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A Biochemical Study of Experimental Q Fever Infection in the Bovine Mammary Gland

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This investigation was made to study several aspects of bovine mammary gland metabolism as they were influenced by experimentally induced infection with *Coxiella burnetii*, the causative agent of Q fever.

The data reported represent only a part of the clinical, epidemiological, and pathological information obtained from a study of six cows. Additional details will be published later (*1*).

Material and Methods

Six young cows in the second and third months of their first lactation period were used in the experiment. Four were experimentally infected and two served as controls. Prior to initiation of the experiment, the cows were free of mastitis, as indicated by leukocyte counts and blood agar cultures.

Representative milk samples from the total morning milking from each quarter of the udder of each cow were taken three times a week until the acute phase of the infection was past. Samples were then taken once a week. Samples were also taken during the 10 days preceding inoculation with *C. burnetii*.

The cows were inoculated with *C. burnetii* (a California strain) by the injection of 20-ml. aliquots of 2.5 percent infected chick yolk-sac tissue via the lacteal duct. The infectious suspension was made up in 0.85 percent NaCl and contained 2.5×10^9 guinea pig infectious doses per milliliter. The infected yolk-sac material came from 13-day-old chick embryos; the corresponding normal yolk-sac suspension used in the control cattle came from 12-day-old chick embryos. The two control cows were injected in the left front and right rear, and the right front and right rear quarters, respectively. The animals receiving the infectious suspension were injected in the left front and left rear quarters in all cases.

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Butterfat determinations were made according to the procedure described by Goss and Rudnick (*2*).

The pH determinations were made with a glass electrode. These determinations were accurate to 0.02 units.

Lactose determinations were made following the Folin-Wu method for blood sugar. To 2 ml. of milk in a 25-ml. volumetric flask, 18 ml. of a mixture of eight parts of N/10 H_2SO_4 and one part of 10 percent Na_2WO_4 was added slowly and with shaking. This was allowed to stand 5 minutes and then diluted to volume with distilled water. The filtrate from this preparation was diluted 3:25, and 1 ml. of the diluted filtrate was used for the lactose determinations (*3*).

Chloride determinations were performed by adding 5 ml. of $AgNO_3$ (2.905 gm/1.) to a 5-ml. aliquot of a filtrate prepared as described in the preceding paragraph. To this was added 5 ml. of concentrated HNO_3 and 0.3 gm. $FeNH_4(SO_4)_2 \cdot 12\ H_2O$. This mixture was titrated in ice water to a 15-second persisting end point with KCNS of such a concentration that 5 ml. was equivalent to 1 ml. of the $AgNO_3$ solution.

Total nitrogen determinations were made by the usual micro-Kjeldahl method and nesslerization of the distillate.

Casein was determined by a method involving the use of KH_2PO_4 as the precipitating agent. To a 25-ml. aliquot of milk diluted 1:50 with distilled water was added 25 ml. of M/4 KH_2PO_4. The mixture was allowed to stand 60 to 90 minutes, during which time the casein was precipitated as a fine flocculate. If precipitation was delayed, the addition of 0.1 ml. of 10 percent HAc caused rapid completion. The mixture was then filtered through Whatman No. 2 paper and washed with M/4 KH_2PO_4 to a total wash volume of about 75 ml. The precipitate was then dissolved by the drop-wise addition of 10 percent NaOH which was also used for rinsing out the original flask. A total volume of 15 ml. of 10 percent NaOH was employed, and the filter subsequently washed with water. The NaOH and wash waters were collected in a 25-ml. volumetric flask and made up to volume. N determinations were made on 1-ml. aliquots by the method previously described. The amount of casein was calculated on the basis of 15.7 percent N content. A comparison of this method of casein determination with other methods will be published at a later date.

Total solids were measured by weighing out approximately 3 ml. of milk into a 6.5-cm. Vycor evaporating dish and drying it at 96° for 24 hours. The dish then was stored in a desiccator until weighed.

Total ash was determined by weighing out approximately 20 ml. of milk in an 8.5-cm. Vycor evaporating dish and adding 6 ml. of concentrated HNO_3. This was digested cautiously over a micro-burner and then ashed at a temperature below redness until free of

carbon. Removal of carbon was accelerated by the cautious dropwise addition of 30 percent H_2O_2. The samples were then cooled and left in a desiccator overnight before weighing.

Hemoglobin was measured colorimetrically as acid hematin. A 0.2-ml. aliquot of heparinized blood made up to 50 ml. with N/10 HCl was allowed to stand for 1 hour at room temperature, and the absorption measured at 540 mμ.

Results

A summary of the most important data is presented in table 1. The data given in this table cover only the period beginning 4 days before infection and the 12 days following infection. The major changes noted took place during this period. Determinations were actually made on the animals beginning 10 days before infection and lasting until the animals were killed—a period of 41 days postinoculation in some cases. Although individual determinations were made on each quarter of the udder of each cow, only the data on the quarters actually injected with normal or infectious yolk-sac suspensions are presented. The effects on the uninjected quarters are noted in the appropriate sections.

Table 1. *Summary of the changes occurring as a result of experimental Q fever infection, including data for the injected quarters of the udders only*

	A	B	C		A	B	C
	Milk volume in ml.				*Butterfat, gm. percent*		
Control	963	415	848		4.2	4.7	3.8
Infected	1148	276	792		3.3	7.7	3.7
	Chloride, mg. percent				*Total nitrogen, mg. percent*		
Control	77	114	65		579	767	449
Infected	87	201	187		500	818	553
	Casein, mg. percent				*Lactose, gm. percent*		
Control	[1] 2380	[2] 1950	2370		6.09	4.28	5.32
Infected	[1] 2040	[2] 1760	2380		5.46	3.39	4.43
	Dry wt., gm. percent				*Total ash, gm. percent*		
Control	12.90	13.53	12.28		[3] 0.96	--------	[4] 0.68
Infected	11.47	16.39	12.78		[3] 1.02	--------	[5] .66
	pH				*Hemoglobin, gm. percent*		
Control	6.49	6.61	6.51	(2 animals)	10.0	11.3	10.6
Infected	6.46	6.65	6.56	(3 animals)	10.3	10.1	11.0

[1] Average value 7 to 11 days before injection.
[2] Complete series of determinations not made. Minimum value during first 8 days postinjection may have been lower. These values were obtained after dry weight values had returned almost to normal.
[3] Determinations made only once, 10 days before injection.
[4] Determinations made 42 days after injection.
[5] Determinations made on one cow only, 42 days after injection.

