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CLINICAL REPORT

Atopy Patch Test – Reproducibility and Elicitation of Itch in Different Application Sites

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We evaluated the reproducibility of atopy patch test reactions and the quality and quantity of itch in 16 patients with atopic eczema and a history of a positive atopy patch test reaction, comparing three different application sites. The allergen was re-applied simultaneously on both forearms and the back. Intensity and quality of pruritus were evaluated using a visual analogue scale and the Eppendorf itch questionnaire, respectively. The atopy patch test reaction was highly reproducible, occurring in 15/16 (94%) patients. Pruritus was reported by 14/16 (88%) patients. There was no significant difference in either the intensity or quality of itch between the two forearms and the back ($p > 0.05$). The mean peak visual analogue scale itch score was comparable across all three test sites (range 28.3–31.9). Regarding quantification of test reactions, a positive reaction was more frequent on the back (94% versus 69% on the arms) and the peak atopy patch test score was higher on the back compared with the arms (right forearm, $p = 0.0018$ and left forearm, $p = 0.0683$). Allergens should preferably be applied on the back for the atopy patch test. However, the atopy patch test can induce atopic itch irrespective of the application site. ***Key words: atopic eczema; atopy patch test; itch.***

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Atopic eczema (AE), a chronic inflammatory disease, is characterized by an age-related distribution and morphology and often appears together with allergic rhinitis and bronchial asthma (1–3).

AE can also be regarded as a prototypic pruritic disease, as itch is often the primary symptom of this disease (2). Aeroallergens play an important role in inducing eczema flares (4, 5). Immediate-type allergens are considered to penetrate the disturbed skin barrier (6) where they become bound to Langerhans cells and presented to T cells (7). The atopy patch test (APT) is an epicutaneous patch test which uses IgE-inducing allergens from, for example, house dust mite, grass pollen, cat dander or birch pollen, with evaluation of an eczematous skin reaction (4). The first systematic investigations with house dust mite allergen in patients with AE were published by Mitchell et al. in 1982 (8).

The aim of this study was to evaluate the reproducibility of APT reactions. In addition, qualitative and quantitative itch scores and the visual APT reactions were compared at different time points and application sites. The Eppendorf itch questionnaire (EIQ) was used by the patients to evaluate the quality of the itch sensation. The EIQ was developed in analogy to the McGill pain questionnaire and has previously been used to characterize the main components of clinical itch in AE (9). It consists of two forms, one presenting 80 randomized descriptors (sensory, affective, emotional) each scored from '0' (not true) to '4' (exactly true) and the other containing temporary and topographic aspects and a visual analogue scale (VAS).

STUDY DESIGN AND METHODS

Sixteen patients (11 men, 5 women, age 18–64 years, mean age 30.3 years) with a history of AE (2, 3), but in remission at the time of the study and positive APT were investigated in a single-centre, within-patient comparison study. Before enrolment in the study, all patients gave their informed consent. The study was approved by the appropriate ethical committee of the TU Munich. Before the study, patients had been shown to develop positive APT skin reactions to at least one of the following aeroallergens: house dust mite (*D. pteronyssinus, D. farinae*), cat dander, grass pollen or birch pollen, with negative tests to the vehicle control. For patients who had reacted to multiple allergens, the allergen that elicited the most intense reaction was selected. All patients were tested with the same series of allergens. Allergens were tested as follows: house dust mite (11 patients), cat dander (2 patients), grass pollen (2 patients) and birch pollen (1 patient). The lyophilized aeroallergen (200 IRg^{-1} in petrolatum, Stallergenes, France) was applied in 12-mm diameter aluminium Finn chambers (Epitest Ltd, Oy, Finland) to untreated, clinically uninvolved skin; one chamber to the back and simultaneously one to each forearm. The Finn chambers were removed after 48 h.

