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# IV. The Louisiana Epidemic

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## Introduction

Definition of Terms: A glossary of terms used in the present publication is included in a previous paper.[1]

In the previous papers of this series, the results of two controlled investigations were reported, one carried out in a hospital nursery in Cincinnati[2] and the other in Atlanta.[3] Both studies demonstrated that the phenomenon of bacterial interference could be employed on a practical scale to prevent newborn colonization with hospital strains of staphylococci.

However, because of low disease rates encountered among control infants in the Cincinnati study and the occurrence of a high incidence of spontaneous cross-infection with *Staphylococcus aureus* strain 502A in the Atlanta nursery, additional observations seemed desirable. Neither one of these factors affected the validity or the interpretation of the data; nevertheless, it was felt that another series of observations under different conditions would be of value.

During March, April, and May of 1962, severe staphylococcal disease was noted

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among infants and postpartum mothers recently discharged from a medium-size hospital in Louisiana, where approximately 150 babies were delivered monthly. A preliminary epidemiologic and bacteriologic survey undertaken in late May revealed that at least six staphylococcal pneumonias, two septicemias, two breast abscesses, and one case of osteomyelitis occurred among babies born at this institution during the preceding three months. Precise information about the incidence of less severe staphylococcal disease was not available, because no records of the occurrence of such illnesses were kept. The incidence of breast abscesses among the mothers was high during the first 25 days in May. For example, eight breast abscesses were surgically drained and four additional maternal breast abscesses were observed and cultured during a two-day investigation.

Five strains of staphylococci obtained from lesions were phage typed during this preliminary survey: four were recovered from breast abscesses and one from the pleural fluid of a three-week-old infant with pneumonia. All five strains were found to belong to phage type 80/81, and antibiotograms suggested their biologic identity. Four nasal carriers of the same staphylococcus type were found among 21 nursery and obstetrical attendants and nurses cultured.

Since these preliminary data indicated that the incidence of disease was high, it was decided to proceed under rigidly controlled conditions with artificial colonization of infants.

## Methods and Materials

*Nursery and Newborn Admitting Procedures.*—The hospital under study maintains an active maternity service, serving primarily charity cases. The labor and delivery suites are separated from each other by a floor-to-ceiling partition. The nursery suite consists of four nurseries, separated by a common nurses' station (Fig 1). Two units (Rooms *A* and *B*), used for full-term babies, are nearly identical, measuring respectively 22×14 ft and 24 ×12 ft, while two smaller units (Rooms *C* and *D*), measuring 18×10 ft each, are used entirely for premature infants and isolation care. The infants present investigation. The daily average census during the course of the study was 24 infants.
in these latter two rooms were not utilized in the

After delivery, the newborn was placed on a sterile sheet in a bassinet. An ungowned nurse, who did not wash her hands prior to handling the newborn, instilled silver nitrate solution in both eyes, footprinted the infant, attached a name bracelet, and then carried the newborn to the nursery.

On admission to the nursery unit, the infant was given a complete bath using a hexachlorophene-containing detergent, weighed on a common scale, and then placed in an individual bassinet, containing the linen supply for his entire hospital stay. When entering the nursery area, personnel wore fresh caps, gowns, and masks and scrubbed their hands with a hexachlorophene-containing detergent for two minutes, with a shorter wash being employed between infants. The regular nursery personnel and a pediatric resident were the only individuals permitted to handle the newborns. Mothers were housed on adjacent wards and were discharged 24 hours after delivery, without having had any direct contact with their baby throughout its nursery stay which averaged four days. Infants delivered without the usual sterile precautions were assigned to the same units as "clean" infants but, in addition to the routine admission procedures, received intramuscularly 50,000 units of aqueous penicillin and