NOTE: Column A gives the average value during the 4 days preceding injection; column B the average maximum or minimum value obtained during the 8 days following injection; column C gives the average value during the eighth to the twelfth days postinjection. In the group designated "Control" each value represents the average value obtained from 4 quarters of the udders (2 from each of 2 cows). In the "Infected" group, each value represents the average value from 6 quarters (3 cows), unless otherwise indicated. The values are for morning milk only, except for the hemoglobin values, which are for blood.

Milk Production

The volume of milk from each of the four quarters of the udder of a given cow varied considerably during the preinoculation period, as well as later. The injection of an infectious yolk-sac suspension into a given quarter resulted in a drastic fall in milk volume from 5 to 20

percent of normal. A similar drop, though not as great, occurred in the uninjected quarters. Beginning with the fourth day postinoculation, volume rapidly rose to near normal levels in both the injected and uninjected quarters. Milk volume never again, in the time period studied, reached the average preinjection level.

Milk flow from the quarters injected with normal yolk-sac suspensions likewise dropped but not as much as in the quarters injected with infectious material. Recovery was more rapid. The uninjected quarters of the control animals showed only a slight drop in milk production.

Butterfat

The percent of butterfat was only slightly affected in the control animals. However, the total butterfat production dropped significantly in these animals, particularly in the inoculated quarters, because the milk production dropped immediately following inoculation. This transitory decrease in butterfat production had largely disappeared by the eighth day following inoculation.

In the experimental animals the butterfat concentration increased markedly, particularly in the inoculated quarters, going as high as 9.9 percent in the inoculated quarters of one cow. Although the butterfat concentration increased markedly in the infected quarters of the experimental animals, there was a decrease in total butterfat much greater than that in the control animals because of the extreme decrease in milk volume in the infected animals.

The average percent of butterfat in the infected quarters rose about twofold and then dropped to normal within the 8 days following infection. A somewhat smaller rise occurred in the uninjected quarters of the experimental animals.

Lactose

A slight drop in lactose concentration following inoculation was noted in both the injected and uninjected quarters of the control animals.

The lactose concentration in the infected quarters of the experimental animals declined 38 percent immediately following inoculation, whereas the control quarters in these animals were relatively unaffected. Within 8 days, postinoculation lactose content was normal or nearly so in all cases although the return to normal values was more protracted than in the control animals.

Chloride

In the control animals a gradual decrease in milk chloride concentration was noted during the 10-day preinoculation period. This was likewise true of the experimental animals. Immediately following inoculation a sharp, but relatively small rise in concentration was

noted in the control animals. This was more marked in the injected quarters.

In the experimental animals the chloride concentration in the infected quarters rapidly rose far above those in the uninfected quarters of the udders of the same animals. The values in the infected quarters equaled the values of the corresponding normal quarters by the seventh day in one animal, and by the fourteenth day in the second, but never did reach the lower chloride concentration of the control quarters in a third, up to the time of sacrifice on the twenty-second day. The average maximum increase in chloride in the infected quarters was twofold.

Total Nitrogen

In the control animals a slight rise in nitrogen concentration was noted in the injected quarters on the day immediately following injection. The level had dropped to control values by the time the next determinations were made, 4 days following injection. The total nitrogen content of all quarters in the control animals fell to a level of about 78 percent of the preinjection value and was subsequently maintained at this level.

In the infected animals a sharp rise in total nitrogen occurred in the milk from the injected quarters, with a peak equaling 164 percent of normal on the fourth day. A fall in the total nitrogen of the corresponding uninjected quarters occurred. The peak nitrogen content in the milk from the injected quarters came simultaneously with the lowest ebb of milk production, and probably reflects protein concentration. NPN determinations were not made so it is not known whether or not the rise in total N reflects only protein concentration or whether an increase in NPN also occurred at this time. It is very likely that a rise in NPN did occur. The total N concentration in the milk from the injected quarters did not drop to the values obtained from the uninjected quarters until 20 days postinjection.

Casein

No evidence of significant fluctuations in casein concentrations in the milk from the control quarters as opposed to the injected quarters was noted in either the control animals or the experimental animals. Nor was the relative casein production in the experimental animals markedly altered as compared with the controls. These data, however, cover only part of the preinoculation period and only the postinoculation period beginning 6 days after inoculation. Since these data do not cover the period in which the greatest fluctuations occurred in the other constituents measured, no conclusion can be drawn concerning the influence of the experimental treatment on the casein content of the milk during the period in which marked changes were most likely to occur.

Dry Weights

Dry weight determinations of milk samples showed a marked but erratic fluctuation in the control animals in the 6 days immediately following injection. In one control animal no change occurred in the uninjected quarters. In the other control animal a marked drop in dry weight values from the injected quarters and a smaller but significant drop in the uninjected quarters occurred.

In the experimental animals, a rise in dry-weight values occurred in all quarters, both injected and uninjected. This rise was maximal 4 days following injection or at the time of minimum milk production and was much greater in the injected quarters than in the uninjected quarters. One cow was a partial exception to this in that the highest dry-weight values in the infected quarters were observed 6 days following injection, at a time when milk production was increasing from its low point on the fourth day. The uninjected quarters in this cow showed a decrease in dry-weight value on the sixth day. The values from the injected quarters as compared to the corresponding control quarters were approximately equal in the control animals by the sixth day following injection. This was likewise true in a second animal. In a third animal, however, the values from the control and infected quarters were never equal up to the time of sacrifice on the eleventh day following injection. In a fourth animal, values were approximately equal by the fifteenth day following injection although true equality did not obtain until the day of sacrifice, 22 days following injection. Dry-weight increases up to 158 percent of normal were observed. The average maximum increase in dry-weight values was 43 percent above normal and occurred in the injected quarters of the infected cows. Total ash measurements made in the preinjection period again just prior to sacrifice indicated no significant differences between the control and the experimental animals.

pH Changes

A small increase in the pH of the milk from the control animals was detected. The injected quarters were indistinguishable from the uninjected quarters in this regard.