Evaluation

The APT reaction was read 48 and 72 h after application of the patches according to the ETFAD (European Task Force on Atopic Dermatitis) key (4). For each anatomical site, the

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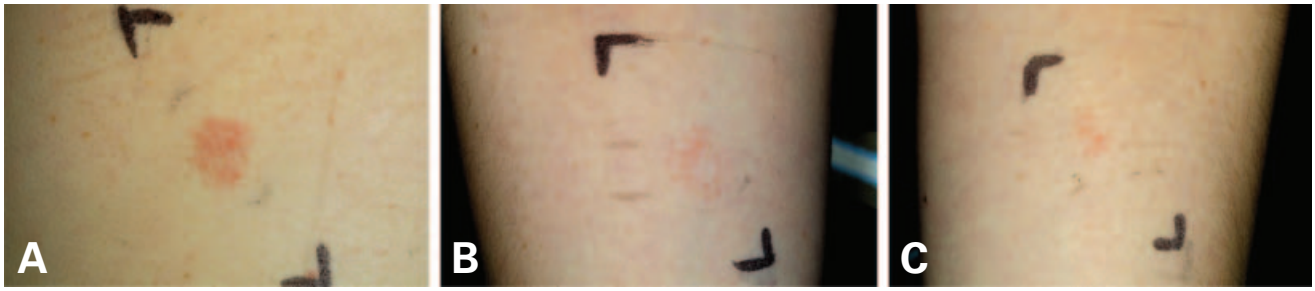

Fig. 1. Moderate atopy patch test reaction to house dust mite (*D. pteronyssinus*/*D. farinae*) in a patient with atopic eczema after 72 h on three different test areas: a, back; b, right forearm; c, left forearm. A lower reaction intensity can be seen on the forearms.

peak APT score and the mean score at the two time points were calculated.

Intensity and quality of pruritus were evaluated by the patients using a 100-mm VAS and the EIQ (9), respectively. For itch intensity, 0 mm on the VAS represents 'not perceptible' and 100 mm represents 'severest itch imaginable' (10). Itch intensity was assessed at baseline and 24, 48 and 72 h after application of the aeroallergen at all three application sites. For each anatomical site, the peak VAS itch score and the mean score at the three time points were calculated. The EIQ was completed 48 and 72 h after allergen application for each site.

In addition, type I sensitizations were evaluated in the skin prick test (SPT) using the same test aeroallergens (Allergopharma, Reinbek, Germany) as in the APT. Levels of IgE antibodies (antigen-specific and total) were also determined using CAP-RAST-FEIA (Pharmacia, Uppsala, Sweden). SPT with a wheal ≥ 3 mm and specific IgE antibodies (against the same aeroallergens as in the APT) with $>0.35\ \mathrm{kUl}^{-1}$ (CAP-Class 1) were defined as positive. The SPT was done and blood was taken on day 0 of the study.

Statistical analysis

Analyses were done using the paired t-test. Differences between pairs of sites (back vs left forearm, back vs right forearm and left forearm vs right forearm) are presented, together with 95% confidence intervals and associated *p* values. All tests were two-tailed and a 5% significance level was used throughout. The statistical package SAS (v8.2) was used as well as the non-parametric tests. In addition, a linear regression analysis was performed to assess the relationship between VAS itch and total Eppendorf score.

RESULTS

Atopy patch test skin reaction

The APT skin reaction showed a high level of reproducibility, as 15/16 (94%) patients who had previously developed a positive APT reaction on the back (average 16 months prior to the study) developed a positive reaction on re-challenge on the back. Only one patient failed to react to the allergen applied on the back but nonetheless this patient developed a positive reaction to the allergen on the left forearm. Thus, a positive APT reaction involving at least one test site was elicited in all patients. An example is shown in Fig. 1.

APT skin reactions were observed on both forearms in 11 patients (69%) at 48 and 72 h. Five patients failed to generate a visible skin reaction at these sites, although itch was recorded for the forearm sites by all five patients.