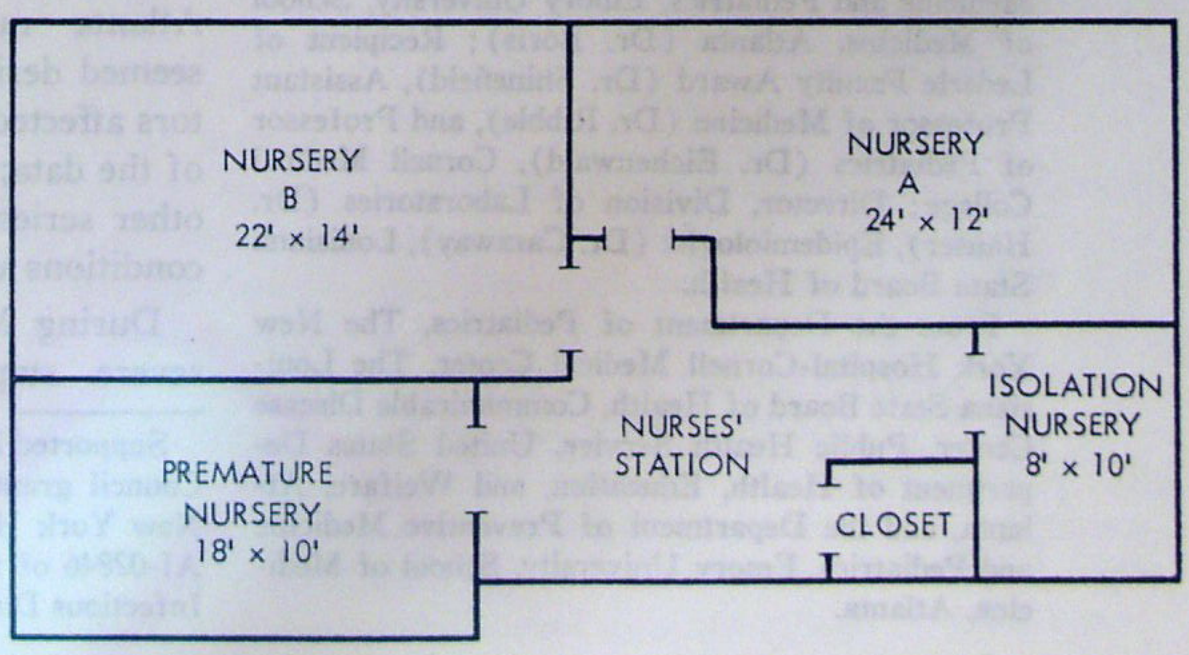


Fig 1.—A simplified floor plan of the nursery unit.

TABLE 1.—*Site and Number of Takes* in Infants Inoculated With Strain 502A*

| Site | No. Inoculated | Takes | % Takes |
|---|---|---|---|
| Nose | 25 | 21 | 84 |
| Umbilicus | 25 | 23 | 92 |

* Take indicates presence of marker 502A strain detected at 24 hours after inoculation.

25 mg/kg of streptomycin twice daily for three days.

The usual admission practice consisted of assigning newborns to one room until it was filled and then to the other; three days before our investigations were begun, newborns were admitted alternately to the two full-term nurseries, a procedure continued throughout the study. No other changes in nursery practice or personnel were made. The four members of the staff known to be carriers of *Staphylococcus aureus* type 80/81 continued to work and were not informed of their colonization status.

*Bacteriologic Methods.*—Cultures from the mother's nose and the infant's nose and umbilicus were obtained in the manner previously described.[3] Colonies for phage typing were selected at the Louisiana State Board of Health Laboratory in Alexandria and sent to the Epidemiology Branch Laboratory at the Communicable Disease Center, Atlanta, Ga. The criteria used for the selection of colonies for phage typing and testing by agglutination and antibiotogram have been described in detail previously.[3] During the present hospital and follow-up study, 5,744 cultures were obtained and a total of 12,932 colonies were typed.

*Artificial Colonization of Infants.*—The strain used for artificial colonization was the coagulase positive, penicillin-sensitive *Staphylococcus* strain 502A described in detail previously.[1,4]

*Techniques of Inoculation.*—Newborn infants were admitted alternately to nursery units *A* and *B*. Every infant admitted to Room *B* was inoculated on the nasal mucosa and umbilical stump with 2,000 to 4,000 organisms of strain 502A within two hours after birth, and the umbilical stump was reinoculated at 12 hours, using the technique previously employed. Babies admitted to Room *A* received the same amount of fluid delivered in an identical manner, except that sterile saline was employed as an inoculum. Twenty-nine infants were artificially colonized, while 36 served as controls. Four inoculated and three control infants born under nonsterile conditions received antimicrobial therapy and are not included in the analysis of the data.

