INTERGENERIC AND INTRAGENERIC INHIBITION BETWEEN STRAINS OF *PROPIONIBACTERIUM ACNES* AND MICROCOCCACEAE, PARTICULARLY *STAPHYLOCOCCUS EPIDERMIDIS*, ISOLATED FROM NORMAL SKIN AND ACNE LESIONS

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More than 80 years after the first observation of micro-organisms in acne lesions (Unna, 1891, cited by Breed, Murray and Smith, 1957; Unna, 1896) there is still not universal acceptance of the part played by them in the aetiology of acne vulgaris. *Propionibacterium acnes* (*Corynebacterium acnes*) which for many years since its first isolation (Sabouraud, 1897) has been considered the likeliest pathogen, co-exists on normal skin, in healthy sebaceous follicles and in acne lesions with members of the Micrococcaceae, particularly *Staphylococcus epidermidis* biotype 1 (Baird-Parker, 1974), and with the yeast *Pityrosporum ovale*. This subject was well reviewed by Rosenberg (1969) and by Marples (1974). The production of inflammatory lesions when *P. acnes* organisms are injected into sterile steatocystomas (Kirschbaum and Kligman, 1963) and the reduction in severity of acne when the numbers of *P. acnes* are reduced by systemic tetracycline (Lane and Williamson, 1969; Marples and Kligman, 1971) indicate involvement of this organism in the development of acne lesions. Whether *S. epidermidis* is involved in pathogenesis or not is unknown. Whatever their exact role, the major question still remains: what causes these bacteria, almost universally found on skin, to express a pathogenic character in acne patients?

It is possible that characters of certain strains, not certain species, determine whether pathogenesis is expressed or not. Differences may exist between the strains present on normal and acne skin. Some strains may be capable of influencing the growth of others and thereby directly determine the components of an individual's flora.

If the production of substances active against *P. acnes* and *S. epidermidis* can be demonstrated in skin micro-organisms, a possible control system may be envisaged in which microbial interactions assist or prevent the development of the acne lesion. The work of Evans *et al.* (1950) and of Selwyn and Ellis (1972) suggests that such interactions occur.

The aims of the present investigation were (1) to re-examine inhibitor production by skin Micrococcaceae and to determine whether any of them inhibit...

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P. acnes; (2) to determine whether strains of P. acnes produce inhibitors of S. epidermidis and of P. acnes; and (3) to determine the frequency of occurrence of inhibitory strains on normal and acne skin.

MATERIALS AND METHODS

Media. Nutrient Broth Number 2, Blood Agar Base, and Reinforced Clostridial Medium were obtained from Oxoid Limited, Southwark Bridge Road, London, SE1 9HF. Brain Heart Infusion Agar was purchased from Difco Laboratories, PO Box 14B, Central Avenue, West Molesey, Surrey.

Source of bacteria. All strains of bacteria were isolated from the faces or backs of 44 subjects attending the acne clinic at the Leeds General Infirmary or from employees of the Department of Microbiology, University of Leeds (table I). All subjects were in the age range 15-30 years, with the exception of one patient aged 39 who had severe acne. There were 29 females and 15 males. Patients were graded according to a modification of the scheme of Burton et al. (1971), increasing the number of grades within the 'low acne' group. Only subjects without evidence of acne on face, back and chest were regarded as controls.

Samples were obtained from the skin surface with swabs soaked in nutrient broth. Organisms were released from the swab by agitation in 10 ml of nutrient broth on a rotamixer for 5 s; 0.1-ml volumes of the undiluted and tenfold diluted samples were plated out on heated blood agar (7% v/v horse blood, Oxoid) for isolation of Micrococcaceae and on Brain Heart Infusion Agar with 0.3% glucose added for isolation of P. acnes. Plates were incubated at 37°C, for 2 days aerobically for Micrococcaceae and 7 days anaerobically for P. acnes in an atmosphere of H₂ plus 10% CO₂ in Baird and Tatlock cold-catalyst anaerobic jars.

Strains for use in inhibition tests were selected on the basis of predominant colonial types of either P. acnes or Micrococcaceae, verified by Gram staining and coded as follows: M = Micrococcaceae; P = Propionibacterium; F = isolated from face; B = isolated from back; hence MF1, MB2, PF1, PB2, etc.

