THE EFFECT OF TOPICAL ANTIBACTERIAL AGENTS ON THE BACTERIAL FLORA OF THE AXILLA*

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Millions of adults apply bacteriostatic substances daily to their axillae in order to restrain the organisms which generate odor by the decomposition of apocrine sweat. The practical success in controlling axillary odor, following Shelley's basic observation (1), seems to have counteracted rather than stimulated further researches, as if odorlessness were a sufficient substitute for inquisitiveness. The near universal use of deodorants in this country has astonishingly not led to any further illumination of our knowledge of the bacteriology of the skin. Yet, there is every reason to believe that the axilla contains many bacteriologic treasures. It is a veritable haven for microorganisms, since it is moist, warm, protected, abundant in appendages and secretions, and utterly unique in its characteristic odor.

A crowd of questions arise in connection with the prolonged topical application of antibacterial substances: 1) Will resistant strains emerge? 2) Will there be an overgrowth of natively resistant bacteria or fungi? 3) Will a shift in the ecology of the native population of microorganisms affect the health of the skin, particularly as regards its vulnerability to infection? 4) Is it possible to keep the axilla permanently at near levels of sterility?

It is inevitable that deodorant suppliers still seek to improve the efficacy of their products by utilizing the most potent agents, notably antibiotics. This problem is already upon us with the over-the-counter availability of neomycin-containing deodorants. Already there are cries against this practice (2). We undertook this study to try to answer the questions which must arise when the axillary flora is artificially disturbed by different kinds of antibacterial agents.

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MATERIALS AND METHODS

1. Methods of application—The subjects were adult male inmates of Holmesburg Prison, mainly Negroes between the ages of 20–40 years. Only subjects with intense axillary odor were selected. The active agents were applied liberally once daily to one axilla, under supervision. The inert vehicle was applied to the opposite control axilla. No restrictions were imposed with regard to activity or washing. The preparations used were as follows:
   a. Neomycin, 0.175%, in an aqueous medium containing 15% alcohol, 3% glycerine, and 0.7% methylcellulose supplied in a roll-on container.
   b. The above preparation plus aluminum chlorhydroxide, 18%.
   c. Aluminum chlorhydroxide, 18%, in the same vehicle.
   d. Streptomycin, 1%, in the same vehicle.
   e. Penicillin G, 2%, in petrolatum.

   Odor was rated daily from 0–4+.
2. Bacterial sampling—The mechanical scrubbing apparatus devised by Pillsbury and Rebell was utilized for quantitative sampling (Fig. 1). The patient was placed in the supine position with his arm extended over his head and his shoulder supported by a pillow to flatten the axillary surface. One axilla was dry-shaved with a disposable razor blade just prior to sampling, which was done never less than eighteen hours after the last application. One cc. of sterile water was pipetted into a one-inch plastic cylinder placed in the axillary vault. A grooved rubber brush was mechanically rotated under a 20 gram weight within the cup for a period of one minute. This force is sufficient to produce a turbid suspension owing to disruption of the stratum corneum. The procedure is moderately discomforting and occasionally produces an erosion. The contents of the cup, 0.4–0.75 cc., were withdrawn and processed within the next few hours. For identification of bacterial species, blood agar plates were streaked with a loopful of the undiluted suspension and incubated at 37° C., aerobically and anaerobically. Staphylococci were tested for coagulase activity. Gram negative organisms were identified by transfer to appropriate media. The proportions of the various organisms were estimated from the original blood plates without actual counting. When Proteus species were significantly present, another plate, phenylethanol agar, was streaked with the refrigerated original sample to prevent spreading. For quantitative analysis, 0.1 cc. aliquots of the original suspension was diluted 100 and 1000-

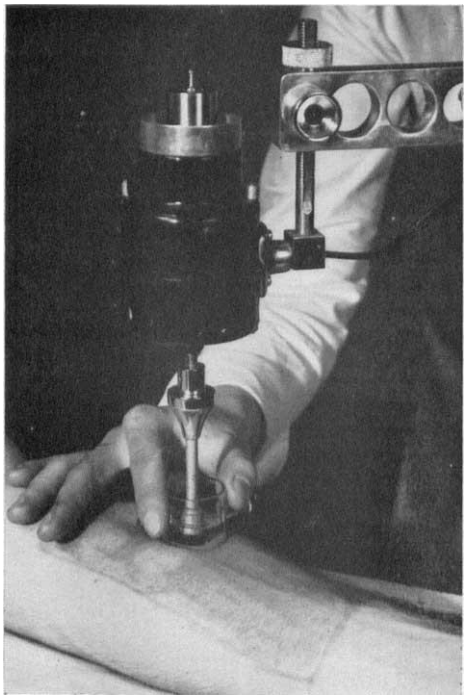

FIG. 1. Mechanical Scrubbing Apparatus

fold and duplicate plates poured. Colonies were counted at 48 hours with a Quebec counter.

