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Quantitative Nasal Cultures from Carriers of *Staphylococcus aureus*: Effects of Oral Therapy with Erythromycin, Rosamicin, and Placebo

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Serial quantitative nasal cultures were performed on 87 healthy nasal carriers of *Staphylococcus aureus*, who were randomly assigned to 7 days of oral therapy with erythromycin, rosamicin (an investigational macrolide antibiotic), or placebo. Staphylococcal carrier rates decreased during therapy with both antibiotics; however, erythromycin was significantly more effective in lowering carrier rates than was rosamicin. The anti-staphylococcal effects of both antibiotics were similar when the mean numbers of *S. aureus* isolated from positive cultures during therapy were compared. Side effects to each regimen were minimal.

The anterior nose is an important reservoir for *Staphylococcus aureus*, which produces clinical disease by dissemination to the skin of nasal carriers (10) or by transmission from person to person (1, 4, 14). The factors responsible for determining nasal carriage of staphylococci have not been defined. Carriers and noncarriers of nasal staphylococci have similar levels of serum and nasal antibodies against several staphylococcal antigens (3). Most studies involving nasal carriers have found that 15 to 40% of healthy individuals harbor staphylococci in the anterior nose, and this percentage increases in insulin-treated diabetics and narcotic abusers (9).

When nasal staphylococcal carriers are treated with an antibiotic to which the staphylococcal strain is susceptible (whether this is locally applied as an ointment or administered systemically), the carrier rates for staphylococci and the numbers of susceptible organisms isolated on quantitative cultures may be temporarily reduced (4, 5, 7, 11, 12, 15). Usually such suppression is temporary and persists only as long as local or systemic antibiotic therapy continues. The original staphylococcal strain or another strain (frequently resistant to the antibiotic employed) may colonize the nose during or after antibiotic therapy (4, 12). Quantitative nasal cultures for staphylococci have been used to evaluate the effects of new antibiotics, including semisynthetic penicillins (7, 13), macrolide antibiotics (15), and anti-staphylococcal enzymes (5, 6). The responses of nasal staphylococci cultured from stable carriers have correlated with in vivo responses of staphylococci to antibiotic therapy (4).

The purpose of the present study was to evaluate effects of oral therapy with erythromycin stearate, rosamicin, and a placebo on nasal *S. aureus*, as evaluated by serial quantitative nasal cultures. Antibiotic susceptibility tests and phage typing were performed on isolates obtained before and after therapy to determine whether resistance was induced by in vivo exposure to either of the antibiotics and to detect whether the original strain was eradicated after therapy.

MATERIALS AND METHODS

The volunteers studied were 95 healthy adults of both sexes, ranging in age from 18 to 43 years, who were randomly assigned to one of the three treatment regimens. These 95 subjects were identified from a larger group of 408 healthy adults who had been screened to determine the presence or absence of nasal *S. aureus*. Written informed consent was obtained according to the guidelines of the Institutional Review Board for Human Research of the Baylor College of Medicine.

Of the 95 nasal carriers, 1 erythromycin-treated subject was dropped because of a bacterial infection requiring alternate antibiotic therapy, and 6 additional subjects (2 each from the three groups) were dropped because one or more pretreatment cultures failed to yield coagulase-positive staphylococci. Also, one subject treated with erythromycin yielded an erythromycin-resistant *S. aureus* from all cultures and was deleted from the data analysis. The remaining 87 subjects comprised the three treatment groups; 30 received rosamicin, 27 received erythromycin, and 30 received placebo. Data from these 87 individuals are analyzed below.

The antibiotic regimens employed were as follows: 1.0 g of erythromycin stearate per day, 1.0 g of rosa-

micin per day, or placebo. All therapy was administered orally in four divided doses for 7 days. Rosamicin was supplied as capsules (a subsequently discontinued dosage form) by Schering Laboratories, Bloomfield, N.J. Erythromycin stearate was obtained from Abbott Laboratories, North Chicago, Ill., as a film-coated tablet containing 250 mg of erythromycin stearate. The placebo was identical in appearance to the rosamicin capsules.

