A pre-analytical challenge to determine estradiol in children: A monovette systematically causing increased estradiol-concentrations in LC-MS/MS analysis

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INTRODUCTION

In children, 17β-estradiol (E2) concentrations are 1000-times lower in comparison to their precursors, the androgens. Depending on gender, age, and pathology plasma concentrations vary in a broad range. The sensitive and specific determination of E2 is a particular challenge in the endocrine analysis.

• female patient
  • 12/6-12 years, Tanner stage B1
  • presumed diagnosis: ovarian insufficiency
  • high E2 concentration of 296 pmol/L (age reference 10-438 pmol/L, 12-14 years) does not suit with the clinical diagnosis and Tanner B1

A different blood collection tube was used than typically preferred by us.

AIM

• The aim was to identify possible interfering factors by blood collection tubes.

METHOD

• E2 was determined by LC-MS/MS.
  • A: Two new blood samples of the patient were collected independently in two different types of tubes. One tube was the same used beforehand (Kabe Labortechnik) and the other one typically preferred by our pediatric laboratory (Sarstedt).
  • B: Tubes were broken down into their components (gel, pellets, and hole tube). The components were incubated independently with either aqua dest. or strip-plasma for one week.

RESULTS

A

• On average ten times higher E2 concentration was determined in the patient using the Kabe tube than using the tube produced by Sarstedt (Table 1).

<table>
<thead>
<tr>
<th>Device</th>
<th>Kabe</th>
<th>Sarstedt</th>
</tr>
</thead>
<tbody>
<tr>
<td>Repetition</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>E2 average [pmol/L]</td>
<td>147.947</td>
<td>15.133</td>
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An unusual broad peak was noticed at the typical position of the E2 peak using the Kabe tubes (Fig. 1 & 2).

B

• After incubating each component of the tubes, the gel chromatogram of the Kabe tubes showed a similar wide peak (Fig. 3).

• Plasma that was incubated with the gel of the Kabe tube shows a wide peak that leads to a high E2 concentration (Fig. 3).

• Possible isomeric interference could be excluded by chromatographic separation of 17α- and 17β-E2 into two separated peaks (Fig 4).

CONCLUSIONS

• The majority laboratory errors (~46–68.2%) originate in the preanalytical phase
  • Typical pre-analytical interfering factors are ischemia and haemolysis
  • Blood collection tubes are an often under-recognized variable in the preanalytical phase of clinical laboratory (Bowen et al 2016)
  • Especially in analysis of E2 which require greater analytical sensitivities
  • Paediatric hormonal measurements should involve paediatric endocrine validation to notice discrepancies
  • Therefore, automated evaluation should not be used for E2 determination in LC-MS/MS.

REFERENCES


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