Evaluation of the mean APT scores at 48 and 72 h demonstrated statistically significant differences between the back and right forearm at both time points with higher scores on the back (mean score differences of 1.0 ($p=0.018$) and 1.2 ($p=0.0031$), respectively). Differences were also observed between mean APT scores for the back and the left forearm (mean differences of 0.8 ($p=0.0929$) and 0.8 ($p=0.0386$), at 48 and 72 h). The mean APT score ranged from 1.7 (right forearm) to 2.8 (back) at 48 h and from 1.3 (right forearm) to 2.5 (back) at 72 h (Fig. 2). In agreement with these findings, the peak APT score was highest on the back with a mean difference of 1.1 against the right forearm ($p=0.0018$) and 0.8 against the left forearm ($p=0.0683$).

Itch intensity (VAS)

An itch sensation was elicited in 15/16 patients (94%) on at least one occasion involving at least one test site. In the one patient failing to report itch, APT skin reactions were observed 48 and 72 h after application of the allergen at all sites. Overall, 14/16 patients (88%) reported pruritus on the back at one or more time points. Similarly, 14/16 patients (88%) had pruritus on one or both forearms at one or more time points (24, 48 and 72 h) (Table I). The mean VAS itch scores at 48 and

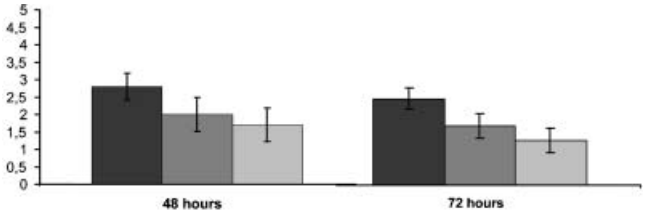

Fig. 2. Atopy patch test (APT) reaction score (mean ± standard error) measured 48 h and 72 h after patch application to the back (■), left arm (■) and right arm (■).

Table I. *Occurrence of pruritus on the back and the forearms of the 16 patients at 24, 48, 72 h after atopy patch test application*

Site	24 h *n* (%)	48 h *n* (%)	72 h *n* (%)
Back	12 (75)	13 (81)	10 (63)
Forearms	13 (81)	13 (81)	9 (56)

72 h showed no significant differences between any of the sites (Fig. 3). The mean VAS itch score ranged from 19.1 (right forearm) to 27.1 (left forearm) at 24 h and from 7.4 (right forearm) to 10.5 (left forearm) at 72 h.

Mean peak VAS itch was similar across all three sites; it ranged from 28.3 mm for the right forearm up to 31.9 mm for the left forearm. A peak VAS itch score ≥ 50 mm was reported by 5/16 patients (31%) following allergen exposure on the back compared with 2/16 patients (12.5%) on both forearms and 5/16 patients (31%) at one or both forearms.

Quality of itch

Evaluation of the quality of itch by the EIQ revealed generally similar findings to the VAS itch intensity data. The mean total Eppendorf score (points) ranged from 22.7 (right forearm) to 25.6 (back) at 48 h and from 5.3 (left forearm) to 6.8 (back) at 72 h. The mean descriptive Eppendorf score (points) ranged from 12.1 (left forearm) to 13.6 (back) at 48 h and from 3.4 (left forearm) to 4.2 (back) at 72 h. The mean emotional Eppendorf score (points) ranged from 10.6 (right forearm) to 12.4 (left forearm) at 48 h and from 1.8 (right forearm) to 2.6 (back) at 72 h. There was some evidence of a positive correlation between VAS itch score and total Eppendorf score at 48 and 72 h (R^2 ranging from 0.37 to 0.64) (Fig. 4). The most frequent items chosen in the EIQ were similar at all three application sites and at both time points: 'itching, disturbing, annoying, unpleasant, tickling, crawling and deterioration in warmth' with the positive control item 'itching' being the first.

