Analytical performance of IgG subclass assays for use on the Optilite® turbidimetric analyser

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Background:

IgG subclass (IgGsc) measurements are used for diagnosis of primary immunodeficiencies and other disorders affecting B cell function; including IgG4-related disease. In this study we assessed the analytical performance of these assays on the Optilite® analyser.

Materials & Methods:

For all IgGsc assays on the Optilite analyser, linearity was assessed using a serially-diluted patient sample pool. Interference was tested with 200 mg/L bilirubin, 5 g/L haemoglobin, 1500 FTU of chyle, or with 125 mg/dL intralipid. To determine within run, between run, between day, between instrument, between batch (for IgG4 only) and total precision a 21 day study was carried out by running 5–10 different IgGsc samples (range IgG1 0.54–29.1 g/L, IgG2 0.08–5.95 g/L, IgG3 0.009–1.8 g/L and IgG4 0.02–4.7 g/L) in duplicate with 2 runs per day, using 3–4 instruments and 1–3 reagent lots. Large comparison studies were performed (IgG1 n= 355, IgG2 n=333, IgG3 n=293, IgG4 n=312) and results compared to the SPAplus® analyser. IgGsc age-specific normal ranges and an IgG4 cut-off of 1350 mg/L were used. For IgG4 antigen excess, capacity was determined by rate following sample addition.

Results:

The assays were shown to be linear over the following ranges (min - max): IgG1 1.2–36.0 g/L, IgG2 0.2–7.8 g/L, IgG3 0.03–2.4 g/L and IgG4 0.02–3.1 g/L with median recoveries of 98–100% (min-max 83%-112%). Precision is shown in Table 1 with total precision between 2.6–12.1%. No clinically significant interference by bilirubin, haemoglobin, chyle or intralipid were detected. Measurements between Optilite® and SPAplus® analysers were highly concordant (bias -1.35–2.66%, slopes 0.90–1.05, rho = 0.98–1.00). Correlation of IgGsc concentration below or above the age-specific normal ranges was highly concordant ($P<0.0001$, >95% agreement). Detecting IgG4 concentrations >1350 mg/L gave 99.7% agreement between analysers, $P<0.0001$. Finally the Optilite IgG4 assay had an antigen-excess capacity of 71.3 g/L.

Conclusions:

The IgGsc immunoassays developed for the Optilite® instrument provide automated, rapid and precise IgGsc quantifications that are highly concordant with results obtained using the SPAplus® analyser. The wide measuring ranges will accurately measure IgGsc in the vast majority of patients, including individuals with IgGsc below, or above normal ranges.

Table 1. Precision study for IgG subclasses on the Optilite® analyser (%CV).					
IgG subclass	Within run	Between run	Between day	Between instruments	Between batch	Total
IgG1	1.2 – 3.1	1.4 – 5.4	0 – 4.2	0.4 – 4.0	N/A	3.0 – 5.5
IgG2	0.7 – 3.3	1.0 – 2.4	2.0 – 4.4	2.5 – 5.6	N/A	2.6 – 5.7
IgG3	2.0 – 8.4	0 – 1.9	5.1 – 8.8	0.4 – 5.5	N/A	5.8 – 12.1
IgG4	1.0 – 4.8	0 – 4.4	1.9 – 8.7	0.9 – 3.3	0.5 – 7.1	4.0 – 10.9