*Community Follow-Up.*—Follow-up epidemiologic data and cultures were obtained by a public health nurse on 56 of the 58 infants included in this study. The seven infants who had received antimicrobial therapy were also followed. Home visits were conducted routinely at two-week intervals for the first eight weeks and then once each month. Follow-up cultures and epidemiologic data were collected in the manner previously described.[3]

## Results

*Incidence of Successful Artificial Colonization.*—Twenty-five infants were inoculated with strain 502A on the umbilicus and in the nose. "Takes" were obtained in the nose of 21 infants (84%) and on the umbilicus in 23 infants (92%) (Table 1). Two of the infants in whom nasal inoculation was unsuccessful were found to be infected with coagulase-positive staphylococci at the time when artificial colonization with strain 502A was attempted.

*Interference at the Nasal Site.*—Excluded from the analysis of the data was one infant who was colonized with both strain 502A and a nontypable staphylococcus within the first 24 hours of life, and the four "no take" infants. Of the 20 successfully inoculated newborns, one infant was colonized with another strain of coagulase-positive staphylococcus (type 80/81) during his hospital stay (Table

TABLE 2.—*Nasal Colonization with Coagulase-Positive Staphylococci Other Than 502A in Control and Successfully Inoculated Infants During Hospital Stay and Initial Follow-Up*

| Infants | Coagulase Positive 80/81 H* | 80/81 F* | Coagulase Positive Not 80/81 or 502A H* | Not 80/81 or 502A F* | Total H* | Total F* |
|---|---|---|---|---|---|---|
| Inoculated takes † | 1/20 ‡ | 1/20 | 0/20 ‡ | 1/20 | 1/20 ‡ | 2/20 |
| Control | 14/33 | 15/31 | 5/33 | 8/31 | 19/33 | 23/31 |

* Indicates infants in category/total number of infants; H indicates hospital stay; F, follow-up at 2 weeks.
† Take: presence of 502A strain detected at 24 hours after inoculation.
‡ One infant colonized with two strains on day 1 of life and four no-take infants excluded.

TABLE 3.—*Umbilical Colonization With Coagulase-Positive Staphylococci Other Than 502A in Control and Successfully Inoculated Infants During Hospital Stay and Initial Follow-Up*

| | Coagulase Positive 80/81 | | Coagulase Positive Not 80/81 or 502A | | Total | |
|---|---|---|---|---|---|---|
| Infants | H * | F * | H * | F * | H * | F * |
| Inoculated takes † | 3/20 ‡ | 1/20 | 0/20 ‡ | 0/20 | 3/20 ‡ | 1/20 |
| Control | 17/33 | 9/31 | 8/33 | 6/31 | 25/33 | 15/31 |

* Indicates infants in category/total number of infants; H indicates hospital stay; F, follow-up.
† Take: presence of 502A strain detected at 24 hours after inoculation.
‡ Three infants colonized with two strains on day 1 of life and two no-take infants excluded.

2). In the control group, the noses of 14 of 33 infants became colonized with *Staphylococcus* type 80/81, and an additional five control infants acquired coagulase-positive staphylococci other than strain 502A or type 80/81. Therefore, 19 of 33 infants in the control group acquired coagulase-positive staphylococci other than strain 502A. The difference between the inoculated and the control groups is significant at $P=0.001$.*

At the initial two-week follow-up visit, 2 of 20 infants in the successfully inoculated group were noted to be nasal carriers of coagulase-positive staphylococci other than the 502A strain. In the control group, 23 of 31 were now nasal carriers of staphylococci other than the 502A strain.

Among the four "no take" infants, no coagulase-positive staphylococci could be isolated from one baby throughout his hospital stay, one infant was found to harbor strain 502A on subsequent cultures, another was infected both with strain 502A and an organism typed by phages 7 and 77, and the fourth baby was colonized with a nontypable coagulase-positive staphylococcus.

