Comparison of 3 Automated Immunoassays for Detection of Anti-Hepatitis A Virus Immunoglobulin M in a Tertiary Care Hospital

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Three automated immunoassay kits for anti-Hepatitis A Virus (HAV) IgM—Architect, (Abbott Laboratories, USA), Elecsys (Roche Diagnostics, Germany), and ADVIA Centaur (Siemens Healthcare Diagnostics Inc., USA)—were compared. We included 178 consecutive samples, for which an anti-HAV IgM test was requested at Seoul National University Hospital from September 2009 to January 2010. Reviewing of medical records, reverse transcription (RT)-PCR for HAV RNA, or total anti-HAV assay were performed on 16 (9.0%) samples with discrepant results. The percent agreements (kappas) of the Architect and ADVIA Centaur, Architect and Elecsys, and ADVIA Centaur and Elecsys kits were 96.6% (0.91), 96.6% (0.92), and 97.8% (0.94), respectively. Eight out of 16 discrepant samples showed gray-zone values in Architect but were nonreactive in the others. Slightly earlier seroconversion was suspected in Elecsys. The 3 assays showed comparable performances with excellent agreements in a tertiary care hospital setting.

Key Words: Anti-hepatitis A virus immunoglobulin M, Immunoassay, Agreement
study was approved by the Seoul National University Hospital Institutional Review Board (E-1110-046-381).

The agreements (kappas) between assays were calculated [4]. Correlations in S/CO values between assays were evaluated by a Spearman’s test, excluding those results exceeding the measurable range using SPSS for Windows (version 12.0; SPSS Inc., Chicago, IL, USA).

Among 178 samples, 45 (25.3%) were reactive and 117 (65.7%) were nonreactive for all 3 kits. When the gray-zone results of Architect and ADVIA Centaur were interpreted as nonreactive, the percent agreements (kappas) between Architect and ADVIA Centaur, Architect and Elecsys, and ADVIA Centaur and Elecsys were 96.6% (0.91), 96.6% (0.92), and 97.8% (0.94), respectively. Among the 16 (9.0%) discrepant sera, 8 (case 1-8, Table 1) showed gray-zone values with Architect, but they were nonreactive with ADVIA Centaur and Elecsys. The negative anti-HAV IgM follow-up tests indicated that cases 1 and 2 were less likely to have HAV infection. For cases 3-8, HAV infection could not be ruled out from additional test results (HAV RT-PCR, negative; total anti-HAV, reactive). Case 9 (Architect, reactive; others, nonreactive) and Case 10 (ADVIA Centaur, reactive; others, nonreactive) were also less likely to have HAV infection considering the negative HAV RT-PCR, although very high levels of AST and ALT were seen.

Cases 11 and 12, confirmed as HAV+ (positive RT-PCR), were nonreactive with ADVIA Centaur but reactive with Elecsys. Cases 13 and 14, confirmed as HAV+ from reactive results with higher S/CO values of follow-up anti-HAV IgM tests in all 3 assays, showed gray-zone results with Architect and were reactive with Elecsys. Case 13 was nonreactive with ADVIA Centaur.

Cases 15 and 16, with infection history (7 and 8 months ago, respectively) (reactive anti-HAV IgM and clinical course consistent with HAV infection) were reactive with ADVIA Centaur and Elecsys and nonreactive and in the gray-zone with Architect, respectively.

Although, these assays were not quantitative, their S/CO values were moderately correlated with each other. Spearman’s correlation coefficient (r) between Architect and the ADVIA Centaur HAV IgM was 0.757 (P < 0.001); Architect and Elecsys, 0.732 (P < 0.001); and Elecsys and ADVIA Centaur, 0.776 (P < 0.001) (Fig. 1).

Here, 3 kits showed excellent overall agreement (kappas: 0.91-

Table 1. Clinical characteristics of cases with discrepant results among Architect, ADVIA Centaur, and Elecsys Anti-HAV IgM assays (N = 16)

