

## HUMAN SKIN SENSITIVITY TO PENICILLINS G, BT, AND O. DEMONSTRATION OF CROSS-SENSITIZATION\*

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**S**ENSITIVITY to penicillin as manifested by urticaria, various skin rashes, fever, and other sequelae is observed quite frequently.<sup>1</sup> While in the majority of cases it presents only an inconvenience to the patient, severe reactions have been recorded.<sup>2, 3</sup> Originally, the sensitizing property of penicillin was attributed to impurities in the amorphous (yellow) form of the drug, but crystalline material also has the capacity to sensitize.<sup>4, 5</sup>

"Penicillin" is a generic term and designates a group of compounds that contain an alanine dimethyl cysteine nucleus but which differ in the side chains. Commercial preparations consist predominantly of penicillin G, characterized by a benzyl group in the side chain. However, a variety of substituted penicillins, so-called biosynthetic penicillins, can be prepared by incorporating appropriate precursors in the media in which *Penicillium notatum* (or chrysogenum) is grown.<sup>6</sup> Two such penicillins,‡ "BT" (*n*-butylthiomethylpenicillin) and "AT" or "O" (allylmercaptomethylpenicillin) have been studied to a limited extent.<sup>7, 8</sup> The chief interest in these penicillins is the possibility of decreasing the incidence of sensitization following their use by reason of the straight chain substituent in their structure replacing the benzene ring of penicillin G.

This study was undertaken to determine whether there were any differences in skin sensitivity to penicillins G, BT, and O.

### SUBJECTS STUDIED

The test subjects were patients on the wards of two general hospitals. Careful inquiry was made as to previous penicillin therapy, allergic background, and drug sensitivity. Only a few patients with dermatoses were tested and no patient was tested who had received penicillin therapy within the preceding 5 days. Histories of allergic reactions to penicillin were carefully investigated and an effort made to obtain confirmation from medical personnel.

For purposes of evaluation, the subjects have been classified into three groups: Group A, those who never knowingly received penicillin§; Group B, those who had received penicillin; and Group C, those who were known to have shown allergic reactions to penicillin. Of the 270 patients who received penicillin (Groups B and C), the previous penicillin exposure was topical (ointment or aerosol) in 5, oral in 4, and parenteral in 261. From no patient in this study

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§"Penicillin" refers to crystalline penicillin G, since other types of penicillin have been available for investigational purposes only and may be assumed to have been used in a very limited number of individuals.

was the history obtained that reaction followed topical application of penicillin. Fourteen persons in Group C had had urticarial reactions and the remaining 17 had had generalized rashes with pruritus. Clinical reactions were attributable to oral administration in 3, and to parenteral administration in 28 patients.

#### MATERIALS USED

The sodium salt of penicillin G, the potassium salt of penicillin BT, and the sodium salt of penicillin AT or O (hereinafter referred to as penicillin O) were dissolved in isotonic saline solution to a concentration of 20,000 units per ml. Because the hypertonic solutions that resulted were irritating, distilled water was later substituted for the saline solution. Solutions were used immediately or stored at  $-20^{\circ}$  C. for periods not longer than 2 weeks. The penicillins of the types used in this investigation were as nearly pure as it is practical to manufacture them. Commercial crystalline penicillin G is highly purified but it contains traces of contaminants in the form of other penicillins. Similarly penicillins BT and O contain contaminating traces of other penicillins.

#### METHODS

Intracutaneous injections of 0.1 ml. containing 2,000 units of the penicillins to be tested were made in widely separated sites on the flexor surface of the forearm. Twenty-four and 48 hours later, erythema and induration at the sites of injection were measured in two diameters. An area of erythema less than 4 mm. in diameter at the end of 24 hours was recorded as a negative skin reaction. Other reactions such as vesiculation, itching, and systemic manifestations were noted.

#### RESULTS

Of the 276 persons tested with penicillins G and BT, a total of 37, regardless of previous exposure, were reactive to G and 29 were reactive to BT (Table I). It is noteworthy that of the 29 persons reacting to BT penicillin, 28 reacted also to penicillin G.