*Interference at the Umbilical Site.*—Excluded from the analysis of the data were three infants colonized with two different staphylococcal strains within the first 24 hours of life, as well as two "no take" infants. Of the 20 infants successfully colonized on the umbilical site, three subsequently were found to have become infected with *Staphylococcus* type 80/81 (Table 3). Among the control group of 33 infants, 25 were colonized on the cord stump with coagulase-positive staphylococci other than strain 502A (17 were type 80/81). The difference in colonization rates with staphylococci other than strain 502A between the inoculated and the control groups is significant at $P=0.001$.

*The Occurrence of Staphylococcal Disease.*

After their discharge from the nursery, 56 study infants were followed by home visits.

* Statistical analyses were performed using $\chi^2$-test with Yates correction term. If numbers totaled less than 50 or contained a zero Fisher's exact test was used.

TABLE 4.—*Relationship of Type of Staphylococcus Carried in Infant's Nose to Development of Lesions in Infant and Household Contacts*

| Lesion | 80/81 | 80/81 & 502A | 502A | Other Coag. & Staph | Noncoag. & Staph |
|---|---|---|---|---|---|
| Infants | | | | | |
| Conjunctivitis | 2 | | | 1 | |
| Impetigo | 4 | | | 1 | 2 * |
| Breast abscess | | | 1 * | | |
| Family | | | | | |
| Maternal breast abscess | 2 | | 2 † | | |
| Maternal vaginal abscess | 1 | | | | |
| Sibling impetigo | 1 | | | | |
| Lesions/index family | 10/12 | 0/6 | 3/24 | 2/9 | 2/5 |

* Infant colonized with 80/81 on umbilicus and 80/81 cultured from lesion.
† Both mothers 80/81 nasal carriers prior to exposure to infant; 80/81 isolated from lesions. Both newborns colonized with only 502A.

During the five-month follow-up period, 17 lesions were noted to have occurred in these infants or their families. Fifteen lesions were present at the time of a home visit, and were therefore cultured. Among the 12 infants nasally colonized only with *Staphylococcus* type 80/81, disease was noted either in the index infant or in a family member on 10 occasions (Table 4). Lesions in the infants consisted of two cases of conjunctivitis and four of impetigo. Among the household members, two maternal breast abscesses occurred as well as one maternal vaginal abscess and one case of impetigo in a sibling. From all 10 lesions, *Staphylococcus aureus* type 80/81 was isolated.

A total of 24 infants carried only the 502A strain in their nose from the hospital into their home since, in addition to the 20 successfully inoculated infants, 3 control and 1 infant initially unsuccessfully inoculated were found to have become colonized with the 502A strain by the time of discharge. One infant nasally colonized with strain 502A developed a breast abscess; type 80/81 was isolated from a culture of this lesion. This infant was known to be an umbilical carrier of type 80/81. In addition, two breast abscesses occurred in mothers of infants who were colonized with strain 502A both in the nose and on the umbilicus. Nasal cultures from these mothers had been obtained while they were still in the hospital, and these had shown that both of them were nasal carriers of *Staphylococcus* type 80/81 before they had been in contact with their infants. Cultures from these abscesses taken at the time of incision and drainage yielded *Staphylococcus aureus* type 80/81 only.

TABLE 5.—*Strains of Staphylococci Isolated From Lesions in Infants and Household Contacts*

| Lesions | 80/81 | 502A | Other Coag. Positive | Not Cultured |
|---|---|---|---|---|
| Infants | | | | |
| Conjunctivitis | 2 | | 1 | |
| Impetigo | 5 | | | 2 |
| Breast abscess | 1 | | | |
| Family | | | | |
| Maternal breast abscess | 4 | | | |
| Maternal vaginal abscess | 1 | | | |
| Sibling impetigo | 1 | | | |
| Total | 14 | 0 | 1 | 2 |

TABLE 6.—*Lesions Observed in Total Test and Control Infants Irrespective of Presence or Absence of Strain 502A*

| Lesion | Inoculated Infants | Control Infants |
|---|---|---|
| Infants | | |
| Impetigo | | 7 |
| Conjunctivitis | | 3 |
| Breast abscess | 1 * | |
| Family | | |
| Maternal breast abscess | 2 † | 2 |
| Maternal vaginal abscess | | 1 |
| Sibling impetigo | | 1 |
| Lesions/index family | 3/25 | 14/31 |

* Infant colonized with 80/81 on umbilicus and 80/81 isolated from lesion.