Similar fluctuations in the pH of the milk from the infected cows occurred. They were more marked in the injected than in the uninjected quarters. In all cases the pH of the milk from injected quarters rose 0.1 to 0.2 units during the 8 days immediately following injection. This was accompanied by a rise in pH in the corresponding uninjected quarters in all cases. The values returned to values closely approximating the preinjection figures within 11 days in the case of two animals. In the third infected animal, the pH of the milk did not return to its preinfection value within 22 days, at which time the animal was killed. The magnitude of the pH changes was slight.

Control (preinjection) values averaged pH 6.46, and the average maximum value obtained in the 8 days following inoculation was pH 6.65.

Hemoglobin

Blood hemoglobin determinations made revealed no significant variations that could be ascribed to Q fever infection. The initial control values for the individual animals varied considerably, and later changes revealed no regular trend in concentration that could be ascribed to the experimentally induced infections.

Discussion

The changes that were noted occurred chiefly during the 8 days following injection. They were essentially similar in the control animals which had been injected with a normal chick yolk-sac suspension and in the experimental animals which had been injected with a corresponding suspension of infectious material. The major difference between the control and the experimental values lay in the magnitude and the duration of the changes, particularly with respect to butterfat and chloride. In the control animals, the changes resulting from the introduction of normal yolk-sac suspension were smaller in magnitude and recovery was rapid. In the experimental animals, the magnitude of the changes was greater and recovery to normal was considerably slower. Even so, most of the abnormal variations in milk composition had disappeared within the 8 days following infection. This 8-day period following injection also covered the appearance and disappearance of fever in the infected animals. No fever occurred in the control animals.

It seems reasonable to conclude that the major cause of the changes observed in the experimentally infected animals was the introduction in the udder of massive amounts of foreign protein. It is also clear, however, that changes in all constituents occurred in these animals above and beyond those nonspecific reactions caused by the yolk-sac components injected. Thus, the magnitude and duration of the changes which occurred in the infected animals can be attributed to the Q fever infection. This was particularly clear in one of the experimental cows in which the differences persisted for 22 days following injection. The most acute deviations from normal in this animal occurred several days after the appearance of the corresponding deviations in the control animals.

This animal, incidentally, represented the only case in which the original infection spread to an uninjected quarter of the udder. The animal was injected in the left front and left rear quarters, and 7 days later the milk from the right rear quarter became positive for Rickettsiae, and remained so until sacrificed, 63 days after injection.

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Table 2 presents a comparison (with the present findings) of the changes that occur during chronic bacterial mastitis (*4*). The most characteristic changes in the chemical composition of milk from udders with bacterial mastitis are increases in chloride, water, noncasein nitrogen, and pH, and decreases in casein, lactose, milk fat, nonfat solids, and ash. There may be a considerable decrease in milk volume also. The changes that occurred following infection with *C. burnetii* differ from those found in chronic bacterial mastitis in that there is a large increase in milk fat, in solids other than fat, and a relatively small increase in pH. The milk taken at the most acute stage was very thick and creamy in appearance, with butterfat values up to 9.9 percent and total solids values up to 19.1 percent.

The infected cows shed Rickettsiae in their milk until they were killed, or for periods of 5, 11, 22, and 63 days, respectively, following injection. The Rickettsiae were demonstrated by guinea pig inoculation. It thus appears that none of the measurements made were critical in indicating the existence of any Q fever infection except the most acute, since the infections persisted long after the milk had returned to normal. The acute Q fever infection, as exemplified by the foregoing data, is reflected in changes which were concerned with cellular synthetic activities, or with the integrity of epithelial cell membranes.

These data indicate that *C. burnetii* infection produces only temporary metabolic changes in mammary gland activity, even when the organism is injected in massive doses. Milk volume, butterfat, casein, total nitrogen, lactose, chloride, and total solids, all assumed relatively normal levels within 3 weeks at most following infection and within 8 days for three out of four experimental animals. One exception may have been the declining milk volume noted from the twenty-eighth to the forty-first day in one infected animal.

No systemic involvement reflected in hemoglobin concentration changes or in blood cell counts was noted in these animals.

Naturally occurring infections of the mammary gland via the

Table 2. *A comparison of the present data on milk composition with that for chronic bacterial mastitis* (*4*)

	Bacterial mastitis	*Q fever infection*	*Injected with normal chick yolk sac only*
NCN	+	+	+
Casein	−	−	−
Chloride	+	+	+
Lactose	−	−	−
Fat	−	+	±
pH	+	+	+
Nonfat solids	−	+	±
Ash	−	?	?
Milk volume	−	−	−
Total N	---	+	+
Water	---	−	−

NOTE: A "plus" sign indicates increase in concentration; a "minus" indicates a decrease in concentration.

lacteal duct might be expected to produce less disturbance than was seen in these animals. The inocula in such cases would probably consist of a much smaller number of organisms and would be accompanied by much less foreign protein, which presumably caused the intense inflammatory reaction noted initially in both the control and the experimental animals. Since this experiment was performed, other data which suggest that naturally occurring mammary gland infections are of hematogenous origin have been published (*5*).

Conclusions

1. The injection of infectious suspensions of *C. burnetii* in the udders of cows resulted in the following changes in the composition of their milk: increases in total nitrogen, noncasein nitrogen, chloride, butterfat, nonfat solids, and pH; decreases in casein, lactose, milk volume, and water. No change in blood hemoglobin resulted.
2. Major fluctuations in these constituents disappeared within 8 days following injection.
3. Similar changes occurred in control animals injected with chick yolk-sac suspension, but the changes were of smaller magnitude and disappeared more quickly.
4. The changes are different from those that occur with chronic bacterial mastitis in that the butterfat and nonfat solids increased in concentration.

