MAJOR ARTICLE

# Predicting the *Staphylococcus aureus* Nasal Carrier State: Derivation and Validation of a "Culture Rule"

**Jan L. Nouwen,[1,2,3] Alewijn Ott,[1,3] Marjolein F. Q. Kluytmans-Vandenbergh,[1] Hélène A. M. Boelens,[1] Albert Hofman,[3] Alex van Belkum,[1] and Henri A. Verbrugh[1]**

Departments of [1]Medical Microbiology and Infectious Diseases and [2]Medicine, Infectious Diseases Section, and [3]Department of Epidemiology and Biostatistics, Erasmus Medical Center, Rotterdam, The Netherlands

***Background.*** To study determinants and risks of *Staphylococcus aureus* nasal carriage, adequate differentiation between the different *S. aureus* carrier states is obligatory. We set out to develop a "culture rule" capable of differentiating between persistent and intermittent or noncarriers that uses a minimum of nasal swab cultures.

***Methods.*** In 51 healthy volunteers (derivation cohort), 12 quantitative nasal cultures were performed to establish *S. aureus* nasal carriage states. Persons with 11 or 12 cultures positive for *S. aureus* were classified as persistent carriers, and those with negative results of all cultures were classified as noncarriers. All other persons were classified as intermittent carriers. By means of logistic regression and receiver operating characteristic (ROC) curves, a culture rule was derived. This culture rule was subsequently validated in 106 participants of an ongoing study in 3882 elderly persons, again with the use of 12 quantitative nasal cultures.

***Results.*** In both cohorts, the positive predictive value of 2 consecutive positive culture results for persistent carriage was 79%. The model best differentiating between persistent and intermittent or noncarriers used the number of positive culture results combined with the amount of *S. aureus* in these cultures. By using the outcome of 2 cultures, the areas under the ROC curves were 0.981 (95% confidence interval [CI], 0.949–1.0) for the derivation cohort and 0.936 (95% CI, 0.881–0.990) for the validation cohort.

***Conclusions.*** Combining qualitative and quantitative results of 2 nasal swab cultures accurately predicted the persistent *S. aureus* carriage state with a reliability of 93.6%. Thus, this culture rule can be used in studies of determinants and risks of *S. aureus* nasal carriage.

*Staphylococcus aureus* nasal carriage is a major risk factor for both community-acquired and nosocomial infections [1–7], and the anterior nares are the primary reservoir of *S. aureus* in humans [8–10]. Three *S. aureus* nasal carriage patterns can be discerned: persistent carriage, intermittent carriage, and noncarriage [11–22]. However, no consensus has been reached on how to exactly identify these different states, but most studies use findings from 10–12 weekly nasal swab cultures [23].

The number of colony-forming units (CFUs) of *S. aureus* isolated from the anterior nares are higher in persistent than in intermittent carriers [24, 25], resulting in more extensive dispersal of staphylococci in the environment [25] and in an increased risk of *S. aureus* infection [26–28]. Bacterial variability (i.e., the number of *S. aureus* genotypes isolated in repeated cultures from one individual) is lower for persistent than for intermittent carriers [15, 22, 29], indicating that the underlying mechanisms determining persistent and intermittent carriage differs. Adequate differentiation between persistent and intermittent carriage is thus relevant for epidemiological studies.

At present, a large study of *S. aureus* nasal carriage in a population aged ⩾60 years is being conducted at Erasmus Medical Center (Rotterdam, The Netherlands). The main objectives are to study determinants and risks of *S. aureus* nasal carriage. This is part of the Rotterdam Study, a population-based prospective study of chronic diseases in the elderly population. The Rotterdam Study started in 1990 with 7983 persons and has just finished its third phase, in which >4000 persons have been included. In this large survey, an efficient and reliable way

Received 16 October 2003; accepted 3 May 2004; electronically published 26 August 2004.

Presented in part: 9th International Society for Staphylococci and Staphylococcal Infections meeting, June 2000, Kolding, Denmark.

Reprints or correspondence: Dr. J. L. Nouwen, Erasmus MC, Dept. of Medical Microbiology and Infectious Diseases, Dr. Molewaterplein 40, 3015 GD Rotterdam, The Netherlands (j.l.nouwen@erasmusmc.nl).