| Case No. | HAV IgM | HAV IgM | F/U HAV IgM | Anti-HAV | Total AST/ALT (IU/L) | Tbil/D.bil (mg/dL) | Clinically suspected diagnosis |
|----------|---------|---------|-------------|----------|---------------------|-----------------|-----------------------------|
|          | Architect (S/CO)* | ADVIA Centaur (S/CO)* | Elecsys (S/CO)* | (days since first bleed) | IgG | IgM | RT-PCR |
| 1        | G (0.9) | N (0.05) | N (0.25) | N (8) | NT | R | N | 1,015/190 | 2.3/1.5 | Common bile duct stone |
| 2        | G (1.0) | N (<0.02) | N (0.24) | N (3) | N | R | N | 532/342 | 1.0/0.4 | Gallbladder stone |
| 3        | G (0.8) | N (0.03) | N (0.26) | NT | R | R | N | 27/42 | 1.0/0.2 | Toxic hepatitis |
| 4        | G (1.0) | N (0.08) | N (0.21) | NT | NT | R | N | 45/74 | 0.7/NT | Leptospirosis |
| 5        | G (1.0) | N (0.07) | N (0.23) | NT | NT | NT | N | 64/306 | 1.1/NT | Diabetes mellitus, hepatitis |
| 6        | G (0.9) | N (<0.02) | N (0.22) | NT | NT | NT | N | 147/204 | 1.6/0.4 | Amyopathic dermatomyositis |
| 7        | G (1.1) | N (0.07) | N (0.30) | NT | NT | NT | N | 316/84 | 2.0/1.1 | Metastatic breast cancer |
| 8        | G (0.9) | N (0.21) | N (0.26) | NT | NT | NT | N | 129/325 | 22.8/16.3 | Toxic hepatitis |
| 9        | R (1.5) | N (0.06) | N (0.23) | NT | NT | NT | N | 1,031/3,467 | 0.7/NT | Toxic hepatitis |
| 10       | N (0.4) | R (4.21) | N (0.20) | N (4) | N | N | N | 15,864/8,340 | 8.0/NT | Alcohol hepatitis |
| 11       | G (0.9) | N (0.66) | R (1.17) | NT | N | R | P | 3,385/2,627 | 1.3/NT | HAV hepatitis |
| 12       | R (1.4) | N (0.77) | R (1.04) | R (1) | N | R | P | 2,150/703 | 2.0/NT | HAV hepatitis |
| 13       | G (0.9) | N (0.22) | R (3.75) | R (4) | NT | R | NT | 2,134/3,053 | 2.4/NT | HAV hepatitis |
| 14       | G (1.1) | R (2.85) | R (4.23) | R (3) | NT | R | NT | 382/1,407 | 3.5/NT | HAV hepatitis |
| 15       | N (0.5) | R (1.50) | R (1.09) | NT | NT | NT | R | 19/18 | 0.8/NT | Resolving HAV hepatitis |
| 16       | G (0.9) | R (1.66) | R (1.14) | NT | R | R | NT | 587/557 | 14.5/8.5 | Resolving HAV hepatitis |

*For Architect HAVab-IgM, specimens with signal-to-cutoff (S/CO) values 0.80-1.20 were considered gray-zone. For ADVIA Centaur HAV IgM, S/CO values ≥0.80 and <1.20 were considered equivocal. Abbreviations: T.bil, total bilirubin; D.bil, direct bilirubin; F/U, follow up; HAV, hepatitis A virus; N, nonreactive or negative; G, gray zone; R, reactive; P, positive; NT, not tested; S/CO, signal-to-cutoff; RT-PCR, reverse transcription-PCR.
0.94) when the gray-zone values of Architect were considered nonreactive (ADVIA Centaur showed no equivocal results). Architect showed gray-zone results in 12 samples: HAV infections, 4; less-likely infections, 2; uncertain for infection, 6. The agreement was slightly lower (kappa values: Architect and ADVIA Centaur, 0.81; Architect and Elecsys, 0.87; data not shown) when the gray-zone values of Architect were considered reactive.

ELISAs can exhibit false-reactive results in various conditions, including autoimmune diseases or renal failure [5]. Rheumatoid factor or heterophilic antibodies can also interfere with immunoassay results [6, 7]. Nonspecific binding of serum IgM to the microparticle bead induces false reactivity in the Liaison system adopting chemiluminescence immunoassay, in the absence of rheumatoid factor or paraprotein; the use of chemical-blocking reagents eliminated this problem [8]. The Architect system adopts a different assay principle (direct coating of HAV antigens on a microparticle bead) from that of the other assays (using streptavidin-coated microparticles and biotinylated mouse anti-human IgM antibodies). Further investigations are needed to determine if gray-zone results, more frequently observed with Architect, could be partially explained by the nonspecific adsorption of proteins to the microparticle bead.

In cases 11-14, in the early phase of HAV infection, the ADVIA Centaur and Architect showed slightly later seroconversions compared to the Elecsys. Two cases with history of HAV infection (~7-8 months ago) were reactive with ADVIA Centaur and Elecsys with low S/CO values (1.09-1.66), whereas Architect showed nonreactive in one sample and gray-zone in another, suggesting a slight difference in the sensitivity for the detection of decreasing anti-HAV IgM in patients who had recovered from previous HAV infection.

Although all 3 kits are not quantitative tests, the S/CO values showed moderate correlations among them. For samples from patients with resolving HAV infection, S/CO values were low (1.09-1.66), suggesting very low anti-HAV IgM levels. Further development of quantitative tests for anti-HAV IgM may be helpful in patients showing atypical disease courses during HAV infection or HAV reactivation after transplantation [9].

In conclusion, 3 automated immunoassay kits showed comparable performances, with excellent overall agreement among them when performed on samples submitted to a tertiary care hospital and can be successfully applied in clinical laboratory practice.

Authors’ Disclosures of Potential Conflicts of Interest

No potential conflicts of interest relevant to this article were reported.

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