ACKNOWLEDGMENT

The data on rickettsiae in the milk of the infected cows were obtained by Drs. H. G. Stoenner and E. J. Bell, and will be presented in detail in a future publication. It is a pleasure to acknowledge also the technical assistance of George Tallent.

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Changes Induced in the Flagellar Antigens of *Salmonella rostock* and *Salmonella california*

By O. A. Peso and P. R. Edwards*

Several instances of induced type transformation in the genus *Salmonella* have been reported. The first such report was that of Edwards, Moran, and Bruner (*1*) in which the intertransformability of *Salmonella simsbury* and *Salmonella senftenberg* was described. Bruner and Edwards (*2*) induced changes in the nonspecific phases of certain types. Bruner (*3*) was able to transform *Salmonella oranienburg* into a form indistinguishable from *Salmonella montevideo*. All of these changes in H antigens were brought about by cultivation of the organisms in semisolid agar to which had been added appropriate agglutinating serum in amount sufficient to immobilize the bacteria inoculated. Antigenic change was indicated by the migration of the bacteria through the medium after one or more transfers.

The present paper deals with changes produced in organisms which possess antigen g of the Kauffmann-White schema, i. e., forms which possess flagellar antigens related to *Salmonella enteritidis*. The methods used were similar to those described in the references cited above. The first change to be described was the transformation of *Salmonella rostock* (IX, XII: g, p, u) into a form indistinguishable from *Salmonella dublin* (IX, XII: g, p). Growth from a single colony of Kauffmann's standard culture of *S. rostock* was inoculated into the semisolid medium of Edwards and Bruner (*4*) to which had been added serum for antigen u of *S. rostock* in a final dilution of 1 to 100. The u serum was produced by absorption of *S. rostock* serum by *S. dublin*. The serum was sterilized by addition of an excess of chloroform. After eight transfers at intervals of 3 days, filmy bulbs extending from the line of inoculation appeared. Upon prolonged incubation, these bulbs gradually spread through the medium. From the spreading growth a form was isolated which was serologically indistinguishable from *S. dublin*. While the induced form retained the biochemical reactions of *S. rostock*, it no longer was agglutinated by factor u serum and it removed all agglutinins from *S. dublin* serum.

The second change to be described was produced in *Salmonella california* (IV, XII: g, m, t). Progeny of a single colony of the original strain of this type was cultivated in semisolid agar which contained *Salmonella oranienburg* (VI, VII: m, t) serum which had been absorbed

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by *Salmonella montevideo* (VI, VII: g, m, s). This serum contained agglutinins for factor t, which is the major antigen of *S. california*. After four transfers, filmy bulbs extending from the line of inoculation were noted. Further transfers yielded a spreading growth from which was isolated an induced phase closely related to *Salmonella essen*. The agglutinative reactions of *S. essen*, *S. california*, and the induced phase are given in the table.

Agglutinative properties of induced variant of S. california

Antigens	Serums					
	Derby (f, g)	Enteritidis (g, m)	Oranienburg (m, t)	Enteritidis absorbed with california (induced phase)	m [1]	t [2]
California (original)	80	80	5,000	<20	±	+++
Essen	2,500	20,000	320	5,000	+	−
California (induced phase)	2,500	10,000	80	<20	+++	−
Enteritidis	2,500	20,000	640	5,000	+	−

[1] *S. oranienburg* antiserum absorbed with *S. berta*+*S. senftenberg*.
[2] *S. oranienburg* antiserum absorbed with *S. montevideo*.

In the transformation of *S. rostock* to a form serologically identical with *S. dublin*, only loss variation is involved. In this instance the observed changes coincide with the theory of White (*5*) that *Salmonella* types arise by loss variation. On the contrary, the change of *S. california* to a form resembling *S. essen* involved a gain of certain antigenic characters as well as a loss of others. The induced form no longer contained demonstrable t antigen, the major component of the parent form. This loss of t antigen was accompanied by a distinct gain in g and m components which exist only in minimal amount in the parent culture of *S. california*. While the induced form was not identical with *S. essen* as shown by agglutinin absorption, nevertheless, by the usual methods employed in the typing of *Salmonella* cultures, the induced form would be identified as *S. essen* to which it is closely related.

The above experiments offer further evidence that it is possible to transform *Salmonella* types by induced variation and indicate probable phylogenetic relationships of the types involved.

Summary

By induced variation, *S. rostock* was converted into a form serologically indistinguishable from *S. dublin*. Likewise, *S. california* was changed to a form closely related to, but not identical with, *S. essen*.

ACKNOWLEDGMENT

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Public Health Service Publications

January–June 1951

This list is issued to provide a complete and continuing record of Public Health Service publications for reference use by librarians, scientists, researchers, and others interested in public health work, and not to offer the publications for indiscriminate free distribution.

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For quantities of any of these publications, except the statistical reports of the National Office of Vital Statistics, order from the Government Printing Office, where they are available at the prices shown, with a 25 percent reduction on orders of 100 or more copies of any single publication. The statistical reports of the National Office of Vital Statistics can be obtained only by writing to the National Office of Vital Statistics, Public Health Service, Washington 25, D. C.

PERIODICALS

Public Health Reports (weekly), January-June, vol. 66, Nos. 1–26, pages 1 to 850. 15 cents a copy.

Extracts from Public Health Reports (monthly), January-June, Tuberculosis Control Issues Nos. 59–64. Average 28 pages each. 10 cents a copy.

The Journal of Venereal Disease Information (monthly), January-June, vol. 32, Nos. 1–6. Pages 1 to 175. 15 cents a copy.

Journal of the National Cancer Institute (bimonthly), February-June, vol. 11, Nos. 4–6, pages 663 to 1324. $1.50 a copy. Subscription price $8 a year (available from the Superintendent of Documents, U. S. Government Printing Office, Washington 25, D. C.).

