Significance of Anti-deoxyribonuclease-B (ADN-B) Determination in Clinical Practice

SATOSHI FUJIKAWA, M.D.¹, SEIICHI KAWAKITA, M.D.², NOZOMI KOSAKAI, M.D.³
TEIICHI ODA, M.D.⁴, MASAHIKO OHKUNI, M.D.⁵, YUICHI SHIOKAWA, M.D.⁶
NOBUO WATANABE, M.D.⁷, AND TOSHIHIKO YAMADA, M.D.⁸

The determination of anti-deoxyribonuclease-B (ADN-B) is very important for the diagnosis of antecedent streptococcal infection because almost all of group A streptococci have this antigen and a strong elevation of the antibody is observed in patients with acute rheumatic fever, acute glomerulonephritis and other streptococcal infections. Moreover, ADN-B titers are elevated in cases with streptococcal skin infection, whereas anti-streptolysin O (ASO) titers are rarely elevated in these cases. So the determination of ADN-B is recommended as the second streptococcal antibody test to ASO detection (manual of WHO). In our study, the upper limits of ADN-B in normal subjects were 1:60 in preschool age, 1:480 in school age and 1:340 in adult age groups. In acute rheumatic fever, ADN-B titers were elevated in 87% of the cases and also 87% of the patients with acute glomerulonephritis had high ADN-B titers. In inactive rheumatic fever, 72% of the patients had high ADN-B titers, while, ASO titers were elevated in only 22% of the cases. In streptococcal carrier states, 39% of the children had high ADN-B titers but ADN-B was positive only 8% in non-carrier children.

GROUP A streptococci produce various kinds of extracellular antigens including streptolysin O, streptokinase, hyaluronidase, deoxyribonuclease. Antibody determinations for these

antigens are clinically designated as anti-streptolysin O (ASO), anti-streptokinase (ASK), anti-hyaluronidase (AHD) and anti-deoxyribonuclease-B (ADN-B).

Group A streptococci produce four types of deoxyribonuclease, namely, A, B, C and D. Type B is the most important among them because almost all of group A streptococci produce this antigen in a larger amount than the others.¹ Thus,

Key Words:

Anti-deoxyribonuclease-B (ADN-B)
Anti-streptolysin O (ASO)
Rheumatic fever
Acute glomerulonephritis

- 1) Department of Pediatrics, Tokyo Kyosai Hospital
- 2) Department of Internal Medicine, Shiga University of Medical Science
- 3) Department of Clinical Pathology, Juntendo University
- 4) Department of Pediatrics, Fukuoka University
- 5) Department of Pediatrics, Nihon University
- 6) Department of Internal Medicine, Juntendo University
- 7) Department of Pediatrics, Kyorin University
- 8) Department of Microbiology, Yamanashi Medical School

This work was carried out as a cooperative study by the Committee for the Prevention of Rheumatic Fever and Rheumatic Heart Disease (Chairman: Seiichi Kawakita) of the Japanese Circulation Society.

Address for correspondence: Satoshi Fujikawa, M.D., Tokyo Kyosai Hospital, 2-3-8 Nakameguro, Meguro-ku, Tokyo 153, Japan

TABLE I DISTRIBUTION OF ADN-B TITERS IN CONTROL SUBJECTS

Age	Number	Titer									
		≥ 60	85	120	170	240	340	480	680	960	≥ 1360
~ 5	40	35 (100.0)	2 (12.5)	(7.5)	(7.5)	(7.5)	1 (7.5)	1 (5.0)	1 (2.5)	(0)	(0)
6 ~ 15	198	47 (100.0)	5 (76.3)	17 73.7)	31 (65.2)	24 (49.5)	26 (37.4)	16 (24.2)	9 (16.2)	7 (11.6)	16 (9.1)
16 ~	40	13 (100.0)	4 (67.5)	7 (57.5)	2 (40.0)	6 (35.0)	2 (20.0)	3 (15.0)	1 (7.5)	1 (5.0)	1 (5.0)
Total	278	95 (100.0)	11 (65.8)	24 (61.9)	33 (53.2)	30 (41.4)	29 (30.6)	20 (20.1)	11 (12.9)	8 (9.0)	17 (7.2)

() cumulative percentage

the determination of ADN-B is particularly important in the diagnosis of streptococcal infections.