TABLE I. COMPARISON OF SKIN SENSITIVITY TO PENICILLINS G AND BT

| RESPONSE TO SKIN TEST | GROUP A<br>NO KNOWN PENICILLIN THERAPY |      | GROUP B<br>PREVIOUS TREATMENT WITH PENICILLIN |      | GROUP C<br>KNOWN REACTION TO PENICILLIN |      | TOTALS          |      |
|-----------------------|----------------------------------------|------|-----------------------------------------------|------|-----------------------------------------|------|-----------------|------|
|                       | NO. OF PATIENTS                        | %    | NO. OF PATIENTS                               | %    | NO. OF PATIENTS                         | %    | NO. OF PATIENTS | %    |
| +G, +BT               | 6                                      | 4.5  | 8                                             | 7.1  | 14                                      | 53.5 | 28              | 10.0 |
| +G, -BT               | 1                                      | 0.7  | 5                                             | 4.4  | 3                                       | 11.6 | 9*              | 3.2  |
| -G, +BT               | 1                                      | 0.7  | 0                                             | 0.0  | 0                                       | 0.0  | 1*              | 0.3  |
| -G, -BT               | 129                                    | 94.1 | 100                                           | 88.5 | 9                                       | 34.9 | 238             | 86.5 |
| Totals                | 137                                    |      | 113                                           |      | 26                                      |      | 276             |      |

\*Deviation in distribution from chance, in these two groups highly significant,  $p = 0.019$ .

Within the separate categories (Groups A, B, C) there were no statistical differences in the incidences of positive reactions to penicillins G and BT. In the entire series only 10 persons reacted differentially, that is, 9 were +G, -BT

and 1 was -G, +BT. The likelihood of obtaining this distribution (9:1) between two alternative responses on the basis of chance alone is very remote ( $p = 0.019$ ). It should also be noted that the incidence of positive reactions to both penicillins paralleled the previous exposure to penicillin. Thus the total incidence of positive skin tests in Group A was 5.9 per cent, Group B, 11.5 per cent, and in Group C, 65.1 per cent.

In Table II are presented the results of testing 224 persons with penicillins G and O. Thirty-six patients were positive to penicillin G, 26 were positive to penicillin O, and 12 reacted differentially. Of the latter group, 11 were +G, -O and only one was -G, +O. Here again, the incidence of differential reactions (11:1) departs significantly from expectancy on the basis of chance alone ( $p = 0.0059$ ). In this series, also, there is an obvious correlation between the incidence of positive skin tests and the previous penicillin exposure, in Group A, 4.4 per cent, Group B, 17.3 per cent, and Group C, 65 per cent.

One hundred patients were tested with all 3 penicillins. Although the data on differential responses are too few to be analyzed statistically, certain observations are noteworthy. In this group, a total of 19 patients showed positive reactions; 13 were sensitive to all 3 penicillins, +G, +BT, +O; 4 were +G, +BT, -O; 1 was -G, -BT, +O; and 1 was -G, +BT, -O.

TABLE II. COMPARISON OF SKIN SENSITIVITY TO PENICILLINS G AND O (AT)

| RESPONSE TO SKIN TEST | GROUP A<br>NO KNOWN PENI-<br>CILLIN THERAPY |      | GROUP B<br>PREVIOUS TREAT-<br>MENT WITH<br>PENICILLIN |      | GROUP C<br>KNOWN REACTION<br>TO PENICILLIN |      | TOTALS             |      |
|-----------------------|---------------------------------------------|------|-------------------------------------------------------|------|--------------------------------------------|------|--------------------|------|
|                       | NO. OF<br>PATIENTS                          | %    | NO. OF<br>PATIENTS                                    | %    | NO. OF<br>PATIENTS                         | %    | NO. OF<br>PATIENTS | %    |
| +G, +O                | 2                                           | 2.2  | 11                                                    | 10.0 | 12                                         | 55.0 | 25                 | 11.2 |
| +G, -O                | 1                                           | 1.1  | 8                                                     | 7.3  | 2                                          | 10.0 | 11*                | 4.8  |
| -G, +O                | 1                                           | 1.1  | 0                                                     | 0.0  | 0                                          | 0.0  | 1*                 | 0.4  |
| -G, -O                | 89                                          | 95.6 | 91                                                    | 82.7 | 7                                          | 35.0 | 187                | 83.6 |
| Totals                | 93                                          |      | 110                                                   |      | 21                                         |      | 224                |      |

\*Deviation in distribution from chance, in these two groups highly significant,  $p = 0.0059$ .