Strains were tested for inhibitory activity immediately after isolation because it was found that they often lost primary characteristics during storage. Maintenance of stock cultures was in 40% (v/v) glycerol/phosphate-buffered saline at -20°C, by a modification of the method of Gore and Walsh (1964).

A total of 241 strains from 36 patients and eight control subjects were used as test strains. A further 32 strains were used as indicators of inhibition. The derivation of these strains is shown in tables I and II. Indicator strains of S. epidermidis were typed by the scheme of Baird-Parker (1974) and only strains of biotype 1 were used. They were also typed by the method of Kloos and Schleifer (1975) and all belonged to the group S. epidermidis. Indicator strains of P. acnes were typed by the method of Marples and McGinley (1974) and comprised 16 strains of P. acnes I, one strain of P. acnes II A and three strains of P. acnes II B.

<table>
<thead>
<tr>
<th>Acne grade</th>
<th>Number of subjects</th>
<th>Number of strains</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Micrococcaceae</td>
<td>P. acnes</td>
</tr>
<tr>
<td>None</td>
<td>8</td>
<td>31</td>
</tr>
<tr>
<td>Mild</td>
<td>11</td>
<td>29</td>
</tr>
<tr>
<td>Moderate</td>
<td>19</td>
<td>63</td>
</tr>
<tr>
<td>Severe</td>
<td>8</td>
<td>25</td>
</tr>
<tr>
<td>Totals</td>
<td>46*</td>
<td>148</td>
</tr>
</tbody>
</table>

* Two patients were sampled at two sites (face and back); only 44 persons were sampled.
INHIBITION OF PROPIONIBACTERIA AND STAPHYLOCOCCI

TABLE II
Derivation of indicator strains

<table>
<thead>
<tr>
<th>Acne grade</th>
<th>Number of strains</th>
<th>S. epidermidis</th>
<th>P. acnes</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>4</td>
<td>6</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td>Mild</td>
<td>3</td>
<td>4</td>
<td>7</td>
<td></td>
</tr>
<tr>
<td>Moderate</td>
<td>2</td>
<td>4</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Severe</td>
<td>3</td>
<td>6</td>
<td>9</td>
<td></td>
</tr>
<tr>
<td>Totals</td>
<td>12</td>
<td>20</td>
<td>32</td>
<td></td>
</tr>
</tbody>
</table>

Detection of inhibition. The method of Jetten and Vogels (1972) was employed with Brain Heart Infusion Agar, pH 6.0) as growth medium. Test strains were inoculated diametrically across the surface of each plate and were pre-incubated. Indicator strains were then inoculated in an "L" design adjacent to the test strain and the plates were re-incubated. Inocula were obtained from overnight nutrient broth cultures of Micrococcaceae and from 2-day Reinforced Clostridial Broth cultures of P. acnes.

Incubation conditions for test plates were 3 days anaerobically with 10% CO₂ for P. acnes test and indicator strains, and 1 day aerobically for Micrococcaceae, all at 37°C.

Interpretation of results. Results were recorded as positive (+), intermediate (±) or negative (−) according to the extent of inhibition of the indicator strain as shown in fig. 1.

The appearance of positive and intermediate inhibition is demonstrated in fig. 2.

Fig. 1.—Appearance of inhibition.
(1) Shows the appearance of positive inhibition.
(2) Shows intermediate inhibition and
(3) Shows normal growth in the absence of any inhibition.
RESULTS

Preliminary experiments

The pH of the skin in acne-bearing regions is generally regarded as being slightly acidic (Noble and Somerville, 1974). Therefore, the pH of the test medium was reduced to 6.0 so that strains capable of inhibitory activity under acidic conditions similar to those on the skin could be identified. The possibility of non-specific inhibition due to hydrogen ions had to be excluded. Twelve strains of S. epidermidis and 12 strains of P. acnes were tested for growth at pH 6.0 and below on Brain Heart Infusion Agar without additional buffer. Plates were incubated as in the inhibition tests. Results are shown in table III.

### Table III

<table>
<thead>
<tr>
<th>Indicator strains (and number tested)</th>
<th>Number capable of growth on solid medium* at pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>4.5</td>
</tr>
<tr>
<td>P. acnes (12)</td>
<td>9</td>
</tr>
<tr>
<td>S. epidermidis (12)</td>
<td>0</td>
</tr>
</tbody>
</table>

* Indicator bacteria were streaked on a range of Brain Heart Infusion Agar media acidified with concentrated HCl and incubated at 37°C as in the inhibition tests.