† Both mothers 80/81 nasal carriers prior to exposure to infant; 80/81 isolated from lesion. Both newborns colonized with only 502A.

Two successfully inoculated and four control infants were found to carry both 502A and 80/81 at discharge. No lesions were uncovered in these six infants. Two of nine infants colonized with coagulase-positive staphylococci other than *Staphylococcus aureus* type 80/81 or strain 502A developed a mild illness; one infant had a conjunctivitis from which a nontypable coagulase-positive staphylococcus was isolated, and the other baby had a mild case of impetigo of short duration from which no cultures were obtained. Among the five infants whose noses were not nasally colonized with any coagulase-positive staphylococci at discharge, two cases of impetigo occurred. In one of these patients no culture was obtained, while type 80/81 was isolated from the lesion and the umbilical cord of the second case.

The differences in lesion rates between infants nasally colonized with strain 502A or type 80/81 are significant at $P=0.003$.

*Staphylococcus* type 80/81 was isolated from 14 of the 17 lesions, while 1 case of conjunctivitis was associated with a nontypable coagulase-positive organism (Table 5). The two remaining patients, both of whom had impetigo, were not cultured. No

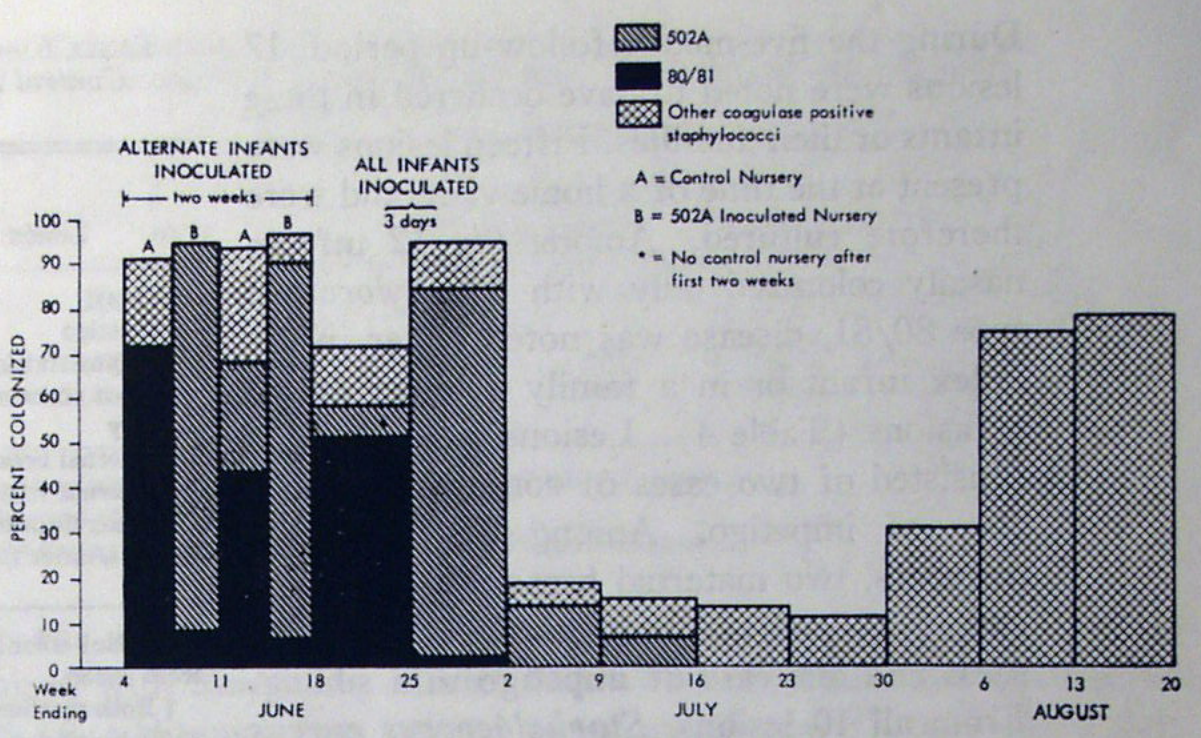


Fig 2.—Staphylococcal colonization in newborns at discharge.

lesions attributable to strain 502A were found.