3. Antibiotic sensitivities The agar plate method was used for convenience in handling large numbers of samples, although it is realized that the sensitivity of this method is far less than the more precise tube dilution method. The antibiotics were incorporated into blood agar plates and inoculated with a replicator using a moderately turbid suspension prepared from 24-hour-old colonies on blood agar. The various organisms were tested against single concentrations of antibiotics, respectively penicillin, 5U/cc.; streptomycin, 15μg/cc.; and neomycin, 30μg/cc. These levels are 2-3 times those necessary to inhibit the majority of gram positive strains isolated from the axilla. It is worth noting here that the concentrations of the topically applied antibiotics were enormously greater than those for sensitivity testing.

RESULTS

1. Normal Axillary Flora

Knowledge of the organisms which normally inhabit the axilla is very scanty owing to lack of systematic study. In a study of 20 subjects, Shelley *et al* (1) regularly isolated coagulase positive staphylococci, coagulase negative *staphylococci*, *corynebacteria*, *Aerobacter sp.*, and *Sarcina lutea*. From 29 subjects, Strauss and Kligman (3) found coagulase negative micrococci and diphtheroids to be predominant with a scattering of gram negatives in a few subjects. According to Borick and Sarra, (4) *Staph. aureus* and *Staph. albus* were the predominant microorganisms in their 11 patients. Also occasionally isolated were *E. coli*, *Aerobacter sp.*, *Proteus vulgaris*, *Streptococcus*, and diphtheroids.

TABLE I

Normal Axillary Flora

Classification	Organism	Incidence
Resident Flora		
Major Gram (+)	Coagulase (−) Staphylococcus	49/50 (98%)
	Diphtheroids	38/50 (76%)
Minor Gram (−)	Aerobacter sp.	22/50 (44%)
	Alkaligenes fecalis	16/50 (32%)
Transient Flora		
Gram (+)	Sarcina	7/50 (14%)
	Staphylococcus aureus	4/50 (8%)
Gram (−)	Proteus vulgaris	5/50 (10%)
	Escherichia coli	3/50 (6%)

We studied the axillary flora in 50 subjects and have arranged the results according to the scheme given in Table I. In classifying an organism as resident, we do not mean that it is found in every individual or even in the majority but rather that it is found consistently in a given subject, time after time in considerable numbers. When only a few colonies are present, and these inconsistently in a small percentage of subjects, the organism is considered a transient. By all odds, the most numerous organisms are the gram positive coagulase negative staphylococci and diphtheroids, the former being found in almost 100% of cases and the latter in more than 75%. These two are usually found in combination, with staphylococci predominating in about half the cases and diphtheroids in the other half. To a lesser degree the resident flora include the gram negative *Aerobacter sp.* and *Alkaligenes fecalis* in almost one-half to one-third of the individuals respectively. The transients comprise the gram positive *Sarcina lutea* and *Staphylococcus aureus*, and the gram negative *Proteus vulgaris* and *E. coli*; these occur in quite small numbers in 6–15% of individuals.

COMMENT

According to customary teaching, the resident cutaneous organisms are of three kinds: 1) coagulase negative staphylococci, 2) aerobic corynebacteria and 3) anaerobic *Corynebacterium* (*Propionibacterium*) *acnes*.

Following Evans' work (5), there was widespread acceptance of his finding that the anaerobic *Corynebacterium acnes* was 10–100 times more numerous than other resident species in practically all areas tested. He was the first routinely to use anaerobic methods of study. Evidently he called every Corynebacterium capable of growing under anaerobic conditions *C. acnes*. In many hundreds of cultures of the axilla, using a variety of specialized media, we have never isolated *C. acnes*, using the criterion of *strict* anaerobiasis. Indeed, we have found that the resident corynebacteria from all body areas are facultative anaerobes capable of growing both under anaerobic and under aerobic conditions. *C. acnes*. on the other hand. *will not* grow aerobically. It is, therefore, always necessary to transfer anaerobically isolated organisms to aerobic plates. *C. acnes*. accordingly. is not to be considered the dominant resident bacterium over the body surface; indeed, it is not even found in most regions.

The resident corynebacteria embrace a number of species presently not classifiable. They grow richly in the axilla. Crissey *et al* (6) designated as *C. tenuis* the organism which they recovered from 28 cases of trichomycosis axillaris. These 28 strains exhibited great variability and no proof is given either that they belong to one species or that they are different from the resident corynebacteria. We isolated no peculiar or unique corynebacteria from our subjects who had trichomycosis axillaris. We consider this condition to be a result of the exuberant overgrowth of resident corynebacteria on the hair shafts.

It is clear that the axillary flora is more diverse than most other skin areas. As elsewhere the gram positive staphylococci and corynebacteria predominate. That Alkaligenes and Aerobacter achieve resident status here is at least partially ascribable to the known high moisture requirement of gram negatives. Studies (7, 8) show a greater susceptibility of gram negative organisms to desiccation and their inability to grow at reduced moisture levels which can support gram positive staphylococci and diphtheroids. Finally, we did not confirm Shelley *et al* (1) and Borick and Sarra's (4) finding of the rather constant presence of *Staphylococcus aureus*. We recovered this organism in only 8% and in trivial numbers. It is misleading to record merely the frequency of recovery as is almost customary. With *Staph. aureus*, as well as with other transients which are constantly alighting on the skin, the important value is not percentage but rather the number of colonies recovered. With zealous sampling, the percentage recovery would doubtlessly be quite large, but usually only a few colonies are found.

