Introduction

17 OH Progesterone is useful in the investigation of adrenal disease. Traditionally it has been measured using radioimmunoassay (RIA), with 22 of the 24 contributing laboratories in the RCPA QAP 2012 Endocrine Program using it. There are a number of disadvantages in using RIA including reagent stability, radioactive waste management (and occupational exposure) as well as the need for manual pipetting. For these reasons we looked for an alternative to our Siemens Coat-a-Count RIA assay.

We trialled an Enzyme-Linked Immunosorbent Assay (ELISA) kit, produced by IBL. The original patient comparison was performed using the Triturus system (with both manual and automated pipetting). The Bio-Rad Evolis replaced the Triturus for the remaining part of the investigation. Advantages of the ELISA method are a longer shelf life for reagents, use of singletons rather than duplicates, stability, radioactive waste management (and occupational exposure) as well as the need for manual pipetting. For these reasons we looked for an alternative to our Siemens Coat-a-Count RIA assay.

Materials and Method

Thirty five patient samples were used in a paired patient comparison of the IBL ELISA 17OH Progesterone assay performed on the Triturus with the SIEMENS coat-a-count 17 OH Progesterone kit (RIA) assay measured using the LKB 1260 Multigamma II gamma counter. Functional sensitivity, linearity and interference studies were performed on the Bio-Rad Evolis which replaced the Triturus system (with both manual and automated pipetting). The Bio-Rad Evolis replaced the Triturus with the SIEMENS coat-a-count RIA assay.

Results

Correlation between the two methods was excellent ($r^2 = 0.97$), although agreement was poor with a slope of 0.54 (see Charts 1 and 2 for the Passing-Bablok and Bland-Altman plots).

The precision when using quality control material (Bio-Rad Immunoassay Plus) was 12% at concentrations of 2.5 and 15 nmol/L which was similar to the RIA method (12% at 5 and 25 nmol/L). The functional sensitivity (the concentration at which the CV is less than 20%) was found to be 0.8 nmol/L (CV=17%).

There was no significant interference from haemolysis (haemoglobin at 50g/L), icteria (bilirubin of 400 µmol/L) or lipaemia (up to triglyceride of 12 mmol/L). Linearity was found to be acceptable (see chart 4)

Conclusions

Absolute concentrations for both patients and IQC (Bio-Rad Immunoassay Plus Levels 1 and 3) were approximately 40 to 50% less using the ELISA method. Notably the package insert reference interval of the IBL kit appropriately reflected this (see Table 1) and when the equivalent reference interval points of the two methods were plotted against each other the slope was also about 0.5.

Imprecision was comparable to the RIA method and was within RCPA allowable limits (+/- 2 up to 10 nmol/L then +/-20% greater than10 nmol/L).

We found that the IBL ELISA assay is an acceptable replacement for our current RIA method however care should be taken in notifying clinicians about the change in absolute values and reference range.

References

1Siemens Coat-a-count 17 OH Progesterone Package Insert, PITKOP-11, 25/10/2010
2IBL 17 OH progesterone Package Insert, V2011_8