Heterophile antibody interference in qualitative urine/serum hCG devices: Case report

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A B S T R A C T

Objectives: This case report investigates the origin of a false positive result on a serum qualitative human chorionic gonadotropin (hCG) device.

Patient and methods: A 46-year-old woman diagnosed with chronic myeloid leukemia presented with nausea and vomiting. A qualitative serum hCG test was interpreted as positive; however, a quantitative serum hCG test was negative (<5 IU/L). To further investigate this discrepancy, the sample was pretreated with heterophilic blocking reagent (HBR). Additionally, the sample was tested on other qualitative hCG devices composed of antibodies from different animal sources. Blocking reagent from an automated quantitative immunoassay was also tested for its ability to inhibit the heterophile antibody interference.

Results: The qualitative test result was negative after pretreatment with heterophilic blocking reagent. Other devices composed of antibodies from different animal sources also demonstrated mixed results with the patient’s sample. Blocking reagent obtained from the automated quantitative assay inhibited the heterophile antibody interference.

Conclusion: This case demonstrates that positive serum point-of-care hCG results should be interpreted with caution and confirmed with a quantitative serum hCG immunoassay when clinical suspicion is raised.

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1. Introduction

Point-of-care hCG testing is often used in emergency departments and outpatient clinics to rule out pregnancy before performing interventions that can cause harm to a developing fetus. Qualitative point-of-care devices are often FDA approved for use with urine or serum. Urine is preferred as it requires no sample processing. The utility of qualitative serum tests has been questioned. First, qualitative serum tests cannot be performed at the point-of-care because of the need for centrifugation, making the turnaround time comparable to quantitative hCG. Second, the analytical sensitivity of qualitative tests is inferior to quantitative tests (10 IU/L vs. 1 IU/L). However, qualitative serum hCG tests are often ordered because of their perceived faster turnaround time [1,2].

As with most immunoassays, there are limitations to both qualitative and quantitative hCG assays, as they are prone to discrepancies due to the high-dose hook effect and heterophile antibody interference [2]. This case demonstrates an example of a false positive hCG result caused by heterophile antibody interference using a qualitative hCG assay. Interestingly, a quantitative assay that contained capture and signal antibodies from the same animal source as the qualitative device did not demonstrate heterophile antibody interference.

2. Case report

A 46-year-old woman with a history of chronic myeloid leukemia was being evaluated for a bone marrow transplant. A qualitative serum hCG test was performed to rule out pregnancy. The qualitative serum hCG result was interpreted as positive; however, a quantitative serum hCG was <5 IU/L (reference interval <5 IU/L).

The presence of heterophile antibodies was suspected in this patient and pretreatment of her serum specimen with heterophilic blocking reagent (HBR) was performed. HBR contains purified animal immunoglobulins. Heterophile antibodies, if present in the sample, bind the animal immunoglobulins and are prevented from interfering in the immunoassay. Because treatment with HBR causes a dilution effect, saline treatment using the same volume was concurrently performed (Fig. 1). The control sample treated with saline demonstrated a positive hCG signal. The HBR-treated sample appeared to block the heterophile antibody interference. Of note, a weaker signal was observed for the control band. This could possibly be due to detergents, altered pH or other properties of the HBR which are not optimal for the lateral flow device. In order to further confirm heterophile antibody interference, we tested additional devices that contained antibodies from different animal sources.

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The qualitative serum hCG device used in Fig. 1 consists of a monoclonal mouse capture antibody and a monoclonal mouse detection antibody. In order to confirm interference from human anti-mouse antibodies (HAMA), we tested three additional devices. Two devices

<table>
<thead>
<tr>
<th>Device</th>
<th>A</th>
<th>B</th>
<th>C</th>
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</thead>
<tbody>
<tr>
<td>Treatment</td>
<td>Neat</td>
<td>Saline</td>
<td>HBR</td>
</tr>
<tr>
<td>Result</td>
<td>Positive</td>
<td>Positive</td>
<td>Negative</td>
</tr>
</tbody>
</table>

Fig. 1. Patient results using a Beckman Icon 20 qualitative hCG device in the presence of heterophilic blocking reagent (HBR). A. Serum with no dilution; B. Serum diluted with 1 part saline and 3 parts patient serum; C. Serum diluted with 1 part HBR and 3 parts patient serum. The table below indicates the treatment and interpreted result for each device.

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<table>
<thead>
<tr>
<th>Device</th>
<th>Cardinal Health hCG Combo</th>
<th>Alere hCG Combo</th>
<th>Cen-Med hCG</th>
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<tbody>
<tr>
<td>Capture Antibody</td>
<td>Goat</td>
<td>Goat</td>
<td>Mouse</td>
</tr>
<tr>
<td>Signal Antibody</td>
<td>Mouse</td>
<td>Mouse</td>
<td>Mouse</td>
</tr>
<tr>
<td>Result</td>
<td>Negative</td>
<td>Negative</td>
<td>Positive</td>
</tr>
</tbody>
</table>

Fig. 2. Patient results using 3 different qualitative devices. A. hCG Combo by Cardinal Health utilizes mouse and goat antibodies; B. The Alere hCG Combo device utilizes mouse and goat antibodies; C. The Cen-Med hCG device consists of mouse/mouse monoclonal antibodies. The table below indicates the capture and signal antibody for each device and the interpreted result.