#### DISCUSSION

Crystalline penicillin G has sensitizing properties that may be of serious clinical consequence, and while early reports indicated that the incidence of allergic reactions varied from 0.5 per cent<sup>9</sup> to 2.8 per cent<sup>10</sup> recent data indicate an incidence of 6 to 12 per cent.<sup>5, 11, 12</sup> The increasing incidence of reactions probably reflects the more widespread use of penicillin. In one series<sup>5</sup> a positive skin test (intracutaneous) to crystalline penicillin G was found in 5.7 per cent of the patients who had never received the antibiotic and an incidence of 13.4 per cent in those who had. Similar observations have been noted in this study; the incidence of positive skin tests was increased in persons with previous penicillin exposure, (compare Groups A, B, and C in Tables I and II).

The intracutaneous skin test is recognized as a useful technique for determining sensitivity to a particular antigen, but its reliability in predicting an individual's response to parenteral administration or ingestion of the antigen

can be seriously questioned. Despite the limitations of skin testing, it is the only means other than the clinical history of previous reactions to the antibiotic agent that is available for attempting to predetermine a patient's reaction to therapy with penicillin. A negative skin test does not always indicate freedom from sensitivity but a positive skin test certainly demands cautious use of parenteral therapy; in such cases it is suggested that 2,000 to 5,000 units be injected intramuscularly as a trial dose. Similarly, a clinical history of reaction to penicillin does not preclude subsequent use of penicillin, but caution in instituting therapy is enjoined.

The observation in one series of 33 patients,<sup>4</sup> that 58 per cent of those showing a positive skin test exhibited a generalized reaction to 100,000 units injected intramuscularly, lends weight to the viewpoint that a positive skin test bespeaks caution.

The advantages of having other penicillins devoid of cross-sensitization with penicillin G are obvious. Rose, Harris, Behrens, and Chen<sup>7</sup> sensitized guinea pigs to penicillin G but were unable to sensitize them to penicillins BT or AT (O). They were also unable to demonstrate in guinea pigs any cross-sensitivity between penicillins G and BT or O. These results do not appear consistent with our observations in humans, since many individuals were found reacting positively to all three agents. In addition, the data in Tables I and II demonstrate that not only does the incidence of positive reactions to penicillin G increase with previous exposure, but the incidence of positive reactions to penicillins BT and O increases similarly. Since it is chiefly penicillin G which is being used clinically, and there is little probability that penicillins BT and O occur naturally, it appears likely that cross-sensitivity among these agents exists.

The occurrence of positive skin tests to crystalline penicillin G in persons who have never been exposed to therapy has been repeatedly observed,<sup>1, 5</sup> but there is no agreement as to the explanation of this phenomenon. Many of these persons have been reported to react to trichophyton extracts, and there is the possibility that previous fungus infections may have some relation to penicillin sensitivity. It has been suggested that fungi causing disease in man may produce substances which have a close antigenic relationship to penicillin and that sensitivity to these substances may be aggravated by penicillin.<sup>13, 14</sup> Thus, persons never previously treated with penicillin may show sensitivity the first time they are exposed to it. It is of interest that in children and women in whom the incidence of trichophyton infections is lower than in men, the incidence of penicillin sensitivity is also lower. Those patients in this study not known to have had penicillin previously (Group A) showed an incidence of positive skin reactions of 5.2 per cent, whereas clinically obvious (in some cases florid) epidermatophytosis in the group of patients studied was much more frequent. It would have been desirable to correlate our studies with trichophyton and oidiomycin skin tests, but this was not done.

There were 12 patients in both series who reacted with positive skin tests and denied having previous therapy (Group A, Tables I and II). Scrutiny of the reactions of these 12 patients showed that 6 were +G, +BT, and 2 were

+G, +O; 1 was +G, -BT; 1 was +G, -O; and 2 patients reacted in a manner difficult to interpret, -G, +BT and -G, +O. The possibility of a contaminant of penicillin G in our penicillins BT and O giving these last 2 results is obviously eliminated by the remaining significant data showing differential responses in Groups B and C. Actually, these 2 tests were not markedly positive and we are inclined to regard them as nonspecific reactions, since it is difficult to understand sensitivity to a biosynthetic penicillin in the presence of a negative response to the naturally occurring type. Apart from these 2 cases, the remaining data are readily interpretable. The majority of persons showing skin sensitivity (72.6 per cent) were reactive to both types of penicillin used. A small number (27.4 per cent) were reactive to penicillin G but were not reactive to the biosynthetic penicillins BT or O. It is suggested that in some persons sensitization is due to the nucleus which is common in the structure of these penicillins, while in others the sensitization is attributable to the entire molecule (nucleus and side chain). In the former situation, the patient will react to all penicillins, in the latter case only to a specific type of penicillin.

