The subject of carpeting in hospitals is timely and controversial and the present report deals with measurement of microflora in hospital carpeting. This is the first of several papers which will deal with microflora in the air, and with colonization rates and infections in newborns.

A Study of Microflora on Tiled and Carpeted Surfaces in a Hospital Nursery

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Introduction

The installation of carpeting in hospitals has led to concern regarding possible effects on ambient microflora and on the incidence of hospital-acquired infections. The present study was undertaken to investigate the microflora on the floor and in the air of carpeted and uncarpeted (tiled) areas. On one hospital floor, one of two identical newborn nurseries was carpeted and the other was left tiled. The entire corridor outside the nurseries was also carpeted. Newborns were randomly assigned to the carpeted and uncarpeted nurseries. The present study was undertaken early in 1969 after completing a pilot study to refine techniques for obtaining and growing the bacterial cultures. The results of this study will be reported in three parts:

- Microflora on the carpeted and tiled floors.
- Microflora in air samples taken in the carpeted and tiled nurseries.
- Colonization rates and infections in newborns housed in the carpeted and tiled nurseries.

Shaffer and Key (1966) studied bacterial counts obtained from plugs of experimental wool carpet installed in a heavy-traffic area and maintained in the same manner as carpeting on the hospital floors. Nine months after the installation, carpet plugs were taken at monthly intervals for six months, and bacterial counts were done on the bottom third of the pile. The authors reported a twofold rise in colony count in the period of the study, and that vacuum cleaning had little or no effect. This was a more general study than the present one, which devotes itself specifically to newborn nurseries. Another study of microbial contamination of wool carpeting and acrilan carpeting was carried out by Anderson (1969). After installation of sterilized carpet microbial counts increased with time and reached a plateau in about four weeks. The carpeting was installed in a laboratory corridor and in pediatric hospital rooms. Plugs were taken at weekly intervals and the full depth of the pile was cultured.

The type of floor covering did not appear to appreciably influence the number of airborne bacteria at the test sites, and there were no significant increases in numbers of airborne bacteria after the installation of carpeting. However, the numbers of airborne bacteria were related to the number of patients and the degree of activity in the area. Seasons were found to have a slight influence on the numbers of airborne bacteria. There were no apparent increases in hospital acquired infections after the installation of the carpeting in patient care areas. Neither Anderson nor Shaffer (1966) found the other methods of carpet sampling (Rodac plates, seive sampler, and probe method) satisfactory. Anderson found no correlation between any of these methods and total microbial counts found with the plug-sampling technique. Walter and Stober (1968) reported strong correlation between probe and plug methods. They also found significant reduction in counts after vacuum cleaning.

Study Design

Carpeting was installed in one of two adjacent nurseries, identical but not connected. The corridor that ran the length of this floor outside the nurseries was also carpeted. As illustrated in Figure 1, each nursery had an adjacent service area, about half the size of the nursery proper. The nurseries were entered from the service areas, which were entered in turn from the corridor. The sampling scheme for taking floor cultures was based on a subdivision of the nurseries, service areas, and corridor sites as shown in Figure 1. These regions were further subdivided in a grid pattern, as a basis for the randomization of samples location. The sample size and period of observation were based on estimates derived from a pilot study of the area and of laboratory capabilities. Techniques for taking cultures from carpeted and tiled floors can not be made sufficiently comparable to allow comparison of colony counts in samples from the two surfaces. However, colony counts from floor samples were compared within the carpeted regions and within the tiled regions. Comparisons were also made of the colony count within each region before and after cleaning. In a subsequent report comparisons between nurseries will be presented with respect to colony counts in air.

Floor Sampling

Randomization

Each nursery was divided into four equal size (10′ x 10′) areas, and the adjacent service area into two comparable areas. Two additional areas were selected in the carpeted corridor, to represent heavy and light traffic areas. A grid pattern, with one foot spacing, was established within each of these sampling areas. A table of random numbers was utilized to determine in advance the coordinates of location of each sample taken within each area.

Tiled Area

In each of six tiled areas two samples were taken before cleaning and two samples after using Rodac plates. This yielded 24 samples per day from the six tiled areas, T_1 through T_6. (See Figure 1) Sampling was carried out on Monday, Tuesday, Thursday, and Friday of each week for a period of six weeks. The total number of samples from the area was 576.

