Pathergy Test: The Comparison of Clinical vs. Histopathological Evaluation

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Objective: *Behcet's disease is an inflammatory disease of unknown etiology. Pathergy test is the only diagnostic test for Behcet's disease. The evaluation can be done either clinically and/or histopathologically. In the present study, we compare the sensitivity and specificity of clinical vs. histopathological evaluation of the pathergy test.*

Material and Method: *This was a retrospective study in patients who underwent pathergy tests at Phramongkutklao Hospital, Thailand between January 1, 2011 and December 31, 2013. Fifty-eight cases met the inclusion criteria and were included into the study. All basic demographic data were obtained from the medical records. The sensitivity, specificity, and accuracy of the test were evaluated.*

Results: *There were 33/58 (56.9%) cases with the final diagnosis as Behcet's disease. The sensitivity, specificity, and accuracy of the clinical evaluation of pathergy test were 30.3%, 64%, and 44.8% respectively. Upon using the histopathological evaluation, the sensitivity, specificity, and accuracy were 100%, 16%, and 63.8%, respectively.*

Conclusion: *Our results showed that the histopathological evaluation of pathergy test helped to improve the accuracy and sensitivity, but the specificity was low. We suggest the use of histological evaluation of the pathergy test in cases where Behcet's disease is highly suspected especially in areas where the disease is uncommon.*

Keywords: *Behcet's disease, Pathergy test, Specificity, Sensitivity*

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Behcet's disease is a multisystem inflammatory disease of unknown etiology[1,2]. The prevalence is highest in the Eastern and Central Asian, and the Eastern Mediterranean countries along the so-called "Silk Road".

The diagnosis is based mainly on clinical signs. Several sets of diagnostic criteria exist. Nevertheless, the most popular one is the criteria from the "International Study Group" (ISG) of Behcet's disease[1,3,4]. The ISG criteria consist of a major criteria of recurrent oral ulcers, at least three times within a 12-month period, and two or more of the minor criteria i.e., recurrent genital ulcer, eye lesions (anterior or posterior uveitis or retinal vasculitis), cutaneous lesions (erythema nodosum-like lesion, papulopustular lesion, pseudofolliculitis, or acneiform lesion), and positive pathergy test[1,3,4].

Pathergy test is the only diagnostic test for Behcet's disease[3-7]. The pathogenesis of pathergy phenomenon is the exaggerated cutaneous inflammatory response to minimal skin trauma. It is explained through the increase or excessive release of cytokines from the cells in the epidermis and dermis[7-9]. These result in the finding of inflammatory cells infiltration in the skin biopsy[8]. This test is not specific since positive pathergy test can be found in other inflammatory diseases such as pyoderma gangrenosum, Sweet's syndrome, and inflammatory bowel disease[7,9-13].

There are two types of pathergy test i.e., oral pathergy and skin pathergy test[7,14]. Skin pathergy test is more popular. The evaluation of skin pathergy test is made at 48 hours after implementing the test, which can be interpreted via clinical and/or histopathological reaction[7,15,19]. Various histopathological findings have been reported. The main histopathologic finding consisted of neutrophils with or without other inflammatory cells involved in the test area[8,15,16,19]. The density and severity of inflammatory cells ranged from perivascular mononuclear cells infiltration with minimal neutrophils infiltration[8], dense perivascular and interstitial mixed cells infiltration with predominantly neutrophils[15], or the presence of leukocytoclastic vasculitis[16]. The difference in histopathological findings can be explained by the diversity of individual immune response to the stimulating agents[17].

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In the present study, we evaluated the accuracy, sensitivity, and specificity of clinical versus histopathological evaluation of pathergy test in the diagnosis of Behcet's disease. In addition, we evaluated the correlation of Behcet's disease activity and the positive pathergy test.

Material and Method

The study included 58 patients suspected of Behcet's disease and that underwent pathergy test between January 1, 2011 and December 31, 2013 at Phramongkutklao Hospital, Bangkok, Thailand.