The incidence of lesions in the entire artificially colonized group of infants irrespective of whether or not they had successful "takes" has been compared to the entire group of control infants, some of whom were spontaneously colonized with strain 502A. In the artificially colonized group, three lesions occurred: one breast abscess in an infant and two breast abscesses in mothers. From all three lesions a type 80/81 staphylococcus was isolated. Among 31 control infants, a total of 14 lesions occurred among the babies or their family contacts (Table 6).

*Nasal Colonization and Lesions in Infants Who Received Antimicrobial Therapy in the Nursery.*—Seven infants, born under "unsterile" conditions, received penicillin and streptomycin therapy during the course of the study. At the time of discharge, two of the four inoculated infants who had received antimicrobial therapy were found to be nasally colonized with type 80/81. The other two in whom artificial colonization had been attempted did not become infected with coagulase-positive staphylococci. Among the control infants receiving antimicrobial therapy, two babies were found to be colonized with type 80/81 and one baby carried no coagulase-positive staphylococci. Three episodes of staphylococcal disease, consisting of two breast abscesses in mothers and one case of impetigo in an infant, were observed in the households of two of the four infants colonized with type 80/81. *Staphylococcus aureus* type 80/81 was isolated from all three lesions.

*Cross Infection With Strain 502A.*—Despite the fact that the artificially colonized

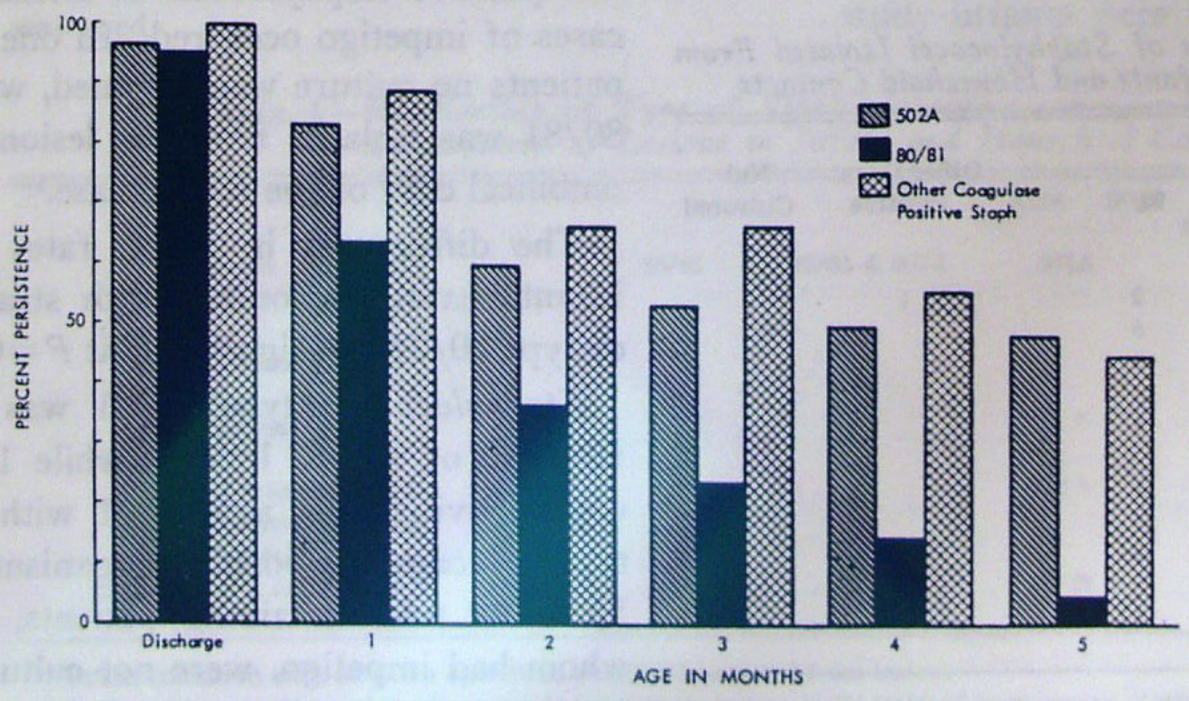


Fig 3.—Comparison of persistence of nasal colonization of the different groups of coagulase-positive staphylococci.