Public Health Engineering Abstracts (monthly), January-June, vol. XXXI, Nos. 1–6. Approximately 30 pages each. No sales stock.

Industrial Hygiene Newsletter (monthly), January-June, vol. 11, Nos. 1–6, 16 pages each. 10 cents a copy. (The name of this periodical was changed to Industrial Health Monthly beginning April 1951).

CDC Bulletin (monthly), January-June, vol. X, Nos. 1–6. No sales stock.

NONPERIODIC PUBLICATIONS

Librarians, Please Note

Nearly all publications on this list carry the designation "Public Health Service Publication No.—." This does not represent a new series, but is a registration system adopted late in 1950 as an internal housekeeping measure. Under the numbering plan, all nonperiodic issuances carry this designation, which is assigned consecutively at the time of going to press. Publications issued prior to adoption of

the numbering plan do not carry this designation and will not be incorporated into the system unless they are reissued.

The following series have been discontinued:

Supplements to Public Health Reports—last issued: No. 213 (1950).
Supplements to Journal of Venereal Disease Information—last issued: No. 23 (1949).
National Institutes of Health Bulletins—last issued: No. 193 (in press).
Public Health Bulletins—last issued: No. 306 (1949). (A bulletin on rural health cooperatives, jointly prepared by the Farm Credit Administration and the Public Health Service which was published by the Farm Credit Administration, was erroneously identified as Public Health Bulletin No. 308.)
Venereal Disease Bulletins—last issued: No. 100 (1949).

Series to Continue. Publications with a subject and field homogeneity will continue to be issued in series. In addition to the registration number given them under the over-all numbering system, they will carry the conventional series notation long familiar to librarians. Publications issued as part of a series are shown first on this list.

Missing Numbers. Under the new numbering system, publications are assigned registration numbers at the time they go to the printer. Variations in printing schedules result in publications being released out of the consecutive order of the Public Health Service Publications number, and some will be temporarily missing. One publication (No. 36), prepared for in-service use and not for general distribution, was inadvertently numbered. No. 36, therefore, will be permanently missing from library files.

PUBLIC HEALTH BIBLIOGRAPHY SERIES

1. Chronic illness. Digests of selected references. By Violet B. Turner. 1951. 216 pages. 50 cents. (PHS Publication No. 10.)
3. Small plant health programs—a bibliography. Compiled and annotated by Walter J. Lear. January 1951. 26 pages. Out of print. (PHS Publication No. 80.)

CANCER MORBIDITY SERIES

2. Cancer illness among residents of San Francisco and Alameda Counties, California, 1947. By William Grodowitz. 1951. 46 pages. No sales stock. (PHS Publication No. 65.)
3. Cancer illness among residents of New Orleans, Louisiana, 1947. By Irving I. Warran. 1951. 52 pages. No sales stock. (PHS Publication No. 67.)

HEALTH INFORMATION SERIES

29. The common cold. Revised 1951. 6-page folder. 5 cents; $1.50 per 100. (PHS Publication No. 106.)
37. Diphtheria. Revised January 1951. 6-page folder. 5 cents; $1.50 per 100. (PHS Publication No. 60.)
58. Gallstones. Revised 1951. 4 pages. 5 cents; $1.00 per 100. (PHS Publication No. 99.)

WATER POLLUTION SERIES

1. Water pollution in the United States. A report on the polluted condition of our waters and what is needed to restore their quality. 1951. 44 pages, illustrated. 35 cents. (PHS Publication No. 64.)
2. Tennessee River Drainage Basin. A cooperative State-Federal report on water pollution. 1951. 121 pages. Limited distribution. No sales stock.
3. Missouri River Drainage Basin. A cooperative State-Federal report on water pollution. 1951. 212 pages. Limited distribution. No sales stock.
4. Great Basin Drainage Basin. A cooperative State-Federal report on water pollution. 1951. 67 pages. Limited distribution. No sales stock.
5. California Drainage Basins. A cooperative State-Federal report on water pollution. 1951. 90 pages. Limited distribution. No sales stock.
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8. Western Gulf Drainage Basin. A cooperative State-Federal report on water pollution. 1951. 67 pages. Limited distribution. No sales stock.

NATIONAL INSTITUTES OF HEALTH BULLETINS

193. Contamination of natural waters and mud with *Pasteurella tularensis* and tularemia in beavers and muskrats in the Northwestern United States. By R. R. Parker, Edward A. Steinhaus, Glen M. Kohls, and William L. Jellison. 1951. 61 pages. 45 cents.

180. Manual for the microscopical diagnosis of malaria in man. By Aimee Wilcox. Second edition, revised. 1950. 49 pages; 16 plates. 65 cents.

ANNUAL REPORT

Annual report of the Federal Security Agency, 1950. Public Health Service. 1951. 73 pages. 25 cents.

POSTER

Public Health Service Poster No. 2. Careers in mental health. 1951. No sales stock. (To accompany booklet Mental Health Series No. 5, Careers in mental health.)

OTHER PUBLICATIONS

Adequate financial support for hospital maintenance and operation. By Louis Block, David H. Spanier, and John V. Berberich, Jr. 1951. 27 pages. No sales stock. (PHS Publication No. 76.)

A draft act governing hospitalization of the mentally ill. 1951. 34 pages. No sales stock. (PHS Publication No. 51.)

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Breast self-examination. 1951. 8-page folder, illustrated. 10 cents; $5 per 100. (PHS Publication No. 48.)

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You'll want to know about your hospital program. Revised May 1951. 2-page folder. 5 cents; $1.50 per 100. (PHS Publication No. 8.)

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International Recommendations on Definitions of Live Birth and Fetal Death, 1950. 11 pages. 5 cents. (PHS Publication No. 39.)

The First Annual Report of the United States National Committee on Vital and Health Statistics, 1950. 23 pages. 15 cents. (PHS Publication No. 40.)

Current Mortality Analysis (monthly), vol. 8, Nos. 10–12, 1950. Vol. 9, Nos. 1–3, 1951.

Monthly Marriage Report (marriage licenses issued in major cities), vol. 4, Nos. 11–13, 1950; vol. 5, Nos. 1–4, 1951.

