



Clinical Significance of Hemagglutination Grades Determined Using the IH-1000 Automated Blood Typing Instrument: Real-world Data

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Dear Editor,

In Korea, many blood banks test ABO blood groups using automated analyzers [1]. This reduces costs and increases the work efficiency compared with manual methods [2]. The IH-1000 (Bio-Rad Laboratories, Hercules, CA, USA) automated analyzer used in Asan Medical Center, Seoul, Korea, functions based on the column agglutination principle. Results obtained using the IH-1000 analyzer reportedly show a good correlation with those obtained using the manual tube method [3]. ABO subgroups are difficult to determine using an automated analyzer. Few studies have investigated this, and most were not conducted in a clinical environment [1, 3]. We compared final blood group typing results generated by the IH-1000 automated analyzer with those obtained using the manual tube method using real-world data. Based on the analysis results, we provide information to assist clinical interpretation of hemagglutination grading when using an automated analyzer. The Institutional Review Board of Asan Medical Center approved this study (2020-0392).

In total, 209,668 samples tested for ABO blood groups between September 2017 and January 2020 were retrospectively investigated. We compared the results determined using the IH-1000 analyzer and the final reading results (i.e., those obtained

using the IH-1000 analyzer only or those confirmed by manual retesting) (Table 1). A final blood group determination by manual retesting was required in the following cases: 1) new patients with no prior blood group data, 2) a grade $\leq +3$ in forward typing, 3) a grade $\leq +2$ in reverse typing, and 4) analyzer results other than positive and negative reactions, and 5) no reading output from the analyzer for any other reason.

When comparing the IH-1000 results with those of the manual testing for the forward typing of antigens A and B in 24,629 samples, 99.9% of the samples showing a reaction grade $\geq 3+$ in the IH-1000 analyzer showed grade 4+ in the manual testing. Moreover, with the same 4+ grade in the manual test, the reaction grades for antigens A and B differed in the IH-1000 analyzer. The samples determined to have grade 3+ by the IH-1000 analyzer were substantially higher for antigen B (59.2%) than for antigen A (24.0%).

The 3+ samples were comprehensively analyzed (Table 2). Most samples showing a grade $\geq 3+$ in both the A and B blood groups were classified as blood group AB in the manual retesting. AB subgroup results were obtained in only a few samples. Among the AB subgroup samples, anti-B antibody was detected by reverse typing in 14 out of 15 A₂B₃ samples, and anti-A1 an-

Received: January 12, 2022
Revision received: March 16, 2022
Accepted: June 15, 2022

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Table 1. ABO blood typing results obtained using the IH-1000 analyzer

ABO group	IH-1000					Total
	Group A	Group B	Group O	Group AB	No output	
Final result						
Group A	86.0% (62,214/72,308)	-	-	-	14.0% (10,094/72,308)	72,308
Group B	-	83.4% (47,307/56,696)	-	-	16.6% (9,389/56,696)	56,696
Group O	-	-	97.8% (55,530/56,781)	-	2.2% (1,251/56,781)	56,781
Group AB	-	-	-	84.3% (19,985/23,715)	15.7% (3,730/23,715)	23,715
Subgroup	-	1.2% (2*/168)	-	0.6% (1 [†] /168)	98.2% (165/168)	168

*B₃, Bw; [†]A₂B.

Table 2. Analysis of samples with a reaction grade 3+ in the IH-1000 analyzer

A/B antigen		N	%	Final interpretation
Automation	Manual			
4+/3+	4+/4+	2,640	99.36	AB
	4+/3+	2	0.08	AB (1), ABw (1)
	4+/2+	15	0.56	A ₂ B ₃
3+/4+	4+/4+	613	100	AB (611), A ₂ B (2)
3+/3+	4+/4+	259	99.62	AB (258), AwB (1)
	2+/4+	1	0.38	AwB
3+/-	4+/-	2,470	100	A
-/3+	-/4+	4,876	99.96	B (4,875), B ₃ (1)
	±/4+	1	0.02	AwB
	-/2+	1	0.02	B ₃

tibody was detected in 1 out of 2 A₂B samples. When antigen A was grade 3+ and antigen B was negative, the final interpretation was that of blood group A. When antigen A was negative and antigen B was grade 3+, most samples were classified as blood group B, and a few were subgrouped as AwB or B₃. We identified 168 cases with an ABO subgroup (0.08%), which is similar to the proportion (0.05%) reported in a previous study of Korean blood donors [4]. The following blood subgroups were determined: 38 A₂B₃, 28 AB₃, 25 ABw, 24 B₃, 16 Aw, 9 Bw, 7 AwB, 6 A₂B, 6 A₃, 4 A₂, 2 A₂Bw, 1 A₃B, 1 AwBw, and 1 Bx.

Noteworthy is the difference between the A and B antigens in forward typing. The proportion of samples with grade 3+ for antigen B was higher when using the IH-1000 analyzer, which is different from the findings in a previous study [3]. While the cause of the difference in reaction grade between antigens A and B when using an automated analyzer remains unclear, the amount of antigen expressed in blood cells, number of blood cells, and amount of antibodies are likely to have been set differently from

those in the manual method. Unlike the previous study, our study was conducted in a real-world environment using actual patient samples and continuously operating instruments, which may explain the differential findings.

In conclusion, a 3+ reaction grade determined using the IH-1000 automated analyzer will generally be assessed as a 4+ grade using a manual method. The proportion of samples with grade 3+ was higher for antigen B than for antigen A. A blood group reading generated by the IH-1000 analyzer can safely be used for transfusion purposes without additional testing. When the antigen reading on this instrument shows a reaction grade 3+, the patient may have an ABO subgroup. Manual retesting and reverse typing are safe choices for blood group determination.

ACKNOWLEDGEMENTS

None.

AUTHOR CONTRIBUTIONS

Park B summarized the data and wrote the manuscript. Kim JS contributed to the data collection. Youk HJ, Chung Y, and Kim H critically revised the manuscript and supported the study. Hwang SH and Oh HB supported and supervised the study. Ko DH designed and supervised the study.

CONFLICTS OF INTEREST

None declared.

FUNDING

This study was funded by Bio-Rad Korea (Seoul, Korea). The

company had no role in the study design, data collection and interpretation, and the decision to submit the findings for publication.

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