During the survey, randomly selected test strains were screened for acid production on the test medium to ensure that the inhibitory activity did not correspond to reduction of the pH of the medium below that at which the indicator strains would grow. The lowest pH recorded after growth for 24 h of staphylococcal test strains and after growth of P. acnes test strains for 3 days was 5.2. Therefore, reduction of pH of the medium was not considered to be the cause of inhibition in any of the cases examined.

As an additional precaution all test strains were screened for auto-inhibition to exclude the possibility of other non-specific inhibitory phenomena such as nutrient depletion, production of autolysins or a localised accumulation of toxic metabolites.

Definitive studies

The 241 test strains were screened for inhibitory activity against the 32 indicator strains; 53 were found to be active (table IV), and the majority of them (72%) were P. acnes. The inhibition spectra of the active strains varied considerably and are shown in table V. P. acnes strains were inhibited 10 times as frequently as S. epidermidis (table VI). Only one active strain did not inhibit P. acnes.
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Intrageneric inhibition between strains of *P. acnes* occurred much more commonly than intergeneric inhibition between strains of *P. acnes* and Micrococcaceae, and more often than intrageneric inhibition between strains of Micrococcaceae (table VII).

The frequency of occurrence of activity among strains derived from acne and non-acne subjects was similar (table VIII). Statistical analysis revealed that \( \chi^2 = 0.197 \) with one degree of freedom which shows that there was no significant difference between the incidence of activity in the two groups.

Test strains derived from acne patients showed a frequency of inhibition of *P. acnes* and *S. epidermidis* indicator strains similar to that of test strains derived from control subjects (table IX). However, the frequency of inhibition of individual *P. acnes* indicator strains varied considerably. Some *P. acnes* were sensitive to many test strains, e.g. strains 12 and 20, whereas some were sensitive to only a few test strains, e.g. strain 6.

### Table IV

**Inhibitory activity detected among 241 isolates**

<table>
<thead>
<tr>
<th>Species (and number tested)</th>
<th>Number (and percentage) of strains with inhibitory activity</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>P. acnes</em> (93)</td>
<td>38 (40.8%)</td>
</tr>
<tr>
<td>Micrococcaceae (148)</td>
<td>15 (10.1%)</td>
</tr>
<tr>
<td><strong>Totals (241)</strong></td>
<td><strong>53 (22.0%)</strong></td>
</tr>
</tbody>
</table>

To determine whether this variation in sensitivity of the indicator strains was correlated with the acne grade from which each strain was derived, regression analyses were done (figs. 3 and 4). The calculated values of \( r \) and the lack of coincidence of the regression lines showed that there was no correlation between the acne grade of the patient from which the indicator strain was obtained and its frequency of inhibition by either *P. acnes* or Micrococcaceae.

The mode of inhibition of the indicator strain is not normally discernible from the assay plates, but for *P. acnes* indicator strains two cases of inhibition were apparently mediated by bacteriophage. This was suggested by the appearance of the inhibited edge of the indicator strain. Inhibition by bacteriophage produced an indented edge of inhibition whereas other modes of inhibition produced an even straight or rounded edge.

The bacteriophage may either have been derived from lysogenic test strains or released from the indicator strains by an inducer substance produced by the test strains. Plaque-formation studies on confluent plate cultures of several different *P. acnes* strains by culture supernates of all *P. acnes* indicator strains failed to reveal any evidence of bacteriophage activity among the indicator strains.
## Table V

Inhibition spectra of active strains

<table>
<thead>
<tr>
<th>Clinical state of donor</th>
<th>Test strain*</th>
<th>S. epidermidis strains</th>
<th>P. acnes strains</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A B C D E F G H I J K L</td>
<td>1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20</td>
<td></td>
</tr>
<tr>
<td>Non-acne</td>
<td>MF205 ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ ++ +</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Fig. 2.—Inhibition of *P. acnes* by a strain of *P. acnes*. Three indicator strains of *P. acnes* are positively inhibited; the fourth strain (upper left) shows intermediate inhibition.
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**TABLE VI**
Frequency of inhibition of indicator strains by the 241 test strains

