Pondichery	C	17					

PLAGUE

[C indicates cases; D, deaths; P, present]

Place							
AFRICA							
Basutoland	C	4					
Belgian Congo—Plague-infected rats		P					
British East Africa:							
Kenya	C	11					
Uganda	C	6			2		
Madagascar	C	17					
Morocco (French)	C	124	74		2	2	
Senegal	C		72			[1] 71	
Dakar	C					7	7
Union of South Africa	C	53					
ASIA							
India	C	1,125	82	4	2	1	
Indochina	C	4					
Palestine	C	8		3			
SOUTH AMERICA							
Peru:							
Lambayeque Department	C	2					
Libertad Department	C	12					
Lima Department	C	3					
Lima	C	1					
Plague-infected rats		P					
Piura Department	C	2					
Venezuela.[2]							
OCEANIA							
Hawaii Territory:							
Hamakua District	D	3	1				
Plague-infected rats		51	[3] 12	1			

[1] For the period June 1–20, 1943.
[2] For the period July 1–14, 1943, 7 cases of plague were reported in Venezuela.
[3] Includes 3 plague-infected mice.

SMALLPOX

[C indicates cases; D, deaths]

Place		January–April 1943	May 1943	June 1943—week ended— 5	12	19	26
AFRICA							
Algeria	C	517	56		53		
Angola	C	507					
Basutoland	C	30					
Belgian Congo	C	562	203	121	38		
British East Africa:							
Mombasa	C	3					
Tanganyika	C	11					
Dahomey	C	28	101				
Egypt	C	118	408	122	152	135	146
French Guinea	C	12	114				
Gold Coast	C	5					
Ivory Coast	C	91	10				
Mauritania	C	1					
Morocco (French)	C	522	57				
Mozambique	C	1					
Nigeria	C	2,389	606	188	98	154	
Niger Territory	C	90	66				
Senegal	C	21	6				
Sierra Leone	C	3					
Sudan (French)	C	741	797				
Union of South Africa	C	221					
ASIA							
Ceylon	C	1					
India	C	9,861	4,394	1,023			
India (French)	C	10					
Indochina	C	[1] 2,726			120		
Iran	C	158					
Iraq	C	159	20				
Palestine	C	29					
Syria and Lebanon	C	605	159	10			
Trans-Jordan	C	11					
EUROPE							
Belgium	C	1					
France	C	1					
Germany	C		1				
Scotland	C	1					
Portugal	C	19	4	10		1	
Spain	C	128	7				
Turkey	C	4,975					
NORTH AMERICA							
Canada	C	1					
Guatemala	C	3	1				
Mexico	C	91	19				
SOUTH AMERICA							
Brazil	C	40					
British Guiana	C			1			
Colombia	C	97	23	14	9		
Ecuador	C	10					
Peru	D	8	1				
Venezuela	C	19	5				

[1] Includes the month of May.

TYPHUS FEVER

[C indicates cases]

AFRICA							
Algeria	C	5,395	1,217		282		
Belgian Congo	C	2					
British East Africa:							
Kenya	C	3	2				
Mombasa	C	1					
Uganda	C	1					
Egypt	C	17,826	11,335	1,520	1,160	1,453	1,175
Gold Coast	C	4			1		
Morocco (French)	C	9,691	1,921				
Morocco (Spanish)	C	59	3				
Nigeria	C	2	1	1			
Rhodesia, northern	C	4					
Senegal	C	1	1				
Sierra Leone	C	3					
Union of South Africa	C	778					

TYPHUS FEVER—Continued

[C indicates cases]

Place		January–April 1943	May 1943	June 1943—week ended— 5	12	19	26
ASIA							
Afghanistan	C	520					
China: Shanghai	C	12					
India	C	965	46	6	20		
Iran	C	4,285	1,328				
Iraq	C	752	488	53	23		
Palestine	C	64	93	6	15	11	
Syria and Lebanon	C	15	8	5			
Trans-Jordan	C	12					
EUROPE							
Bulgaria	C	235					
France—Seine Department	C		2				
Germany	C	[1] 800					
Hungary	C	436	160		32		[2] 30
Irish Free State	C	19					
Portugal	C	3	2				
Rumania	C	4,473	1,112	159	176	194	148
Slovakia	C	192	63	19			
Spain	C	230	174				
Turkey	C	1,614	698				
NORTH AMERICA							
Guatemala	C	396	45				
Jamaica	C	9	2			1	
Mexico	C	581					
Puerto Rico	C	2					
SOUTH AMERICA							
Chile	C	110			4		
Ecuador	C	107	19				
Peru	C	5	2				
Venezuela	C	6					
OCEANIA							
Australia	C	30	22				
Hawaii Territory	C	8	2		1		

[1] For the first 7 weeks of 1943.
[2] For 2 weeks.

