Control of a Staphylococcal Outbreak in a Nursery

Use of Bacterial Interference

Iraun J. Light, MD; James M. Sutherland, MD; and Jean E. Schott, MS

An outbreak of pathogenic staphylococci was recognized in a newborn nursery and was controlled when conventional control measures failed. The phenomenon of bacterial interference was exploited to terminate the outbreak by controlling the spread of the pathogen. Bacterial interference was found to be a successful method of controlling the outbreak.

Material and Methods

Nursery Unit.—The nursery for full-term infants and the procedures used in the care of these infants have been described.² Infants weighing less than 2,200 gm (4.8 lb) at birth were housed in a separate unit in another building. Total floor space in this low-birth-weight unit is 1,100 sq ft. The unit consists of an admission room with 19 incubators, two graduation rooms—each with six bassinets—and an observation (isolation) unit with two beds. The average census in this unit is 20 infants and the capacity is 25 infants. Procedures are similar to those in the full-term nursery.

The outbreak in the nursery was characterized by the finding that two infants died each day during the first week of the outbreak. On the second day each week, bacteriologic survey for staphylococci was carried out by culturing specimens from the noses of all infants being discharged that day and of all infants more than 7 days of age. In the full-term nursery, no staphylococci of the bacteriophage type 80/61 were isolated in the first 16 weeks immediately preceding Jan 1, 1964. Furthermore, staphylococci of bacteriophage type 80/61 were cultured from only six infants in the 80 weeks preceding this time. The organisms were cultured from single infants during the 17th, 23rd, and 27th weeks. A second outbreak of three infants during the 24th week prior to this date.

During the period following Jan 1, 1964, staphylococci of the bacteriophage type 80/61 were isolated during 14 of 17 weeks. Organisms were isolated from one to five infants on these intervals. In 36 surveys, 36 such infants were identified. Nineteen infants or mothers with severe pyogenic lesions were noted during this 17-week period. One infant with pyrhythm of the hip, one infant with supplicative parotitis, and one infant with cellulitis of the arm. Many infants had purulent skin lesions during hospitalization or after hospital discharge.

Four unsuccessful attempts were made to control the outbreak. The nursery was closed to new admissions until infants had been discharged and the nursery emptied and cleaned. Meanwhile, new...
infants were admitted to a nursery opened in another part of the department. Specimens (from all infants) were cultured and all infants with suspicious pyogenic lesions were removed from the nursery.

The Outbreak in the Premature Nursery.—In the premature nursery no type 80/81 staphylococci were detected during a period of 21 weeks prior to Jan. 1, 1984. Thereafter, five infants with positive nasal cultures were detected in a three-week period, though no significant pathogenic lesions were noted.

The Inoculum.—The organism employed for artificial colonization was a coagulase-positive, penicillin-sensitive, relatively nonpathogenic Staphylococcus designated 502A.

The coagulase-positive Staphylococcus was

nated 502A A is a nonpenicillinase-producing organism. This strain is extremely sensitive to penicillin G. As tested by disc sensitivity methods it is sensitive to 2 units of penicillin, 2 mg of erythromycin, 2 mg of clindamycin, 5 mg of kanamycin, 10 mg of novobiocin, and 10% of tetracycline. It is lysed by some of the group III bacteriophages, usually one or more of bacteriophages 7, 47, 53, 54, and 77. Serologically the organism has been designated (C) B, but the strong C reaction is most useful in its typing.

An inoculum in tryptic soy broth (BBL 01-162) was grown for 18 hours and then diluted with broth (BBL 01-162) so that 0.01 cc contained approximately 5,000 organisms (a 1:200 dilution of the 18-hour broth). The diluted broth culture was then divided into 1.0 cc aliquots in separate test tubes and stored in the refrigerator. Broth cultures were prepared daily from a reference slant. The antibiotic disc sensitivity pattern was determined daily on the broth culture to assure identity of the inoculum.

Technique of Colonization.—Specimens from all infants in the premature nursery on the initial day of the study were cultured and then a single colonization was performed on each infant. All infants initially admitted to either the premature or to the full-term nursery after the beginning of the study were colonized within two hours of birth.