Weekly Mortality Index, vol. 21, Nos. 52–53, 1950; vol. 22, Nos. 1–25, 1951.

Weekly Morbidity Report, vol. 1, Nos. 51–52, 1950; vol. 2, Nos. 1–25, 1951.

Communicable Disease Summary, weeks ending January 6, 1951–June 30, 1951.

Vital Statistics—Special Reports, Vol. 33, Selected Studies

No. 9. Estimated average length of life in the death-registration States. 163–170 pages.

Vital Statistics—Special Reports, Vol. 36, National Summaries

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No. 2. Summary of marriage and divorce statistics, United States, 1949 11–26 pages.

No. 3 Marriages, United States, each State and county, 1949. 27–50 pages.

No. 4. Births by race and by urban and rural areas, United States, each division and State, 1949. 51–60 pages.

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*Available only from the National Office of Vital Statistics, Public Health Service, Washington 25, D.C.

Incidence 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

Announcement

A New *Morbidity and Mortality Weekly Report*

The new monthly PUBLIC HEALTH REPORTS (see back cover) will publish from time to time, as appropriate, reports, tabulations, and articles dealing with morbidity statistics, both domestic and foreign. The present weekly "Incidence of Disease" section, however, will be discontinued as of December 31, 1951.

Current provisional morbidity data on notifiable diseases for the United States now appear in summary form and in tabulations by States and cities in the *Weekly Morbidity Report* issued by the National Office of Vital Statistics of the Public Health Service. Beginning on January 11, 1952, the Public Health Service through the National Office of Vital Statistics will issue a *Morbidity and Mortality Weekly Report* presenting these morbidity data as well as certain mortality data for selected cities.

Libraries and agencies that have depended upon PUBLIC HEALTH REPORTS for current morbidity statistics for the United States may continue to receive the same data by writing to the National Office of Vital Statistics, Public Health Service, Washington 25, D. C., requesting that they be placed on the mailing list for the new *Morbidity and Mortality Weekly Report.* Individuals who wish to be placed on the mailing list should indicate how and to what extent they will make use of this publication.

Since the *Weekly Epidemiological Record* and other publications of the World Health Organization, Geneva, Switzerland, contain morbidity data for foreign countries, tabulations of notifiable diseases occurring outside the United States and its Territories will not appear regularly in National Office of Vital Statistics publications.

UNITED STATES

Reports From States for Week Ended December 1, 1951

The number of reported cases of measles increased from 3,401 for last week to 5,138 for the current week. Incidence increased in all sections of the country except in the West South Central, Mountain, and Pacific States. In 1950 the Mountain States was the only group in which a relatively high incidence rate was reported. The reported case rates show wide variations in the various parts of the country which probably are the result of differences in completeness of reporting. Thus, the relatively high rates of incidence which are common in epidemic years in the New England, Middle Atlantic, East North

Central, Mountain, and Pacific States probably are a reflection of more complete reporting. In other areas, such as the West North Central, Middle Atlantic, and East South Central States, reporting of measles probably is much less complete even in epidemic years when compared with the former group.

The incidence of diphtheria for the current week was 20 percent under that for the same week last year. The usual seasonal rise has not been evident in any part of the country except in the southern States.

The number of cases of meningococcal meningitis for the current week (84) is well above that for the same week last year (60) and the 5-year median (60). Since the seasonal low week early in September, 728 cases have been reported as compared with 661 for the same period last year.

A total of 29 cases of malaria in civilians was reported, 21 of which were in Wisconsin. A total of 117 cases in military establishments was reported. One military case in California, reported to have been infected in a southern State, is said to be a recurrent infection which has been treated since 1939.

One case of psittacosis was reported by Illinois, and two cases of smallpox were reported by Nebraska.

Epidemiological Reports

Serum hepatitis

Dr. C. R. Freeble, Ohio Department of Health, has reported an outbreak presumed to be homologus serum jaundice which has occurred in a State institution and was investigated in retrospect. From September 1950 through August 1951 there was a total of 13 cases of hepatitis admitted to the institution hospital, all having jaundice. Each patient had had a febrile illness which started with general malaise and, usually, abdominal pain. In most cases the liver was palpable and each had an elevated icteric index. The cases had their onsets as follows: September 1950, one case; February 1951, one case; May, one case; June, two cases; July and August, four cases each. The patients were scattered at random throughout the institution, and it was not possible to show any correlation between exposure to known cases and onset of illness. However, during February and March 1951, all inmates were subjected to routine blood typings by the finger puncture method. One spring lancet was used for all finger punctures, and was "sterilized" each time by dipping in isopropyl alcohol. The patients who had onsets of illness in June, July, and August remembered the month in which the puncture was done, and in all cases the illness fell within the general accepted incubation period for homologus serum jaundice. In September it was learned that a case of hepatitis occurred in a person who had been a patient

in another hospital where he had received a transfusion of blood on May 2. The blood donor was an inmate of the institution referred to above, but had not been recognized as a case of hepatitis.

Botulism

The food which was found to be implicated in the three fatal cases of botulism in Los Angeles, as reported last week, was home-canned greens, *Portulaca oleracea*. Type A toxin was demonstrated by animal inoculation.

Gastroenteritis

Dr. W. R. Giedt, Washington State Department of Health, has reported a small family outbreak of food intoxication in which commercially canned kippered herring was eaten. Bacteriological examination of the opened can showed it to contain hemolytic *Staphylococcus aureus*, but a specimen of another can of the same consignment was bacteriologically negative.

J. L. Rowland, Missouri Department of Public Health, has reported an outbreak of staphylococcus food poisoning which resulted from eating a cream-filled product known as "Long Johns." Of 40 persons known to have eaten this food, 34 became ill. On the day the cream filling was made, a few pastries were filled. The mix was refrigerated between fillings and overnight, but persons purchasing the pastry on the second day were more violently ill than those purchasing and eating the food on the first day.

