Epidemiological Surveillance of *Serratia marcescens* Infections by Bacteriocin Typing

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During 10 months, 155 isolates of *Serratia marcescens* were cultured from 105 patients, of whom 49 were considered to have significant infection. The 155 isolates were typed by bacteriocin sensitivity, and 137 (88.4%) were assigned to 37 provisional bacteriocin groups; 18 isolates were nontypeable. No major outbreaks of nosocomial infection were demonstrable; however, there were four chronologically separate minor episodes of cross-infection that involved two or three patients per room or unit, respectively.

Lately, a variety of opportunistic microorganisms have gained notoriety by causing serious infections in debilitated patients. *Serratia marcescens*, long considered a nonpathogen, is now among these (3–5, 12, 21, 23, 24) and has caused outbreaks of nosocomial infection (1, 2, 6, 14, 16). The increased prevalence of this organism was noted within our hospital during the past 2 years. We recently developed a procedure for typing *S. marcescens* by bacteriocin sensitivity (20); we now wish to show the use of the procedure in defining the epidemiology of *S. marcescens* infections in our hospital.

MATERIALS AND METHODS

From 1 June 1970, through 15 April 1971, 155 isolates of *S. marcescens* were cultured from 105 patients. The isolates were identified as previously described (19), as were 24 isolates that had been encountered a year earlier, frozen, and stored at −65°C (20). The isolates were typed by bacteriocin sensitivity as previously reported (20).

Patient charts and laboratory records were reviewed to differentiate between significant infection and colonization by the organism. The criteria of Thoburn et al. (18) served to categorize the patients.

To determine the prevalence of the organism in the upper respiratory tract of laboratory personnel, throat swabs (tonsils and posterior pharynx) from 72 persons were inoculated into tryptic soy broth (TSB, Difco Laboratories, Detroit, Mich.) containing polymyxin B (50 μg/ml; Aerospin, injectable; Burroughs Wellcome and Co., Inc., Tuckahoe, N.Y.) and incubated at 35°C overnight; tubes with growth were subcultured and further processed for isolation of *S. marcescens*. On 15 April 1971, 8 patients and 31 personnel (nurses, physicians, medical students, nursing aides, and maids) of the intensive care unit were examined; on this date the unit contained no patients known to have *S. marcescens* infection. Environmental samples were taken as follows. Respirators were sampled immediately before and after use, and after decontamination with 2% aqueous glutaraldehyde for 20 min, followed by successive rinses with “vinegar water” [15 ml of commerical vinegar in 2 gal (ca. 7.6 liters) of water] and hot water. Utensils, floors, and walls of the surgical and urological operating suites and of the intensive care unit were sampled by the swab technique.

RESULTS

The sources of *S. marcescens* from patients are shown in Table 1. The organism was often isolated from the respiratory tract. Of the five patients with questionably significant infection, three isolates were cultured from patients in outside hospitals; the chart of one patient was not available for review, and one patient had *S. marcescens* in his urinary bladder urine at the time of necropsy.

The age and sex distributions of the 100 patients with significant and insignificant infection are listed in Table 2. The organism appeared to have a predilection for middle-aged and elderly patients in both groups. The sex ratio was five males to one female among the patients with significant infection, but less than 2:1 in patients with insignificant infection. All but three patients were Caucasian. The underlying illnesses of the patients are shown in Table 3. The patients with significant infection had a variety of diseases which predispose to infection with opportunistic pathogens. We judged that *S. marcescens* contributed to mortality in 9 of the 49 patients with significant infection. The organism colonized in the respiratory tract of several patients with chronic bronchitis. *S. marcescens* colonized and remained colonized in a number of patients with predisposing illnesses.

Table 4 shows the distribution of all the patient
TABLE 1. Clinical sources of Serratia marcescens isolates

| Source                          | No. of isolates clinically |
|---------------------------------|-----------------------------|
|                                 | Significant | Questionable | Insignificant |
| Respiratory tract               |             |             |              |
| Sputum                          | 25          | 2           | 40           |
| Tracheal aspirate               | 2           | 1           |              |
| Bronchial washing               | 1           |             |              |
| Blood                           | 3           |             |              |
| Catheter (intravenous) tip      | 1           |             |              |
| Urine                           |             |             |              |
| Clean-voided                    | 5           | 4           |              |
| Catheterized                    | 2           |             |              |
| Genital tract (cervix)          |             |             |              |
| Wounds, abscesses               | 9           | 1           | 1            |
| Skin                            | 1           |             |              |
| Peritoneal fluid                | 1           |             |              |
| Eye                             |             |             |              |
| Stool                           |             |             |              |
| Autopsy specimens               |             |             |              |
| Left ventricular blood          |             |             |              |
| Urinary bladder urine           | 1           |             |              |
| Total                           | 49          | 5           | 51           |