In each case, the pathergy test was performed on the flexural aspect of the forearm. A sterile disposable sharp needle for injection No. 21 gauge was inserted obliquely at an angle of 45 degrees to a depth of 3 to 5 mm without any prior application of disinfectant[2,7]. Intradermal injection of 0.1 ml isotonic saline solution was done[2,7,18]. The evaluation was done at 48 hours via clinical evaluation and skin biopsy was done simultaneously for histopathological evaluation. The clinical positive pathergy test was defined as the presence of erythematous papule or pustule. Erythema without skin infiltration is considered negative[2]. The histopathological evaluation was done in every case after the clinical evaluation. The positive histopathology test was defined as the presence of neutrophilic infiltration in the dermis or subcutaneous tissue with or without leukocytoclastic vasculitis[8,16,19].

For each patient, a retrospective review of the available medical records was obtained. Basic demographic data included age, gender, clinical presentation, indication for pathergy test, clinical and histopathological evaluation of pathergy test, final diagnosis, patient status, and disease status. The diagnosis of Behcet's disease from the medical records was based mainly on the ISG criteria as mention above. The patients who did not fulfilled the ISG criteria were diagnosed as non-Behcet's disease.

The present study was approved by the Institutional Ethical Committee. All patients included were notified and consents were obtained.

Statistical methods

Sensitivity, specificity, and accuracy of the clinical evaluation and histopathological evaluation were calculated with respective of 95% confidence interval. The correlation of Behcet's disease activity and the positive or negative pathergy test were calculated with Mann-Whitney test. All the statistics were analyzed by SPSS, version 17.0.

Results

The basic demographic data from the 58 cases were shown in the Table 1. The age at first visit ranged from 28 to 50 years, median 39.25 years. Twenty-seven (27/58, 46.6%) were male and thirty-one (31/58, 53.4%) were female (male:female = 0.87). Oral ulcers were the most common clinical presentation, followed by eye lesions, which were uveitis (anterior, posterior, or panuveitis), scleritis, and vasculitis (Table 1). Eye lesions were the most frequent presenting symptom that physicians requested the patients to undergo pathergy tests. Of the 58 cases who underwent pathergy test, 33 cases (56.9%) had final diagnosis as Behcet's disease (Table 1). The median time of follow-up period in all cases was 24.29 months (0.26-183.15 months, 1.14-786.43 weeks).

The clinical positive pathergy test was found in 19/58 (32.8%) cases. Histopathological positive result was found in 54/58 skin biopsies (93.1%). Eighteen cases (18/58, 31.03%) had positive results in both clinical and histopathological evaluation.

Table 1. The basic demographic data of the cases

Variables	n (%) or median (range)
Age at first visit (years)	39.25 (28.82-50.14)
Sex (n = 58)	
Male	27 (46.6)
Female	31 (53.4)
Clinical presentations (n = 58)	
Oral ulcer	35 (60.3)
Eye lesions	28 (48.3)
Arthritis/musculoskeletal symptoms	15 (25.86)
Skin lesions (EN-like, Sweet-like, pseudofolliculitis/acneiform)	13 (22.4)
Genital ulcer	10 (17.2)
Small/medium vessel involvement, not related tovital organs*	8 (13.8)
Major vessel involvement, related to vital organs*	7 (12.1)
GI symptoms	3 (5.71)
Vital organs* involvement	2 (3.4)
Clinical reason for pathergy test (n = 58)	
Eye lesions	26 (44.8)
Oral ulcer	19 (32.8)
Skin lesions	5 (8.6)
Arthritis/musculoskeletal symptoms	4 (6.9)
GI symptoms	2 (3.4)
Genital ulcer	1 (1.7)
Final diagnosis of Behcet's disease (n = 58)	
Yes	33 (56.9)
No	25 (43.1)

EN = erythema nodosum; GI = gastrointestinal
* Vital organs included brain, lung, heart, and kidney

Thirty-six cases (36/58, 62.06%) had negative clinical evaluation but positive histopathological evaluation. The negative results from both clinical and histopathological evaluation were found in three cases (3/58, 5.17%).

In the 33 cases of Behcet's disease, only 10 cases had positive clinical evaluation while all of them had positive histopathological evaluation (Table 2). None of the negative histopathologic evaluation had final diagnosis as Behcet's disease. On the other hand, 23 cases of negative clinical evaluation had final diagnosis as Behcet's disease (Table 2).