Initially colonization was carried out by utilizing an 0.25 cc syringe and administering a measured 0.01 cc to each of the external nares and to the base of the umbilicus. Colony counts on dilutions after colonization, and then at one-week intervals. Lactate and glucose cultures were obtained, for virtually all cultures obtained on admission and prior to inoculation were negative for staphylococci. Specimens from full-term infants were cultured on the day of discharge from the hospital, usually at 4 to 6 days of age. From infants remaining in the nursery seven days or longer, cultures were obtained at weekly intervals. Umbilical cultures were obtained from the base of the umbilical cord or from the skin of the umbilical site. Nasal cultures were obtained by inserting a dry cotton swab into one nostril. Swabs streaked directly onto mannitol salt agar (BBL 01-319) were incubated for 48 hours, and examined for colonial morphology, pigmentation, and mannitol fermentation. All staphylococci were tested for coagulase production by a standard slide test with rabbit plasma (BBL 73-036). A test tube coagulase test was performed if the result of the slide test was equivocal or if the slide test did not parallel mannitol fermentation. Antibiotic disc sensitivity patterns were performed on all mannitol-positive coagulase-positive strains of staphylococci (5 aureus) using the following discs: penicillin, 2 units; tetracycline, 5 mg; lincomycin, 5 mg; clindamycin, 5 mg; kanamycin, 5 mg; novobiocin, 5 mg. The 502A strain was consistently sensitive to all discs but the 5 mg of tetracycline. The latter part of the study bacteriophage typing was limited to coagulase-positive staphylococci not demonstrating the typical 502A sensitivity pattern.

Results

Full-term Nursery.—A total of 940 cultures were obtained from 470 artificially colonized full-term infants. (Table 1). A "take" was defined as the recovery of one or more colonies of 502A strain on cultures taken 24 hours or more after colonization.

There were 95% successful "takes" in the nose and 94% successful "takes" at the umbilical site.

A daily epidemiologic survey of all infants in the nursery was compiled from available data (Fig 1). If infants were discharged before one month of age it was assumed that the infant harbored from birth the organism which was isolated at discharge. If infants remained in the nursery for one month of age, it was assumed that the organism isolated from the weekly or the discharge culture had been present throughout the interval since the preceding culture. Following the onset of artificial colonization the 502A organism displaced from the nursery all other coagulase-positive staphylococci including those of bacteriophage type 80/81 from both the nose and cord. The 80/81 organism reappeared when colonization was discontinued. Artificial colonization was reinstated and the 502A strain again became the dominant Staphylococcus, displacing all other coagulase-positive staphylococci from the nursery. Colonization was discontinued after seven consecutive weeks. Thereafter, other coagulase-positive organisms reappeared as the 502A strain slowly disappeared. The 502A organism isolated in the postcolonization period were both artificially and spontaneously colonized infants. Infants who had been purposefully implanted with the 502A strain were still in the nursery. Two hundred and thirty-six full-term infants were admitted to the nursery in the 21 days after artificial colonization had been discontinued. Of these infants 159 (67%) spontaneously acquired the 502A organism.

Premature Nursery.—A total of 226 cultures for full-term infants were obtained from 114 premature infants at intervals of one to seven days after colonization (Table 1). There were 74% successful "takes" in the nose and 69% at the umbilical site.

The results of cultures are graphically represented on a weekly basis (Fig 2). In the nose, coagulase-positive staphylococci including bacteriophage type 80/81 were displaced by the 502A strain, though other coagulase-positive staphylococci persisted in 5% to 20% of infants. Following cessation of artificial colonization 502A slowly disappeared from the nursery. Only 80% to 85% of the infants car...
The present study demonstrated the lack of pathogenicity of the 502A strain in premature infants as well as in full-term infants. In addition, in the premature group a larger number of 502A staphylococci were used for colonization than had been used in previous infant studies. Despite the inadequate understanding of the phenomenon of bacterial interference several factors were shown to influence the colonization of newborn infants with staphylococci.

Preexisting Flora.—Preexisting staphylococci prevent the subsequent acquisition of other strains of coagulase-positive staphylococci. In the present study infants already colonized with S. aureus did not easily "take" or acquire the 502A Staphylococcus. This finding is similar to the observations of Shinsfield and associates. When these workers first isolated the 502A strain, they noted that infants who spontaneously acquired this organism did not later acquire another Staphylococcus. In their first inoculation studies they found that infants already colonized with S. aureus would not accept the 502A inoculum. Similar findings were made in adults subjects and were extended by challenging the 502A carriers with bacteriophage type 89/81 organisms. The infant is an ideal candidate for artificial colonization since he may be considered bacteriologically sterile at birth. This is in contrast to the adult subject where pretreatment with antimicrobial therapy before obtaining a successful inoculation.