2. Bacterial Counts

It is appreciated that no method of sampling a surface as variable as the skin can claim absolute precision and reproducibility; nonetheless, the mechanical scrubbing method does give acceptably constant results and in our hands is considerably more reliable than swabbing or stripping with Scotch tape. With three consecutive one-minute scrubs in five subjects, it was found that 85% of the three-minute total was removed during the first minute. This indicates the superficial position of the organisms and justifies the usefulness of one-minute scrubs for quantitative analysis.

The numbers of organisms were quantitatively determined in both axillae of 25 individuals. The results are given in Table II. As is known, individuals differ greatly in quantities of organisms recovered. By and large, there is good correspondence between the counts in the right and left axillae. The average counts of the right and left axillae of these 25 subjects are almost identical, 2.36 millions/cm^2 and 2.59 millions/cm^2 respectively. A high count in one axilla is generally matched by a similar count in the opposite one with occasional striking exceptions, probably attributable to technical errors. In the same 25 subjects, quantitative bacterial counts were done at 0, 2, 4, and 8 weeks with the results displayed in Table III. Here again it may be observed that some individuals have consistently low counts, while others have high ones. This may be considered a basic individual characteristic. Fifty-two per cent of these subjects had an average of 2 million or less/cm^2, while only fifteen per cent had more than 4 million/cm^2. On the whole, individual variations from week to week are tolerably small while the average weekly counts are strikingly constant. Therefore, it is possible

TABLE II

Comparison of bacterial counts in left and right axillae (Millions/cm²)

Subject	Left	Right
1	0.68	1.24
2	0.78	1.42
3	0.50	0.65
4	0.66	0.31
5	0.52	1.72
6	1.48	0.31
7	0.53	0.36
8	5.93	5.66
9	4.78	4.05
10	4.33	4.19
11	3.20	4.97
12	3.33	3.38
13	3.28	5.27
14	0.58	0.38
15	4.13	2.42
16	0.36	0.38
17	1.05	0.83
18	0.92	4.37
19	1.12	1.31
20	5.84	6.07
21	3.44	2.67
22	0.63	0.96
23	3.68	3.19
24	3.07	4.68
25	4.29	4.13
Average	2.36 Millions/cm²	2.59 Millions/cm²

to assess with some confidence changes in counts under the impact of antibacterial agents.

3. Antibiotograms

Table IV shows the sensitivities of the axillary organisms to single concentrations of penicillin, streptomycin, and neomycin. There is uniform sensitivity to 30 ug/cc. of neomycin, except for Alkaligenes and Proteus. Streptomycin has a similar spectrum of activity except for the frequent resistance of *Staph. aureus*. Penicillin is not active at all against the gram negatives. It is emphasized that all three antibiotics are active against the gram positive residents.

4. Effect of Neomycin on the Axillary Flora

Neomycin was applied daily to one axilla of 20 subjects for 15 weeks. Quantitative bacterial analyses and antibiotic sensitivities were done at 2, 6, and 14 weeks as well as 4 weeks post-treatment. An additional 30 persons received neomycin for 4 months with studies only at the beginning and end. During treatment the samples were plated into antibiotic-containing Sabouraud's medium for the detection of fungi, especially Candida. Figure 2 shows the results in the first 20 subjects in graphic form. The proportions of gram positive to gram negative bacteria are, of course, an estimate. Moreover, individual differences, which were at times considerable, are blended into the average.

Within a few days, there was a sharp suppression of odor which persisted without change as long as the antibiotic was given. At 2 and 6 weeks there was a great numerical reduction of organisms with almost complete obliteration of the gram positives. These were indeed completely eradicated in many subjects. At 14 weeks there was definite expansion of the gram negatives which were now clearly dominant and present in greater numbers than originally. Nonetheless, the total count was still greatly below that of the

TABLE III

Successive bacterial counts of the same axilla (Millions/cm²)

