

SALMONELLAE IN FERAL PINNIPEDS OFF THE SOUTHERN CALIFORNIA COAST

WILLIAM G. GILMARTIN,[□] PATRICIA M. VAINIK,[□] and VIOLA M. NEILL[□]

Abstract: Rectal swabs were collected from 90 Northern fur seal (*Callorhinus ursinus*) and 50 California sea lion (*Zalophus californianus*) pups on San Miguel Island for *Salmonella* screening. Three serotypes (newport, heidelberg, and oranienburg) were recovered from 33% of the fur seals and 40% of the sea lions.

INTRODUCTION

Many species of *Salmonella* have been recovered from animals in the marine environment. Isolates from marine mammals include a beluga whale (*Delphinapterus leucas*),¹ a bottlenosed whale (*Hyperaodon rostratum*),¹¹ young California sea lions (*Zalophus californianus*),¹³ and Northern fur seal pups (*Callorhinus ursinus*)⁹ (Doyle, unpubl.).

In 1951, cultures of blood and tissues of 5 of 12 dead fur seal pups on the Pribilof Islands yielded *S. enteritidis*.⁹ The authors of the report suggested that mortality in the pups may be a result of salmonellosis. In another study of fur seal pup mortality later in that decade, *S. enteritidis* was isolated from the enlarged mesenteric lymph nodes of only 1 of 58 animals examined bacteriologically (Doyle, unpubl.). This author also suggests that salmonellosis may be related to the problem but cautions against assigning too much importance to the finding.

Sick and injured California sea lions are picked up at local beaches (vicinity of San Diego, California) for treatment at the laboratory of one of the authors (WGG) and release. Salmonellae fre-

quently are isolated from the stool of these animals and in one case the organism was recovered from a bronchial swab culture.¹³ An increasing incidence of isolation of *Salmonella* spp. from these sick beached animals led to the study reported here in which a survey for *Salmonella* in feral California sea lion and Northern fur seal pups was conducted on San Miguel Island to determine the prevalence in the normal feral populations.

MATERIALS AND METHODS

In October 1975, rectal swabs were taken from 90 Northern fur seal (*Callorhinus ursinus*) and 50 California sea lion (*Zalophus californianus*) pups. These animals were sampled at the rookery on the northwest point of San Miguel Island off Southern California in conjunction with animal tagging procedures conducted by the National Marine Fisheries Service. The pups, all 3-5 months old, appeared to be in generally good health at the time of sampling.

The swabs were used to inoculate plates of Levine Eosin Methylene Blue Agar,[□] Salmonella-Shigella Agar[□] and Selenite-F Broth.[□] After 24 h of incuba-

[□] Biosciences Department, Naval Ocean Systems Center, P.O. Box 997, Kailua, Hawaii 96734, USA.

[□] Water Utilities Department, City of San Diego, San Diego, California 92101, USA.

[□] Biology Department, California State College, Hayward, California 94542, USA.

[□] BBL, Division of Becton, Dickinson & Co., Cockeysville, Maryland 21030, USA.

tion the broth enrichments were streaked onto Salmonella-Shigella Agar plates. All plated media were examined after 24 h of incubation for colonies suggestive of non-lactose fermenters. Several of such colonies were selected from each plate and inoculated onto Christensen Urea Agar.⁵ All organisms giving a positive urease reaction were discarded. The urease negative cultures were further tested for bio-chemical activity on API²⁰ E test strips. Cultures showing typical salmonellae reaction patterns were selected and maintained on slants of Trypticase Soy Agar.⁶

Serological characterization with specific antisera⁶ was performed using standard procedures.⁶ All organisms were grown on Trypticase Soy Agar⁶ prior to typing for somatic antigens. These were subsequently passed through Motility Test Medium⁷ before inoculating a broth consisting of 1:1 Trypticase Soy Broth and Brain Heart Infusion Broth.⁸ This broth culture was then formalized to provide the antigen for flagellar typing.

The virulence of nine of the *Salmonella* isolates was determined in LD₅₀ trials with seven-week-old Swiss Webster mice. LD₅₀ titers were obtained using the method of Reed and Muench.¹² Spleen, liver, and intestinal contents were cultured from the mice that died.

RESULTS AND DISCUSSION

Three *Salmonella* serotypes were recovered in this investigation: *S. newport*, *S. oranienburg* and *S. heidelberg*. They were found in 40% of the California sea lion pups and 33% of the Northern fur seal pups as detailed in Table 1. In contrast to previous attempts at isolation from pinnipeds, these cultures were collected from apparently healthy animals rather than sick or dead pups.

Thus prompting a question as to the virulence of the isolates.

LD₅₀ studies with several of the cultures indicated a wide range of pathogenicity for mice, from a high LD₅₀ of 9.4×10^4 cells to 1.3×10^7 cells, injected intraperitoneally. Post-mortem cultures of selected tissues from the mice which died yielded *Salmonella* in all cases.

The absence of any obviously sick animals in this sampling may indicate these serotypes produce only a mild gastroenteritis followed by a prolonged carrier state or that most of the animals, due to passively acquired maternal antibody or early immune competence, do not experience the disease but are simply transient asymptomatic carriers. In a large epidemic of *S. newport* in a human population, two-thirds of the patients were found to be only carriers; only 2% developed severe illness.^{3,10}

The high prevalence of a potential pathogen such as these salmonellae in a pinniped population is not surprising as the rookeries are heavily contaminated with fecal material. The western gull (*Larus occidentalis*) also is present in the rookeries and may be responsible to some extent for spreading the salmonellae about the beaches since gulls of several species have been implicated as carriers.^{2,14,15}

Mortality due directly to *Salmonella* in these young pups does not appear to be a problem, yet it may have significance as a disease problem in pups or older animals which become debilitated for other reasons. Consideration here must be given to nutritional deficiencies at the time of weaning and the potential of a parasitic pneumonia in animals once they begin feeding for themselves.

Salmonellae are responsible for abortion in some species (i.e. cattle⁷ and horses⁴). A high rate of abortion exists in

⁵ Analytab Products, Inc., Planview, New York 11803, USA.

⁶ Difco Laboratories, Detroit, Michigan 48232, USA.

TABLE 1. Number of individuals of each pinniped species from which the respective serotype(s) were isolated.

| | California sea lion | Northern fur seals |
|----------------------------|---------------------|--------------------|
| Salmonella Serotypes | | |
| newport | 16 | 22 |
| heidelberg | — | 4 |
| oranienburg | 3 | — |
| newport and oranienburg | 1 | 2 |
| oranienburg and heidelberg | — | 1 |
| newport and heidelberg | — | 1 |

the California sea lion population at the San Miguel Island rookery,^{5,8} but to date selective culture techniques for *Salmonella* isolation have not been used in the study of the problem.

An effort is now underway to investigate the possibility that these salmonellae may be involved in the abortions and to further study the disease in young pinnipeds.

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