Antimicrobial Drugs.—The acquisition of staphylococci by the newborn infant is influenced by the use of antimicrobial drugs. Among hospitalized adults the acquisition of drug-resistant staphylococci increased among patients receiving antimicrobial therapy. In the present study antibiotics were frequently, though not routinely, administered to premature infants. By administering these drugs and thereby eliminating the drug-sensitive nonpathogenic strains, and interfering with interspecies relationships, spontaneous colonization with drug-resistant organisms became possible.

Site and Age Differences.—Persistence of all coagulase-positive staphylococci, including those of the 502 strain, differs in the nose and at the umbilical site. In the nose the incidence of coagulase-positive staphylococcal carriers remains constant from 1 to 12 weeks of age. On the umbilical stump a similar incidence of coagulase-positive staphylococcal carriers is initially observed. Thereafter, the coagulase-positive staphylococci steadily disappear from the umbilical site and are replaced by coagulase-negative organisms. This shows that changes occur at the umbilical site with aging and the changes are detrimental to the survival of coagulase-positive staphylococci. Perhaps these organisms require the warm, moist mucous membranes of the umbilical stump for survival. Then as the cord becomes mummified and the umbilical cord heals, the coagulase-positive staphylococci are replaced.

Therefore, most of the differences noted in the present study between full-term and premature infants...
In Vitro Susceptibility of Shigella to Sodium Sulfadiazine and to Eight Antibiotics

Kenneth C. Hall, M.D., and John D. Nelson, M.D.

Materials and Methods

Identification of Cultures—Three hundred and forty recently isolated Shigella strains obtained from patients with acute diarrhea were studied. Two hundred and twenty-two were isolated in Dal-
las during the seven-month period from July 1963 through January 1964. Seventy additional strains had been obtained during a previous 23-month period (January 1961 through June 1963).

All cultures were examined in this laboratory to ensure their purity and to confirm their identification. All strains acidified 1% glucose without the production of gas and did not blacken triple sugar iron agar. Urease was not produced and growth did not occur on Simmons citrate medium. Tests for the deamination of lysine and the production of phenylpyruvic acid were negative. All strains were nonmotile. Serologic grouping was performed by the slide agglutination method using Shigella polyvalent antisera.

S. flexneri strains comprised 78% of the cultures; S. sonnei, 19%; S. boydii, 2%; and S. dysenteriae, 1%. Cultures were maintained in paraffin-sealed nutrient agar slants at room temperature. Media.—The following media were used: Mueller-Hinton agar and Mueller-Hinton broth, 1% tryptone water, oxiid sensitivity test agar, and oxiid sensitivity test broth.

Sulfadiazine Testing—Aqueous injectable sodium sulfadiazine was used. According to the manufacturer, the concentration of 250 mg/ml is accurate. (Personal communication from A. T. Laboratories.) This was prepared with 0.1% sodium sulfate which does not affect growth of shigelle.

Serial twofold dilutions of sulfadiazine were prepared aseptically in sterile phosphate-buffered normal saline (pH 7.2 to 7.4). Dilutions were freshly prepared for each batch of media. From these dilutions were unsatisfactory because precipitation occurred on thawing.

Plate Dilution Method.—One milliliter of a given

kanamycin sulfate (Kanutes), neomycin (Mycri-
fradin), sodium pentolinic G, streptomycin
sulfate, and tetracycline hydrochloride (Achro-
mycin) were used.

300 more recently isolated Shigella strains were tested for susceptibility, to sodium sulfadiazine by tube and plate dilution techniques. With a low inoculum (100 organisms), 69%, of Shigella flexneri and 87% of S sonnei were sulfadiazine resistant. All strains were resistant to high inoculum testing (2 x 10^6 organisms).

Antibiotic susceptibility testing by the plate dilution method using high inocula demonstrated by the following percentages of resistance: potassium pentolinic G, 95%; streptomycin, 17%; tetracycline hydrochloride, 12%; chloramphenicol, 11%; ampicillin, 6%; sodium colistin sulfate, 2%; kanamycin sulfate, 1%; and neomycin sulfate, 1%. These studies indicate that sulfadiazine is no longer an appropriate drug for initiating treatment of shigelllosis. Ampicillin and three orally administered non-absorbable antibiotics (kanamycin, kanamycin and neomycin) are the most effective drugs against shigellosis in vitro.

Sulfadiazine has been the most widely used drug for the treatment of shigellosis since Hardy and co-workers established its effectiveness two decades ago. At the present time several standard medical textbooks recommend as the initial therapy for the treatment of shigellosis* while others favor a broad spectrum antibiotic such as tetracycline.**

Ref: In the late 1960s, the development of the De-