**Clinical Infectious Diseases 2004; 39:806–11**



1058-4838/2004/3906-0009$15.00

to assess *S. aureus* nasal carriage was obligatory. It would be impossible to perform 10–12 weekly nasal swab cultures in all participants. Thus, we developed a "culture rule" to discriminate reliably between persistent carriage and noncarriage or intermittent carriage, with a minimum of nasal swab cultures.

Our main questions were as follows: (1) how many quantitative nasal swab cultures are needed to accurately predict persistent carriage in a cohort of healthy adult volunteers, and (2) does the derived culture rule correctly predict persistent carriage in the elderly cohort of the ongoing Rotterdam Study?

## PATIENTS AND METHODS

### Patient Cohorts and Microbiological Investigations

***Derivation cohort.*** In 1988, a cohort of healthy volunteers (staff members of the departments of medical microbiology and infectious diseases and virology at Erasmus Medical Center) was formed to investigate bacterial and human factors associated with *S. aureus* nasal carriage [23]. During the period of September 1995 through March 1996, a total of 51 volunteers agreed to participate in this study. Nasal swab cultures were performed weekly for 12 weeks. All nasal swab samples were obtained for culture by one study physician (M.F.Q.K.-V.), according to the protocol below.

***Validation cohort.*** On the basis of the results of the derivation cohort, 2 quantitative nasal swab cultures of samples obtained at 1-week intervals were performed in 3882 participants of the Rotterdam Study. While this study was ongoing, 106 participants entering the study during the period of October 1997 through April 1998 agreed to be included in the validation cohort. Persons with 2 positive or 2 negative nasal swab culture results were oversampled to estimate the predictive value of these cultures for persistent carriage and noncarriage, or for intermittent carriage. One trained research assistant visited the participants at home and performed 10 additional nasal swab cultures at 1-week intervals, according to protocol.

The study was approved by the Medical Ethics Review Committee of the Erasmus Medical Center, University Medical Center, Rotterdam. Informed consent was obtained from all participants.

### Definitions

*S. aureus* nasal carriage state was assessed by means of the results of nasal swab cultures 3–12, as follows: persistent carrier, 9 or 10 of 10 cultures were positive for *S. aureus;* noncarrier, no positive culture results; and intermittent carrier, all intermediate numbers of positive culture results.

### Microbiological Procedures

Nasal swab cultures were performed according to a standard operating procedure, as described elsewhere [23]. Nasal swabs specimens were obtained with sterile cotton-wool swabs (Transwab; Medical Wire and Equipment). Both the left and right anterior nares were swabbed by rubbing the swab 4 times around the inside of each nostril while applying an even pressure and rotating the swab without interruption. The swabs were immediately placed in Stuart transport medium and kept at 4°C until inoculation (within 24 h).

Swabs were then cultured quantitatively on phenol-red mannitol salt agar (PHMA) and in phenol red mannitol salt broth (PHMB). The flasks with transport media containing the nasal swab were vortexed for 15 s. The swab was then pressed firmly against the wall of the flask with a sterile pincette and cultured in 8 mL of PHMB. Subsequently, 500 $\mu$L of the remaining bacterial suspension was inoculated evenly onto a large PHMA culture plate (diameter, 14 cm). Another PHMA culture plate (diameter, 8.5 cm) was divided into 3 sectors, which were inoculated with 10 $\mu$L of the original bacterial suspension, 10 $\mu$L of a 1:10 diluted bacterial suspension, and 1 $\mu$L of the 1:10 diluted bacterial suspension, respectively. The PHMB was incubated at 37°C for 7 days; the PHMA culture plates were incubated at 37°C for 48 h and at room temperature for 5 days. Both were interpreted after 7 days of incubation. If, after 7 days, no *S. aureus* had grown on the PHMA but the PHMB demonstrated a yellow color, a PHMA culture plate (diameter, 8.5 cm) was inoculated with 10 $\mu$L of PHMB and incubated as before. Culture results were recorded as 0 (no *S. aureus*), 1 (*S. aureus* only on the PHMB culture plate), 2 (2–9 CFU), 3 (10–99 CFU), 4 (100–999 CFU), or 5 ($\geqslant$1000 CFU).