The sensitivity and specificity of the clinical evaluation were 30.3% and 64% respectively, while the accuracy was 44.8% (Table 3). In contrast, sensitivity, and specificity of histopathological evaluation were 100% and 16% respectively, while the accuracy was 63.8% (Table 3).

Of the 25 cases of non-Behcet's disease, 22 cases had positive pathergy tests with either clinical or histopathological evaluation alone. Their final diagnoses in these cases were shown in Table 4. The most common diagnosis was eye diseases, followed by aphthous ulcers and other inflammatory skin diseases.

Table 2. Pathergy test, clinical and histopathological evaluations and final diagnosis of Behcet's disease

	Behcet's disease (n = 58), n	
	Yes (n = 33)	No (n = 25)
Clinical evaluation (n = 58)		
Positive (n = 19)	10	9
Negative (n = 39)	23	16
Histopathological evaluation (n = 58)		
Positive (n = 54)	33	21
Negative (n = 4)	0	4

Discussion

Pathergy test is the only test in the diagnostic criteria for Behcet's disease. It was first described by Blobner in the year 1937[20]. Since then, the advantage of pathergy test had been confirmed[4,6]. Studies showed an increase in sensitivity after adding pathergy test in the diagnostic criteria of Behcet's disease[4]. Moreover, there were decrease in specificity and accuracy after disregarding the pathergy test[6]. Various specificity and sensitivity of pathergy test have been reported, ranging from 18 to 80% depending on race and countries[6,7,21,22,24-26]. The rate of positive pathergy test is highest in the Middle East, approximately 60% and lowest among western countries[27].

Table 3. Sensitivity, specificity and accuracy of pathergy test, clinical and histopathological evaluation

	Clinical evaluation (%)	95% CI		Histopathological evaluation (%)	95% CI	
		Lower limit	Upper limit		Lower limit	Upper limit
Sensitivity	30.3	0.2	0.5	100.0	0.9	1.0
Specificity	64.0	0.4	0.8	16.0	0.0	0.4
Positive predictive value	52.6	0.3	0.7	61.1	0.5	0.7
Negative predictive value	41.0	0.2	0.6	100.0	0.4	1.0
Agreement (accuracy)	44.8			63.8		

95% CI = 95% confidence interval

Table 4. The final diagnoses of non-Behcet's disease with positive pathergy test (either clinical histopathological evaluations)

Final diagnoses	n = 22, n (%)
Eye diseases (uveitis, retinal vasculitis, toxoplasma retinitis)	6 (27.27)
Oral aphthous ulcers	3 (13.64)
Other inflammatory skin diseases (prurigopigmentosa, pemphigus vulgaris, suppurative lobular panniculitis)	3 (13.64)
Rheumatologic diseases (panlindromic, myofacial pain)	2 (9.09)
Vasculitis	2 (9.09)
Reactive arthritis/psoriatic arthritis	2 (9.09)
Unidentified connective tissue diseases	1 (4.54)
Pyodermagangrenosum	1 (4.54)
Non-specific genital ulcers	1 (4.54)
Inflammatory bowel diseases	1 (4.54)

Pathergy test can be evaluated clinically and/or histopathologically. However, only clinical evaluation was included into the ISG criteria for Behcet's disease[1,3]. Our study showed an obvious difference in sensitivity between clinical and histopathological evaluation. The sensitivity of histopathological evaluation was substantially higher comparing to clinical evaluation (Table 3). Although the specificity of histopathological evaluation was low, the accuracy was high. Hence, our results supported the use of histopathological evaluation over the clinical evaluation. A recent study from Akmaz et al[19] found no difference in sensitivity between clinical and histopathological evaluation, however the procedure was different i.e., the use of non-disposable/blunt needle technique. In our study, the use of a disposable/sharp needle could result in lower positive rate in clinical evaluation comparing to the use of blunt needle[19,29]. The clinically negative but histologically positive results could be explained by the minimal skin change after minor trauma that was difficult to be seen by the naked eyes, however the process of inflammation could have evolved and was detectable histopathologically[8].