The changing pattern of antibody to DN-B is comparable to that of ASO in any given case. However, ADN-B determination offers the following advantages: 1) A strong elevation of the antibody is observed in cases with streptococcal skin disease, whereas ASO titer rarely shows high levels^{2,3} 2) ADN-B level shows a more continuous increase than ASO level in acute rheumatic fever, acute glomerulonephritis and other streptococcal infections^{4,5} Hence the ADN-B detection plays an important role in the diagnosis of Sydenham's chorea or chronic valvular heart disease without obvious onset^{4,5}

Consequently, the determination of ADN-B is recommended as the second streptococcal antibody test to ASO detection by the WHO manual⁶

The determination of ADN-B is widely used in the United States and some European countries, but it is not popularly used in Japan as yet.

This paper is the results of the cooperative study on ADN-B by the members of the Committee for the Prevention of Rheumatic Fever and Rheumatic Heart Disease of Japanese Circulation Society.

The comparison of ADN-B titers between streptococcal carriers and non-carriers are also discussed.

MATERIALS AND METHODS

The serum samples used in this study were obtained from patients with rheumatic fever (94 cases), acute glomerulonephritis (65), acute respiratory streptococcal infections (210), myeloma with high ASO titers (4) and healthy

controls (278). Serum samples both "acute" rheumatic fever and glomerulonephritis were taken from the patients within 3 months after the onset of the diseases or from the patients who were in the active stage clinically.

The ADN-B titer was determined at the institute of each member of the Committee using Hemoprobe-B Test (Wampole-Kyorin Tube Test).

RESULTS

ADN-B titers in healthy controls were determined in various age groups. The distribution of the titers is shown in Table I. Upper limits of normal controls were determined by division of upper 20% from the lower 80%.^{7,8}

By this definition, the upper limits in normal control group of ADN-B titers were 1:60 under the age of 5, 1:480 at the age of 6 to 15 and 1:340 over 16 years age group.

In patients with acute rheumatic fever, 87% of the cases showed positive ADN-B titers and 71% showed positive ASO titers.

In patients with inactive rheumatic fever 3 months after the onset of the disease, ADN-B titers were in the elevated range in 72% of the cases but ASO titers were positive in only 22%. This shows ADN-B titers remained at a high level for a longer period than ASO titers.

In patients with acute glomerulonephritis, 87% of the cases had positive ADN-B titers and 55% of the cases showed high ASO titers. The renal involvement in our patients of acute nephritis was not secondary to obvious skin infection.

In culture positive streptococcal infections, elevated ADN-B were observed in 49% and elevated ASO in 47% respectively.

TABLE II CORRELATION OF ADN-B AND ASO WITH CLINICAL AND BACTERIOLOGICAL FINDINGS

Subjects	Number	Elevated*	
		ADN-B	ASO
<i>Rheumatic fever</i>		%	%
<i>active</i>	46	87.0	71.7
<i>inactive</i>	48	72.8	22.9
<i>Glomerulonephritis</i>	65	87.7	55.4
<i>Acute streptococcal infections</i>	51	49.0	47.1
<i>Myeloma</i>	4	0	100.0
<i>Healthy controls</i>	278	21.9	18.7
<i>Throat culture</i>			
<i>positive</i>	58	39.7	36.2
<i>negative</i>	25	8.0	0

*Elevated	ADN-B	ASO
under 6 yrs.	≥ 85	≥ 320
6 ~ 16	≥ 480	≥ 320
over 16 yrs.	≥ 340	≥ 250

TABLE III CORRELATION BETWEEN TITERS FOR ADN-B AND ASO TITERS

			Rheumatic fever	Glomerulonephritis	Controls
<i>Agreement</i>	ADN-B	+	62.2	49.2	10.1
	ASO	+			
	ADN-B	—	2.2	6.2	62.0
	ASO	—			
			64.4	55.4	72.2
<i>Disagreement</i>	ADN-B	+	24.4	38.5	12.7
	ASO	—			
	ADN-B	—	11.1	6.2	15.2
	ASO	+			
			35.5	44.6	27.8

Numbers indicate percent.

From patients with myeloma with high ASO titers, none had high ADN-B titers.

Regarding the carrier state of streptococci, 39% with positive throat swabs had high ADN-B titers and 36% had positive ASO titers. In contrast, only 8% of children, whose throat swabs were negative, had elevated ADN-B and none had high ASO titers (Table II).