TABLE 2. Age and sex of patients

| Age (years) | Significant infection | Insignificant infection |
|-------------|-----------------------|-------------------------|
|             | Male | Female | Total | Male | Female | Total |
| 0           | 1    | 1      | 1     | 1    | 1      | 2     |
| 1–10        | 2    | 2      | 4     |       |        |       |
| 11–20       | 1    | 1      | 1     | 2    | 3      | 5     |
| 21–30       | 2    | 2      | 2     | 1    | 1      | 2     |
| 31–40       | 3    | 2      | 1     | 3    | 1      | 4     |
| 41–50       | 5    | 4      | 9     | 6    | 4      | 10    |
| 51–60       | 7    | 1      | 8     | 3    | 1      | 4     |
| 61–70       | 15   | 10     | 25    | 4    | 3      | 7     |
| 71–80       | 7    | 3      | 10    | 3    | 1      | 5     |
| 81–90       | 1    | 1      | 2     | 1    | 1      | 2     |
| Total       | 41   | 32     | 73    | 19   | 19     | 38    |

TABLE 3. Underlying illnesses of patients

| Underlying illness                                         | Significant infection | Insignificant infection |
|-----------------------------------------------------------|-----------------------|-------------------------|
| Respiratory disease (chronic bronchitis with or without obstructive pulmonary emphysema) | 6         | 22                      |
| Diabetes mellitus                                           | 4         | 3                       |
| Renal insufficiency (chronic glomerulonephritis, pyelonephritis) | 2         | 2                       |
| Chronic alcoholism                                           | 2         |                         |
| Acute myocardial infarction, arteriosclerotic heart disease | 1         | 3                       |
| Major surgery (thoracic, abdominal, cranial)                | 13        | 6                       |
| Severe burns                                                | 4         |                         |
| Prematurity, neonatal period                                | 1         | 2                       |
| Hematologic disorders (leukemia, lymphoma, thrombocytopenia, nonthrombocytopenic purpura) | 4         | 6                       |
| Malignant neoplasms (carcinoma of lung, colon, rectum, gull bladder, prostate; brain tumor; multiple myeloma) | 7         | 3                       |
| Gastrointestinal and hepatic disease (rectal bleeding, ruptured ileum, perforated gastric ulcer, ulcerative colitis, hepatic cirrhosis) | 3         | 2                       |
| Miscellaneous                                               |           |                         |
| Wegener's granulomatosis                                    | 1         |                         |
| Myasthenia gravis                                           | 1         |                         |
| Periartheritis nodosa                                        | 1         |                         |
| Total                                                      | 49        | 51                      |

isolates into 37 bacteriocin sensitivity patterns. The majority of the isolates constituted bacteriocin groups 1, 4, 9, 14, 16, 17, 18, 21, 22, and 26. The isolates comprising these 10 groups were cultured from 62 of the 105 patients. Bacteriocin groups 1, 4, 9, 14, and 18 were most common. Among the isolates cultured in 1969, five and six isolates belonged to bacteriocin groups 4 and 9, respectively. On the other hand, S. marcescens strains of bacteriocin groups 5, 6, 8, and 10, that had been present in our hospital in 1969, were not encountered in 1970 and 1971. In contrast, strains of bacteriocin groups 1 and 11 through 37 were not prevalent in 1969.

Of the 100 patients, 20 had required intensive care; 16 of these had significant infection with S. marcescens. Of 31 isolates of S. marcescens cultured from these patients, 28 isolates belonged to 12 different bacteriocin groups, and three could not be typed. Once, on the same day (12 July 1970), two patients in the intensive care unit had sputum cultures that yielded S. marcescens of bacteriocin types 4 and 14, respectively.