Comparative Data for Cases of Specified Reportable Diseases: United States

[Numbers after diseases are International List numbers, 1948 revision]

Disease	Total for week ended—		5-year median 1946–50	Seasonal low week	Cumulative total since seasonal low week		5-year median 1945–46 through 1949–50	Cumulative total for calendar year—		5-year median 1946–50
	Dec. 1, 1951	Dec. 2, 1950			1950–51	1949–50		1951	1950	
Anthrax (062)	1			(1)	(1)	(1)	(1)	57	42	47
Diphtheria (055)	115	145	218	27th	1,820	2,441	4,298	3,828	5,569	8,909
Encephalitis, acute infectious (082)	33	18	5	(1)	(1)	(1)	(1)	973	933	603
Influenza (480–483)	581	848	974	30th	6,350	9,887	9,887	122,405	148,651	137,825
Measles (085)	5,138	2,515	2,277	35th	25,282	15,243	14,873	494,193	303,414	578,346
Meningitis, meningococcal (057.0)	84	60	60	37th	728	661	653	3,789	3,460	3,184
Pneumonia (490–493)	1,012	1,443	(2)	(1)	(1)	(1)	(1)	55,132	74,697	(2)
Poliomyelitis, acute (080)	428	597	433	11th	26,379	30,855	26,321	27,591	31,986	26,671
Rocky Mountain spotted fever (104)				(1)	(1)	(1)	(1)	328	451	555
Scarlet fever (050) [3]	1,276	1,158	1,551	32d	10,132	10,479	14,061	63,518	50,649	70,143
Smallpox (084)	2	4	1	35th	3	11	9	14	37	52
Tularemia (059)	13	23	23	(1)	(1)	(1)	(1)	593	825	899
Typhoid and paratyphoid fever (040, 041) [4]	52	51	51	11th	[5] 2,500	2,745	3,186	[5] 2,935	3,254	3,671
Whooping cough (056)	1,307	1,931	1,931	39th	9,784	14,997	14,997	63,559	112,192	91,503

[1] Not computed.
[2] Data not available.
[3] Including cases reported as streptococcal sore throat.
[4] Including cases reported as salmonellosis.
[5] Deduction: Arkansas, week ended Oct. 27, 1 case.

Reported Cases of Selected Communicable Diseases: United States, Week Ended Dec. 1, 1951

[Numbers under diseases are International List numbers, 1948 revision]

Area	Diphtheria (055)	Encephalitis, infectious (082)	Influenza (480–483)	Measles (085)	Meningitis, meningococcal (057.0)	Pneumonia (490–493)	Poliomyelitis (080)
United States	**115**	**33**	**581**	**5,138**	**84**	**1,012**	**428**
New England	**1**	**1**	**1**	**939**	**3**	**87**	**2**
Maine			1	161		28	
New Hampshire				32		13	
Vermont				87			1
Massachusetts	1			386	3		1
Rhode Island				93		4	
Connecticut		1		180		42	
Middle Atlantic	**14**	**11**	**8**	**2,042**	**13**	**95**	**35**
New York	8	11	(1)	888	6		19
New Jersey	1		8	422	2	33	2
Pennsylvania	5			732	5	62	14
East North Central	**9**	**3**	**4**	**708**	**12**	**78**	**50**
Ohio	5			176	4		13
Indiana				42	1		1
Illinois		2	3	242	6	62	16
Michigan	3	1	1	136		16	10
Wisconsin	1			112	1		10
West North Central	**8**	**2**	**9**	**281**	**8**	**78**	**42**
Minnesota	2		1	15	3	29	15
Iowa				2	1		
Missouri	5		4	6	1	1	10
North Dakota				225	2	40	3
South Dakota		1		6			1
Nebraska	1			16			3
Kansas		1	4	11	1	8	10
South Atlantic	**44**		**31**	**491**	**10**	**145**	**25**
Delaware							
Maryland			2	239		22	3
District of Columbia				20		18	
Virginia	8			61	2	47	2
West Virginia	5			69			5
North Carolina	6			3	2		1
South Carolina	9		5	3		6	
Georgia	11		24	82	5	52	10
Florida	5			14	1		4
East South Central	**23**	**2**	**1**	**264**	**6**	**54**	**31**
Kentucky	5			49	2	10	8
Tennessee	4			18	1		5
Alabama	10	1		185	1	25	5
Mississippi	4	1	1	12	2	19	13
West South Central	**15**	**3**	**169**	**46**	**17**	**343**	**75**
Arkansas	4		84	3	2	35	10
Louisiana	7		2		1	42	8
Oklahoma	1	1	83	1	9	14	11
Texas	3	2		42	5	252	46
Mountain		**1**	**255**	**190**	**4**	**57**	**39**
Montana		1	13	35	2		7
Idaho				10			
Wyoming				23		2	3
Colorado			74	55	1	29	13
New Mexico				21		2	
Arizona			168	4	1	24	
Utah				41			15
Nevada				1			1
Pacific	**1**	**10**	**103**	**177**	**11**	**75**	**129**
Washington			71	51	3	6	16
Oregon			26	21		29	12
California	1	10	6	105	8	40	101
Alaska							
Hawaii			172	599		2	

[1] New York City only.

Anthrax: Arkansas, 1 case.

Psittacosis: Illinois, 1 case.

