

Conjunctival provocation test: high clinical reproducibility but little local temperature change

Rimås M, Gustafsson PM, Kjellman N-IM, Björkstén B. Conjunctival provocation test: high clinical reproducibility but little local temperature change.

Allergy 1992; 47: 324–326.

Ten atopic and five healthy individuals participated in eight conjunctival provocation tests (CPT) in an effort to improve the CPT procedure. Results were evaluated by thermography and the use of conventional criteria under various conditions. Due to considerable spontaneous variability and change in temperature already after instillation of diluent, thermography was not sensitive enough to identify positive CPT results. The precision of the CPT was within one 10-log step in all tests and in all patients.

Itching appeared before erythema in 83% of subjects and within 5–8 min after instillation of the allergen. This was true also when only one eye was used for the CPT. We therefore conclude that the CPT procedure can be used in one eye every 5–8 min when indicated in clinical routine and that it is a safe, precise method.

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Key words: atopic; conjunctival provocation test; erythema; itching; thermography.

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Accepted for publication 7 November 1991

The conjunctival provocation test (CPT) is a valuable tool in clinical practice, e.g. to confirm allergy to a defined allergen, to estimate the degree of sensitivity in a shock organ (2, 5), and to measure the effects of immunotherapy and pharmacological treatment (6, 7). The CPT is simple to perform and has good precision if performed outside the season for the particular allergen (5). In an effort to improve the CPT we have assessed whether measurement of temperature changes in the eyes during the tests makes the method quicker and more sensitive.

Ten skin prick test (SPT)-positive individuals registered at the Allergy Clinic of our department and five healthy SPT-negative subjects volunteered for this methodological study. The nine boys and the six girls with a mean age of 16 years (range 13–17), were divided into three groups of five subjects each based on the clinical diagnosis. In Group 1 all participants had allergic rhinoconjunctivitis due to deciduous-tree or grass pollen. Group 2 comprised allergic individuals with no pollen-induced rhinoconjunctivitis. Group 3 comprised healthy non-atopics.

The conjunctival provocation tests (CPT) were performed once a week on eight occasions during the winter (Table 1). The allergens used were either birch ($n = 9$) or timothy pollen ($n = 6$) (Pharmalgen®

Pharmacia Diagnostics AB, Uppsala, Sweden). Albumin Diluent®, Pharmacia (sodium chloride 0.9%, phenol 0.4%, human serum albumin 0.03%) was used for reconstitution of allergen preparations and as a negative control. The extracts were delivered freeze-dried in four concentrations, stored at $+4^{\circ}\text{C}$, and used within 1 week after reconstitution. The CPT was started by administering one drop of Albumin Diluent into the conjunctival sac of the right eye. If no reaction occurred within 10 min (in one test within 20 min, Table 1), then one drop of the lowest allergen concentration, i.e. 100 BU/ml, was instilled. The allergen doses were then increased with 10-log steps until a positive reaction occurred, or the top concentration of 1 million BU/ml was reached. Results were evaluated by a 7-grade itching score and by measuring thermal changes on the eye surface (thermography). Conventional criteria, i.e. redness of at least 50% of the conjunctiva, itching, tears and local swelling of the caruncle, were used to define a positive reaction (7).

A Thermovision Camera Model 782 SW (AGEMA Infrared Systems AB, Danderyd, Sweden) with video tape recording and a Digital Image Processing System (DIPS) were used to record thermal changes in the orbito-ocular area during each CPT. The DIPS is a complete hardware and soft-

Table 1. Test procedures in 8 CPT performed in 15 individuals. Table shows type and temperature of the solutions used and the interval between administration of drops

Test no.	1	2	3	4	5	6	7	8
Eye tested	right	right	right	right	right	right	alternate	right
Temperature +C°	22	4	4	4	4	4	4	4
No. of test drops with Alb. Dil.	1	1	4	1	1	1	1	1
Interval between drops (min)	10	10	5	20	10	5	10	10
Assessed after (min)								
2	+	+	+	+	+	+	+	+
5	+	+	+	+	+	+	+	+
8	+	+	n.d.	+	+	n.d.	+	+
10	+	+	n.d.	+	+	n.d.	+	+

n.d = not done

ware package, allowing easy analysis and handling of the thermal images by means of a standard microcomputer. A photographic set for white and black thermograms calibrated at 29°C was used as a constant reference temperature. Infrared energy emitted from the eye surface is converted into a visible image, which is displayed on a cathode ray tube and stored on disks for later retrieval. Regions of equal temperature are mapped by a specific thermal setting for display as isotherms, allowing bilateral thermal comparison. The instrument can detect differences of 0.1°C.

The environmental conditions in the room in which the CPT were performed were standardized to 50% relative humidity and 22°C temperature. The patients were adapted to this environment for 15 min before starting the tests (5). The girls were asked not to use make-up on the test day. On each occasion, patients were tested about the same time of the day (a.m.-p.m.) The thermovision camera unit was placed 1–1.2 m in front of the patient, who sat in a comfortable chair in the same position during all the tests. Itching, redness and eye temperature were recorded before CPT and after 2, 5, 8 and 10 min if not otherwise stated.

Student's t-test was used for comparison between the groups and for evaluation of change in temperature after the different instillations. A change in eye temperature by thermovision was defined as the 95th percentile of delta-temperature occurring after Albumin Diluent instillation. The specificity, sensitivity and efficacy of a delta-temperature in relation to standard criteria for the CPT were calculated according to Galen (3).

The study was performed according to the Helsinki Declaration and approved by the Human Research Ethics Committee at Linköping University.