Identification of *S. aureus* was based on colony morphology on the PHMA culture. Suspected colonies were cultured overnight on Columbia blood agar plates (Becton-Dickinson). A catalase test and a latex agglutination test (Staphaurex Plus; Murex) were then performed. All *S. aureus* isolates were stored at −70°C in glycerol-containing liquid media.

### Statistical Analysis

Percentages and continuous data were compared using Fisher's exact test and the Mann-Whitney test, respectively. Logistic regression was performed, and receiver operating characteristic (ROC) curves were constructed for different tests and combinations of tests (number of positive cultures, $^{10}$log-transformed CFUs [$^{10}$log {CFU + 1}] and the geometric mean CFUs of $\geqslant$2 cultures [e.g., {3}/2]) to study their ability to discriminate between persistent carriage and noncarriage or intermittent carriage [30]. Culture results of the derivation cohort were added as independent covariates to a logistic regression model with our "gold standard" diagnosis of persistent carriage or not (derived from 10 consecutive cultures) as binary outcome variate.

The right side of the regression equation was [$\beta 0 + \beta 1 \times$ number of positive cultures $+ \beta 2 \times$ geometric mean of CFUs]. Fitting the model gave us $\beta 0$ to $\beta 2$. Then we calculated the odds of persistent carriage for all persons of the validation cohort by adding their respective culture outcomes in the formula: odds =

[e($\beta 0 + \beta 1 \times$ number of positive cultures $+ \beta 2 \times$ geometric mean of CFUs)]. Subsequently, the probability of persistent carriage was obtained by [odds/(1 + odds)]. We choose the midpoint between 0 and 1 as the cut point. Areas under the ROC curves (AUC) and the corresponding SE were estimated by a nonparametric method (2-sample Wilcoxon test) [31, 32]. Differences between AUCs of the different test combinations were compared by the method of Hanley and McNeil [33].

## RESULTS

Fifty-one persons were included in the derivation cohort (19 men [37%] and 32 women [63%]), with a mean age 29 years (range, 20–52 years). Twenty (39%) participants were classified as noncarriers, 16 (31%) were classified as intermittent carriers, and 15 (29%) were classified as persistent carriers (derivation cohort; table 1). Positive predictive values for persistent carriage, derived from regression models that included the results of cultures 1 and 2, ranged from 0.79 in a model containing the qualitative outcome only, to 0.88 in a model including both qualitative and quantitative results (figure 1*A*). The use of the results of only 1 culture (either 1 or 2) produced a positive predictive value of only 0.69.

The validation cohort consisted of a subset of 106 participants of the Rotterdam Study cohort (44 men [42%] and 62 women [58%]), with a mean age 73 years (range, 62–89 years). For the present study, persons with 1 positive and 1 negative culture result were less informative. Two positive culture results could either indicate persistent or intermittent carriage. Possibly, the number of CFUs of *S. aureus* cultured could differentiate between persistent and intermittent carriage. Persons with 2 negative culture results could help to assess the predictive value for true noncarriage. Therefore, after initial random inclusion of participants, we decided to oversample persons with 2 positive or 2 negative screening culture results. Fifty-seven participants (54%) were classified as noncarriers, 17 (16%) were classified as intermittent carriers, and 32 (30%) were classified as persistent carriers (validation cohort; table 1). In 1 participant, both screening culture results were negative, and the results of cultures 3–12 were all positive. The most probable explanation for this would be either sample handling mistakes or a laboratory error. Because exclusion of this person did not significantly alter the data, and because mistakes happen in real life, it was decided not to exclude this person's data from analysis. The positive predictive value derived from regression models that included the results of cultures 1 and 2 was 0.79 in a model containing the qualitative outcome only, as well as in a model including also the quantitative results (figure 1*B*). The use of the results of only 1 culture (either 1 or 2) produced a positive predictive value of 0.74.

The numbers of CFUs of *S. aureus* were significantly higher in the validation than in the derivation cohort (figure 2). The median geometric mean in intermittent and persistent carriers were 1.4 (range, 0.3–3.3) and 3.6 (range, 1.9–3.9) in the validation versus 1.0 (range, 0.3–2.0) and 1.8 (range, 0.9–3.2) in the derivation cohort ($P = .001$ and $P < .001$), respectively. Persistent carriers had significantly higher numbers of CFUs of *S. aureus* in their positive nasal swab cultures than did intermittent carriers (figure 2): 1.8 CFUs (range, 0.9–3.2 CFUs) versus 0.98 CFUs (range, 0.30–2.0 CFUs; $P = .001$) in the derivation cohort and 3.6 CFUs (range, 1.9–3.9 CFUs) versus 1.4 CFUs (range, 0.30–3.3 CFUs; $P < .001$) in the validation cohort (figure 2).