The positive pathergy test was reported to be at 33% for Behcet's disease in Thailand[28]. However, the evaluation method of the pathergy test was not clearly delineated and the tests were not done in all of the recruited cases (only 9/23 cases). Our study, in practically the same population, showed higher rate of positive pathergy test (56.89%) as evaluated histopathologically for Behcet's disease. We believe the inclusion of positive pathergy test, as evaluated by histopathology, should be used instead of the clinical evaluation as indicated in the ISG criteria for Behcet's disease among the Thai population.

The correlation between positive pathergy test and disease activity is shown in Table 5. Of the 33 Behcet's disease cases, 10 cases were lost to follow-up. Therefore, the disease status was available in 23 cases. The statistical result showed there was no correlation between positive pathergy test as evaluated clinically and the activity of the disease (p = 0.142). Owing to the zero number of the negative case, the correlation of disease activity and positive pathergy test as evaluated histopathologically could not be calculated. However, previous studies showed the relationship of histopathological positive pathergy test with both the disease activity[16,18,19] and clinically active vascular involvements[18,27]. The discrepancy in these findings could probably be explained by the difference in racial groups. Some studies have been done in the Middle East population that had higher incidence of Behcet's disease and therefore, higher rate of positive pathergy test[16,19,27]. In addition, there were differences in the study methods[16,18,19,27]. Certain prior studies were done in patients who were already diagnosed as Behcet's disease. Our study looked retrospectively in a population that had never been diagnosed as Behcet's disease. Most of the cases in our study did not fulfill the ISG criteria for Behcet's disease when they underwent the pathergy test. Another difference is the technique used in doing the pathergy test i.e., the use of non-disposable/blunt needle vs. our use of disposable/sharp needle as mentioned above.

Twenty two of our cases with positive pathergy test were not diagnosed as Behcet's disease (22/58, 37.94%) (Table 5). All of them had inflammatory diseases. These findings support the non-specificity of the pathergy test. As in many prior reports, positive pathergy test could be found in many inflammatory diseases, especially the neutrophilic dermatoses[7,9-13]. All our cases with negative pathergy test both clinically and histopathologically were not diagnosed as Behcet's disease. We can assume from our study that these negative pathergy cases are less likely to become Behcet's disease.

Although eye lesions were the most common presenting symptoms that clinicians requested for pathergy test in our study (Table 1), oral ulcer was the most common clinical presentation. It was found in more than 60% of the cases. These results get along well with the ISG criteria whereby oral ulcer is the only major criteria.

The limitations of the present study were the relatively small number of subjects, data lost due to lost follow-up, single-center site, and relatively short follow-up period.

Table 5. The follow-up of Behcet's disease activity and positive pathergy test (n = 23*)

Pathergy test	Active (n = 18)		Non-active (n = 5)		p-value
	n	%	n	%	
Clinical evaluation					
Positive	4	57.1	3	42.9	0.142
Negative	14	87.5	2	12.5	
Histopathological evaluation					
Positive	18	78.3	5	21.7	NA
Negative	0	0	0	0	

NA = not available

* 33 cases of Behcet's disease, 10 cases were lost to follow-up

Conclusion

The present study helps to confirm the advantage of histopathological evaluation of the pathergy test, which can improve both sensitivity and accuracy in the diagnosis of Behcet's disease. We would recommend the use of histopathological evaluation of the pathergy test in every suspected case of Behcet's disease, especially in areas where the disease is uncommon.

What is already known on this topic?

Pathergy's test can be evaluated in both clinical evaluation and/or histopathological evaluation. Nevertheless, clinical evaluation of pathergy test is the only laboratory test in the ISG criteria for diagnosis Behcet's disease.

What this study adds?

Histologic evaluation can improve the sensitivity and accuracy for the diagnosis of Behcet's disease.

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Potential conflicts of interest

None.