There were four episodes of cross-infection involving two, in one instance three, patients of the same room or unit (Table 5). In all instances, the durations of hospitalization over-
lapped. On 15 April 1971, there was one previously undetected patient (S.C.G.) in the intensive care unit who had acquired infection with *S. marcescens* of bacteriocin group 37; patient B.E.Q. had been shown to be infected with *S. marcescens* of the same bacteriocin type several days earlier. Subsequently, patient N.J.S., who developed postsurgical bronchopneumonia attributable to *Staphylococcus aureus* and *Escherichia coli* infection, had *S. marcescens* of identical bacteriocin type in the right lower lobe of the lung at the time of necropsy on 24 April 1971. However, none of 31 workers of the intensive care unit was a pharyngeal carrier of the organism.

At any one time, there were never more than five patients with significant infection or colonization by *S. marcescens* among the roughly five hundred patients of our hospital. If infection were of endogenous origin, this would suggest a low carrier rate. Therefore, we attempted to assess the carrier rate among laboratory personnel with little, if any, contact with patients. Only one of seventy-two persons (1.4%) was found to be a pharyngeal carrier of the organism. The isolated strain was bacteriocin group 10;
TABLE 5. Epidemiology of four minor episodes of cross-infection with Serratia marcescens

| Episode | Bacteriocin (marcescin) group | Isolate no. | Date cultured | Patient (initials) | Source | Location (room or unit) | Ward | Admission date | Discharge or death |
|---------|-------------------------------|-------------|---------------|-------------------|--------|------------------------|------|------------------|-------------------|
| 1       |                               | 28          | 6/28/70       | D.G.R.           | Wound  | 225                    | Surg-S | 5/26/70         | 7/8/70            |
|         |                               | 29          | 6/13/70       | D.G.R.           | Wound  | 225                    | Surg-S |                |                   |
|         |                               | 34          | 6/21/70       | H.N.S.           | Sputum  | 225                    | Urol-S | 6/16/70         | 6/30/70           |
| 2       |                               | 121         | 11/30/70      | H.H.             | Sputum  | 334                    | Med-S  | 11/22/70        | 12/5/70           |
|         |                               | 128         | 12/19/70      | W.E.H.           | Sputum  | 334                    | Neuros-S | 12/3/70         | 12/18/70          |
| 3       |                               | 83          | 11/5/70       | E.C.P.           | Sputum  | ICU*                   | Surg-P | 10/17/70        | 11/24/70*         |
|         |                               | 94          | 11/12/70      | E.C.P.           | Sputum  | ICU*                   | Surg-P |                |                   |
|         |                               | 96          | 11/11/70      | E.C.P.           | Sputum  | ICU*                   | Surg-P |                |                   |
|         |                               | 97          | 11/14/70      | E.C.P.           | Sputum  | ICU*                   | Surg-P |                |                   |
|         |                               | 111         | 11/16/70      | A.L.B.           | Sputum  | ICU*                   | Surg-P | 11/5/70         | 11/28/70          |
|         |                               | 114         | 11/25/70      | A.L.B.           | Sputum  | ICU*                   | Med-P  |                |                   |
|         |                               | 115         | 11/27/70      | A.L.B.           | Sputum  | ICU*                   | Med-P  |                |                   |
| 4       |                               | 37          | 4/9/71        | B.E.Q.           | Sputum  | ICU*                   | Surg-P | 4/1/71          | 4/14/71*          |
|         |                               |             | 4/15/71       | S.C.G.           | Throat swab | ICU*                   | Surg-P | 4/11/71        | 4/16/71*          |
|         |                               |             | 4/24/71       | N.J.S.           | Right lung | ICU*                   | Surg-P | 3/29/71        | 4/24/71*          |

* Intensive care unit.

b Deceased.

c Transferred to another service.

TABLE 6. Human carriers of Serratia marcescens

| Patient (initials, age, sex, race) | First hospitalization | Second admission | S. marcescens isolate no. | Bacteriocin type | Date isolated | Underlying illness |
|----------------------------------|-----------------------|------------------|---------------------------|-----------------|---------------|-------------------|
| W.R.B., 50 years old, male, Caucasian | 10/5/70–10/17/70 | 11/20/70–11/30/70 | 51 17 | 10/5/70 | Chronic bronchitis with obstructive pulmonary emphysema |
| J.E., 35 years old, male, Caucasian | 12/21/70–12/26/70 | 2/24/71–3/1/71 | 131 29 | 12/26/70 | Chronic bronchitis, bronchial asthma |
| C.T., 81 years old, female, Caucasian | 2/9/71–3/14/71 | 5/4/71 | 163 18 | 3/7/71 | Poorly diff. granulocytic leukemia |

* In each case, S. marcescens cultures were taken from the patient's sputum. In no case was infection found to be significant.

only one other isolate of this type had been detected previously. The carrier denied a history of recent or chronic respiratory illness. Among the 51 patients with insignificant infection, there were three patients who had the organism in sputum cultures upon readmission to the hospital. The isolates belonged to the same respective bacteriocin group as did the isolates that had been cultured previously (Table 6). Seventy per cent of the patients of both groups had received respiratory assistance. However, all respirators were free of bacteria after decontamination, despite the fact that several of these that had been employed for patients with significant infection had become grossly contaminated with the organism during use.