**Table 1. Classification of the *Staphylococcus aureus* nasal carrier state based on results of the first 2 cultures, compared with results of cultures 3–12, for derivation and validation cohorts.**

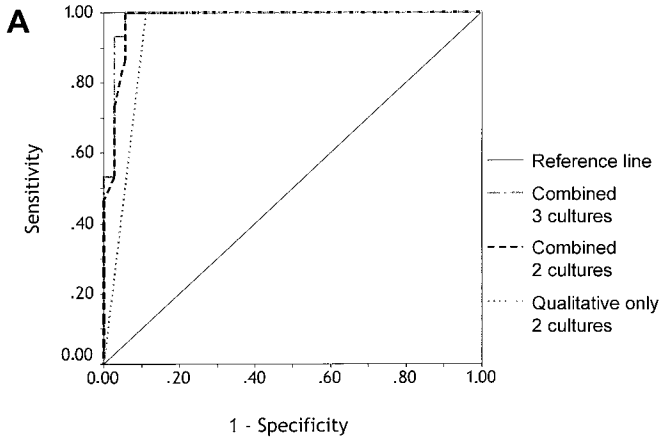
| Cohort | Results of cultures 1 and 2: Both negative | 1 Positive and 1 negative | Both positive | Total |
|---|---|---|---|---|
| Derivation cohort | | | | |
| Noncarrier | 19 | 1 | … | 20 |
| Intermittent carrier | 7 | 5 | 4 | 16 |
| Persistent carrier | … | … | 15 | 15 |
| Total | 26 | 6 | 19 | 51 |
| Validation cohort | | | | |
| Noncarrier | 53 | 4 | … | 57 |
| Intermittent carrier | 7 | 2 | 8 | 17 |
| Persistent carrier | 1 | … | 31 | 32 |
| Total | 61 | 6 | 39 | 106 |

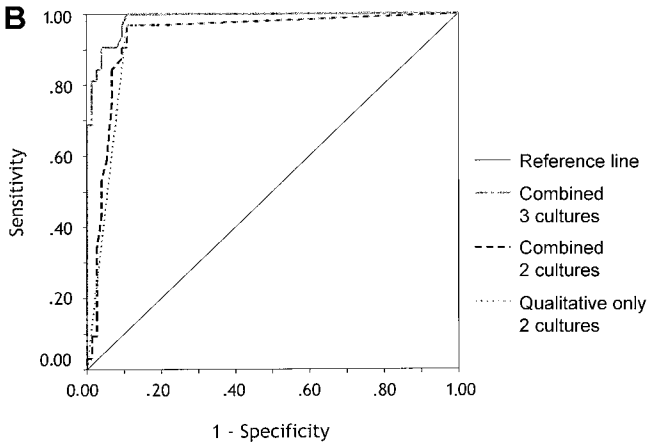
**NOTE.** Data are no. of subjects. *S. aureus* carrier status is based on results of cultures 3–12. For the validation cohort, persons for whom the results of both culture 1 and 2 were positive or negative were oversampled (see Patients and Methods). Therefore, the distribution of the different carrier states does not represent the population prevalence.

In the derivation cohort, logistic regression showed that the model best differentiating between persistent carriage and noncarriage or intermittent carriage used qualitative culture results in combination with quantitative data. The model that used the results of 2 cultures performed significantly better than a model that used the results of only 1 culture. Adding the results of a third or fourth culture did not significantly improve the model. Results from the ROC analysis showed that all tests used had good performance (all AUCs were >0.9), with the combined model being slightly—but not significantly—better than the qualitative result of 2 nasal swab cultures (figure 1*A*).

In the validation cohort, 2 qualitative culture results (positive or negative) discriminated similarly between persistent carriage and noncarriage or intermittent carriage as the combined qualitative and quantitative results. All logistic regression models were significantly improved by adding data on a third culture. However, in the ROC analysis, the differences between the models were small. Adding data on a third (but not a fourth) culture only significantly improved the model when both qualitative and quantitative culture results were used (figure 1*B*).