"Rodac" plates were prepared with 18.5 ml. of Standard Methods Agar containing lecithin and Tween 80. The plates were applied to the specified floor locations for approximately two seconds. They were then incubated at 35° for 48 hours and the total number of colonies on each plate counted on a Quebec Colony Counter.

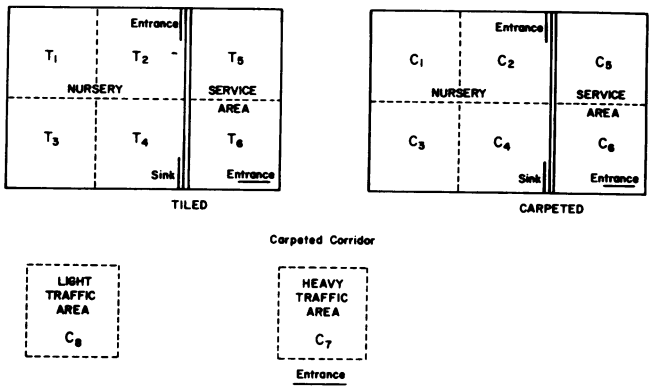
Carpeted Area

Each day samples were taken in two of the eight carpeted areas, C_1 through C_8. (See Figure 1) Thus, in the four sampling days of each week all eight areas were represented. Area-days were randomized. In the period of six weeks a total of 192 samples was obtained. Each sample consisted of five plugs of carpeting taken at each specified location. The plugs, obtained with a cork borer, were eight mm. in diameter and included the entire thickness of the pile (4-5 mm.) and backing (2-3 mm.). One plug was taken at the precise intersect of the grid lines. The remaining four plugs were taken at 3 inch distances from the initial plug, in north, south, east, and west directions. The five plugs were pooled and homogenized in 100 ml. of trypticase soy broth in an Omnimixer,* for two minutes at speed setting 6. An aliquot of 1 ml. of the homogenate was removed and 10-fold serial dilutions made with trypticase soy broth. One ml. of each dilution was inoculated on duplicate plates of Standard Methods Agar containing lecithin and Tween 80. The plates were incubated at 35° for 48 hours and the total number of colonies on each plate counted. The mean of the two plates was used to calculate the number of organisms per ml. of the initial homogenate.

Surface Cleaning Methods

Both the tiled and carpeted nurseries were cleaned each day during the 10 a.m. feeding period. The tiled floor was cleaned with a wet mop using the detergent Vesphene† (1/2 oz. per gallon). Once a month the floor was stripped of wax, rinsed with clean water, allowed to dry, and covered with a coat of "No-Slip" wax. A second coat was applied after the first coat dried.

Fig. 1 SAMPLING LOCATIONS FOR FLOOR STUDIES IN THE CARPETED AND TILED NURSERIES



The carpeted floor was cleaned with an American Cyclonic Vacuum cleaner (Model 380). The wand was passed over each area of the floor at least three times. Once a month the carpet was shampooed. The procedure included vacuuming followed by spray with ammonia water and application of shampoo (101 V.G. von Schrader, Racine, Wisconsin) at a concentration of 14 ounces per gallon. Prior to shampooing, the carpet was spot-cleaned with an acetone cleaner.

Walls and other surface areas of both nurseries were cleaned once a month with Vesphene at a concentration of one ounce per gallon.

In the present study, the floor samples were taken within 15-30 minutes before and within 15-30 minutes after the daily cleaning process.

Statistical Analysis

The floor study was carried out within the framework of a factorial analysis of variance. The variables of classification were location and cleaning. The dependent variable was colony count. The study consisted of six replications, each representing one week of sampling. It was thus possible to test for differences in location, cleaning, and replications, and for the interactions among these variables. When F ratios were significant, comparisons between areas were made by t-test for study hypotheses and by the Newman-Keuls procedure in all other cases.

Results

Table 1 presents the colony counts obtained from the floor cultures in the carpeted area. Table 2 presents an analysis of variance with respect to location and cleaning in the nursery service area, and corridor sites, in the period of the study. Data obtained from cultures of the carpet showed significant differences with respect to location. No significant differences or interactions were found with respect to cleaning. The service area showed significantly greater colony counts than the nursery proper, and the area of heavy traffic in the corridor showed significantly greater colony counts than each of the other carpeted areas. These data

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represent six replications, each comprising one week of sampling.

