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INCIDENCE OF SALMONELLA CARRIERS AMONG MAMMALS IN THE LANSING AREA

Thesis for the Degree of M. S.

MICHIGAN STATE COLLEGE
Josefa Campodonico
1948

THESIS

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This is to certify that the

thesis entitled

Incidence of Salmonella Carriers
Among Mammals in the Lansing Area

presented by

Josefa Campodonico

has been accepted towards fulfillment
of the requirements for

Master's degree in Bacteriology

N. J. Stafseth
Major professor

Date August 3, 1948

M-795

INCIDENCE OF SALMONELLA CARRIERS AMONG MAMMALS IN THE LANSING AREA

A THESIS

By

Josefa Campodonico

Submitted to the School of Graduate Studies of Michigan State College of Agriculture and Applied Science in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

Department of Bacteriology

1948

THESIS

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ACKNOWLEDGMENT

The writer wishes to acknowledge her sincere appreciation to Doctor H. J. Stafseth under whose supervision these studies were planned and executed. She also wishes to thank Miss Lisa Neu who helped in one way or another to make this study possible.

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INTRODUCTION

A carrier is an individual who harbors specific organisms of a disease in his body without manifesting any clinical symptoms. Human and animal Salmonella carriers are a potential danger to susceptible individuals in the neighborhood. Human carriers are a serious public health menace, because of their many contacts with other people, especially if they are employed as food handlers. Animal carriers endanger the health of other animals, and pets and food animals which harbor organisms pathogenic for human beings are likewise serious threats to public health.

Salmonella carriers may have subclinical infections or they may be convalescent carriers or chronic carriers. In some cases persons and animals are considered as carriers because they harbor some extra-intestinal localized infection. Such infections are not accompanied by intestinal disorder. Because of the importance of Salmonella carriers, the following experiment was undertaken for the purpose of obtaining some information concerning the incidence of Salmonella carriers among mammals in the Lansing area.

REVIEW OF LITERATURE

Human Carriers

Wolff (1947) stated that only a few Salmonella types occur exclusively in man. The majority of the known types occur primarily as animal and bird pathogens but attain importance in human medicine because of their obvious adaptation as human pathogens.

Welch, Dehler, and Havens (1925) examined 1,076 persons in the dairy industry in Alabama and found 55 carriers (5.1 per cent). The incidence of infection was as follows: Salmonella typhosa 39, Salmonella paratyphi 13 and Salmonella schottmülleri 3. Of the typhoid and paratyphoid carriers, 18 and 5, respectively, were discharging the organism in the urine. Salmonella schottmülleri was isolated only from feces of the 3 carriers.

Schiff and Saphra identified (1941) Salmonella havana from an outbreak of meningitis in a maternity hospital in Havana. Twenty-one infants born within the hospital and an additional number born outside developed symptoms of severe purulent meningitis. The source of the infection could not be determined. Sera of three servants of the hospital agglutinated the organism in a dilution 1:150, but the organisms were not isolated from the stools.

Bornstein, Saphra, and Strauss (1941) at the New York Salmonella Center isolated the following organisms from the stools

of healthy carriers, three of whom were food handlers: S. schottmülleri, S. typhosa, Salmonella oranienburg, Salmonella derby, Salmonella typhimurium, Salmonella chester, Salmonella montendeo.

Stone (1943) reported that from two thousand food handlers in the Panama Canal Zone, 14 species of Salmonella were isolated from 44 of the individuals.

Galton and Quan (1943) in Florida isolated the following Salmonellae from normal persons: S. derby from a man, age 57; Salmonella anatum from a foodhandler and from the feces of a normal female applying for a job as foodhandler.

Edwards and Bruner (1943) reported a study of 3,000 Salmonella cultures isolated from man and animals in the United States and its possessions between 1934 and 1941. The following species were isolated from normal human carriers:

No. of Salmonella Carriers	
paratyphi B	5
paralyphi B var java	14
typhi-murium	7
derby	2
california	1
bredeney	2
oranienburg	2
bareilly	3
montevideo	8
tenessee	1
newport	18
oregon	1
manhattan	1
litchfield	1
bonariensis	1
typhi	6
panama	2
javiana	1
give	1
anatum	4
nemington	31
senftenberg	8
simsbury	1
northington	1
mississippi	1

Seligmann, Saphra, and Wassermann (1943) reported that bacteriological examination of 1,000 persons by the New York Salmonella Center, showed 89 to be healthy carriers. Of these, 44 were contact infections. They represent one group of healthy resistant individuals (among them were two babies in the first month of life.)