The AUCs that used the combination of qualitative culture




| Test | No. of cultures | Predictive values | | ROC analysis | |
|---|---|---|---|---|---|
| | | PV+ | PV- | AUC | 95% CI |
| No. of positive culture results | 1 | 0.69 | 0.95 | 0.903* | 0.810-0.968 |
| | ***2*** | ***0.79*** | ***1.0*** | ***0.944*** | ***0.881-1*** |
| | 3 | 0.88 | 1.0 | 0.972 | 0.927-1 |
| Geometric mean no. of CFUs | 2 | 0.85 | 0.89 | 0.973 | 0.936-1 |
| | 3 | 0.92 | 0.92 | 0.983 | 0.954-1 |
| Combined | ***2*** | ***0.88*** | ***1.0*** | ***0.981*** | ***0.949-1*** |
| | ***3*** | ***0.88*** | ***1.0*** | ***0.985*** | ***0.957-1*** |

| Test | No. of cultures | Predictive values | | ROC analysis | |
|---|---|---|---|---|---|
| | | PV+ | PV- | AUC | 95% CI |
| Qualitative culture results (no. of positive culture results) | 1 | 0.74 | 0.95 | 0.901 | 0.840-0.960 |
| | ***2*** | ***0.79*** | ***0.99*** | ***0.929*** | ***0.873-0.985*** |
| | 3 | 0.88 | 0.96 | 0.967 | 0.934-1 |
| Quantitative culture results (geometric mean no. of CFUs) | 2 | 0.83 | 0.91 | 0.936 | 0.881-0.990 |
| | 3 | 0.84 | 0.93 | 0.976 | 0.953-0.999 |
| Combined | ***2*** | ***0.79*** | ***0.99*** | ***0.936*** | ***0.881-0.990*** |
| | ***3*** | ***0.85*** | ***0.96*** | ***0.986**** | ***0.971-1*** |

**Figure 1.** *A* , Receiver operating characteristic (ROC) curve illustrating the predictive value of different tests for the persistent *Staphylococcus aureus* nasal carrier state in the derivation cohort. *Area under the ROC (AUC) of 2 versus 1 cultures ($P < .05$). *B,* ROC curve illustrating the predictive value of different tests for the persistent *S. aureus* nasal carrier state in the validation cohort. *AUC of 3 versus 2 cultures in the combined test ($P < .05$). PV+, positive predictive value; PV−, negative predictive value.

results and the geometric mean CFUs of 2 cultures were 0.981 (95% CI, 0.949–1) for the derivation cohort and 0.936 (95% CI, 0.881–0.990) for the validation cohort, respectively (figure 1). The logistic regression equation that uses the combination of qualitative culture results and the geometric mean of CFUs from 2 cultures could be written as follows: probability of persistent *S. aureus* nasal carriage = $e(\beta 0 + \beta 1 \times$ number of positive cultures $+ \beta 2 \times$ geometric mean of CFUs$)/1 + e(\beta 0 + \beta 1 \times$ number of positive cultures $+ \beta 2 \times$ geometric mean of CFUs). In the derivation cohort, the respective values of $\beta 0$, $\beta 1$, and $\beta 2$ were −20.171, 9.341, and 1.661. In the validation cohort these values were −4.572, 2.563, and 0.274, respectively.

When a cutoff of 0.50 was used, above which probability persons were classified as persistent carriers, it followed from the logistic regression equation from the derivation cohort that a person was a persistent carrier only if both cultures were positive with a geometric mean of ⩾0.9 (~8 CFUs per culture). This culture rule, when applied to the validation cohort, had a positive predictive value of 0.78, a negative predictive value of 0.96, and an AUC of the corresponding ROC curve of 0.936 (95% CI, 0.881–0.990).

## DISCUSSION

We examined the diagnostic value of 2 weekly quantitative nasal swab cultures to predict the *S. aureus* nasal carriage state and developed a culture rule to enable adequate differentiation between persistent carriage and intermittent carriage among those individuals with 2 positive screening culture results.

We used logistic regression and ROC analysis to derive a culture rule under ideal laboratory circumstances in a cohort of healthy adult volunteers. Strictly speaking, the derivation cohort actually was more of an exploratory data set to help select the variables in the model, but not the actual predictions. The culture rule was subsequently validated under real-life conditions in a subset of elderly participants of the Rotterdam Study.