TABLE 7.—*Persistence* of Nasal Colonization of 61 Staphylococcal Strains in 55† Infants Followed From One to Five Months*

| Group | Two-Week Colonization Status | 1 | 2 | 3 | 4 | 5 |
|---|---|---|---|---|---|---|
| 502A Strain | 29/30 (97) | 25/30 (83) | 18/30 (60) | 16/30 (53) | 15/30 (50) | 14/29 (48) |
| 80/81 Strain | 21/22 (95) | 14/22 (64) | 8/22 (36) | 5/21 (24) | 3/21 (14) | 1/21 ( 5) |
| Other coagulase-positive staph | 9/9 (100) | 8/9 (89) | 6/9 (67) | 6/9 (67) | 5/9 (56) | 4/9 (44) |

* Number with persistence/number colonized initially with strain (%).
† Includes four infants colonized with 80/81 strain who received antibiotics during nursery stay because of "unclean" delivery.

infants were maintained in a nursery unit physically separate from that of the control infants, 7 of the 31 control babies (23%) became spontaneously colonized on the nasal mucosa with strain 502A.

*Surveillance of the Nursery Following the End of the Period of Artificial Colonization.* —Nasal and umbilical cultures were obtained three times weekly from all infants in the nursery for a period of ten weeks after the end of the study. Several days after artificial colonization of babies with strain 502A had been terminated, 50% of the infants were still found to be colonized with type 80/81 Fig 2). Because of this, all newborns delivered over a three-day period were artificially colonized with strain 502A at birth. Immediately following this procedure, *Staphylococcus* type 80/81 disappeared from the nursery and could not be recovered from infants at discharge. Interestingly enough, the colonization rate with all types of coagulase-positive staphylococci fell to a level of 18% at about this time and this low level persisted for a period of four weeks, even though cross colonization with strain 502A had virtually disappeared two weeks earlier, perhaps because no nursery personnel had become carriers of the 502A strain. Six weeks after artificial colonization of newborns was stopped, 80% of the infants were again colonized: this time with nontypable coagulase-positive staphylococci.

*Persistence of Nasal Colonization With Coagulase-Positive Staphylococci.*— Data were available on the rate of persistence of nasal colonization with coagulase-positive staphylococci in 55 newborn index infants who were followed from one to five months after discharge from the nursery. The 55 infants included the 4 babies who were nasally colonized with the 80/81 strain at discharge but who were excluded from the analysis of the interference data because they were born under "unsterile" conditions and arbitrarily received prophylac-

TABLE 8.—*Relationship of Prior Nasal Colonization With Staphylococcus Aureus Among 55 Mothers to Subsequent Acquisition of Staphylococci From Their Nasally Colonized Infants*

| Prior Maternal Nasal Colonization | Total No. at Risk | Acquired Nursery Strain | Did NOT Acquire Nursery Strain |
|---|---|---|---|
| Coagulase-positive staph isolated | 16 | 1 | 15 |
| No coagulase-positive staph isolated | 39 | 19 | 20 |
| Total | 55 * | 20 | 35 |

* Includes four infants colonized with 80/81 strain who received antibiotics during nursery stay because of "unclean" delivery.

TABLE 9.—*Relationship of Prior Nasal Colonization With Staphylococcus Aureus Among Sibling Contact to Acquisition of Staphylococci From 55* Nasally Colonized Index Babies*

| Prior Sibling Nasal Colonization | Total No. At Risk | Acquired Nursery Strain | Did NOT Acquire Nursery Strain |
|---|---|---|---|
| Coagulase-positive staph isolated | 65 | 1 | 64 |
| No coagulase-positive staph isolated | 100 | 24 | 76 |
| Total | 165 | 25 | 140 |

* Includes four infants colonized with 80/81 strain who received antibiotics during nursery stay because of "unclean" delivery.

tic penicillin and streptomycin during their nursery stay.