Subject	0	2 weeks	4 weeks	8 weeks
1	0.68	0.44	0.65	0.81
2	0.78	0.59	1.20	0.83
3	0.50	0.41	0.69	0.41
4	0.66	0.29	1.11	0.58
5	0.52	0.58	0.96	0.66
6	1.48	1.02	1.06	0.83
7	0.53	1.20	1.44	2.01
8	5.93	6.31	4.72	5.72
9	4.78	5.78	4.26	4.22
10	4.33	3.04	4.40	3.39
11	3.20	2.91	2.86	2.34
12	3.33	2.17	3.57	3.13
13	3.28	2.24	3.93	3.19
14	0.58	0.83	1.19	0.84
15	4.13	2.68	3.80	2.43
16	0.36	0.46	0.35	0.30
17	1.05	0.76	1.02	1.25
18	0.92	0.75	1.23	0.86
19	1.12	2.07	1.52	1.15
20	5.84	5.09	3.88	3.47
21	3.44	1.87	2.54	2.07
22	0.63	1.69	1.14	1.35
23	3.68	3.79	3.34	2.80
24	3.07	3.30	3.25	3.72
25	4.29	4.98	3.43	4.10
Average	2.36	2.21	2.30	2.10

original level. The marked change in the composition of the axillary flora is depicted in Table V. No organisms have entered which were not present originally, but the balance of power has shifted. *Alkaligenes* fecalis is the predominant organism in 90% of cases. Although found much less frequently (15%), Proteus is present in sufficient numbers as to be elevated to resident (major) status. It should be noted that while Figure 2 shows the presence of small proportions of gram positives, this is an average rather than literal, since the staphylococci were completely eliminated in 55% and the diphtheroids in 85% of individuals. In these particular subjects the gram negative organisms increased to a larger extent than when a residue of gram positives remained. At the first post-treatment sampling at one month, the flora had reconstituted itself in types and numbers so as to resemble the original, except that the gram negatives appeared to be somewhat more numerous than at the start. From the fact that there was not a complete restitution of odor for about 7–10 days after stopping treatment, it may be judged that the effects of neomycin are long-lasting.

No originally sensitive organisms became resistant in this total of 50 subjects treated for 4 months.

TABLE IV

Antibiotograms

Organisms	Penicillin 5 u/cc	Streptomycin 15 μg/cc	Neomycin 30 μg/cc
Coagulase (−) Staphylococcus	S (occ. R)	S or R	S
Diphtheroids	S	S	S
Aerobacter sp.	R	S	S
Alkaligenes	R	R	R
Sarcina	S	S	S
Staphylococcus aureus	R	R (occ. S)	S
Proteus vulgaris	R	R	R (occ. S)
E. Coli	R	S	S

5. Effect of Neomycin Combined with Aluminum Chlorhydroxide

The structure of this study was similar to neomycin alone. The results differ in one important detail, namely that when the antibacterial activity of aluminum is added to that of neomycin, the gram positive organisms are totally eliminated in all subjects. At 2 weeks the results are comparable to neomycin alone, but at 6 weeks when the gram positives have been extinguished, a remarkable ecological change occurs. The now uniformly gram negative population expands greatly and numerically reaches almost the total of the original population of gram positives and gram negatives. This change is an enduring one as long as the combination is applied. Four weeks after discontinuation, the flora returns to its original state (Fig. 3). The change in the composition of the flora is graphically summarized in Table VI. There is a narrowing down to three organisms: Alkaligenes, Proteus, and Aerobacter. Alkaligenes is clearly the dominant organism. The other two now occur in the majority of subjects.

The odor declined promptly and did not return until a week or two after discontinuation. No organism became resistant to neomycin.

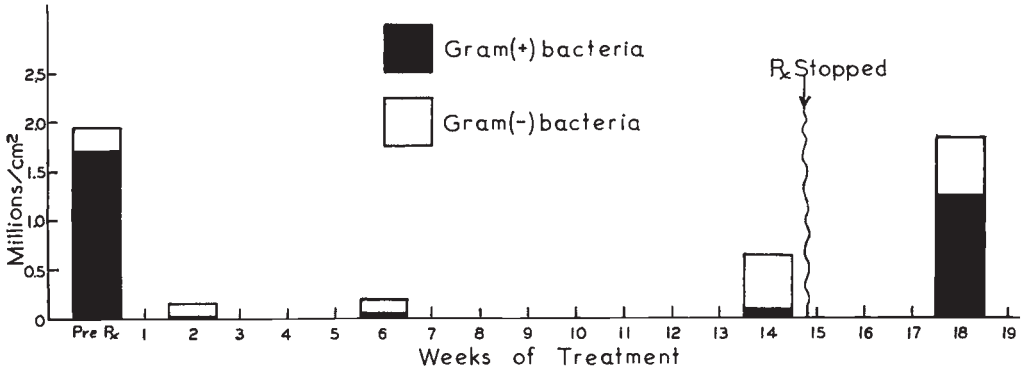

FIG. 2. Graphic Representation of Bacterial Flora of Axilla Following Treatment with Topical Neomycin.

COMMENT

These findings with neomycin and the combination of neomycin and aluminum have much to teach. The objections which have been raised against the use of neomycin in deodorants deal principally with the hazards of sensitization, emergence of resistant strains, overgrowth with pathogenic bacteria and fungi, and superimposed infection. In these studies in which 60 subjects were treated for about 4 months with liberal application of aqueous solutions containing 0.175% of neomycin, not one instance of sensitization occurred. Clinical experience indicates that neomycin sensitivity is comparatively uncommon, and this is certainly borne out in this research. The low concentration used also reduces the probability of sensitization, although an individual already sensitized will undoubtedly react.