In the derivation cohort, the best test combined qualitative culture results (number of positive culture results) with quantitative data (geometric mean number of CFUs of *S. aureus* in nasal swab cultures). In the validation cohort, however, the simple qualitative culture result when data on 2 cultures were used performed as well as the more complicated culture rule. The culture rule performed slightly less well in the validation

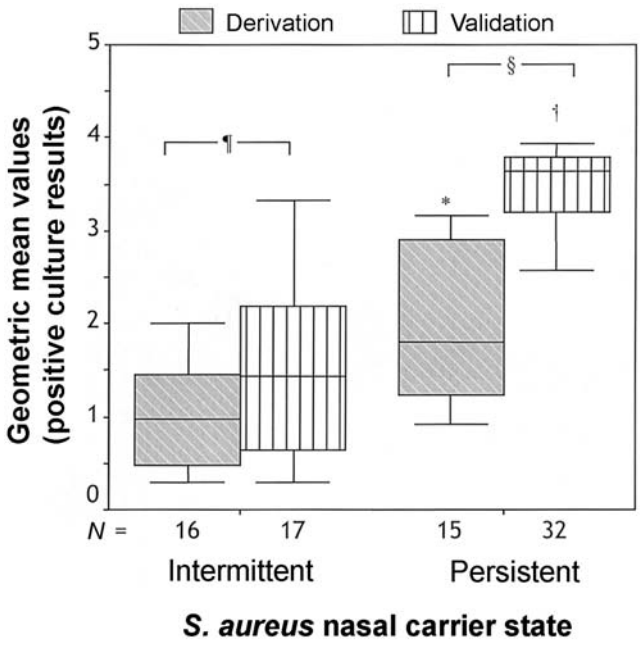



**Figure 2.** Geometric mean ($^{10}$log) number of colony-forming units (CFUs) of *Staphylococcus aureus* in positive cultures in intermittent versus persistent carriers from both cohorts. *Boxes,* median, quartile, and extreme values; *persistent versus intermittent carriers in derivation cohort ($P = .001$); †, persistent versus intermittent carriers in validation cohort ($P < .001$); ¶, intermittent carriers, derivation versus validation cohort ($P = .001$); §, persistent carriers, derivation versus validation cohort ($P < .001$).

cohort (AUC, 0.981 in the derivation and 0.936 in the validation cohort, respectively). In the ideal laboratory situation, one trained physician performed all nasal swab cultures in a cohort of healthy individuals. In the real-life situation of large-scale epidemiologic surveys, misclassification of the carrier state could have occurred for a variety of logistic reasons, such as differing nasal culturing techniques of study physicians, sample-handling mistakes, and laboratory errors. In theory, many of these "errors" are preventable but can never be totally eradicated. The fact that in the validation cohort the first 2 cultures were obtained at the Rotterdam Study research center by various study physicians, whereas cultures 3–12 in the validation cohort were performed by one trained person, certainly affected culture results: when cultures 3 and 4 of the validation cohort were used, instead of cultures 1 and 2, the AUC was increased from 0.936 to 0.996.

Misclassification of the carrier state could also have occurred because of factors associated with individual participants of the Rotterdam Study. Culture results will potentially have been influenced by the use of medication (recent courses of antibiotic therapy), institutionalization (recent hospital admissions), and underlying diseases, as well as other unknown determinants.

We confirm earlier data that showed that the number of CFUs of *S. aureus* in the anterior nares was higher in persistent carriers than in intermittent carriers [24, 25]. We also found a striking difference in the amount of *S. aureus* in the nose of persistent carriers between young, healthy volunteers and healthy, elderly participants. No previous data are available regarding age and the number of CFUs of *S. aureus* in the noses of persistent carriers. From the Rotterdam Study (3851 persons), the high numbers of CFUs (median geometric mean, 2.8) in elderly persistent carriers are confirmed (data not shown), but the underlying mechanisms of this finding remain to be elucidated. The differences in the number of CFUs in persistent carriers in both cohorts will have affected the performance of the derived culture rule in the validation cohort. When applying this culture rule to other patient populations, it will need to be validated in the specific population first, when possible.