References

1. Moschella SL, Davis M DP. Neutrophilicdermatosis. In: Bolognia JL, Jorizzo JL, Schaffer JV, editors. Dermatology. 3rd ed. New York: Elsevier Sauders; 2012: 423-38.
2. Zouboulis CC. Adamantiades-Behcet disease. In: Goldsmith LA, Katz SI, Gilchrest BA, Paller AS, Leffell DJ, Wolff K, editors. Fitzpatrick's dermatology in general medicine. 8th ed. New York: McGraw-Hill; 2012: 2033-41.
3. Criteria for diagnosis of Behcet's disease. International Study Group for Behcet's Disease. Lancet 1990; 335: 1078-80.
4. Davatchi F. Diagnosis/classification criteria for Behcet's disease. Patholog Res Int 2012; 2012: 607921.
5. Tunc R, Uluhan A, Melikoglu M, Ozyazgan Y, Ozdogan H, Yazici H. A reassessment of the International Study Group criteria for the diagnosis (classification) of Behcet's syndrome. Clin Exp Rheumatol 2001; 19 (5 Suppl 24): S45-7.
6. Davatchi F, Sadeghi AB, Chams-Davatchi C, Shahram F, Ghodsi Z, Nadji A, et al. Impact of the positive pathergy test on the performance of classification/diagnosis criteria for Behcet's disease. Mod Rheumatol 2013; 23: 125-32.
7. Sequeira FF, Daryani D. The oral and skin pathergy test. Indian J Dermatol Venereol Leprol 2011; 77: 526-30.
8. Gül A, Esin S, Dilsen N, Konice M, Wigzell H, Biberfeld P. Immunohistology of skin pathergy reaction in Behcet's disease. Br J Dermatol 1995; 132: 901-7.
9. Varol A, Seifert O, Anderson CD. The skin pathergy test: innately useful? Arch Dermatol Res 2010; 302: 155-68.
10. Su WP, Davis MD, Weenig RH, Powell FC, Perry HO. Pyoderma gangrenosum: clinicopathologic correlation and proposed diagnostic criteria. Int J Dermatol 2004; 43: 790-800.
11. Bi XL, Gu J, Yan M, Gao CF. A case of Sweet's syndrome with slack skin and pathergy phenomenon. Int J Dermatol 2008; 47: 842-4.
12. Hsu PJ, Huang CJ, Wu MT. Pathergy in atypical eosinophilic pustular folliculitis. Int J Dermatol 2005; 44: 203-5.
13. Hatemi I, Hatemi G, Celik AF, Melikoglu M, Arzuhal N, Mat C, et al. Frequency of pathergy phenomenon and other features of Behcet's syndrome among patients with inflammatory bowel disease. Clin Exp Rheumatol 2008; 26: S91-S95.
14. Sharquie KE, Al Araji A, Hatem A. Oral pathergy test in Behcet's disease. Br J Dermatol 2002; 146: 168-9.
15. Ergun T, Gurbuz O, Harvell J, Jorizzo J, White W. The histopathology of pathergy: a chronologic study of skin hyperreactivity in Behcet's disease. Int J Dermatol 1998; 37: 929-33.
16. Jorizzo JL, Solomon AR, Cavallo T. Behcet's syndrome. Immunopathologic and histopathologic assessment of pathergy lesions is useful in diagnosis and follow-up. Arch Pathol Lab Med 1985; 109: 747-51.
17. Gilhar A, Winterstein G, Turani H, Landau J, Etzioni A. Skin hyperreactivity response (pathergy) in Behcet's disease. J Am Acad Dermatol 1989; 21: 547-52.
18. Chang HK, Cheon KS. The clinical significance of a pathergy reaction in patients with Behcet's disease. J Korean Med Sci 2002; 17: 371-4.
19. Akmaz O, Erel A, Gurer MA. Comparison of histopathologic and clinical evaluations of

