

SKIN TESTING: COMPARISON OF VARIOUS TECHNIQUES

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SKIN testing is one of the basic investigative procedures used by most allergists. The preference for any one test varies greatly among the different workers. Four different tests were evaluated in this study: puncture, intracutaneous, conjunctival, and serial dilution intracutaneous threshold test. There was a total of 215 patients who were clinically sensitive to ragweed or grass pollens. These patients were tested with the specific aqueous antigen indicated from their clinical history. Each patient was tested on the same day with the puncture, standard intracutaneous, conjunctival threshold, and serial dilution intracutaneous threshold tests. They were tested approximately 4 months before the onset of the pollinating season. The following is a brief description of the four techniques employed.

1. *Puncture Test.*—This test was performed on the volar surface of the forearm, using 5 per cent solution of antigen in 50 per cent of glycerin. Only one strength was used in contrast to the other three tests, in which varying concentrations were used. Readings were taken at the end of 20 minutes. The results were recorded as 0, + (5 mm. wheal), ++ (10 mm. wheal), +++ (15 mm. wheal), and ++++ (20 mm. wheal).

2. *Intracutaneous Test.*—Two hundredths of a cubic centimeter of the aqueous antigen was injected intracutaneously on the lateral aspect of the arm. Readings were made at the end of 15 minutes and the results were recorded as 0, +, ++, +++, and ++++, depending upon the size of the reaction. The starting concentration was 100 PNU per cubic centimeter; if this was negative, 1,000 U., 10,000 U., and 20,000 U. per cubic centimeter were employed. Occasionally this test is referred to as the "Standard Intracutaneous Test" to differentiate from the fourth test, "The Serial Dilution Intracutaneous Threshold Test."

3. *Conjunctival Threshold Test.*—The aqueous testing antigen was prepared in double fold dilutions starting at 10,240 PNU per cubic centimeter, 5,120 PNU, 2,560 PNU, and so forth, down to 10 PNU per cubic centimeter. Starting at the weakest strength and gradually increasing, one drop of antigen was placed on the everted lower lid. Readings were taken every 5 minutes. The end point was determined when the caruncle became bright red. When the ocular con-

junctival blood vessels became injected, in addition to caruncular reddening, the end point had been exceeded. At the completion of the test, the conjunctival sac was flushed with normal saline and, occasionally, one drop of epinephrine, 1:1,000, was instilled.

4. *Serial Dilution Intracutaneous Threshold Test.*—Two hundredths of a cubic centimeter of ten or one-hundred fold increasing dilutions were injected

TABLE I. COMPARISON OF REACTIONS BETWEEN PUNCTURE TEST AND INTRACUTANEOUS TEST*

		INTRACUTANEOUS (PNU/C.C.)						
		100				1,000	10,000	20,000
		++++	+++	++	+			
PUNCTURE	0	000	00000 00	00000 00000 0000	00000 00000 00000 00000 000	00000 00000 00000 00	00000 00000 0000	0000
	+	00	00000 0000	00000 0000	00000 00000 00000 00000 000	00000		
	++	000	00000 00000 00	00000 00000 0	00000 0	0		
	+++	00	00000 00000 00000 0	00000 00	000			
	++++	00000 0	00000 000	00000	00			

*Tables I through VI compare the reactivity or end point as obtained by one test with the end point obtained from another technique. The zeros represent each patient studied.

TABLE II. COMPARISON OF REACTIONS BETWEEN PUNCTURE TEST AND SERIAL DILUTION INTRACUTANEOUS TEST

		SERIAL DILUTION INTRACUTANEOUS (PNU/C.C.)													
		20,000	10,000	1,000	100	10	1	0.1	0.01	0.001	0.000,1	0.000,01	0.000,001	0.000,000,1	0.000,000,01
PUNCTURE	0	000	00000 00000	00000 00000 00000 00000 0	00000 00000 00000	000	00000 00		00000 00000 000		00000		000		00
	+			00 00000 00000 00000 00000 0	00000 00000 00000 00000 00	00000 0	00000 00		00000 0000		00000 00		000		
	++			0 00000 0000	00000 00000 00000 00		0		00000 00000		00000 00000 00000		00000		0
	+++				000		0000		00000 00		00000 0		0		0
	++++				0		000		00		00000 000		000		00

DISCUSSION

In recent years new forms of pollen therapy have been developed, such as alum-precipitated pyridine pollen extracts and mineral oil emulsions. The purpose of these newer forms of therapy is to reduce the number of required

TABLE V. COMPARISON OF REACTIONS BETWEEN CONJUNCTIVAL THRESHOLD TEST AND PUNCTURE TEST

		PUNCTURE				
CONJUNCTIVAL THRESHOLD (PNU/C.C.)		0	+	++	+++	++++
	40					000
	80		0	0	000	000
	160		0000	0000	000000	0000000 0
	320	0000	0000	0	000000	000000
	640	000	00	000000	0000	
	1,280	0	000000	000000	000000	0
	2,560	0	00	00000	0	
	5,120	000	00000	00		0
	10,240	00000	00	0000		
	> 10,240	0000000000000000 0000000000000000 0000000000000000 0000000000000000 00000	0000000 0000000 0000000 000	00000	0	0