Hormaeche, Surraco, Feluffo, and Aleppo (1943) reported that from 3,592 children, admitted to hospitals for all causes

without regard to intestinal disorders, 29 different types of Salmonella were isolated from 489 patients. The most prevalent types were S. typhimurium, Salmonella newport, Salmonella montevideo, S. anatum, S. schottmülleri and Salmonella bredeney. Thirty-four (9.04 per cent) of the 489 persons infected with Salmonella were carriers.

Posch, Stofer, and Hirscfield isolated Salmonella choleraesuis as the specific factor in an indolent, subcutaneous abscess in an aged Negro laborer who had suffered for 40 years as a result of a severe contusion a few inches above the wrist.

Animal Carriers

The role played by the Salmonella in animal disease is an important one as all species of domestic mammals are in varied degree susceptible to infection.

The work of Edwards and Bruner (1943), showed that swine were by far the most important source of Salmonella organisms in mammals. Salmonellae have also been isolated from healthy cattle, dogs, rats, mice and birds.

Wolff (1947) stated that S. cholerawsuis was the most prevalent of more than 30 Salmonellae isolated from swine. The second most frequently occurring type was S. typhimurium.

Rubin, Scherago, and Weaver (1942) traced many outbreaks of Salmonella food poisoning in man to the eating of pork from swine which had undergone rigid antemortem and postmortem veterinary inspection.

Hormaeche and Salsamendi (1936) isolated Salmonella from the mesenteric lymph nodes of apparently healthy hogs slaughtered for meat purposes. They examined materials from 36 lots of hogs consisting of 20 animals each, and were able to isolate Salmonella organisms from 22 (47.9 per cent) of these lots. These organisms were: S. typhimurium, S. anatum, S. derby, S. newport, S. montevideo, Salmonella muenchen, S. schottmülleri, S. bredeney, Salmonella Berta, Salmonella Carran, and Salmonella cerro.

Monteverde (1942) isolated Salmonella bonariensis from the mesenteric gland of a normal pig in Buenos Aires.

Rubin, Scherago and Weaver (1942) examined 40 lots of hogs, each lot consisting of 25 animals, and found the incidence of Salmonella organisms to be 47.5 per cent. Of the 50 hogs examined individually, 5 (10 per cent) yielded Salmonellae. From these positive lots and individual animals, 242 strains yielded 13 types of Salmonella.

Cherry, Scherago and Weaver (1943) investigated the incidence of Salmonella organisms in retail market meats. They found a higher incidence rate in pork products than in other meats. Of the 250 meat samples examined, 13 (5.2 per cent) yielded Salmonella and 10 (5 per cent) of these were isolated from 170 pork products. The Salmonellae found in pork samples were: S. typhimurium, S. newport, S. anatum, Salmonella give, S. bredeney, Salmonella newington, S. derby, and Salmonella senftenberg.

Bruner and Edwards (1946) reported the isolation of 11 types of Salmonella from 60 bovine outbreaks. The most frequent types found were Salmonella dublin, S. typhimurium, S. choleraesuis and Salmonella enterilidis.

Guerrero (1943) stated that carrier cows are usually the source of Salmonella infection on farms where the disease is not prevalent. These cows usually abort their calves from Salmonella infection at successive pregnancies.

Johnson and Graham (1944) reported the isolation of Salmonella choleraesuis var. kunzendorf from a bovine fetus originating from a brucellosis free herd.

Wolff (1947) stated: "Meat from cattle has often been incriminated in outbreaks of Salmonella infection, though less often than pork. In fact, the first recorded Salmonella food infection was traced to meat of a diseased cow, in which S. enteritidis was isolated from the patients and from the suspected meat."

Cherry, Scherago and Weaver (1943) examined 64 beef samples. S. typhimurium was found in one sample and S. senftenberg in two of the samples.

Salmonella infection of the horse is quite common, but there is insufficient information about Salmonella carriers among horses.

Jordan and Burrows (1945) give S. abortus equi as the cause of infectious abortion of mares. S. typhimurium has also occasionally been reported in horses.

Wolff (1947) stated that there have been reports of horse meat as a source of infection with Salmonella abortus equi, S. typhimurium and other varieties.

Newson and Cross (1924, 1930, 1935) investigated several outbreaks of gastroenteritis in lambs caused by S. typhimurium. They regarded the long railway journeys the lambs had to make and the long periods of fasting as predisposing factors.

Henning (1936) described a new type of Salmonella isolated from a sheep at Onderstepoort, South Africa, and was named Salmonella Onderstepoort.