TABLE V

Axillary Flora Post Neomycin

Classification	Organism	Incidence
Major		
Gram (−)	Alkaligenes fecalis	18/20 (90%)
	Proteus vulgaris	3/20 (15%)
Minor		
Gram (+)	Coagulase (−) Staphylococcus	9/20 (45%)
	Diphtheroids	3/20 (15%)
Gram (−)	Aerobacter sp.	6/20 (30%)

As regards emergence of resistance, experience has established that this happening is proportional to usage, particularly in respect to staphylococci. In closed hospital populations, the percentage of resistant strains increases when given antibiotics are widely used and decreases when they are not exhibited. Staphylococcal resistance to neomycin can be readily induced *in vitro*, although there is a definite feeling that this does not often happen clinically. Gocke and Finland (9) found that *Staph. aureus* originally sensitive to 25 ug/cc. became resistant to 800 ug/cc. of neomycin after 40 subcultures. Similarly, Hall (10) was able to rapidly adapt most strains of staphylococci to high concentrations of neomycin; yet after topical treatment of 109 patients with a variety of skin infections using either neomycin, bacitracin, or a combination, Livingood *et al* (11) found that there was no change in the antibiotogram of *Staph. aureus*. This compares to the resistance which developed in 16 out of 136 patients treated either with erythromycin or tetracycline ointment. Thus, while staphylococci, including *Staph. albus*, in our experience has the potentiality *in vitro* of becoming strongly resistant to neomycin, this does not appear to occur with significant frequency in actual use, either in the treatment of skin infections or in the deodorization of normal skin. Still this issue cannot be considered entirely closed since there are reported instances of neomycin-resistant staphylococci. Quie and coworkers (12), until 1959, never found resistant

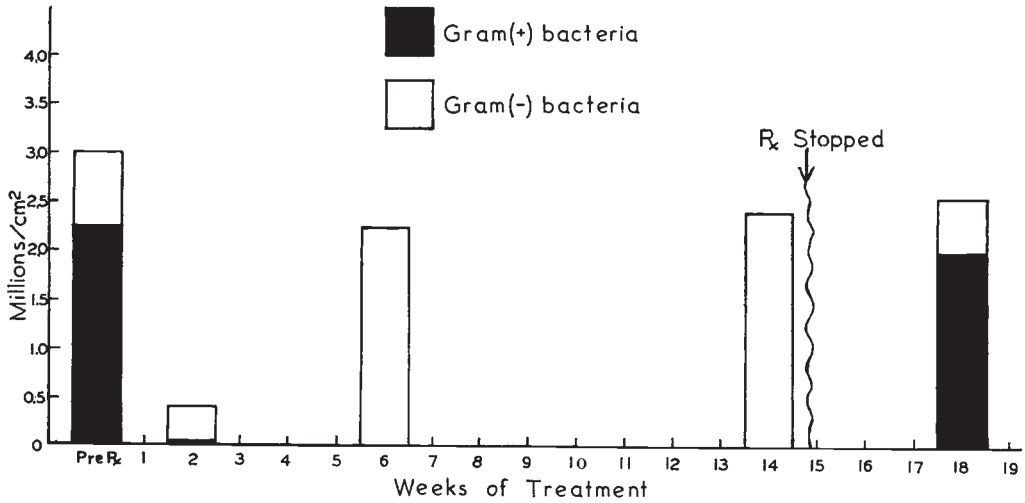

FIG. 3. Graphic Representation of Bacterial Flora of Axilla Following Treatment with Topical Neomycin and Aluminum chlorhydroxide.

TABLE VI

Axillary Flora Post Neomycin + Aluminum

Classification	Organism	Incidence
Major Gram (−)	Alkaligenes fecalis	10/10 (100%)
	Proteus vulgaris	7/10 (70%)
Minor Gram(−)	Aerobacter sp.	5/10 (50%)

staph., but after that year 20% of 1248 cultures of *Staphylococcus aureus* were neomycin-resistant. Peck and Kantor (13), found half of the staphylococci isolated from 22 cases of pyoderma or pustular acne to be neomycin and penicillin resistant. A study which more closely bears on our own is that of Jordan (14) who found that diphtheroids became resistant to neomycin in 12 subjects after 3 months of daily use of toilet soap containing 0.1% of neomycin. The same soap in the same 12 subjects did not lead to neomycin resistance of any component of the axillary flora. In our 60 subjects, no instances of pyoderma or moniliasis occurred. Livingood *et al* (11) consider that the treatment of pyoderma with neomycin leads to an increased incidence of moniliasis in the treated sites. Others have worried about this possibility, but we personally have seen no proved cases. As a matter of fact, we did not once recover any species of Candida using appropriate media for fungi. In summary, we did not encounter an overgrowth of bacteria which were not originally present, and no fungi.