Skin prick test and specific IgE

The skin prick test (wheal) was positive in 14/15 (93%) patients for *D. pteronyssinus*, in 13/15 (87%) patients for

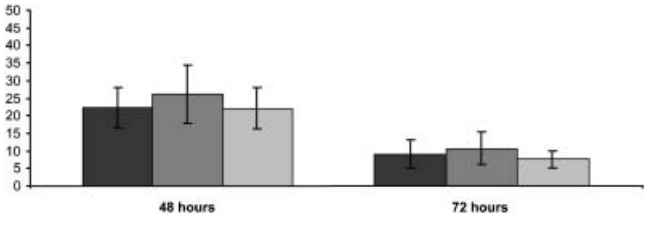

Fig. 3. VAS itch score (mean ± standard error) measured 48h and 72h after patch application to the back (■), left arm (■) and right arm (■).

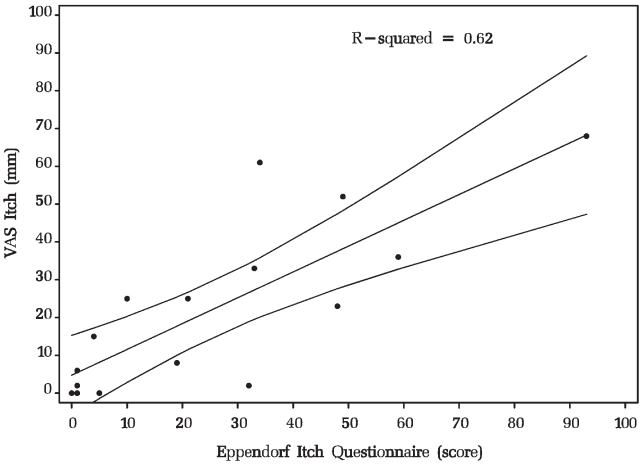

Fig. 4. VAS itch score versus total Eppendorf itch questionnaire score measured at 48 hours after patch application on the back.

D. farinae, in 14/15 (93%) patients for cat dander, in 15/15 (100%) patients for grass pollen and in 12/15 (80%) patients for birch pollen. Overall, 15/16 (94%) patients had elevated total IgE antibody titres, ranging from 69 kUl^{-1} to 14380 kUl^{-1} with a mean value of 3254 kUl^{-1}. There was an agreement between positive skin prick tests and specific IgE antibody titres. Moreover there was a concordance of positive APT with the allergen-specific IgE antibody titre in 12/15 (80%) patients.

DISCUSSION

The reproducibility of an APT may depend on several factors such as concentration, type and number of allergens (4, 11, 12). From classic contact patch testing we know that the reproducibility decreases the more positive reactions have occurred simultaneously at the first test, when tested in close proximity to another positive allergic reaction ('spillover') and when a strong irritant is included in the test series (13, 14). These can be reasons for induction of a generalized state of hyper-reactivity, the so-called 'angry back' syndrome (or excited skin syndrome, ESS). In this state of hyper-reactive skin, doses of substances that normally cause no reaction may lead to an inflammation. Also, a sub-clinical inflammation where eczema is not visible can cause a hyper-reactivity of the skin which again provokes an ESS, which may lead to positive patch tests on initial testing that are negative on retesting (14). All known factors eliciting an ESS were considered in our study, which may have contributed to the high reproducibility. Most of the patients were tested to house dust mite allergen, which exhibits a proteolytic enzyme activity similar to papain and serine proteases (15, 16). This could elicit irritant reactions, which may add to the allergic reactions and explain the higher frequency of positive APT to house dust mite allergen