YELLOW FEVER

[C indicates cases; D, deaths]

AFRICA							
Belgian Congo:							
Bondo	D	1					
Leopoldville	C		1				
Stanleyville	D	1					
Yanonge	C	1					
Gold Coast: Kibbi	C						[1] 1
Sierra Leone: Freetown	C				[1] 1		
SOUTH AMERICA							
Colombia:							
Cundinamarca Department	D	1					
Intendencia of Meta	D	2					

[1] Suspected.

COURT DECISIONS ON PUBLIC HEALTH

Sewage disposal—stream pollution by city—order of State stream control commission upheld.—(Michigan Supreme Court; *People ex rel. Stream Control Commission* v. *City of Port Huron et al.*, 9 N.W.2d 41; decided April 6, 1943.) The Michigan Stream Control Commission ordered the city of Port Huron to construct a sewage treatment plant to permit treatment of the city's sewage before its discharge into State waters. The city failed to comply with this order and the commission brought a proceeding to enforce its order and to restrain the city from discharging untreated sewage into the Black and St. Clair Rivers. In the lower court there was a decree in favor of the city, and an appeal was taken to the State supreme court.

The latter court took the view that there was sufficient evidence to substantiate the State's contention that the present raw sewage disposal method was a constant menace to the health and well-being of the down-river communities and tourists. According to the court this evidence clearly justified the commission's order and it was no defense to a statutory charge of river-water pollution that others had contributed or were contributing to that condition.

With respect to the doctrine of comparative injury, the appellate court stated that the instant case was not a proper one for the application of that doctrine even if there should be concurrence with the trial court in its conclusion that "a balancing of equities" favored the city. The doctrine "should be confined to those situations where the plaintiff can be substantially compensated" and "should not be invoked to justify the continuance of an act that tends to impair public health."

The city also raised the question of its financial inability to comply with the commission's order but to no avail. After quoting from a New Jersey case in which the same question had been raised and held to be no defense, the supreme court pointed out that the statute creating the commission was under the police power vested in the State and that the order was not arbitrary or unreasonable but became necessary because of the city's previous refusal to stop polluting the rivers.

In holding that the evidence justified the order and in vacating the lower court's decree, the appellate court stated that it was not unmindful of the situation caused by war conditions and of the fact that the city would have difficulty in complying with the commission's order "due to necessary materials now required for war purposes." Proceeding, the court said: "This, however, does not, and should not, prevent the city from immediately taking those steps necessary to insure the carrying out of the mandate of the commission, but a rea-

sonable time should be allowed for completion of the project. We apprehend that the State and city can agree upon the time that is necessary, and if they cannot, this is a matter which can be determined by the trial judge upon proper proofs."

Liability of physician for failure to use prophylactic in infant's eyes at birth.—(Kentucky Court of Appeals; *Walden* v. *Jones*, 158 S.W.2d 609; decided January 13, 1942, rehearing denied March 3, 1942.) An action against a physician was brought by an infant to recover damages for the loss of the plaintiff's eyesight allegedly caused by the negligence of the physician in failing to place nitrate of silver in the plaintiff's eyes at the time of his birth. A jury found for the plaintiff and the judgment entered upon such verdict was appealed from by the defendant.

Regarding the question as to whether negligence was established, the Court of Appeals of Kentucky stated: "Certainly the evidence that the defendant failed to place a prophylactic in the eyes of the newborn child is sufficient to conclusively establish negligence on the part of the physician, in the light of the uncontradicted medical testimony that in all localities physicians ordinarily use silver nitrate or some other prophylactic in the eyes of a child at birth, and that reasonable care and diligence require such to be done." The court concluded that the defendant's negligence was clearly proved and said that under the proof in the case it was not proper to submit to the jury the question as to whether the failure of defendant to drop the prophylactic in the child's eyes constituted an act of negligence.

On the question as to whether the established negligence of the defendant was the proximate cause of the injury, the appellate court's conclusion was that the trial court properly submitted the case to the jury. The judgment of the lower court was, however, reversed because of a statement made in argument by the plaintiff's counsel, which statement was held by the court of appeals to be improper.