Since aluminum salts have been demonstrated to be a fairly good bacteriostatic agent (15), there remains to be explained how the gram negative organisms could proliferate so extensively. The answer evidently is to be found in its feeble bacteriostatic effect against gram negatives. For example, Blank *et al* (15) found staphylococci to be killed after three-minute exposure to 1:100,000 dilution of aluminum chlorhydroxide, whereas even a 1% solution was ineffective against a coliform organism after 14 minutes.

The above findings are particularly revealing with regard to bacterial ecology. Next to nothing is known about the forces which determine the composition of the cutaneous microflora. We ourselves do not subscribe to the belief that the skin possesses an important degerming or self-sterilizing faculty which wards off transient organisms. More likely, the resident organisms are entrenched because local conditions are more favorable to them than to competitors. There is no previous study to indicate what might happen if an artificial interference were exerted upon the normal relationships allowing certain minor organisms to become dominant. Is there an optimal balance among the members of the bacterial community which is so delicate that an important shift in proportions will have an adverse effect on the health of the skin? We can now begin to answer some of these questions.

In the first place, one would have predicted from the neomycin antibiotogram which of the original organisms would likely gain ascendency—namely, Alkaligenes fecalis and Proteus vulgaris, both being resistant to neomycin. The elimination of dominant gram positive competitors probably permits these organisms to proliferate. There is indeed an overgrowth of organisms but only those which already had some kind of foothold in the axilla and which were not susceptible to the expulsive force of the antibiotic.

There is an informative contrast between neomycin alone and neomycin combined with aluminum. In the former case in which the gram positives are suppressed but not eliminated, the gram negatives expand, but only moderately, by no means replacing the gram positives numerically; but when the presence of aluminum totally abolished the gram positives, the gram negatives grow luxuriantly until they numerically replace the original population. This would seem to be a simple matter of competitive population pressure. As long as some gram positives are present they can exert a restraining influence on a somewhat larger number of gram negatives. Another way of putting this is that the gram positives are better adapted to the axilla and hold the gram negatives in check. This restraining influence is not relieved until there is a complete extinction of the gram positives which occurs with the greater antibacterial effectiveness of the neomycin and aluminum combination. This is nicely seen at 2 weeks with this combination when the bacterial count is still quite low owing to the persistence of a small gram positive component. At 6 weeks, in the absence of gram positives, the gram negatives flourish and fill up the population vacuum left by the gram positives. How a small population of gram positives restrains a

larger population of gram negatives is an ecological problem well worth solving.

The high moisture of the axilla undoubtedly contributes to the overgrowth of the gram negatives, though it does not explain why other species occasionally found in the axilla, *i.e. E. coli*, do not become established. As long as one is dealing with mixtures of organisms, these ecological preferences can hardly be worked out.

Odor

As long as neomycin alone or in combination with aluminum was used there was effective odor suppression, even though there was a great increase of gram negatives with the combination. This fact makes it necessary to amend a commonly held view concerning the organisms responsible for axillary odor. Following Shelley's basic work (1), Strauss and Kligman (3) inoculated apocrine sweat *in vitro* with a variety of organisms and came to the erroneous conclusion that any of the organisms present in the axilla could generate the typical odor, including gram negatives. How they came a cropper is now apparent. The gram negatives have a putrid odor of their own *in vitro*. This odor, however, is differenciable from the typical axillary odor by experience. Indeed, this very gram negative odor can often be faintly detected in axillae treated with the neomycin-aluminum combination. This clinically inapparent odor is that of the organisms themselves and not a consequence of the decomposition of apocrine sweat. Clearly, it is the resident gram positive organisms which are responsible for the typical axillary odor, though whether it is the staphylococci, the corynebacteria, or both, cannot be declared.

6. Aluminum Chlorhydroxide

Eighteen per cent aluminum chlorhydroxide in the aqueous vehicle was applied to one axilla of ten subjects for 14 weeks. The previous procedures were followed. At 2 weeks there was a sharp depression in the total count. In this instance, however, the gram positives still exceeded the gram negatives and there was no expansion of the latter. Much the same situation prevailed at 6 and 14 weeks, except that there was a modest increase in the gram positives (Fig. 4).

Throughout there was relatively good suppression of odor but not so completely as the combination with neomycin or with neomycin alone.

COMMENT

Aluminum chlorhydroxide strongly suppresses the axillary flora, exerting its main effects on the gram positive organisms. There is no expansion of the gram negatives. This is attributable to the presence of moderate numbers of gram positive organisms. It will be recalled that aluminum is rather ineffective against gram negatives (15). The reduction in the gram positive bacterial population is great but less than that achieved with neomycin. There is good correlation between odor in the treated axilla and the number of gram positives, hence, the lesser deodorant powers of aluminum salts alone.