compared with other allergens. However, most of the patients in our study were positive for house dust mite in skin prick test and CAP-RAST as well. Furthermore, the tests were repeated on the back, which seems to give more reproducible responses than the forearms, and in the screening period only four aeroallergens were applied which could contribute to the high reproducibility. Memon & Friedmann (17) elicited a similar high reproducibility rate of 90–95% in nickel allergic patients using classical (not APT) patch testing on the back. Heinemann et al. (18) recently described a rather low reproducibility rate of 56.3% in their APT model. They tested aeroallergens on the back and retested the same allergen on the forearms 4–12 weeks later. In our study, the frequency and intensity of positive APT reaction were statistically significantly higher on the back than on the forearms, but were similar on both arms. Memon & Friedmann (17) also found that the forearms were clearly less responsive than the back (40% of the sequential testings with nickel on the forearms were non-reproducible). Magnusson & Hersle (19) explain such differences by pressure variations on the patches at the various application sites. Increased pressure on the back by lying in bed may enhance a patch test of an allergen. Another reason could be a higher percutaneous absorption through back skin due to higher density of sebaceous glands and hair follicles.

To maximize the reproducibility of the allergen-specific APT reaction, patches should be applied to the back. As with classical patch tests, reactions were more frequent and more intense on the back compared with the arms.

Relating to the reliability of the APT the validity of the test results should also be discussed. APT is a model for AE (12), a pruriginous disease (2, 3). Atopic itch can be induced by this model in different locations and with defined time course.

In contrast to the APT reactions, there was no significant difference according to the quality of itch (mean EIQ scores) and to itch intensity (VAS mean itch score and the peak VAS itch score) between the application sites. Five patients failed to generate a positive APT reaction on both arms. Nevertheless three of them recorded mild pruritus at these test sites. It may be speculated that subclinical inflammation occurs at an APT site to trigger pruritus without visible signs of a reaction. On the other hand, it is possible that the adhesive tape per se elicited an itch sensation which for the patients was not distinguishable from aeroallergen-induced pruritus. Fig. 3 shows that there was a tendency to higher itch ratings on the left compared with the right arm, but this was not statistically significant. Mean scores for the APT reaction at each site were somewhat lower at 72 h than at 48 h. These findings agree with those of Langeveld-Wildschut et al. (12) and our own earlier results (11) evaluating some of the variables influencing the outcome of different APT models. The maximal number of positive APT results was recorded at 48 h. In contrast to the APT reaction the reduction of quality and quantity of itch from 48 to 72 h was significant. A correlation between the quantity and the quality of itch could be demonstrated both for AE and for the APT as inflammation model. A correlation between VAS itch scores and qualitative EIQ items could also be shown in a histamine model for pruritus (10). Perhaps the inflammation and release of mediators decreases despite a persisting visible APT reaction, resulting in a decrease of pruritus. Another possibility is an adaption and habituation to the pruritus.

This study involved 11 patients with reproducible APT reactions to house dust mite. The question as to clinical relevance of these reactions can only be answered by larger trials using specific provocation and avoidance measures; however, these are of controversial efficacy (20, 21). Nevertheless, the APT may be a valuable instrument in characterizing patients who benefit more from allergen avoidance.

To our knowledge this is the first study investigating the quality of itch at APT. We used both qualitative (EIQ) and quantitative (VAS) scales to measure itch intensity. The quality of itch remained the same regardless of the test location and the two time points, as judged by the choices of describing adjectives. Similarly the intensity of itch was comparable on all three application sites. The intensity of the APT skin reactions on the left arm was marginally higher than on the right arm, whereas EIQ scores were similar for both locations. The intensities of itch and the visual APT reaction decreased from 48 to 72 h after allergen application. These observations again correspond to the EIQ scores. These data contribute to the validation of the EIQ for use in clinical studies of pruritus associated with AE.