Combining qualitative results with quantitative data is, in our opinion, conceptually the best choice. Incorporating quantitative data makes it possible to refine associations between potential determinants and *S. aureus* nasal carriage because not only carriers are compared with noncarriers, but carriers with low CFUs can also be compared with carriers with high CFUs in their anterior nares. Incorporating quantitative data will also make it possible to refine associations between carriage state and morbidity and mortality. However, in large-scale epidemiologic studies, simplicity will often prevail because of logistic reasons and resources. It is therefore reassuring that, in the validation cohort, the simple qualitative culture results performed as well as the more complicated culture rule.

Thus, 2 nasal swab culture of samples obtained at a 1-week interval can indeed provide sufficient information to adequately predict the *S. aureus* nasal carriage state. The use of only 1 nasal swab culture to predict the carriage state, as is often done, cannot be recommended on the basis of our data because it will lead to misclassification of the carriage state. On the other hand, the addition of a third or fourth quantitative nasal swab culture only minimally improved test performance. Of importance, no persons whose first 2 culture results were positive were found to be noncarriers. The finding of 2 negative screening culture results in 1 person with subsequent positive culture results is difficult to explain but may be attributable to sample handling mistakes or laboratory error. These results were included in the evaluation, however. One negative screening culture result virtually excludes persistent carriage. Predicting the noncarrier state from 2 nasal swab cultures is more difficult because $\geqslant$7 nasal swab cultures would be needed to distinguish intermittent carriers from noncarriers.

At present, data on determinants of persistent *S. aureus* nasal carriage in elderly patients in the Rotterdam Study are being analyzed by means of this culture rule. This is the first study to validate the potential of a limited number of nasal swab cultures in predicting the *S. aureus* carrier state. Because the incidence of *S. aureus* infections has increased substantially, and because of

the dramatic worldwide increase in antibiotic resistance (methicillin and, recently, even vancomycin resistance) in *S. aureus*, prevention is now more important than ever. Apart from its role in the Rotterdam Study, we hope that the presented culture rule will prove to be a helpful tool in identifying determinants of *S. aureus* nasal carriage and infections, as well as in identifying high-risk patient populations and the implementation of new methods in the prevention of *S. aureus* infections.