Correlations between titers for ASO and ADN-B test is shown in Table III. Agreement of

both titers were observed in 64% patients with acute rheumatic fever and 55% with acute glomerulonephritis. On the other hand, disagreement of ASO and ADN-B titers was recognized in 35% patients with acute rheumatic fever and 46% with acute glomerulonephritis respectively. In these cases, ADN-B titers were elevated in 24% of the rheumatic fever patients and 38% of those with acute nephritis. ASO titers, on the other hand, were elevated only in 11% of the rheumatic

fever cases and 6% of those with glomerulonephritis.

These results show that ADN-B titers maintain a high level for a longer period than that of ASO titers.

DISCUSSION

It is difficult to define the normal range of streptococcal antibodies because they may vary with age, season of the year, geographic area and other factors. In our series of healthy controls, upper limit of normal ADN-B titers were 1:60 in preschool age, 1:480 in school age and 1:340 in adult age groups. These results are different from previously reported titers especially in the school age group in which 1:170 was upper limit of normal.⁹ If a titer of 1:170 is considered to be the upper limit of normal, 49% of the children in our study were in the abnormal range.

Kaplan and his co-workers¹⁰ demonstrated the differences of antibody responses to streptococcal antigens in carrier state. In their report, 35% of non-symptomatic cases with positive cultures for group A streptococci had high ASO titers and 27% had elevated ADN-B titers.

In our series of carrier children, ADN-B titers were elevated in 39% and ASO in 36% respectively.

On the other hand, only 8% of the cases whose throat swabs were negative, showed high ADN-B. None of them had high ASO titers in the present study.

In patients with acute rheumatic fever and acute glomerulonephritis, approximately 80 to 90%, showed elevation of ADN-B titer, ASO titer or both.^{4,5,7} Moreover, in cases with acute glomerulonephritis following streptococcal impetigo, an elevation of ASO titer is not observed usually; on the other hand, 75% of the cases showed an elevation of ADN-B titer.^{2,3}

In our studies of patients with acute rheumatic fever and glomerulonephritis, 87% patients showed elevated titers of ADN-B. On the other hand, 71% of rheumatic patients and 55% of nephritis patients showed high ASO titers.

In patients with inactive rheumatic fever whose disease began more than 3 months ago,

73% of the cases showed high ADN-B titers, but only 22% of the patients showed high ASO titers. This indicates that ADN-B titers remain at a high level longer than that of ASO titers. So the determination of ADN-B titer is important for the diagnosis of suspected Sydenham's chorea or chronic valvular heart diseases with obscure onset.

Moreover, ADN-B determination may play an important role in cases with nonspecific high ASO titer. Patients with myeloma sometimes show significantly high ASO level. In our studies 4 cases with myeloma showed high ASO titers, but ADN-B titers of these patients were not elevated.

REFERENCES

1. WANNAMAKER LW: The differentiation of three distinct desoxyribonuclease of group A streptococci. *J Exp Med* 107: 797, 1958
2. DILLON HC, REEVES MSA: Streptococcal immune responses in nephritis after skin infection. *Am J Med* 56: 333, 1974
3. BISNO AL, NELSON KE, WAYTZ P, BRUNT J: Factors influencing serum antibody responses in streptococcal pyoderma. *J Lab Clin Med* 81: 410, 1973
4. STOLLERMAN GH, LEWIS AJ, SHULTZ I, TARANTA A: Relationship of immune response to group A streptococci to the course of acute, chronic and recurrent rheumatic fever. *Am J Med* 20: 163, 1956
5. AYOUB EM, WANNAMAKER LW: Evaluation of the streptococcal desoxyribonuclease B and diphosphopyridine nucleotidase antibody tests in acute rheumatic fever and acute glomerulonephritis. *Pediatr* 30: 527, 1962
6. ROTA J, FACKLAM RR: Manual of microbiological diagnostic methods for streptococcal infections and their sequelae. WHO/BAC 1980
7. WANNAMAKER LAW, KAPLAN EL: The modern face of rheumatic fever. *Cardiovasc Clin* 5: 1, 1973
8. COPE JB, REDYS JJ, RANDOLPH MF: A comparison of two streptococcal antibody levels in post treatment carriers of group A streptococci. *Clin Pediatr* 15: 1120, 1976
9. KLEIN GC, BAKER CN, JONES WL: "Upper limits of normal" antistreptolysin O and anti-desoxyribonuclease B titers. *Appl Microbiol* 21: 999, 1971
10. KAPLAN EL, FRANKLIN HR, TOP FH: Diagnosis of streptococcal pharyngitis: Differentiation of active infection from the carrier state in the symptomatic child. *J Infect Dis* 123: 490, 1971