Before enrollment in the study, a preliminary history and physical examination were performed, and all subjects were found to be healthy and free of significant acute or chronic medical problems. Blood and urine samples were obtained twice before and 1 day after completion of therapy to monitor clinical laboratory tests appropriate to detect possible adverse effects of the drug on hepatic, renal, or hematological function. Standard 12-lead electrocardiograms were obtained before therapy and on day 7 of therapy.

A diary of side effects and complaints was maintained by each subject during the 7 days of medication. A diary of drug administration was recorded in relation to meals and other activities during the 7 days of therapy. Compliance was good, with 95% of planned doses taken during the week of therapy.

Serial quantitative nasal cultures included two pretreatment cultures, two cultures during therapy, and three posttreatment cultures, which spanned the interval from 1 day to 4 weeks after completion of therapy. Compliance of the subjects was excellent, and 100% of the planned cultures were obtained. *S. aureus* nasal carriers were defined as subjects whose pretreatment nasal cultures consistently yielded pigmented, coagulase-positive staphylococci. Normal rabbit plasma was employed in the coagulase tube test.

The quantitative nasal cultures were performed by the method of White et al. (12) with the modification previously reported (15) by using dilutions of materials swabbed from the anterior part of the nose. Commercially prepared cotton applicators were moistened in sterile Trypticase soy broth from Baltimore Biological Laboratory, Cockeysville, Md., and used immediately to swab both nares by a circular motion. The swabs were placed into tubes (13 by 100 mm) containing 3 ml of sterile broth and shaken on a Kahn shaker for 5 min. Tenfold serial dilutions were made, and 0.1 ml each of undiluted inoculum and 1:10 and 1:100 dilutions of inoculum were dropped on the surface of Trypticase soy agar (Baltimore Biological Laboratory) and Vogel and Johnson agar (Baltimore Biological Laboratory) plates which had been dried at 37°C to remove surface moisture. The Trypticase soy agar plates were incubated overnight at 37°C and then held at room temperature to allow development of pigment. The Vogel and Johnson plates were incubated at 37°C and examined at 24 and 48 h for the appearance of characteristic black colonies.

To determine the effects of antibiotic therapy on the strain of *S. aureus* isolated, three colonies of coagulase-positive staphylococci were selected and stored from the immediate pretreatment cultures from each subject. From the posttreatment cultures of subjects who reacquired staphylococci or who continued to have staphylococci cultured from the nose three colonies were also obtained for in vitro testing. Pretreatment and posttreatment *S. aureus* isolates were tested for antibiotic susceptibility by the Kirby-Bauer disk susceptibility method. Strains were considered susceptible to erythromycin or to rosamicin if a zone diameter of 18 mm or greater was measured around the 15-μg disk (Baltimore Biological Laboratory) for either antibiotic.

Phage typing of pre- and posttherapy isolates was performed by the State Public Health Laboratory in Austin, Tex.

Statistical analyses were performed by using the CLINFO data analysis system and by the Schering-Plough Research Division. In calculating the mean log values of subjects, only cultures which were positive for *S. aureus* were considered.

RESULTS

The stability of the staphylococcal nasal carrier state for the subjects included in this study was confirmed by culture results in placebo-treated individuals. Of the 30 subjects treated with placebo, *S. aureus* was isolated from 232 of 240 cultures (96.7%). Of the antibiotic-treated subjects, the carrier rates showed the maximum reduction on cultures taken 1 day after completion of therapy (Table 1). Erythromycin was significantly more active than rosamicin in producing nasal cultures which no longer yielded *S. aureus* (only 25.9% of erythromycin-treated subjects were positive, whereas 56.7% of subjects treated with rosamicin had positive cultures ($P < 0.02$). Subjects from both treatment groups gradually reacquired nasal staphylococci, until around 75% of both erythromycin- and rosamicin-treated individuals were nasal carriers at the completion of the study on day 29 after therapy.

