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CHANGING PHARYNGEAL BACTERIAL FLORA OF HOSPITALIZED PATIENTS*

Emergence of Gram-Negative Bacilli

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Abstract The prevalence of gram-negative bacilli among the oropharyngeal bacterial flora was low in physiologically normal subjects despite hospital exposure but rose markedly in patients with illnesses of varying severity. This increased prevalence was not correlated with antibiotic administration or inhalation therapy, was not dependent on duration of hospitalization and correlated best with the clinical severity of illness. Increased exposure to these

organisms alone does not adequately explain these findings, suggesting that pharyngeal clearance mechanisms are impaired in these patients. Since most bacterial pneumonias begin with the aspiration into the lung of bacteria present in the upper respiratory tract, this alteration in the pharyngeal flora of ill patients may represent an important initial step in the pathogenesis of pneumonia due to gram-negative bacilli.

PNEUMONIA due to gram-negative bacilli represents a major threat to the hospitalized patient.^{1,2} Although a number of vehicles responsible for the spread of nosocomial infection have been described, other extensive studies have not revealed a gram-negative flora unique to the hospital or identified common gram-negative pathogens in the environment, suggesting that the infecting agents are endogenous to the patient.^{3,4}

Most bacterial pneumonias probably result from the entry into the lung of bacteria present in the upper respiratory tract. The mechanism of entry into the lung may be by aspiration of fluid from the pharynx, a phenomenon known to occur in healthy persons during sleep.⁵ Thus, the pharyngeal flora may be an important determinant of the etiology of pneumonia and, depending on the bacterial species, may in part determine the risk that pneumonia will develop. On the basis of this concept, the presence of gram-negative bacilli in the oropharynx might serve as a harbinger of pneumonia of this origin.

The present study was undertaken to ascertain the prevalence of gram-negative bacilli in the oropharynx of normal subjects and hospitalized patients with illnesses of varying severity.

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METHODS

Five groups of adult subjects were selected for study: nonhospital-associated normal subjects; hospital-associated normal subjects; physically normal hospitalized patients; moderately ill hospitalized patients; and moribund patients. The normal-nonhospital-associated subjects were 82 members of the Dallas Fire Department. Forty-seven physicians and hospital-ward personnel served as hospital-associated normal subjects. The physically normal, hospitalized group consisted of 20 patients on the psychiatry service. No normal subject had received antibiotics in the two weeks before the culture period. The moderately ill, hospitalized group included 81 patients on the orthopedic-surgery service. Their mean age was 39 years, and the mortality during the study and for two weeks thereafter was 1.2 per cent. Twenty-three critically ill patients on the medical service were selected because they appeared moribund. The mean age was 60 years, and 91 per cent died during the observation period.

Oropharyngeal cultures were obtained with an area-sampling device.⁶ Aliquots of the specimen were spread on eosin-methylene blue agar and trypticase soy agar containing 5 per cent sheep's blood. The plates were incubated for 48 hours, and representative colonies of the gram-negative bacilli isolated were identified by means of standard technics.⁷

RESULTS

The prevalence of gram-negative bacilli in pharyngeal cultures on single-culture surveys is shown in Table 1. No differences in prevalence were ob-

TABLE 1. *Results of Single-Culture Surveys.*

STUDY GROUP	NO. OF SUBJECTS	CULTURES CONTAINING GRAM-NEGATIVE BACILLI %
Normal subjects:		
Nonhospital associated	82	2
Hospital associated	47	2
Patients:		
Psychiatry service	20	0
Moderately ill	81	16
Moribund	23	57

served between the physically normal groups despite a gradation in exposure to the hospital environment from none, through intermittent, to the continual exposure of psychiatry patients. Moderately ill (orthopedic) and moribund patients had strikingly increased prevalence rates.

