Berkshire & Surrey Pathology Services
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NHS Pathology Solutions

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Determination of detection capability parameters for the Faecal Immunochemical Test

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Introduction

The faecal immunochemical test (FIT) for faecal haemoglobin (fHb) is used for patients presenting with symptoms of colorectal cancer. Thresholds at the lowest analytical capability of the analysers are used for referral.

Assessment of limit of blank (LOB), limit of detection (LOD) and limit of quantification (LOQ) with patient samples is challenging. fHb is unstable; samples have to be transferred immediately into buffer. Samples collected into poo-pots should not be used because they will contain Hb-degradation products. Published studies use manufacturer provided control material or haemolysate spiked into faeces. Neither of these replicate analysis of samples provided by patients. These challenges mean laboratories often quote manufacturer reported thresholds for LOB, LOD and LOQ.

This study followed recently published proposals[1] to assess the detection capability of five OC-SENSOR PLEDIA (Eiken Chemical Co. Ltd., Japan) analysers using patient samples.

LOB definition

LOB is the highest apparent analyte concentration when blank samples with no analyte present are tested.[2]

LOD definition

LOD is the lowest analyte concentration that can be reliably distinguished from the LOB.[2]

LOQ definition

LOQ is the lowest concentration at which analyte can be detected with predefined goals for bias and precision met.[2]

Method

LOB: 20 blank faecal sample collection devices were analysed using 5 OC-SENSOR PLEDIA analysers.

LOD and LOQ: Patient samples with 20-40 µg Hb/g faeces (µg/g) were pooled and diluted to give 16 samples with a concentration range 0-15 µg/g. Samples were analysed 10 times.

LOB, LOD and LOQ were calculated individually for each analyser:

LOB=mean[blank]+1.645(SD[blank])

LOD=LOB+1.645(SD[low concentration of sample])

LOQ=lowest concentration with CV≤10%.

A single assigned value for the 5 analysers used in the laboratory was then set for each parameter.

Results

LOB, LOD, LOQ were determined individually for each analyser (Figure 1). Data for LOB calculation are shown in Table 1.

Figure 1: OC-SENSOR PLEDIA (Eiken Chemical Co. Ltd., Japan) analyser

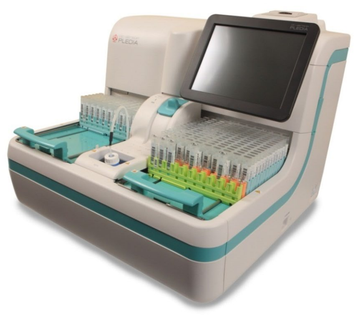


Table 1: LOB results showing mean concentration, SD and calculated LOB for each analyser

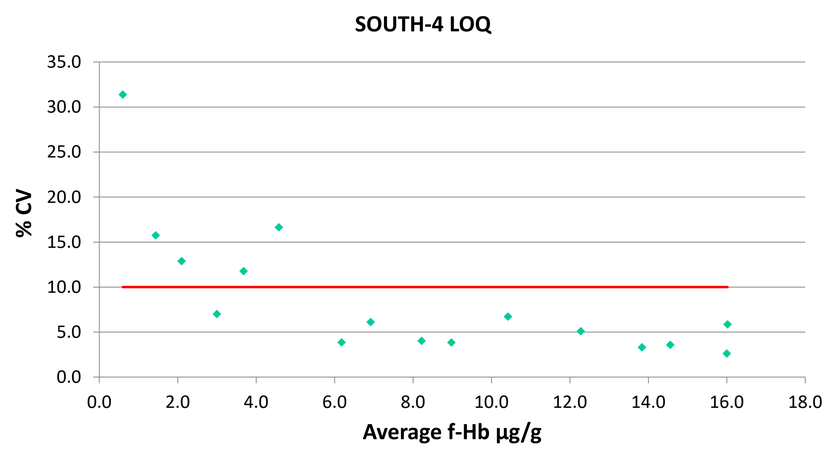
Analyser	Mean Concentration (µg/g)	SD	LOB
South 1	0.13	0.22	0.49
South 2	0.18	0.22	0.55
South 3	0.21	0.29	0.68
South 4	0.03	0.10	0.19
South 6	0.00	0.00	0.00

Table 2: LOD/LOQ concentration gradient results for South 4. Each pool was analysed 10 times and mean value calculated. This was used to calculate SD, %CV and LOD.

Expected Concentration (µg/g)	Mean Measured Concentration (µg/g)	SD	%CV	LOD
0	0.6	0.2	31.4	0.9
1	1.4	0.2	15.8	1.0
2	2.1	0.3	12.9	0.7
3	3.0	0.2	7.0	0.2
4	3.7	0.4	11.8	1.3
5	4.6	0.8	16.7	1.6
6	6.2	0.2	3.9	0.7
7	6.9	0.4	6.1	1.1
8	8.2	0.3	4.0	0.9
9	9.0	0.3	3.9	0.7
10	10.4	0.7	6.7	1.6
11	12.3	0.6	5.1	1.5
12	13.8	0.5	3.3	1.3
13	14.6	0.5	3.6	1.4
14	16.0	0.4	2.6	1.3
15	16.0	0.9	5.9	1.7

LOD on South 4 was determined as 1.6 µg/g, this was the highest LOD value calculated from low value samples, results are shown in Table 2.

Figure 2: LOQ chart for South 4



LOQ for South 4 is 6 µg/g, Figure 2 demonstrates this is the lowest concentration with consistent CV≤10%.

After calculating parameters for each analyser, results were compared in Table 3 to assign values for each parameter to the laboratory. LOB was <0.7 µg/g on each analyser and used to calculate LOD on respective analysers, which was <1.6 µg/g across the analysers. The laboratory LOD was set as 2 µg/g because fHb is reported in whole numbers. The LOQ was set at 6 µg/g.

Table 3: LOB, LOD, and LOQ results for each analyser and single assigned values for the laboratory for each parameter

Analyser	LOB (µg/g)	LOD (µg/g)	LOQ (µg/g)
South 1	0.49	1.34	3
South 2	0.55	1.07	3
South 3	0.68	1.47	6
South 4	0.19	1.58	6
South 6	0.00	1.59	6
Laboratory assigned value	1	2	6
Manufacturer quoted value	Not given	Not given	10

Conclusion

We have demonstrated that it is possible to derive laboratory based fHb detection capability parameters using patient collected samples. This should support other laboratories in deriving such parameters for their FIT obviating the need to use manufacturer quoted ranges and enabling lower thresholds to be set.

References

1) Fraser C, Benton S. Detection capability of quantitative faecal immunochemical tests for haemoglobin (FIT) and reporting of low faecal haemoglobin concentrations. Clin Chem Lab Med. 2019 Apr 24;57(5):611-616

2) Armbruster D, Pry T. Limit of Blank, Limit of Detection and Limit of Quantitation. Clin Biochem Rev. 2008 Aug;29(1):49-52