Tables 3 and 4 present similar information for the tiled area. Significant differences appeared with respect to location, cleaning, and the six replications. The service area showed greater colony counts than the nursery proper. The region of entry into the service area (T_6) showed greater colony counts than each of the other tiled areas. There were also significant differences with respect to the colony counts before and after cleaning, those before being greater. The entire sampling scheme for the area was replicated each week, for a period of six weeks. While there were significant differences in mean colony counts between replications in the tiled area, the carpeted area demonstrated no such variation. In the tiled area, the first and sixth week counts were highest.

It was not within the scope of the present study to identify all of the colonies that appeared in the carpet samples. However, colonies representing gram-positive cocci and gram-negative rods were noted. There appeared to be no consistency in the frequency of various types of colonies.

Discussion

In each nursery the pattern of relationship of traffic to contamination was similar. The service area was significantly more contaminated than the nurseries proper and within the service areas the sections closest to the corridor doors were most contaminated. However, within the carpeted nursery the area closest to the corridor door (C_6) did not show significantly higher colony counts than other areas. This may have been due, in part, to the relatively small sample taken in each carpeted area—24 specimens as against 96 in comparable tiled areas.

Within the nurseries proper, the areas about the sinks (C_4 and T_4) were the most contaminated, although these differences did not achieve statistical significance. It was the practice of the nurses to go to the sink and wash their hands each time they entered the nursery and immediately after handling each of the newborns. For this reason, these areas probably represent the heaviest traffic within the nurseries proper.

Of all carpeted areas sampled, a section of the carpeted corridor (C_7), representing the overall heaviest traffic, was the most contaminated. Colony counts from this section were two to five times higher than other carpeted areas. Unfortunately, there was no tiled corridor in which a similar comparison could be made.

The carpet was cleaned by the method suggested by the carpet manufacturer, but this procedure seemed to have no significant effect in reducing the number of bacteria

Table 1—Carpeted Nursery Colony Counts* by Location, Cleaning, and Replication

Nursery areas	N	Mean	Service areas	N	Mean	Corridor areas	N	Mean	Cleaning	N	Mean	Replications	N	Mean
C_1	24	4402	C_5	24	7481	C_7	24	19849	Before	96	7207	1st	32	9619
C_2	24	3702	C_6	24	8708	C_8	24	8353	After	96	8923	2nd	32	6686
C_3	24	4331	Combined		8095							3rd	32	6671
C_4	24	7697										4th	32	11317
Combined		5033										5th	32	6637
												6th	32	7462
												Combined		8071

* Colony counts per square inch of floor area (ca 13 plugs = 1 square inch)

Table 2—Carpeted Nursery Colony Counts: Analysis of Variance with Respect to Location, Cleaning, and Replication

Sources of variation		df	Variance estimate x10⁶	F ratio	P
Location	(L)	7	638.5	10.57	P<.001
Cleaning	(C)	1	141.3	2.34	N.S.
Replications	(R)	5	123.1	2.04	N.S.
Interactions					
L x C		7	28.8	0.48	N.S.
L x R		35	40.5	0.67	N.S.
C x R		5	99.2	1.64	N.S.
L x C x R		35	73.6	1.22	N.S.
Within		96	60.4		
Total		191			

Log transformations of colony counts produced similar results

Significant location differences:

Service Area > Nursery

c_7 > each of other C areas

Table 3—Tiled Nursery Colony Counts* by Location, Cleaning, and Replication

Nursery area	N	Mean	Service area	N	Mean	Cleaning	N	Mean	Replications	N	Mean
T_1	96	7.2	T_5	96	9.6	Before	288	10.1	1st	96	10.7
T_2	96	8.7	T_6	95	11.7	After	288	8.3	2nd	96	6.5
T_3	96	8.6	Combined		10.7				3rd	96	7.6
T_4	96	9.3							4th	96	9.3
Combined		8.5							5th	96	8.2
									6th	96	13.0
									Combined		9.2

* Per square inch of tiled floor area. This unit of area is used for convenient reporting of counts. (Not to be compared directly to carpet counts obtained from plugs of the full depth of pile and backing, and subjected to different microbiological techniques.)