When phage types of staphylococci isolated before and after therapy were compared, four subjects from the erythromycin-treated group and three from the rosamicin-treated group had acquired new strains on cultures either 8 or 29 days after therapy was completed. Therefore, most individuals in both groups of antibiotic-treated carriers became colonized in the nose with the same strain they had carried before exposure to antibiotics. All pretreatment isolates were susceptible to rosamicin, and only one isolate (from a rosamicin-treated subject) was resistant to erythromycin. When the staphylococcal strains were retested for susceptibility to erythromycin and rosamicin, no change in antibiotic resistance was documented after therapy.

The effects of therapy can also be evaluated by comparing quantitative counts of staphylococci isolated from nasal cultures which remained positive (Table 2). Each subject had at least two quantitative nasal cultures before receiving medication. Similar numbers of staphylococci were cultured from the noses of subjects in each of the three treatment groups before

TABLE 1. *Positive nasal cultures for S. aureus*

Time of culture	Erythromycin			Rosamicin			Placebo	
	No. positive/No. tested	%	*P* value[a]	No. positive/No. tested	%	*P* value	No. positive/No. tested	%
Screening (not quantitative)	27/27	100	NS[b]	30/30	100	NS	30/30	100
Pretreatment day								
1	27/27	100	NS	30/30	100	NS	30/30	100
2	27/27	100	NS	30/30	100	NS	30/30	100
Treatment day								
2	23/27	85.2	NS	29/30	96.6	NS	29/30	96.6
6	9/27[c]	33.3	<0.005	22/30	73.3	NS	27/30	90
Posttreatment day								
1	7/27[c]	25.9	<0.005	17/30	56.7	<0.005	28/30	93.3
8	17/27	62.9	<0.005	23/30	76.7	<0.01	30/30	100
29	21/27	77.8	NS	23/30	76.7	NS	28/30	93.3

[a] *P* value was calculated by a Student's *t* test comparison between antibiotic-treated and placebo-treated groups.
[b] NS, Not significant (Student's *t* test value was >0.05).
[c] Value was lower for the erythromycin-treated group than for the rosamicin-treated group ($P < 0.02$).

TABLE 2. *Distribution by treatment group versus change in mean log counts of positive S. aureus nasal cultures*

Time of culture	Erythromycin (n = 27)			Rosamicin (n = 30)			Placebo (n = 30)	
	Mean	Difference[a]	*P* value[b]	Mean	Difference	*P* value	Mean	Difference
Pretreatment (mean of two cultures)	5.146	0	NS[c]	5.272	0	NS	5.238	0
Treatment day								
2	4.339	−0.807	<0.005	4.444	−0.828	<0.003	5.138	−0.1
6	3.201	−1.945	<0.001	3.742	−1.53	<0.001	5.1313	−0.107
Posttreatment day								
1	3.506	−1.64	<0.001	3.653	−1.62	<0.001	5.202	−0.031
8	4.342	−0.804	<0.005	4.256	−1.016	<0.003	5.135	−0.103
29	5.308	+0.162	NS	5.388	+0.116	NS	4.921	−0.317

[a] Difference was the mean pretreatment value minus the observed value.
[b] *P* value was calculated from Student's *t* test, comparing value for antibiotic-treated subjects with simultaneous values for placebo-treated subjects.
[c] NS, Not significant (Student's *t* test *P* value was >0.05).

therapy. After initiation of oral therapy and persisting through day 8 after therapy, significantly lower numbers of staphylococci were isolated from both antibiotic-treated groups than from the placebo-treated subjects. However, by the final culture day (day 29 after therapy), the numbers of staphylococci isolated from the noses of the antibiotic-treated subjects were again comparable to the numbers isolated from the subjects treated with placebo. No differences were noted in the numbers of staphylococci isolated from subjects treated with erythromycin or rosamicin during any of the treatment or posttreatment periods. Some fluctuations occurred in the quantities of staphylococci cultured from the placebo-treated individuals, with values slightly lower than base line being attained on all posttreatment cultures. The maximum mean decrease in the placebo-treated group was 0.3 of a log, which was not significantly lower than the mean pretreatment value. Consequently, in evaluating the quantitative effects of therapy on nasal staphylococci, erythromycin was no more effective than rosamicin, although both antibiotics produced significant decreases in the nasal staphylococcal counts.