Reported Cases of Selected Communicable Diseases: United States, Week Ended Dec. 1, 1951—Continued

[Numbers under diseases are International List numbers, 1948 revision]

Area	Rocky Mountain spotted fever (104)	Scarlet fever[1] (050)	Smallpox (084)	Tularemia (059)	Typhoid and paratyphoid fever[2] (040, 041)	Whooping cough (056)	Rabies in animals
United States		**1,276**	**2**	**13**	**52**	**1,307**	**179**
New England		**84**			**3**	**127**	
Maine		3				8	
New Hampshire		5				6	
Vermont					1	24	
Massachusetts		46			2	70	
Rhode Island		19					
Connecticut		11				19	
Middle Atlantic		**229**			3	244	36
New York		135				86	16
New Jersey		24				73	
Pennsylvania		70			3	85	20
East North Central		**366**		**1**	**4**	**217**	**20**
Ohio		108			2	25	5
Indiana		45				44	9
Illinois		55		1	1	25	4
Michigan		114				63	2
Wisconsin		44			1	60	
West North Central		**54**	**2**	**2**	**3**	**36**	**30**
Minnesota		21				1	23
Iowa		5				5	5
Missouri		11		2	1	15	2
North Dakota						1	
South Dakota		1				6	
Nebraska		2	2				
Kansas		14			2	8	
South Atlantic		**171**		**4**	**10**	**144**	**27**
Delaware		3				1	
Maryland		21		1	1	7	
District of Columbia		8					
Virginia		18				15	3
West Virginia		26				74	3
North Carolina		63		1		4	5
South Carolina		8		1	2	7	9
Georgia		15		1	5	30	7
Florida		9			2	6	
East South Central		**69**		**1**	**1**	**108**	**29**
Kentucky		18		1		53	12
Tennessee		41				17	6
Alabama		7				24	11
Mississippi		3			1	14	
West South Central		**38**		**3**	**9**	**290**	**37**
Arkansas		3			1	24	5
Louisiana		4				14	
Oklahoma		9			2	11	3
Texas		22		3	6	241	29
Mountain		**41**		**2**	**4**	**53**	
Montana		10				2	
Idaho		1			1	5	
Wyoming		1		2		1	
Colorado		3			1	15	
New Mexico		1			2	9	
Arizona		6				20	
Utah		13				1	
Nevada		6					
Pacific		**224**			**15**	**88**	
Washington		26				7	
Oregon		16			3	3	
California		182			12	78	
Alaska		5					
Hawaii		2			1		

[1] Including cases reported as streptococcal sore throat.
[2] Including cases reported as salmonellosis.

FOREIGN REPORTS

CANADA

Reported Cases of Certain Diseases—Week Ended Nov. 17, 1951

Disease	Total	Newfoundland	Prince Edward Island	Nova Scotia	New Brunswick	Quebec	Ontario	Manitoba	Saskatchewan	Alberta	British Columbia
Brucellosis	1						1				
Chickenpox	1,071	1		43	1	161	448	59	57	156	145
Diphtheria	10					10					
Dysentery:											
Amebic	2					2					
Bacillary	1							1			
German measles	76			8		6	14		13	30	5
Influenza	30			24			2	2			2
Measles	807	3		50	6	134	32	21	12	287	262
Meningitis, meningococcal	7	1				1	2		1	2	
Mumps	413	3			1	71	225	33	6	21	53
Poliomyelitis	25			3		7	9	4			2
Scarlet fever	376	3			2	90	20	41	32	38	150
Tuberculosis (all forms)	162	1		2	7	65	32	16	9	12	18
Typhoid and paratyphoid fever	3					1	1			1	
Venereal diseases:											
Gonorrhea	288	4		13	6	78	54	35	19	30	49
Syphilis	67	4		5	4	28	7	2		7	10
Primary	6					3	2			1	
Secondary	3	1			2						
Other	58	3		5	2	25	5	2		6	10
Whooping cough	219	3				79	59	30	6	25	17

CUBA

Reported Cases of Certain Diseases—4 Weeks Ended Oct. 27, 1951

Disease	Total	Pinar del Rio	Habana		Matanzas	Santa Clara	Camaguey	Oriente
			Habana City	Total				
Cancer	113	6		17	13	31	27	19
Chickenpox	1			1				
Diphtheria	19	1	5	12	1	1		4
Leprosy	47			10	3	18	8	8
Malaria	81	1	1	1			2	77
Measles	10	3	4	5	1	1		
Poliomyelitis	1							1
Tuberculosis	206	2	1	1	13	131	47	12
Typhoid fever	43	6	11	12	1	11	6	7

NORWAY

Reported Cases of Certain Diseases—July 1951

Disease	Cases	Disease	Cases
Diphtheria	6	Pneumonia (all forms)	1,688
Dysentery, unspecified	2	Poliomyelitis	106
Encephalitis, infectious	2	Rheumatic fever	66
Erysipelas	300	Scabies	448
Gastroenteritis	3,818	Scarlet fever	71
Hepatitis, infectious	49	Tuberculosis (all forms)	232
Impetigo contagiosa	1,254	Typhoid fever	4
Influenza	1,329	Venereal diseases:	
Malaria	2	Gonorrhea	146
Measles	1,164	Syphilis	30
Meningitis, meningococcal	11	Other forms	3
Mumps	121	Whooping cough	1,395

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

The following reports include only items of unusual incidence or of special interest and the occurrence of these diseases, except yellow fever, in localities which had not recently reported cases. All reports of yellow fever are published currently.

Cholera

Pakistan. The incidence of cholera in East Bengal Province rose from 52 cases for the week ended October 6 to 272 cases for the week ended October 27. In Dacca two cases were reported for the week ended November 24. The seaport of Chalna reported seven cases during the week ended November 17.

Smallpox

Indonesia. Two cases of smallpox were reported in Ambon, Ceram, for the week ended October 13. For the week ended November 17, smallpox was reported in Java as follows: Bandoeng, two cases; and Surabaya, nine.

Sudan, Anglo-Egyptian. An outbreak of smallpox has occurred in the Anglo-Egyptian Sudan. The number of cases reported for the week ended November 17 was 25 as compared with only 2 for the previous week.

Venezuela. During September, six cases of alastrin were reported.

Typhus Fever

Ceylon. One case of typhus fever was reported in the Western Province for the week ended September 22. This was the first case since June 24.

India. During the week ended November 24, two cases of typhus fever were reported in the seaport of Cochin.

Iraq. For the week ended November 24, four cases of typhus fever were reported. Of these, two were in Baghdad and one in Mosul.

Mexico. During the period October 28 to November 10, four cases of typhus fever were reported in Mexico City.

Yellow Fever

Gold Coast. During the period November 7–12, four cases of yellow fever were reported. Two cases were in African males, ages 8 and 27, at Akwatia. The other two cases were in the Oda District.