Table 4—Tiled Nursery Colony Counts: Analysis of Variance with Respect to Location, Cleaning, and Replication

Source of variation		df	Variance estimate	F ratio	P
Location	(L)	5	212	4.57	$P < .001$
Cleaning	(C)	1	474	10.21	$P < .01$
Replications	(R)	5	532	11.47	$P < .001$
Interactions					
L x C		5	55	1.18	N.S.
L x R		25	54	1.17	N.S.
C x R		5	100	2.14	N.S.
L x C x R		25	44	0.95	N.S.
Within		504	46		
Total		575			

Significant location differences:
Service area > Nursery
T_6 > each of other T areas

Significant cleaning differences:
Before Cleaning > After Cleaning

recovered. In contrast, consistent with other studies, cleaning appeared to have a significant effect in decreasing contamination on the tiled floor.

The carpet cleaning phenomenon has been observed in several other studies and may be due to the lack of effective extraction of bacteria from the depth of the carpet pile by the vacuum cleaning process. Such reduction as may occur on vacuuming may be insufficient to demonstrate a change. On the other hand, the cleaning procedure may cause dispersion of aggregates of bacteria, producing a number of smaller particles. On recovery, the presence of these more numerous smaller units might mask an absolute decrease in the total number of bacteria. In any event, the mechanism governing the failure of cleaning to reduce bacterial counts in carpet has not been demonstrated.

While this investigation shows no detectable short-term effects of cleaning carpeted areas, it is not to be inferred that vacuuming and other methods of cleaning have no role in bacterial control. No test was made of the value of such cleaning in controlling the rate of bacterial build-up in carpeting over time.

It is not valid to compare directly the colony counts from tiled and carpeted areas, as methods for obtaining these counts are simply not comparable. The Rodac plates used to obtain samples from the tiled floor were applied to a flat surface area, while the carpet was sampled by the method of Anderson, in which plugs were removed with a cork borer. It seemed impractical to calculate the surface area of the fibers included in the carpet plugs. In any case, the surface qualities of carpet fibers and of tiled floor are quite different. There remained, however, the possibility of developing a sampling pattern that offered consistency and replicability. Valid comparisons could then be made within the carpeted area and within the tiled area, but not between the two areas. It was necessary to rely on studies of bacteria in the air of the two nurseries to demonstrate possible differences in the effects of carpeted and tiled floors. Reports of these studies are in preparation.

It is reasonable to assume that floor maintenance has some effect on the bacterial colony counts in the overall environment. It was demonstrated by Gable (1966) that the cleaning of tiled areas with detergent germicides has the effect of reducing bacterial counts. Comparable studies of the effects of germicides in carpet cleaning are not known to the authors. As stated above, the data obtained in the present study show no immediate effects of cleaning on bacterial counts in the carpet, when the cleaning was carried out in accordance with the manufacturer's directions.

Shaffer (1966), Shaffer and Key (1966, 1969) and Anderson tried a variety of sampling procedures. It ap-

peared that methods of obtaining samples, other than the plug technique, did not allow valid conclusions regarding colony counts in carpet. Anderson studied carpeting that was installed in a laboratory corridor and in patient rooms, while the present study addressed itself to sampling on a hospital floor that included two nurseries, one tiled and the other carpeted. The present study may not be comparable to that of Anderson, because of these differences as well as differences in the type of fiber, the carpet construction, and the depth of pile.

In addition, the culture medium used by Anderson, trypticase soy agar, was different from that used in the present study. Standard Methods Agar was employed as the culture medium, since preliminary tests indicated that it yielded more reproducible results.

Reporting colony counts per square inch of carpet does not take into account the surface area of the fibers per se. This refers only to the floor area covered by the carpet. It seems clear that changing the depth of the pile, or the density of loops per unit area, might significantly influence the bacterial count. Nevertheless, to establish uniformity of reporting, the authors decided to express the results in colony counts per square inch.

The present study design called for six complete replications, each representing one week of sampling. The differences reported herein appeared repeatedly throughout the replications giving evidence of the reliability of the findings. The design is applicable to the comparison of bacterial contamination in different areas of the same carpet, or to the comparison of different varieties of carpeting. Despite the differences between this study and the earlier studies discussed, the general conclusions with respect to the effects of traffic and cleaning appear to be similar.

It has been suggested that the specific methods employed in the maintenance of floor covering is a critical factor in the control of contamination. However, each type of floor covering has unique characteristics that determine the efficacy of available and feasible methods of cleaning. The present design may also serve in the comparison of different cleaning methods on the same floor covering, or of the same cleaning method on different types of floor covering.

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