7. Effect of Streptomycin and Penicillin

These will be discussed together since the results were almost identical. Penicillin was used

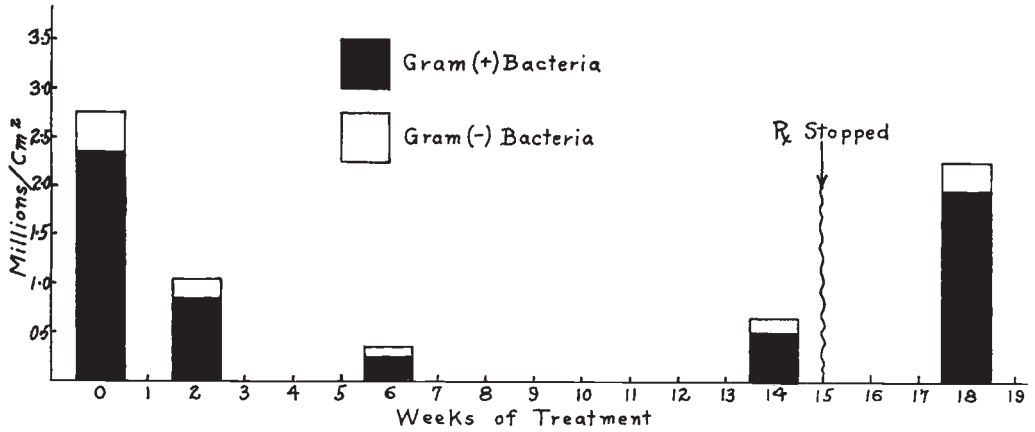

FIG. 4. Graphic Representation of Bacterial Flora of Axilla Following Treatment with Aluminum Chlorhydroxide.

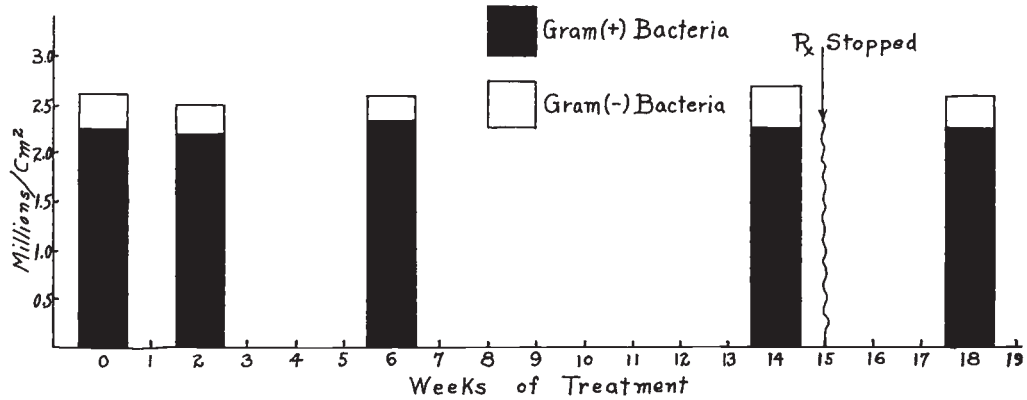

FIG. 5. Graphic Representation of Bacterial Flora of Axilla Following Treatment with Topical Streptomycin.

in a 2% concentration in petrolatum and streptomycin in a 1% concentration in the above aqueous vehicle in 10 subjects each. These studies were carried out in the routine fashion. Subjects were selected whose gram positive organisms were demonstrated to be sensitive to streptomycin and penicillin.

Results

Within the first few days there was a marked suppression of odor which can be taken indirectly to reflect diminution of gram positive organisms. To our surprise, with both agents the deodorant effectiveness waned in about 10 days. Neither antibiotic could be considered an effective deodorant after 2 weeks. The bacterial analysis at 2 weeks was not very different from the pretreatment sample as regards species and numbers. This situation remained unchanged during the course of the study as shown in Figure 5 for streptomycin.

The antibiotograms at 2 weeks and thereafter were interesting indeed. The staphylococci were uniformly resistant to penicillin and streptomycin. We unfortunately did not determine the degree of resistance acquired but only that the organisms grew in the presence of 5 U/cc. of penicillin and 15 ug/cc. of streptomycin when they could not do so before. The sensitivity of diphtheroids did not change and these were relatively suppressed. The total count remained the same owing to a corresponding increase in the staphylococci. Four weeks after cessation of treatment, the staphylococci were still resistant. There were two exceptions in the above account in 2 streptomycin-treated subjects. In these odor was suppressed throughout the course of treatment and resistant staphylococci did not develop.*

COMMENT

Streptomycin and penicillin are effective as deodorants for a period of about one week. The return of odor parallels the emergence of resistant staphylococci and the reconstitution of the gram positive flora to its original level. In this case there was a differential effect against diphtheroids, which owing to continued sensitivity, are moderately suppressed allowing a corresponding increase in the resistant staphylococci.