## References


1. Chow JW, Yu VL. *Staphylococcus aureus* nasal carriage in hemodialysis patients: its role in infection and approaches to prophylaxis. Arch Intern Med **1989**; 149:1258–62.
2. Corbella X, Dominguez MA, Ayats J, et al. *Staphylococcus aureus* nasal carriage as a marker for subsequent staphylococcal infections in intensive care unit patients. Eur J Clin Microbiol Infect Dis **1997**; 16:351–7.
3. Kluytmans JA, Mouton JW, Ijzerman EP, et al. Nasal carriage of *Staphylococcus aureus* as a major risk factor for wound infections after cardiac surgery. J Infect Dis **1995**; 171:216–9.
4. Luzar MA, Coles GA, Faller B, et al. *Staphylococcus aureus* nasal carriage and infection in patients on continuous ambulatory peritoneal dialysis. N Engl J Med **1990**; 322:505–9.
5. von Eiff C, Becker K, Machka K, Stammer H, Peters G. Nasal carriage as a source of *Staphylococcus aureus* bacteremia. N Engl J Med **2001**; 344:11–6.
6. Archer GL, Climo MW. *Staphylococcus aureus* bacteremia—consider the source. N Engl J Med **2001**; 344:55–6.
7. Martin GS, Mannino DM, Eaton S, Moss M. The epidemiology of sepsis in the United States from 1979 through 2000. N Engl J Med **2003**; 348:1546–54.
8. Casewell MW, Hill RL. Elimination of nasal carriage of *Staphylococcus aureus* with mupirocin ("pseudomonic acid")—a controlled trial. J Antimicrob Chemother **1986**; 17:365–72.
9. Moss B, Squire JR, Topley E. Nose and skin carriage of *Staphylococcus aureus* in patients receiving penicillin. Lancet **1948**; 251:320–5.
10. Williams REO. Skin and nose carriage of bacteriophage types of *Staphylococcus aureus.* J Pathol Bacteriol **1946**; 58:259–68.
11. Kluytmans J, van Belkum A, Verbrugh H. Nasal carriage of *Staphylococcus aureus:* epidemiology, underlying mechanisms, and associated risks. Clin Microbiol Rev **1997**; 10:505–20.
12. Williams REO. Healthy carriage of *Staphylococcus aureus :* its prevalence and importance. Bacteriol Rev **1963**; 26:56–71.
13. Armstrong-Esther CA. Carriage patterns of *Staphylococcus aureus* in a healthy non-hospital population of adults and children. Ann Hum Biol **1976**; 3:221–7.
14. Goslings WRO, Buchli K. Nasal carriage rate of antibiotic-resistant staphylococci. Arch Intern Med **1958**; 102:691–715.
15. Gould JC, McKillop EJ. The carriage of *Staphylococcus pyogenes* var *aureus* in the human nose. J Hyg (Lond) **1954**; 52:304–10.
16. Hoffler U, Bulanda M, Heczko PB, Pulverer G. A comparison of staphylococcal nasal carrier rates in Germany and Poland. Med Microbiol Immunol **1978**; 164:285–90.
17. Hu L, Umeda A, Kondo S, Amako K. Typing of *Staphylococcus aureus* colonising human nasal carriers by pulsed-field gel electrophoresis. J Med Microbiol **1995**; 42:127–32.
18. Lamikanra A, Olusanya OI. A long-term study of the nasal carriage of *Staphylococcus aureus* in healthy Nigerian students. Trans R Soc Trop Med Hyg **1988**; 82:500–2.
19. Maxwell JG, Ford CR, Peterson DE, Mitchell CR. Long-term study of nasal staphylococci among hospital personnel. Am J Surg **1969**; 118: 849–54.
20. Miles AA, Williams REO, Clayton-Cooper B. The carriage of *Staphylococcus (pyogenes) aureus* in man and its relation to wound infection. J Pathol Bacteriol **1944**; 56:513–24.
21. Miller DL, McDonald JC, Jevons MP, Williams REO. Staphylococcal disease and nasal carriage in the Royal Air Force. J Hyg Camb **1962**; 60:451–65.
22. Riewerts Eriksen NH, Espersen F, Thamdrup Rosdahl V, Jensen K. Carriage of *Staphylococcus aureus* among 104 healthy persons during a 19 month period. Epidemiol Infect **1995**; 115:51–60.
23. VandenBergh MF, Yzerman EP, van Belkum A, Boelens HA, Sijmons M, Verbrugh HA. Follow-up of *Staphylococcus aureus* nasal carriage after 8 years: redefining the persistent carrier state. J Clin Microbiol **1999**; 37:3133–40.
24. White A. Quantitative studies of nasal carriers of staphylococci among hospitalized patients. J Clin Invest **1961**; 40:23–30.
25. White A, Hemmerly T, Martin MP, Knight V. Studies on the origin of drug-resistant staphylococci in a mental hospital. Am J Med **1959**; 27: 26–39.
26. Bruun JN. Post-operative wound infection: predisposing factors and the effect of a reduction in dissemination of staphylococci. Acta Med Scand Suppl **1970**; 514:1–89.
27. Calia FM, Wolinsky E, Mortimer EA Jr, Abrams JS, Rammelkamp CH Jr. Importance of the carrier state as a source of *Staphylococcus aureus* in wound sepsis. J Hyg (Lond) **1969**; 67:49–57.
28. White A. Increased infection rates in heavy nasal carriers of coagulase-positive staphylococci. Antimicrob Agents Chemother **1963**; 161:667–70.
29. Van Belkum A, Riewerts Eriksen NH, Sijmons M, et al. Coagulase and protein A polymorphisms do not contribute to persistence of nasal colonisation by *Staphylococcus aureus.* J Med Microbiol **1997**; 46:222–32.
30. Metz CE. Basic principles of ROC analysis. Semin Nucl Med **1978**; 8: 283–98.
31. Hanley JA, McNeil BJ. The meaning and use of the area under a receiver operating characteristic (ROC) curve. Radiology **1982**; 143:29–36.
32. Bamber D. The area above the ordinal dominance graph and the area below the receiver operating graph. J Math Psychol **1975**; 12:387–415.
33. Hanley JA, McNeil BJ. A method of comparing the areas under receiver operating characteristic curves derived from the same cases. Radiology **1983**; 148:839–43.