<table>
<thead>
<tr>
<th>Test species</th>
<th>Number (and percentage) of test strains inhibiting</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>P. acnes</em></td>
<td>52 (21.6%)</td>
</tr>
<tr>
<td><em>S. epidermidis</em></td>
<td>5 (2.1%)</td>
</tr>
<tr>
<td>Both</td>
<td>4 (1.7%)</td>
</tr>
</tbody>
</table>

**TABLE VII**
Incidence of inter- and intra-generic inhibition

<table>
<thead>
<tr>
<th>Test species</th>
<th>Percentage of stated test species with inhibitory activity against</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>P. acnes</em></td>
<td><em>S. epidermidis</em></td>
</tr>
<tr>
<td>Micrococcaceae</td>
<td></td>
</tr>
<tr>
<td><em>P. acnes</em></td>
<td>40.8</td>
</tr>
<tr>
<td>Micrococcaceae</td>
<td>9.5</td>
</tr>
<tr>
<td><em>S. epidermidis</em></td>
<td>1.1</td>
</tr>
<tr>
<td>Micrococcaceae</td>
<td>2.7</td>
</tr>
</tbody>
</table>

**TABLE VIII**
Frequency of isolation of active strains from acne and non-acne subjects

<table>
<thead>
<tr>
<th>Clinical state of donor</th>
<th>Number of test strains</th>
<th>Number (and percentage) with inhibitory activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acne</td>
<td>191</td>
<td>43 (22.5%)</td>
</tr>
<tr>
<td>Non-acne</td>
<td>50</td>
<td>10 (20.0%)</td>
</tr>
</tbody>
</table>

**TABLE IX**
Patterns of inhibitory activity of strains isolated from acne and non-acne subjects

<table>
<thead>
<tr>
<th>Test strains</th>
<th>Clinical state of donor</th>
<th>S. epidermidis</th>
<th>P. acnes</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Micrococcaceae</em></td>
<td>Acne</td>
<td>1.7</td>
<td>9.4</td>
</tr>
<tr>
<td></td>
<td>Non-acne</td>
<td>6.5</td>
<td>9.7</td>
</tr>
<tr>
<td><em>P. acnes</em></td>
<td>Acne</td>
<td>1.4</td>
<td>41.9</td>
</tr>
<tr>
<td></td>
<td>Non-acne</td>
<td>0</td>
<td>36.8</td>
</tr>
</tbody>
</table>
FIG. 3.—Correlation of frequency of inhibition of *P. acnes* indicator strains by test strains of *P. acnes* with acne grade of donor. The graph shows the frequency of inhibition of each indicator strain plotted against the acne grade of the subject from which that strain was derived. The regression lines have been plotted. The value of *r*, the correlation coefficient, is +0·256.
Fig. 4.—Correlation of frequency of inhibition of *P. acnes* indicator strains by test strains of Micrococcaceae with acne grade of donor. The value of *r* is +0.055.
Analysis of the results of our studies on the inhibitory activity of the isolates from 44 subjects yielded the following facts:

(a) 31 of 44 subjects (70.5%) possessed at least one inhibitory strain of \( P. acnes \), but only 12 (27.3%) possessed inhibitory strains of Micrococcaceae;
(b) 36 subjects (81.8%) possessed strains of \( P. acnes \), Micrococcaceae or both that were inhibitory to \( P. acnes \) indicator strains;
(c) among the subjects possessing inhibitory strains, the majority possessed either active \( P. acnes \) or active Micrococcaceae; only six possessed active strains of both;
(d) 11 subjects had more than one inhibitory strain;
(e) 8 subjects (18.2%) yielded no inhibitory strains.

**DISCUSSION**

The first demonstration of antimicrobial activity by a member of the skin microflora was in 1950 when Evans et al. showed the inhibition of a strain of \( P. acnes \) by two strains of Micrococcaceae isolated from the same individual. Two years later Pillsbury and Rebell (1952) failed to demonstrate any inhibitory activity among 100 strains of skin Micrococcaceae. This phenomenon was not studied again until Selwyn and Ellis (1972) found that 20% of people possessed strains of micro-organisms capable of inhibiting the in-vitro growth of other skin micro-organisms. In a further study Selwyn (1975) demonstrated an increase in the occurrence of inhibitory strains in dermatological lesions and postulated a protective role for these organisms against secondary infection.