Henning (1939) stated that Salmonella abortus ovis is the most common pathogenic Salmonella for sheep.

Wolff, Henderson and McCallum (1948) examined 100 dogs. Eighteen of these animals were found to be excreting different Salmonella types: Salmonella manhattan, S. newport, Salmonella minnesota (both monophasic and diphasic varieties), S. oranienburg, S. typhimurium, S. bredeney, S. worthington, Salmonella give, Salmonella cubana, S. cerro, Salmonella kentucky, Salmonella illinois and Salmonella meleagrides.

Wolff (1947) stated that more than 20 Salmonella types, all of which are known human pathogens, may be isolated from various conditions in dogs.

Wolff (1947) stated that S. choleraesuis and S. paratyphi have been isolated from cats.

Litch and Meyer (1921) described an epidemic infection among rabbits caused by S. schottmülleri.

There is little information in the literature about Salmonella carriers among minks, rabbits and goats.

Coburn, Armstrong and Wetmore (1942) reported S. typhimurium infection in chinchillas (chinchilla laniger). The organism was isolated from 32 of the 44 animals examined. Carrier animals were not detected by repeated cultural tests of fecal material. Four rodents of the domestic species of mice were found to be infected. The authors were of the opinion that wild rodents were the source of the infection in the chinchillas.

There are many reports in the literature about Salmonella carriers among rats and mice because of the importance of these animals in the spread of Salmonellosis.

Duthie and Mitchell (1931) reported that several strains of the genus Salmonella appeared capable of inciting the formation of pseudo tubercles in rodents.

Many attempts to isolate organisms of the Salmonella group from rat and mouse excreta have been made because of food poisoning outbreaks. Meyer and Matsumura (1927) examined 775 rats in the city of San Francisco, California, 58 rodents were infected either with S. enteritidis (28 cases) or S. typhimurium (30 cases).

Krumwiede and Salthe (1924) were successful in isolating S. typhimurium from rodent excreta obtained in a bakery where prepared cream filling was found to be the cause of a food poisoning outbreak.

Welch, Ostrolenk, and Bartram (1941) made a study of rodent feces collected from a large area of the United States without regard to history of intestinal disease. They found that only a small percentage (1.2 per cent) of these animals were excreting food poisoning organisms of the Salmonella type. While performing the same experiment, they found that excreta of rats naturally infected with S. enteritidis and held at room temperature may contain viable organisms for at least 148 days.

Bartram, Welch, and Ostrolenk (1940) found that the majority of infected animals ceased to pass the organisms by the 15th day. Approximately 20 per cent continued to excrete Salmonella for as long as 7 weeks after infection which was the duration of their experiment. They suggest this as indicating, as in the case of human enteric carriers, that a small percentage of infected animals become carriers.

Rubenstein (1944) stated that about 2 per cent of all persons infected with Salmonella excrete these organisms for a period longer than one year, while the clinically manifested disease, if present at all, does not last over a few weeks or days. He also stated that some strains are much more likely to

produce carrier status than are others. Five per cent of all persons infected with Salmonella schottmülleri become carriers, while human carriers of S. choleraesuis have never been observed.

EXPERIMENTAL WORK

Materials

In this study, 170 specimens were collected from feces, intestines, colons, and lymph nodes of humans and of animals as shown in Table I.

TABLE I

Individuals	Sources of Samples				
	Feces	Intestine	Lymph Nodes	Colon	Total
Humans	20				20
Pigs	2	2	4	1	9
Cattle	20	1			21
Horses	14	1			15
Sheep	6	1			7
Goats	4				4
Dogs	30	6			36
Cats	14				14
Rabbits	6				6
Minks		11			11
Chinchilla		1			1
Mice	11				11
Rats	6	8			14

The human samples were obtained from Michigan State College students. The animal materials were taken from the following sources: Michigan State College Veterinary Clinic, Michigan State College barns, and private farms around East Lansing. All the rat and mice specimens were collected from the Department of Chemistry and from the Department of Physiology and Pharmacology of Michigan State College.

Procedure

Fecal samples about as large as a pea were dropped into tetrathionate broth and incubated for 24 hours at 37°C. Some of the fecal specimens were diluted with enough sterile water to make the samples soft, and from each of these streak inoculations were made on one McConkey plate and on one SS agar plate. The plates were incubated with the tetrathionate broth cultures.

When intestines, colons, and lymph nodes were used, a piece of the sample was dropped into tetrathionate broth, and another piece of the original sample was streaked on one McConkey plate and on one SS plate. All were incubated for 24 hours at 37° C. Up to the first 38 samples the tetrathionate broth cultures were streaked upon SS only, after that the rest of the tetrathionate cultures were streaked on McConkey and SS plates.