When repeated cultures were obtained, the cumulative percentage of subjects in all groups with at least one positive culture increased (Table 2). The wide discrepancy in percentage of positive cultures

TABLE 2. *Results of Multiple-Culture Surveys.*

STUDY GROUP	NO. OF SUBJECTS	NO. OF CULTURES	NO. OF CULTURES/ SUBJECT	CULTURES CONTAINING GRAM-NEGATIVE BACILLI %	SUBJECTS WITH 1 OR MORE CULTURES WITH GRAM-NEGATIVE BACILLI %
Normal subjects	33	139	4.2	3	6
Patients:					
Psychiatry service	18	88	4.9	2	6
Moderately ill	75	303	4.0	22	35
Moribund	11	27	2.5	63	73

obtained from physically normal subjects and patients with illnesses of varying severity was maintained. The overall percentage of positive cultures in each group remained essentially unchanged.

Exposure to the hospital environment, as measured by duration of hospitalization before the first culture, was associated with an increased prevalence of gram-negative bacilli. However, 19 per cent of moderately ill patients and 55 per cent of the

moribund patients had positive cultures within 96 hours of admission (Table 3).

The administration of antibiotics had no significant effect on the prevalence of gram-negative bacilli in the physically ill patients (p greater than 0.10) (Table 4). Similarly, patients receiving intermittent positive-pressure breathing treatments or continuous assisted ventilation showed no increase

TABLE 4. *Prevalence of Gram-Negative Bacilli in Relation to Antibiotic Therapy.**

STUDY GROUP	PATIENTS RECEIVING ANTIBIOTICS			PATIENTS NOT RECEIVING ANTIBIOTICS		
	NO.	NO. WITH GRAM-NEGATIVE BACILLI	% WITH GRAM-NEGATIVE BACILLI	NO.	NO. WITH GRAM-NEGATIVE BACILLI	% WITH GRAM-NEGATIVE BACILLI
Moderately ill patients	14	5	36	67	21	31
Moribund patients	10	8	80	13	8	62

*Patients who had received antibiotics within preceding 2 wk.

in prevalence over those not receiving such therapy (p greater than 0.10) (Table 5). The gram-negative bacilli isolated from the moderately ill and moribund patients are listed in Table 6.

The frequency of isolation of *serratia* species and *proteus* species from moribund patients was significantly higher.

DISCUSSION

Several lines of evidence suggest that bacteria present in the upper respiratory tract frequently enter the lung. In nonanesthetized rabbits, pneumococci instilled in the nose appeared in the lung within minutes.⁸ When tracheobronchial clearance is impaired in dogs by occlusion of a distal bronchus with a sterile cotton plug, pharyngeal organisms can be readily recovered distal to the occlusion.⁹ Bronchoscopic culture technics, with precautions to avoid contamination by the upper airways, have shown that secretions from the distal bronchi are apparently sterile in persons with normal lungs but contain organisms of the pharyngeal flora in patients with chronic bronchopulmonary disease.¹⁰⁻¹² These observations are also consistent with diminished clearance of small numbers of normal pharyngeal organisms that may have been aspirated.⁵ Furthermore, autopsy studies have shown that the bacterial

TABLE 3. *Prevalence of Gram-Negative Bacilli in Relation to Duration of Hospitalization.*

STUDY GROUP	HOSPITAL DAY OF 1ST CULTURE					TOTALS
	ADMISSION	1-3	4-7	8-21	>21	
Moderately ill patients:						
No. of subjects	0	26	11	29	15	81
No. with gram-negative bacilli	—	5 (19%)	2 (18%)	12 (41%)	7 (47%)	26 (35%)
Moribund patients:						
No. of subjects	3	8	5	5	2	23
No. with gram-negative bacilli	3 (100%)	3 (38%)	5 (100%)	4 (80%)	1 (50%)	16 (73%)

TABLE 5. *Prevalence of Gram-Negative Bacilli in Relation to Intermittent Positive-Pressure Breathing Therapy.*

STUDY GROUP	PATIENTS RECEIVING THERAPY			PATIENTS NOT RECEIVING THERAPY		
	NO.	NO. WITH GRAM-NEGATIVE BACILLI	% WITH GRAM-NEGATIVE BACILLI	NO.	NO. WITH GRAM-NEGATIVE BACILLI	% WITH GRAM-NEGATIVE BACILLI
Moderately ill patients	19	7	37	62	19	31
Moribund patients	18	11	61	5	5	100

flora of the lung reflects that of the pharynx.¹³⁻¹⁵ Whereas these findings have been attributed to the agonal aspiration of pharyngeal contents,¹⁶ they are also compatible with the terminal failure of clearance mechanisms. Thus, the pharyngeal bacterial flora is an important consideration in the pathogenesis of pneumonia, and the development of a pharyngeal flora due to gram-negative bacilli in hospitalized patients may be the initiating factor leading to nosocomial pneumonia caused by these organisms.