During the course of the present work some of the problems of experimental design and of interpretation have become clear. The failure of Pillsbury and Rebell (1952) to detect inhibitory strains was probably due to their choice of a "non-skin organism" as indicator strain and to the use of a simultaneous antagonism technique which did not allow pregrowth of the producer strain. Indicator organisms would have grown before sufficient inhibitor had been produced by the active strains. Selwyn (1975) succeeded in demonstrating simultaneous antagonism with a very dilute inoculum of indicator bacteria which consequently took longer to grow than did the producer strain. In addition, Selwyn used as test medium nutrient agar plus 0.5% glucose at pH 6.5. Preliminary experiments in this laboratory with Reinforced Clostridial Agar containing 0.5% glucose at pH 6.8 showed that sufficient acid was produced by the test strains to inhibit the growth of indicator strains solely by the production of a reduced pH. Therefore, our experiments were performed on media containing less glucose and we took care to monitor acid production. The preliminary experiments reported here excluded the possibility of acid inhibition in the test system used.

The original aims of this investigation have been satisfied. The re-examination of intrageneric inhibition by strains of Micrococcaceae detected a very low frequency of activity (2.7%). This is considerably lower than all previously
reported results, the lowest of which was the 5.9% recorded by Hsu and Wise-
man (1967) who examined 119 strains of *S. epidermidis* derived from skin for
inhibition of the Oxford staphylococcus. However, in our study, when the
same strains of Micrococcaceae were tested against strains of *P. acnes* as
indicators of inhibitory activity, the frequency rose to 9.5%. Because *P.
acnes* outnumbers Micrococcaceae on the skin by about ten to one (Leyden et
al., 1975; Puhvel, Reisner and Amirian, 1975) the production of inhibitors of
*P. acnes* may be of survival advantage to the Micrococcaceae.

Our studies of inhibitor production by *P. acnes* show that intergeneric in-
hibition is much less common than intrageneric inhibition; this is the converse
of what is known in respect of the Micrococcaceae. Therefore, most inhibi-
tory activity on the skin seems to be directed against *P. acnes*, and indeed we
found that 81.8% of the 44 subjects sampled possessed bacteria inhibitory to
*P. acnes*. The significance of these findings is not known.

The differences in the activity spectra of the *P. acnes* test strains and the
various susceptibilities of the indicator strains suggest that *P. acnes* could be
typed according to sensitivity to a standard set of inhibitors as in bacteriocide
typing of other bacterial species (Reeves, 1972). It would first be necessary to
determine the nature of the inhibitors involved and to define any phage activity.

The importance of *P. acnes* inhibitors, including phage, in the bacterial
ecology of the skin requires further study. In terms of the prevalence of
inhibitory strains of *P. acnes* and Micrococcaceae and of their spectra of
activity we found no significant differences between normal and acne-affected
skin. The present findings suggest that the possession of inhibitory strains or
conversely the possession of sensitive strains does not predispose to acne.

**Summary**

Two hundred and forty-one strains of resident skin bacteria comprising 93
isolates of *Propionobacterium acnes* and 148 of Micrococcaceae derived from 36
acne patients and 8 control subjects were screened for their ability to inhibit
32 indicator strains, including 20 strains of *P. acnes* and 12 strains of *Staphy-
lococcus epidermidis* derived from patients with all grades of acne and from
normal skin. Fifty-three strains (22%) showed some activity against at least
one indicator strain. Both broad- and narrow-spectrum inhibition was detected.
Inhibitory isolates of *P. acnes* outnumbered inhibitory Micrococcaceae by four
to one. There was a low frequency of inhibition of *S. epidermidis* by Micro-
coccaceae (2.7%) and by *P. acnes* (1.1%) and a higher frequency of inhibition of
*P. acnes* by Micrococcaceae (9.5%) and by *P. acnes* (40.8%). Furthermore,
81.8% of the subjects sampled possessed strains inhibitory to *P. acnes*. The
significance of this finding is, as yet, unknown. No difference in the prevalence
of active strains in normal (20%) and acne (22.5%) skin was detected. These
findings suggest that the possession of inhibitory strains and conversely the
possession of sensitive strains does not predispose to acne.

The financial support of the Medical Research Council is gratefully acknowledged.

REFERENCES