After the 24 hour incubation, all the plates were examined and the non-lactose-fermenting colonies were selected and inoculated into Kligler's iron agar slants. The plates were returned to the incubator, re-examined at 48 hours and kept two more days in the incubator for observation. Gram stains were made from the Kligler slant cultures. Gram negative organisms showing salmonella-like reactions in Kligler's media (acid and gas in the butt, alkaline slant with or without H_2S) were inoculated into the following carbohydrates: dextrose, lactose, sucrose,

maltose, mannite, sorbitol and salicin. Tryptone broth, urea and lactose motility media were also inoculated. After 48 hours incubation, the carbohydrate media were observed for the production of acid and gas. If acid and gas were not present the incubation was continued and observations were made daily. The tryptone broth cultures were tested for indol reaction and the lactose motility tubes were observed for motility and lactose fermentation. Proteus organisms were detected on the basis of urease activity.

If acid and gas were produced in dextrose, maltose, mannite, sorbitol and not in lactose, sucrose and salicin; indol was not produced; urea was not decomposed; and hydrogen sulphide was produced or not; and the organism was motile, it was tentatively identified as a member of the Salmonella group. For determination of the species further fermentation studies with the necessary rare carbohydrates were done. Cultures were submitted to the Salmonella typing Station, Bureau of Laboratories, Michigan State Department of Health, for antigenic analysis.

Results

The results from the 170 samples are as shown in Table II. S. typhimurium was the only Salmonella species isolated. This culture was isolated from the feces of a 5 year old dog in the Veterinary Clinic of Michigan State College. The dog had a whipworm infestation.

TABLE II

Animal	Age	Sex M	Sex F	Source of Sample: Feces	Intestine	Lymph nodes	Colon	Total No. Samples	No. of Salmonella found
Humans	20-30		17	17				17	0
Pigs				2	2	4	1	9	0
Cattle	Up to 3	3	4	6	1				0
Cattle	3-6	2	12	14				21	0
Horses	Up to 7	5	3	7	1				0
Horses	7-14	5	2	7				15	0
Sheep				6	1			7	0
Goats				4				4	0
Dogs	Up to 3	15	7	16	6				0
	Unknown			13					0
	5	1		1				36	1*
Cats		11	3	14				14	0
Rabbits				6				6	0
Minks	Up to 2	9	2	11				11	0
Chinchillas	1½	1			1			1	0
Mice				11				11	0
Rats				6	8			14	0

*Salmonella typhimurium

DISCUSSION

Assuming that the technic employed in this work was adequate for the detection of Salmonella, it is obvious that the incidence of Salmonella carriers in the Lansing area is not great.

Selective media are toxic in some degree to all organisms and the use of tetrathionate broth and SS in succession for 38 of the samples may have resulted in inhibition of growth of Salmonellae.

In a thorough examination for Salmonellae, one ought to employ a variety of combinations of media for each sample. Time and facilities did not permit such a far reaching investigation but these studies should be supplemented by further work of more intense nature.

It is also possible that not enough cultures were made.

SUMMARY

1. One hundred and seventy samples of feces, intestines, colons, and lymph nodes obtained from humans and animals were examined bacteriologically for Salmonella.

2. S. typhimurium was isolated from the feces of a 5 year old male dog which was infested with whipworms. All other samples failed to yield Salmonellae.

BIBLIOGRAPHY

Bartram, M. T., Welch, H., and Ostrolenk, M. 1940 Incidence of members of the Salmonella group in rats. J. Infectious Diseases, 66, 222-226.

Bornstein, S., Saphra, I., and Strauss, L. 1941 Frequency of occurrence of Salmonella species. J. Infectious Diseases, 69, 59-64.

Bruner, D. W., Edwards, P. R. 1946 Salmonellosis of domestic animals. Proc. 50th. Ann. Meet. U. S. Livestock Sanitary Association ps 194-198.

Cherry, W. B., Scherago, M., and Weaver, R. H. 1943 The occurrence of Salmonella in retail meat products. Am. J. Hyg., 38, 173-177.

Coburn, D. R., Armstrong, W. H., and Wetmore, P. W. 1942 Observations on bacteria treatment of Salmonella typhi-murium infection in chinchillas. American J. Vet. Research, 3, 96-99.

Duthie, R. C., and Mitchell, C. A. 1931 Salmonella enteritidis infection in guinea pigs and rabbits. J. Am. Vet. M. Assoc., 78, 27-41.