DISCUSSION

We consider it amply demonstrated that the axilla is a fascinating theater for bacteriologic researches. Apart from the practical application of developing better deodorants, this region with its diverse and abundant bacterial population provides an excellent opportunity for the study of ecological relationships. Antibiotics which are effective against some of the normal

* Since submitting the manuscript, the penicillin-streptomycin study was repeated with determination of antibiotic sensitivities two and four weeks after starting treatment. The outcome was the same as before except that streptomycin resistance developed earlier and to a greater degree than penicillin; within two weeks the resident staphylococci were able to grow in a concentration of streptomycin exceeding 100 ug/ml. As before the corynebacteria remained sensitive. With penicillin, moderately resistant staphylococci emerged in about 50 per cent of the group in two weeks. By four weeks, the resistances were 50 ug/ml. for about half of the group and over 100 ug/ml. for the rest.

organisms but not others impose a selective force which inevitably changes the composition of the flora. The presence of even a small number of gram positives has been shown to exert a limiting effect on the growth of the gram negatives. With total abolition of the gram positives, numerical replacement with gram negatives occurs. If one may generalize from this experience, it would seem that the population deficit created by the elimination of one group will be made up by an overgrowth of another group insensitive to the agent applied. It is probably impossible to keep an area at near sterile levels using antibiotics with selective activity. A possible exception is the utilization of a chemotherapeutic chemical which is more or less uniformly poisonous to all forms of microbial life. It is probably undesirable to eliminate bacteria completely, at least with antibiotics, since this might pave the way for colonization by fungi.

A major change in the native bacterial population has no apparent effect on the health of the skin. It is doubtful that surface bacteria significantly influence the underlying living tissue.

The effectiveness of neomycin as a deodorant bears out the clinical judgment that it is one of the best topical agents for suppressing gram positive organisms. The index of sensitization is low and organisms do not readily become resistant. We were at pains to demonstrate that the effectiveness of neomycin was not ascribable to the bacteriostatic effect of residues in the collected specimens. Even in the undiluted original sample, after sterilization, there was not enough neomycin present to inhibit susceptible organisms.

Why do axillary organisms rapidly develop resistance to penicillin and streptomycin but not to neomycin? There is evidence that neomycin is substantive to skin so that repeated applications build up a constant inhibitory level, lessening the opportunity for resistant strains to develop. Jordan (14) found that the use of a 0.1% neomycin soap led to the fixation of small amounts of the antibiotic to the uppermost layers of the stratum corneum. Moreover, neomycin is quite stable in aqueous solution, unlike penicillin and streptomycin.

On the face of it, the axilla would appear to be a useful place to test the probability of organisms becoming resistant to topically applied antibiotics. Here too it is the staphylococci which display the resistance tendency so strikingly. With the 3 antibiotics so far tested, the resistance pattern is similar to the experience of systemic chemotherapy.

The correlation between odor level and the number of gram positive organisms in the treated axilla is so good that one can without facetiousness stress the value of the educated nose as an instrument for estimating the number of gram positive organisms.

SUMMARY

The resident axillary flora consists mainly of gram positive organisms (coagulase negative staphylococci and diphtheroids) and to a lesser extent gram negative organisms (*Aerobacter sp.* and *Alkaligenes fecalis*).

The bacterial flora is quantitatively stable from week to week. A high or a low count is a fundamental individual characteristic.

Prolonged topical application of neomycin alone and neomycin combined with aluminum chlorhydroxide markedly suppresses the gram positive organisms with overgrowth of the gram negative ones.

Gram negative organisms are not responsible for axillary odor.

Bacterial resistance, sensitization, superinfection, and colonization by fungi did not occur following topical neomycin. Contrariwise, staphylococcal resistance to topical penicillin and streptomycin develops rapidly.

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DISCUSSION

Dr. Theodore Cornbleet, Chicago, Ill.: Several years ago we conducted a set of parallel experiments as those presented except that we did not use aluminum salts. Although our observations were not as detailed, our results were different from that of the essayists. We used both bacitracin and neomycin in separate experiments on a half dozen patients in each category. After eight weeks continuous trial of each antibiotic, we found that the axillary odor returned as it was originally after being suppressed, and that in spite of the fact that the dominant organisms previously found were gone, and continued to be absent for some time. In spite of their banishment the axillary odor did return. Apparently there was some organism which we did not plate out and eluded us, and we do not know which one it possibly was, but evidently it (they?) was the cause of the return of axillary odor.

Dr. A. J. Reiches, St. Louis, Mo.: This work of Drs. Shehadeh and Kligman is interesting and informative. I am somewhat surprised that Corynebacterium acnes, which is the same organism, that we formally called the acne bacillus of Unna and Engman was not found. All the bacterial organisms found and cultured are apparently saprophytic and no pathogenic organisms are reported in this study of the axilla.

Dr. Najib Shehadeh, (in closing): In answer to Dr. Cornbleet's question, we have also noted that after treatment with neomycin plus aluminum hydroxide for six to eight weeks, the same odor appeared in the axilla. This odor, however, is very different from the one which is present following the decomposition of apocrine sweat and this odor can also be smelled on the plate which is overgrown with gram negative organisms. We believe that this odor is the natural odor of gram negative organisms rather than due to apocrine sweat decomposition.

In answer to Dr. Reiches' question, we were not able to recover Corynebacterium acnes from the axilla in over 130 subjects. This is different from the view of Dr. Evans who believes that these organisms are very common. We have failed to find these organisms except in areas where acne is ordinarily found.