TABLE VI. COMPARISON OF REACTIONS BETWEEN CONJUNCTIVAL THRESHOLD TEST AND SERIAL DILUTION INTRACUTANEOUS TEST

		SERIAL DILUTION INTRACUTANEOUS (PNU/C.C.)												
CONJUNCTIVAL THRESHOLD (PNU/C.C.)		20,000	10,000	1,000	100	10	1	0.1	0.01	0.001	0.000,1	0.000,01	0.000,001	0.000,000,1
	40						0		0		00		0	0
	80						000		0		0000		000	000
	160				0000		000		00		0000 0000 0000 00		0000	
	320				00		00		0000 000		0000 0000 0		0000 0	
	640				0000 0		0		0000 0	00			0	
	1,280				0000	0			0000 0000 0		0000		0	
	2,560				0000	0	00	0	0000		000			
	5,120				0000 0				000		0			
	10,240			00	0000 00	00	0	0	00		00			
	> 10,240	0000	0000 0000 00	0000 0000 0000 0000 0000 0000 00	0000 0000 0000 0000 0000 0000 0000	0000 0000 00	0000 0000 00	000	0000 0000 00		0		0	

injections. Larger doses are given in one or few visits. Constitutional reactions are a concern with all forms of hyposensitization, especially when larger doses are employed in a single injection. The value of preliminary testing in predicting safe acceptance of dosage is questionable. Some believe that preliminary testing is worthless.² Loveless³ places great value on the conjunctival threshold test in determining safe dosage; in her work on repository treatment of pollinosis, patients who reacted conjunctivally to very low concentrations of pollen received smaller doses. In her experience, constitutional reactions were more apt to occur in patients who reacted with a conjunctival threshold of 320 PNU per cubic centimeter or less. She found constitutional reactions almost nonexistent above the level of 320 PNU per cubic centimeter in repository therapy with the administration of 10,000 PNU in a single injection. Other workers employ the puncture or intracutaneous test⁴ as an aid in determining dosage in this field. Because of the controversy of skin testing in relation to acceptance of emulsion dosage, and the need to relate the different forms of preliminary skin tests, this project was undertaken. The relationship between constitutional reactions following repository therapy as related to preliminary testing will be reported in another paper. This article is devoted to the comparison of end points in the puncture, intracutaneous, conjunctival threshold, and serial dilution intracutaneous threshold test.

For those who were unaccustomed to the conjunctival threshold test, it was difficult to know what a conjunctival threshold test, for example 640 PNU cubic centimeter, was in terms of the intracutaneous test. Also, it was difficult to know what a three plus reaction to a strength of 100 PNU cubic centimeter was in terms of the conjunctival test, etc. Furthermore, because of the criticisms involved in the conjunctival threshold test, the serial dilution intracutaneous test was performed to find a substitute for the conjunctival test.

In this study, 215 patients who were clinically sensitive to either ragweed or grass pollen were tested with the respective antigen by means of four preliminary tests, that is, puncture, intracutaneous, conjunctival threshold, and serial dilution intracutaneous threshold. In the puncture test, only one concentration of solution was used (5 per cent solution of pollen in 50 per cent glycerin). In the other three tests, varying concentrations of the antigen were employed. The results of the four preliminary tests showed that which has been long known,^{5, 6} that is, the intracutaneous test is more sensitive than the conjunctival test, and the conjunctival test more sensitive than the puncture test. This present study also shows that patients who are hyperreactive with one technique, generally show hyperreactive end points in the other three preliminary tests. In addition, the tables show the wide range when one test is compared with another. The degree of reaction as obtained by one method of testing could not be predicted to give similar reactions with the other methods. More than one type of preliminary test would be advisable if testing of skin reactivity should be helpful in determining safe acceptance of dosage in the newer forms of therapy. The author found that the conjunctival threshold test and the serial dilution intracutaneous test were two important preliminary tests in repository therapy.⁷ This work will be reported in another paper.

CONCLUSIONS

Two hundred and fifteen patients who were sensitive to grass or ragweed were tested with the respective antigen by means of four preliminary tests: puncture, intracutaneous, conjunctival threshold, and serial dilution intracutaneous threshold test. Patients who were hyperreactive with one technique were generally hyperreactive with another technique. Conversely, patients who were hyporeactive were similarly less reactive with the other tests. However, there was a wide range of end results when one test was compared with another. Any one patient who was hyperreactive with one technique could be equally, more, or less reactive with other tests. Conversely, any individual who was hyporeactive with one technique, was not necessarily as hyporeactive with another type of test. Several preliminary tests give more information than any single testing technique.

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