Edwards, P. R., and Bruner, D. W. 1943 The occurrence and distribution of Salmonella types in the United States. J. Infectious Diseases, 72, 58-67.

Galton, M. M., and Quan, A. L. 1943 Varieties of Salmonella isolated in Florida during 1942. Am. J. Hyg., 38, 173-177.

Guerrero, R. P. 1943 Salmonellosis of calves in tropical countries. J. Am. Vet. M. Assoc., 103, 152.

Henning, M. W. 1936 Salmonella onderstepoort: A new type of Salmonella from a sheep. J. Hyg; 36, 525-530.

Henning, M. W. 1939 The antigenic structure of Salmonella obtained from domestic animals and birds in South Africa. Onderstepoort J., 13, 79-189.

Hormaeche, E., Surraco, W. L., Feluffo, C. A., and Aleppo, P. L. 1943 Causes of infantile summer diarrhea. Am. J. Dis. Children, 66, 539-551.

Hormaeche, E., and Salsamendi, R., 1936 Sobre la presencia de Salmonellas en los ganglios mesentericos de cerdos normales. Arch. Urug. Med. Cir. y Espec., 9, 665.

Johnson, L. E., and Graham, R. 1944 Results of bacteriological examination of aborted bovine fetuses from brucellosis-free herds Cornell Veterinarian, 35, 36-40.

Jordan, E. O., and Burrows, W. 1945 Textbook of bacteriology 14th ed. W. B. Saunders Co., Philadelphia and London. p. 395.

Krumwiede, C. 1924 Studies on the Paratyphoid-Enteritidis group, VIII, An epidemic of food infection due to a paratyphoid Bacillus of rodent origin. Am. J. Hyg., 4, 23-32.

Litch, V. M., Meyer, K. F. 1921 A spontaneous epidemic among laboratory rabbits caused by a paratyphoid B. Bacillus related to the rodent group. J. Infectious Diseases, 28, 27-42.

Meyer, K. F., Matsumura, K. 1927 The incidence of carriers of B. aertrycke and B. enteritidis in wild rats of San Francisco. J. Infectious Diseases, 41, 395-404.

Monteverde, J. J. 1942 A new Salmonella type. Nature, 149, 472.

Newson, I. E., and Cross, F. 1924 An outbreak of paratyphoid dysentery in lambs. J. Am. Vet. Med. Assoc., N.S.V. 19, 289-300.

Newson, I. E. and Cross,F. 1930 Paratyphoid Dysentery in lambs. J. Am. Vet. Med. Assoc., N.S.V. 29, 91-92.

Newson, I. E. and Cross, F. 1935 Third outbreak of paratyphoid dysentery in lambs. J. Am. Vet. Med. Assoc., N.S.V. 39, 534-536.

Posch, J. L., Stofer, B. E., and Hirshfield, J. W. 1944 Salmonella choleraesuis as a cause of indolent subcutaneous abscess (Human) J. Am. Vet. Med. Assoc., 104, 85.

Rubin, H. L., Scherago, M., and Weaver, R. H. 1942 The occurrence of Salmonella in the lymph glands of normal hogs. Am. J. Hyg., 36, 43-47.

Rubenstein, A. D., Feemster, R. F., and Smith, H. N. 1944 Salmonellosis as a public health problem in wartime. Am. J. Pub. Health, 34, 841.

Schiff, F., and Saphra, I. 1941 A new Salmonella type: Salmonella havana. J. Infectious Diseases, 68, 125-127.

Seligmann, E., Saphra, I. and Wassermann, M. 1943 An analysis of 1,000 cases bacteriologically identified by the New York Salmonella Center. Am. J. Hyg., 38, 226-247.

Stone, W. S. 1943 Food handlers in the army and their relationship to Salmonella food poisoning. Am. J. Pub. Health, 33, 706-708.

Welch, S. W., Dehler, S. A., and Havens, L. C. 1925 The prevalence of typhoid carriers in a general population. J. Am. Med. Assoc., 85, 1036-1039.

Welch, H., Ostrolenk, M., and Bartram, M. T. 1941 Role of rats in the spread of food poisoning bacteria of the Salmonella group. Am. J. Pub. Health, 31, 332-340.

Wolff, A. H., 1947 The public health significance of animal Salmonella infections. J. Am. Vet. Med. Assoc., 111, 474-480.

Wolff, A. H., Henderson, N. D., McCullam, G. L. 1948 Salmonella from dogs and the possible relationship to Salmonellosis in man. Am. J. Pub. Health, 38, 403-408.

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