REFERENCES

1. Ring J. Atopy: condition, disease or syndrome? In: Ruzicka T, Ring J, Przybilla B, eds. Handbook of atopic eczema. Berlin: Springer, 3–8.
2. Hanifin JM. Basic and clinical aspects of atopic dermatitis. Ann Allergy 1984; 52: 386–395.
3. Hanifin JM, Rajka G. Diagnostic features of atopic dermatitis. Acta Derm Venereol 1980; Suppl 114: 146–148.
4. Darsow U, Ring J. Airborne and dietary allergens in atopic eczema: a comprehensive review of diagnostic tests. Clin Exp Dermatol 2000; 25: 544–551.
5. Morren MA, Przybilla B, Bamelis M, Heykants B, Reynaers A, Degreef H. Atopic dermatitis: triggering factors. J Am Acad Dermatol 1994; 31: 467–473.
6. Gondo A, Saeki N, Tokuda Y. Challenge reactions in atopic dermatitis after percutaneous entry of mite antigen. Br J Dermatol 1986; 115: 485–493.
7. Maurer D, Ebner C, Reininger B, Fiebiger E, Kraft D, Kinet JP, et al. The high affinity IgE receptor mediates IgE-dependent allergen presentation. J Immunol 1995; 154: 6285–6290.

8. Mitchell EB, Crow J, Chapman MD, Jouhal SS, Pope FM, Platts-Mills TA. Basophils in allergen-induced patch test sites in atopic dermatitis. Lancet 1982; I: 127–130.
9. Darsow U, Scharein E, Simon D, Walter G, Bromm B, Ring J. New aspects of itch pathophysiology: component analysis of atopic itch using the 'Eppendorf Itch Questionnaire'. Int Arch Allergy Immunol 2001; 124: 326–331.
10. Darsow U, Ring J, Scharein E, Bromm B. Correlations between histamine-induced wheal, flare and itch. Arch Dermatol Res 1996; 288: 436–441.
11. Darsow U, Vieluf D, Ring J. Atopy patch test with different vehicles and allergen concentrations – an approach to standardization. J Allergy Clin Immunol 1995; 95: 677–684.
12. Langeveld-Wildschut EG, van Marion AM, Thepen T, Mudde GC, Bruijnzeel PL, Bruijnzeel-Koomen CA. Evaluation of variables influencing the outcome of the atopy patch test. J Allergy Clin Immunol 1995; 96: 66–73.
13. Brasch J, Henseler T, Aberer W, Bäuerle G, Frosch PJ, Fuchs T, et al. Reproducibility of patch tests. A multicenter study of synchronous left- versus right-sided patch tests by the German Contact Dermatitis Research Group. J Am Acad Dermatol 1994; 31: 584–591.
14. Mitchell J, Maibach HI. Managing the excited skin syndrome: patch testing hyperirritable skin. Contact Dermatitis 1997; 37: 193–199.
15. Deleuran M, Ellingsen AR, Paludan K, Schou C, Thestrup-Pedersen K. Purified Der p1 and p2 patch tests in patients with atopic dermatitis: evidence for both allergenicity and proteolytic irritancy. Acta Derm Venereol 1998; 78: 241–243.
16. Shah D, Hales J, Cooper D, Camp R. Recognition of pathogenically relevant house dust mite hypersensitivity in adults with atopic dermatitis: a new approach? J Allergy Clin Immunol 2002; 109: 1012–1018.
17. Memon AA, Friedmann PS. Studies on the reproducibility of allergic contact dermatitis. Br J Dermatol 1996; 134: 208–214.
18. Heinemann C, Schliemann-Willers S, Kelterer D, Metzner U, Kluge K, Wigger-Alberti W, et al. The atopy patch test – reproducibility and comparison of different evaluation methods. Allergy 2002; 57: 641–645.
19. Magnusson B, Hersle K. Patch test methods II. Regional variations of patch test responses. Acta Derm Venereol 1965; 45: 257–261.
20. Koopman LP, van Strien RT, Kerkhof M, Wijga A, Smit HA, de Jongste JC, et al. Prevention Incidence of Asthma and Mite Allergy (PIAMA) Study. Placebo-controlled trial of house dust mite-impermeable mattress covers: effect on symptoms in early childhood. Am J Respir Crit Care Med 2002; 166: 307–313.
21. Gutgesell C, Heise S, Seubert S, Seubert A, Domhof S, Brunner E, et al. Double-blind placebo-controlled house dust mite control measures in adult patients with atopic dermatitis. Br J Dermatol 2001; 145: 70–74.