There were no laboratory abnormalities attributable to therapy with either antibiotic, and electrocardiograms done on day 7 of therapy exhibited no changes from pretreatment.

No course of therapy was interrupted because of side effects or toxicities. Analysis of symptom diaries showed that there was a tendency toward more gastrointestinal complaints in the antibiotic-treated subjects. No single gastrointestinal side effect was encountered more frequently in the erythromycin- or rosamicin-treated subjects than in the placebo-treated controls. However,

when all of the gastrointestinal complaints were considered together (stomach pain, nausea, loose stools, diarrhea) more rosamicin-treated subjects (13/30, $P < 0.05$) had one or more of these than did subjects treated with erythromycin (7/27) or placebo (5/30). In many instances, these complaints occurred early in therapy, were mild in degree, and ceased despite continued treatment.

DISCUSSION

Most subjects who become nasal *S. aureus* carriers harbor the same strain of staphylococcus in the nose for weeks or months (4, 5, 7, 11, 12, 15). Thus, the staphylococcal carrier is particularly suited as a subject for exploring the in vivo effectiveness of anti-staphylococcal therapy.

When an antimicrobial agent is administered to which the *S. aureus* strain carried in the nose shows in vitro resistance, there is little effect on the nasal cultures. In fact, the numbers of staphylococci may increase with the use of an antibiotic to which the organisms are resistant (4). One explanation for the increased numbers and colonization rates with resistant staphylococci may be the alteration in the normal nasal flora, which ordinarily compete with staphylococci (6).

In the present study, only staphylococcal strains which were susceptible to the therapy used were evaluated. Oral therapy with either of the macrolide antibiotics, erythromycin or rosamicin, was accompanied by a decrease in the percentage of positive cultures and in the numbers of staphylococci recovered on quantitative cultures during and immediately after therapy. At the same oral dose erythromycin was more effective than rosamicin in reducing carrier rates (Table 1), although the mean numbers of staphylococci recovered on quantitative cultures which remained positive were not different with the two antibiotics (Table 2). This comparatively favorable result in reducing carrier rates may be a consequence of greater potency of erythromycin, which is about twice as active as rosamicin when tested in vitro against staphylococcal isolates (2, 8), or of higher concentrations of erythromycin than rosamicin in the nasal secretions during therapy (although these were not measured in the present group of subjects).

As expected, carrier rates rose toward original levels in both antibiotic-treated groups once therapy was stopped. At the time of the final culture (almost 1 month after therapy was completed), only about 25% of the individuals in both treatment groups continued to have negative nasal cultures. Staphylococcal strains which colonized the nose after therapy remained uniformly susceptible to the antibiotics employed, indicating that 1 week of outpatient therapy with either of these antibiotics is unlikely to favor colonization of the nose with resistant organisms. However, when antibiotics are administered to hospitalized patients, the risk of acquiring colonization with multiple-drug-resistant strains is considerably enhanced (1, 4, 12). In healthy, ambulatory subjects, the fact that the original strain of staphylococcus (as determined by phage typing) is usually found in the nose after therapy is discontinued indicates that these individuals probably continue to harbor the organism (in small numbers in the nasopharynx or other sites), allowing the strain to become reestablished once selective pressure of the antibiotic is removed. Of the *S. aureus* carriers at the end of the study, 17 of 21 (81%) treated with erythromycin and 20 of 23 (87%) treated with rosamicin carried their original strain in the nose.

None of the volunteers had clinical staphylococcal infection during the period of therapy or follow-up. Consequently, from the present study, no statement can be made concerning the relative effectiveness of erythromycin versus rosamicin in the therapy of established staphylococcal infections.

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