A COMPARATIVE STUDY OF TWO THIRD-GENERATION ANTI-HEPATITIS C VIRUS ELISAs
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ABSTRACT
The sensitivity and specificity of two third-generation screening tests for the detection of antibody to hepatitis C virus (anti-HCV) was evaluated in a side-by-side study (Abbott HCV EIA 3.0/Ortho HCV ELISA 3.0). Specimens that were reactive in either ELISA were then retested by the other ELISA, and confirmed by RIBA-3. The screening of 15,540 serum samples from healthy blood donors showed a significantly lower number of reactive cases tested by Ortho (21 out of 8,805, 0.24%), than tested by Abbott (35 out of 6,735, 0.52%). In the side-by-side comparison, we found that significantly (p=0.005) more Ortho-positive samples were also reactive in Abbott (14/21, 66.6%), than in Abbott followed by Ortho (7/35, 20%). Moreover, the RIBA analysis revealed a significantly (p=0.014) higher number of RIBA-positive specimens among those reactive in Ortho 10/21 (47.6%), than among those reactive in Abbott 6/35 (17.2%), thus the former provides a greater positive predictive value. However, we did not observed differences in the sensitivity between Abbott and Ortho, because all RIBA-positive samples demonstrated reactivity in both ELISAs.

Key words: hepatitis C, diagnosis, ELISA, virus, third-generation ELISA, screening, hepatitis C virus

Results and Discussion
Table 1 summarizes our findings in the screening of 15,540 sera. Using the Abbott EIA, 35 out of 6,735 screened samples (0.52%) were found positive. In comparison, a significantly lower number of positive cases (21 out of 8,805, 0.24%) was detected within the group tested by Ortho. Interestingly, when cases reactive in either ELISA were retested by the other, we found that only 7/35 (20%) of the specimens positive in Abbott also provided reactivity in Ortho (14/21, 66.6%), than in Abbott followed by Ortho (7/35, 20%). Moreover, the RIBA analysis revealed a significantly (p=0.014) higher number of RIBA-positive specimens among those reactive in Ortho 10/21 (47.6%), than among those reactive in Abbott 6/35 (17.2%). No significant differences were detected in the number of RIBA-indeterminate (p=0.54) and RIBA-negative (p=0.094) results between these groups.

In Table 2 we present the reactivity provided in
both ELISAs of all samples analyzed by RIBA. As shown, all RIBA-positive samples demonstrated reactivity in both Abbott and Ortho EIA. Among RIBA-negative samples (false positives from EIA), 3 provided reactivity in both ELISAs, 17 were reactive only in Abbott EIA, and 4 were reactive only in Ortho. Finally, within the RIBA-indeterminate group, 2 samples were detected as positive in both ELISAs, 11 were positive only by Abbott, and 3 were positive exclusively in the Ortho EIA. As concerns the antigen specificity of RIBA-indeterminate samples, we observed that the 3 RIBA-indeterminate specimens detected only by Ortho and the 2 RIBA-indeterminate samples detected in both ELISAs were reactive on c33c. By contrast, the 11 RIBA-indeterminate samples in the group positive only in Abbott demonstrated reactivity to either NS5 (8/11, 73%), c22p (2/11, 18%), or c100p (1/11, 9%) antigens.

The results of our study suggest that 3rd generation anti-HCV ELISAs of Abbott and Ortho have a similar sensitivity, because all RIBA-positive specimens demonstrated reactivity in both ELISAs. However, the positive predictive value (PPV) of the Ortho EIA was significantly higher than (p=0.01) the corresponding value of Abbott. Thus, 47.6% of the reactive specimens detected by Ortho were confirmed in RIBA-3, whereas only 17.2% of the reactive samples in the Abbott EIA were RIBA-positive. The reasons for the lower PPV of the Abbott assay are unclear. Since most Abbott positive-RIBA indeterminate specimens reacted against the NS5 antigen, unspecific reactivity induced by the presence of this anti-

gen in the Abbott EIA could be speculated. Indeed, the inclusion of the NS5 antigen in third-generation screening assays has been controversial because of its limited contribution in improving sensitivity in the detection of anti-HCV.

In addition, its impact on the specificity of the HCV screening in low-risk donors is uncertain, since there are sera that have been identified as positive in RIBA 3.0 against the NS5 recombinant protein, but that are PCR negative. Nevertheless, the elimination of the NS5 antigen in an attempt to exclude false-positive reactions may result in an increased risk of false-negative interpretations. One case of early seroconversion to NS5 in post-transfusion hepatitis has been reported.

In our study, we detected 8 Abbott-positive sera which only reacted against the NS5 antigen in RIBA. These samples were found negative in Ortho EIA 3.0, and may correspond to the Ortho false-negatives. This argument is also valid for the 2 Abbott-positive samples that were reactive only against the c-22p antigen that did not demonstrate reactivity in Ortho. On the other hand, of the 5 Ortho positive-RIBA indeterminate samples reactive only against the c33c, 3 were not detected by Abbott. These results are in agreement with the findings of an earlier trial by Couroucé et al. comparing the sensitivity of several anti-HCV screening tests including Abbott 3.0 and Ortho 3.0. These authors reported two false-negatives in Abbott corresponding to c33c reactive specimens, and one Ortho false-negative with anti-c22p reactivity.

In conclusion, our results show that the 3rd-generation anti-HCV ELISA of Ortho provides a greater PPV than the Abbott EIA 3.0, although the sensitivity of both techniques is similar.

### References