Previous studies have shown that gram-negative bacilli are infrequently found in pharyngeal cultures from normal subjects.^{17,18} Our data confirm these observations but indicate that the prevalence of gram-negative bacilli among ill patients is strikingly increased. This increased prevalence could be the result either of increased exposure to gram-negative bacilli or of a diminished capacity to clear such organisms from the pharynx.

Intense exposure of the respiratory tract may occur when contaminated aerosols are generated by inhalation-therapy equipment, and the role of such equipment in the pathogenesis of nosocomial pneumonia has been documented.¹⁹⁻²³ However, even under these circumstances, only a small percentage of persons exposed become colonized,²⁴ and necrotizing pneumonia due to gram-negative bacilli is infrequent.²⁵ Furthermore, massive exposure of normal subjects to these organisms does not result in colonization of the upper respiratory tract.²⁶⁻²⁸ We have subsequently found that during part of this study some of the aerosol medications being used

in the hospital were contaminated with *serratia* and that such medications might have been administered to these patients.²⁹ However, none of the medications were contaminated with strains of *proteus* species. Eighteen of the moribund patients, six of whom showed colonization with *serratia*, had received aerosol medications that may have been contaminated with *serratia*. Ten of the moderately ill patients had received the same medications without colonization. No potential environmental source of *proteus* or gram-negative bacilli other than *serratia* was established. Although environmental exposure to *serratia* may have resulted in colonization of some of the moribund patients, exogenous exposure to gram-negative bacilli does not seem to be an adequate explanation for these overall observations.

It seems reasonable to believe, therefore, that defective pharyngeal clearance mechanisms account for the prevalence of gram-negative bacilli in our patients. The implantation of transient bacteria may be inhibited by the normal pharyngeal flora.³⁰ Suppression of the normal bacterial flora by antibiotics may allow emergence of a gram-negative flora.³¹ The administration of antibiotics could not explain the prevalence of these organisms found in our hospitalized patients (Table 4); furthermore, since many patients demonstrated colonization on admission, the presence of antimicrobial agents in the environment was not a significant factor. In addition, Ritchie³² and Burn³³ each found gram-negative bacilli in the lungs of 60 per cent of patients coming to autopsy in the preantibiotic era.

Other factors, such as mechanical removal or the physicochemical or immunologic environment, may be important in the elimination of transient invaders from the oropharynx. On the basis of our data, we are unable to assess the importance of these individual factors. Of the factors examined, the appearance of a gram-negative bacillary flora in our patients correlated best with a clinical impression of the degree of illness. Transport mechanisms and membrane potential abnormalities have likewise been associated with a clinical assessment of severity of illness.^{34,35} Thus, the emergence of a gram-negative bacillary flora may be a readily demonstrable manifestation of the "sick cell."³⁴

Our data are compatible with the hypothesis that the pharyngeal clearance of gram-negative bacilli is impaired in ill patients. The susceptibility of these patients to pneumonia caused by gram-negative bacilli may be explained by the colonization of the pharynx alone or, more probably, by the added parallel impairment in pulmonary clearance as well.

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TABLE 6. *Frequency of Gram-Negative Bacilli Isolated from Oropharyngeal Cultures from Hospitalized Patients.*

BACTERIAL GENUS	STUDY GROUP			
	MODERATELY ILL PATIENTS		MORIBUND PATIENTS	
	no. of isolates	% of total	no. of isolates	% of total
<i>Klebsiella</i>	23	26	5	15
<i>Enterobacter</i>	22	25	4	12
<i>Escherichia</i>	19	22	2	6
<i>Proteus</i>	0	0	8	24*
<i>Serratia</i>	1	1	9	26*
<i>Pseudomonas</i>	15	17	4	12
Others	8	9	2	6
Totals	88		34	

*p < 0.001, Yates modification of chi-square analysis.

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