pathergy test in Behcet's disease. Int J Dermatol 2000; 39: 121-5.
20. Blobner F. Zur rezidivierenden hypopyoniritis. Z Augenheilkunde 1937; 91: 129-39.
21. Friedman-Birnbaum R, Bergman R, Aizen E. Sensitivity and specificity of pathergy test results in Israeli patients with Behcet's disease. Cutis 1990; 45: 261-4.
22. Benamour S, Zeroual B, Bennis R, Amraoui A, Bettal S. Behcet's disease. 316 cases. Presse Med 1990; 19: 1485-9.
23. Al Rawi ZS, Sharquie KE, Khalifa SJ, Al Hadithi FM, Munir JJ. Behcet's disease in Iraqi patients. Ann Rheum Dis 1986; 45: 987-90.
24. Davatchi F, Shahram F, Akbarian M, Davatchi CC, Shams H, Nadji A, et al. Behcet's disease-analysis of 3443 cases. APLAR J Rheumatol 1997; 1: 2-5.
25. Zouboulis CC, Kotter I, Djawari D, Kirch W, Kohl PK, Ochsendorf FR, et al. Epidemiological features of Adamantiades-Behcet's disease in Germany and in Europe. Yonsei Med J 1997; 38: 411-22.
26. al Dalaan AN, al Balaa SR, el Ramahi K, al Kawi Z, Bohlega S, Bahabri S, et al. Behcet's disease in Saudi Arabia. J Rheumatol 1994; 21: 658-61.
27. Alli N, Gur G, Yalcin B, Hayran M. Patient characteristics in Behcet disease: a retrospective analysis of 213 Turkish patients during 2001-4. Am J Clin Dermatol 2009; 10: 411-8.
28. Arromdee E, Tanakitivirul M. Epidemiology of Behcet's disease in Thai patients. J Med Assoc Thai 2006; 89 (Suppl 5): S182-6.
29. Dilsen N, Konice M, Aral O, Ocal L, Inanc M, Gul A. Comparative study of the skin pathergy test with blunt and sharp needles in Behcet's disease: confirmed specificity but decreased sensitivity with sharp needles. Ann Rheum Dis 1993; 52: 823-5.

***Pathergy test:* การประเมินผลทางอาการแสดงเทียบกับการประเมินทางจุลพยาธิวิทยา**

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วัตถุประสงค์: *โรค Behcet เป็นโรคที่เกิดจากการอักเสบเรื้อรังของร่างกายเกิดได้กับหลายอวัยวะโดยที่ไม่ทราบสาเหตุ ไม่มีการตรวจทางห้องปฏิบัติการหรือการตรวจพิเศษใด ๆ ที่จำเพาะและใช้ในการวินิจฉัยโรคนี้ ยกเว้นการตรวจด้วยวิธีที่เรียกว่า pathergy test ซึ่งการประเมินผลในการตรวจวิธีนี้สามารถประเมินโดยอาการแสดงทางคลินิก และ/หรือ ร่วมกับการประเมินทางจุลพยาธิวิทยา การศึกษานี้ได้ศึกษาเปรียบเทียบความไวความจำเพาะและความถูกต้องในการประเมินผล pathergy test ทั้ง 2 วิธี*

วัสดุและวิธีการ: *การศึกษานี้เป็นการศึกษาแบบย้อนหลังในผู้ป่วยที่ได้รับการทำ pathergy test ที่โรงพยาบาลพระมงกุฎเกล้า ประเทศไทย ในระหว่างวันที่ 1 มกราคม พ.ศ. 2554 ถึง 31 ธันวาคม พ.ศ. 2556 ซึ่งพบผู้ป่วยที่มีเกณฑ์เข้าได้ทั้งหมด 58 ราย ข้อมูลทั้งหมดเกี่ยวกับอาการ อาการแสดงผลการตรวจทางห้องปฏิบัติการ การวินิจฉัยของผู้ป่วยได้ถูกรวบรวมไว้นำมาวิเคราะห์และคำนวณเปรียบเทียบความไวและความจำเพาะในการประเมินผล pathergy test*

ผลการศึกษา: *พบว่าการประเมินผลทางจุลพยาธิวิทยามีความไวและความถูกต้องมากกว่าการประเมินผลทางอาการแสดงแต่จะมีความจำเพาะต่ำกว่า*

สรุป: *การศึกษานี้แสดงว่าในการตรวจ pathergy การประเมินผลด้วยวิธีทางจุลพยาธิวิทยาช่วยในการเพิ่มความไวและความถูกต้องของการประเมินผล การศึกษานี้สนับสนุนให้ใช้การประเมินทางจุลพยาธิวิทยาร่วมด้วยในการตรวจ pathergy*