The rate of persistence of colonization with strain 502A gradually fell from 96% at discharge to 48% at the five-month follow-up period (Fig 3, Table 7). This relatively high persistence rate was in sharp contrast to the persistence in colonization noted with the 80/81 strain which fell from 95% at discharge to 36% by the time the baby was two months old. At five months of age only 1 of 21 infants (5%) initially colonized with 80/81 was still found to carry this strain. The persistence rate with other coagulase-positive staphylococci was of the same order as that observed with the 502A strain. The difference in persistence of strain 502A and strain 80/81 at five months of age is significant at $P=0.001$.

*Spread Within Households of the 502A and 80/81 Strains.*—Of the 55 mothers who were exposed at home to index infants carrying hospital strains of staphylococci, 20 subsequently acquired a nursery staphylococcal strain on their nasal mucosa (Table 8). All but one mother who acquired the nursery strain of staphylococcus had no coagulase-positive staphylococci isolated from her nasal mucosa prior to her exposure to an index infant. The difference in acquisition of a nursery strain of staphylococci as related to maternal nasal colonization status with a coagulase-positive staphylococcus is significant at $P<0.01$.

The same relationship was found among siblings of index infants. Of the 25 siblings who acquired a staphylococcus from an index baby, 24 did not have coagulase-positive staphylococci prior to their exposure to the index infant (Table 9). The difference in acquisition between siblings who were colonized with coagulase-positive staphylococci prior to acquiring a nursery strain and those who were not colonized with a coagulase-positive staphylococci prior to acquiring a nursery strain is significant at $P<0.005$.

## Comment

A highly significant degree of protection against *Staphylococcus aureus* phage type 80/81 as well as staphylococcal disease was obtained by the process of artificial colonization of infants' noses and cord stumps with strain 502A without any other changes being made in the nursery routine. The epidemic phage type 80/81 encountered in the present study produced lesions of greater severity than those observed in the previous epidemics where artificial colonization with strain 502A was undertaken.[2,3]

No host factors could be implicated which would explain the increased pathogenicity of this particular strain of phage type 80/81. It is of interest to note that of the 15 lesions cultured, 14 were associated with type 80/81 infection, while no disease could be attributed to strain 502A.

Spontaneous spread of strain 502A again occurred during this epidemic, despite the fact that artificially colonized and noninoculated control infants were physically separated. The degree of cross-colonization was similar to the spontaneous spread of strain 502A observed during nonepidemic conditions.[1]

It seems reasonable to assume that both artificial colonization and spontaneous cross-colonization with strain 502A contributed to the decrease in incidence of colonization of newborns with hospital strains of coagulase-positive staphylococci. The ability of strain 502A to survive and colonize newborns after artificial colonization is discontinued does not appear to be great. The organism rapidly disappeared from the nursery once it was no longer being continuously introduced.

## Summary

Artificial colonization of the nasal mucosa and the umbilicus of newborns with the staphylococcal strain 502A interfered with colonization at these sites by hospital staphylococci during a nursery epidemic caused by *Staphylococcus* type 80/81.

In the group of 56 infants infected by various staphylococci and followed after discharge from the hospital, 17 lesions were encountered in index infants or their household contacts. The staphylococci isolated from 14 were type 80/81. One strain was

nontypable, and two lesions were not cultured. No lesion could be attributed to the 502A strain.

Despite physical separation of the two nursery units under study, 23% of the infants in the control area became spontaneously infected with strain 502A.

Two weeks after artificial colonization of infants was stopped spontaneous colonization of infants with 502A could no longer be detected. The relationship between artificial colonization and the elimination of the 80/81 staphylococcal strain from the nursery is discussed.

At five months of age a striking difference was noted in rates of nasal persistence in infants colonized with various strains of staphylococcus; persistence rates of strain 502A and strain 80/81 were 48% and 5%, respectively.

Colonization of the nasal mucosa with coagulase-positive staphylococci of household members of index infants interfered with subsequent acquisition of staphylococcal strains carried into the household by newborn infants.

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