Environmental sampling yielded one isolate of S. marcescens (bacteriocin type 9) from a
drain beneath the examination table of the urology cystoscopy room on 9 March 1971. A patient with known urinary tract infection attributable to S. marcescens bacteriocin group 9 had been identified 5 months previously.

**DISCUSSION**

Typing *S. marcescens* by bacteriocin sensitivity is simple and reliable. Over 85% of the isolates were typable, a number similar to that obtained by serotyping (22). We found that certain bacteriocin types of *S. marcescens* were more prevalent than others: strains of bacteriocin types 1, 4, 9, 14, and 18. This finding is similar to the observation of Wilfert et al. (22) who found that four serotypes of *S. marcescens* were prevalent at Boston City Hospital. It would be of interest to determine whether isolates of a particular bacteriocin type are of identical serotype. If not, then simultaneous bacteriocin and serotyping would result in greater epidemiologic sensitivity.

The incidence of cross-infection attributable to *S. marcescens* appeared to be low. There were four chronologically separate instances of cross-infection, but each was confined to a single room or unit; two of these episodes occurred in the intensive care unit.

*S. marcescens* colonized in half of the patients from whom the organism had been cultured (11). The majority of the patients with significant infection attributable to this organism probably carried *S. marcescens* in their upper respiratory tract or elsewhere, or both, at the time of hospital admission. Only through appropriate stresses, such as attendant antimicrobial drug therapy or respiratory assistance, was the organism capable of eliciting its pathogenic potential. We should like to refer to this as “iatrotechnical” (Gr. *iatros*, physician; Gr. *technē*, art) infection in contrast to “nosocomial” infection, a term that often is used synonymously with cross-infection. The majority of patients had received some form of respiratory assistance which implied an association between inhalation therapy and infection or colonization. However, the data obtained from bacteriocin typing ruled out this mode of acquisition, which was in agreement with the fact that the respiratory equipment was sterile. We recommend that future reports describing outbreaks of nosocomial infection be documented by specific typing of bacterial isolates (bacteriocin or bacteriophage typing or serotyping) to rule out potentially misleading epidemiological conclusions.

We do not know why the organism had an affinity for elderly, seriously ill males (23). This relationship might have been fortuitous, as our sample was small; furthermore, the male-female sex ratio was less than 2:1 in the case of those patients who were colonized by the organism. The prevalence of *S. marcescens* among elderly patients probably is a function of the higher incidence of predisposing illnesses in that age group (7). In addition, others have shown that the increased incidence of gram-negative rods in the pharynx of hospitalized patients correlated best with the clinical severity of the underlying illnesses (10). However, we are not aware of how and when the patients acquired *S. marcescens*. The organism is known to be ubiquitous in nature (8) and may gain access to, and subsequently colonize in, the human host in the community or in the hospital. There was only one pharyngeal carrier of *S. marcescens* among the seventy-two persons (1.4%) examined, which compared favorably with the incidence of *S. marcescens* in patients (1%). The three patients who originally had been colonized had strains of identical bacteriocin susceptibility upon readmission to our hospital 1 and 2 months later. This finding was evidence for the human carrier state of *S. marcescens*. Other workers found a low incidence of pharyngeal carriers of Enterobacteriaceae among hospital personnel (13); our data appear to support this observation because none of our intensive care workers had positive throat swabs. Admittedly, no effort was made to demonstrate hand contamination by *S. marcescens* as a possible mode of dissemination in any of the four minor incidents of cross-infection (15). We cannot explain why some patients, who had classically predisposing illnesses and in whom *S. marcescens* remained colonized, did not eventually come down with serious infection. We can only speculate that there are *S. marcescens* strains of low-grade virulence, as distinct from strictly saprophytic strains. Little work has been done with the organism in experimental animals (9, 17); consequently, very little is known about its host-parasite relationships.

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