Results

The mean temperature in the eyes before challenge was 35.2°C for all groups (range 34.2°C to 35.8°C). The temperature changes after the initial Albumin Diluent instillation on the eight test occasions in the

15 individuals were recorded after 5 min ($n = 120$). There were no differences in delta-temperature with Albumin Diluent at 4°C and 22°C. Further, no significant difference in delta-temperature reaction was seen between the three groups of test subjects, either between the tests performed in the right eye, or in those performed in alternate eyes. The 95th percentile of delta-temperature in the 120 tests was 0.4°C. A delta-temperature of more than 0.4°C in comparison with baseline was regarded as a significant change when further evaluating the different challenge conditions. No significant change in temperature was noticed during the challenge procedure, either in the challenged, or unchallenged eye. The atopics in Groups 1 and 2 did not differ significantly from the non-atopics in Group 3 at any Albumin Diluent test condition. None of the subjects showed signs of reactions, such as redness of the conjunctiva, itching or swelling of the caruncle after administration of Albumin Diluent. The interval between the Albumin Diluent instillations did not significantly influence the delta-temperature when administered at 4°C or at room temperature (22°C) in any test condition.

The allergen threshold concentrations giving positive CPT were within one 10-log step in all patients on all four occasions when they were tested with the relevant allergen. All healthy subjects tolerated the maximum allergen dose (i.e. 1 million BU/ml) without any reaction, as measured by standard criteria in all tests. In those who had a positive CPT reaction, itching appeared in 81% (30/37) on the test occasions before any obvious erythema, and in 19% (7/37) it appeared at the same time. In 46% (17/37) erythema appeared at the next higher step after the appearance of itching, and in 35% (13/37) at an allergen concentration 100 times higher. The interval between drops did not influence the temperature or the appearance of symptoms. Thermovision identified 24.3% of the positive CPT reactions during allergen provocations. The specificity was 90%. Three falsely positive reactions were noticed, all in control subjects.

The temperatures in the right and left eye showed

Table 2. Efficiency of a significant delta-temperature ($\geq 0.5^\circ\text{C}$) as an indicator of a positive test result in comparison with standard criteria for positive CPT. Results of 67 experiments are summarized

Temp. change	Total		Group 1 Allergic rhinitis		Group 2 Other atopic dis.		Group 3 Healthy	
	CPT pos	CPT neg	CPT pos	CPT neg	CPT pos	CPT neg	CPT pos	CPT neg
$\geq 0.5^\circ\text{C}$	9	3	8	0	1	0	0	3
$\leq 0.4^\circ\text{C}$	28	27	14	1	14	9	0	17
Sensitivity (%)	24.3		36.4		6.7		0	
Specificity (%)	90.0		100.0		100.0		85.0	
Efficacy (%)	53.7		39.1		41.7		85.0	

only minor differences before the start of CPT (range 0.0°C – 0.6°C). After Diluent the differences were 0.0°C – 0.4°C and after allergen 0.1°C – 0.7°C (NS). The sensitivity, specificity and efficacy of thermovision in relation to the standard criteria for positive CPT reactions during allergen provocation in all test groups are shown in Table 2.

Discussion

Allergic subjects did not demonstrate greater sensitivity to Albumin Diluent than healthy subjects. This indicates that the atopics with and without allergic rhinoconjunctivitis were not hyperreactive in the eyes outside the pollen season. The satisfactory reproducibility of the CPT was confirmed (5), as all allergic individuals reacted to an allergen concentration within one 10-log on each of 5 occasions. Our finding that the threshold dose was unaffected by the temperature of the test solutions, and intervals between dose increments from 5 to 20 min, may have implications for clinical routine. Thermovision, even when used in a standardized manner, did not improve the evaluation of CPT results.

There are specific features which are possible

sources of disturbance (1, 4). The temperature on the surface of the eye may be reduced by evaporation of the surface tear film. On the other hand, metabolism, vascularity and inflammation produce hyperthermia, resulting in an increased temperature.

Conventional measurement of itching and erythema is easier to evaluate than thermography. Itching appeared before redness in most of the patients and within 8 min after instillation at a concentration giving a positive reaction. If itching had been the only criterion for a positive reaction, so as to shorten the test time and lessen the patient's discomfort, then the threshold dose would have been one step lower in 17 of 39 tests.

Acknowledgments

This study was supported by AGEMA Infrared Systems AB, Danderyd, Sweden.

References

- ALIO J, PADRON M. Normal variations in thermographic pattern of the orbito-ocular regions. *Diagnostic Imaging* 1982; 51: 93–8.
- BONINI S, BERRUTO A, TOMASSINI M, CARLESIMO S, BUCCI M, BALANSO F. Conjunctival provocation test as a model for the study of inflammation in humans. *Int Arch Allergy Appl Immunol* 1988; 88: 144–8.
- GALEN RS, GAMBINO SR. In: Beyond normality: the predictive value and efficiency of medical diagnosis. New York: John Wiley, 1975: 10–11.
- GUIBOR P. Thermography and ophthalmology. 1970: 74: 1032–43.
- MÖLLER C, BJÖRKSTÉN B, NILSSON G, DREBORG S. The precision of the conjunctival provocation test. *Allergy* 1984; 39: 37–41.
- PIPKORN U, BENDE M. Topical levocabastine, a selective H_1 antagonist, in seasonal allergic rhinoconjunctivitis. *Allergy* 1987; 42: 512–15.
- RIMÅS M, KJELLMAN N-IM, BLYCHERT L-O, BJÖRKSTÉN B. Topical levocabastine protects better than sodium cromoglycate and placebo in conjunctival provocation tests. *Allergy* 1990; 45: 18–21.

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