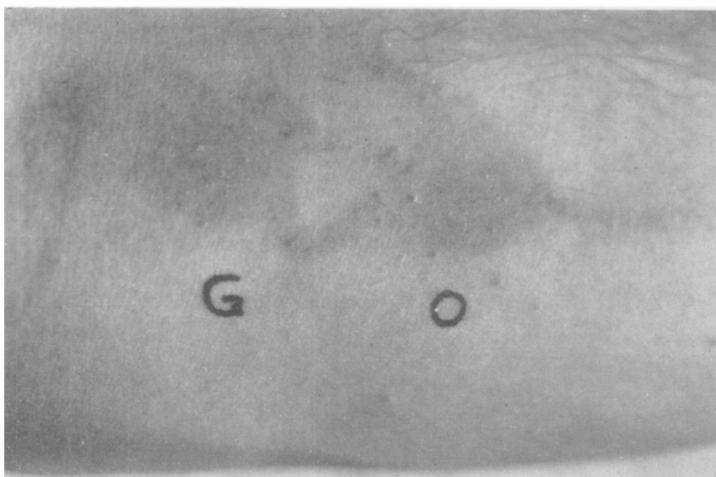


Fig. 1.—Results of intracutaneous skin tests with 2,000 units of penicillins G and O. Patient was a 25-year-old student who had had a generalized reaction to penicillin G 3 months previously. Photograph was taken 14 days after tests were performed. Note pseudopodia radiating from sites of both tests.

It is generally recognized that the severity of skin responsiveness to a test antigen cannot be used to predict the severity with which the patient may react to the antigen when administered by other routes. Many skin reactions to penicillin G were more marked than those obtained with BT or O, but in other instances there was either no difference or, in a few cases, the response to G was less marked. It is noteworthy, however, that persons who showed strongly positive tests to penicillin G (erythema and induration 10 to 20 mm. in diameter) were always positive to penicillins BT and/or O (Fig. 1). On the other hand, in those persons who showed differential responses, skin tests were less markedly positive and in the range of 5 to 10 mm. in diameter.

In no instance was vesiculation or central necrosis seen in a positive reaction. In 2 patients mild generalized reactions with pruritus and rash occurred following skin tests with all 3 penicillins. Both had had previous reactions to intramuscular penicillin therapy; 1, 3 months before and the other, 1 week prior to testing. It is worthy of note that a total of 6,000 units of penicillin elicited a generalized reaction in patients sensitive to the antibiotic.

The necessity of administering penicillin to patients in Group C, those who have already had a generalized reaction, presents the most serious problem that might be solved by having available penicillin that lacked sensitizing properties. The data from Group C (Tables I and II) show that only 2 of 21 patients tested with penicillins BT and G, and only 3 of 26 tested with penicillins G and O manifested a differential response that offered the possibility of substituting penicillins BT or O for the G type. Seventy-three patients, regardless of previous therapy, reacted to penicillin G and of this number, 20 (27.4 per cent) did not react to penicillins BT or O. Thus, the availability of a penicillin that could be used as a substitute for penicillin G in the event of allergic reactions might have definite value in this limited number of instances. It is noteworthy that many patients who exhibit a generalized reaction of rash or even urticaria following penicillin treatment may be continued on therapy, but in some patients continuation of treatment can be disastrous, resulting in exfoliative dermatitis and death. The advent of aureomycin and chloramphenicol which may be substituted for penicillin for therapeutic purposes would seem to lessen the need for such penicillins as BT and O, but in some situations where penicillin is the drug of choice it would be highly desirable to have a biosynthetic penicillin to which patients were not sensitive that could be substituted for penicillin G.

#### SUMMARY

1. Intracutaneous skin tests were performed on 276 persons using penicillins G and BT and on 224 persons using penicillins G and O (AT).
2. The incidence of positive skin reactions to all 3 penicillins showed an increase in persons with previous exposure to penicillin G, and a greater increase in those with a history of previous general reactions to penicillin G.
3. Positive skin tests to all 3 penicillins were observed in persons who denied ever having received penicillin therapy as well as in those who had received therapy. These data suggest that from the clinical standpoint cross-sensitivity to these penicillins exists.
4. A statistically significant number of differential responses were observed when the same persons were skin tested with penicillins G and BT or with penicillins G and O. The number of instances in which these differential responses occurred was small.

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