Heart of England NHS Foundation Trust

Verification and implementation of faecal calprotectin using the BÜHLMANN fCAL® turbo assay

Emma L. Evans, Helen Wiggins, Craig Webster

Heart Of England NHS Foundation Trust

Introduction

Faecal calprotectin (FCAL) is an indicator of inflammatory processes and has an important role in the diagnosis and monitoring of inflammatory bowel disease (IBD). NICE (DC11) recommends FCAL testing in primary care to aid in the differential diagnosis of IBD and irritable bowel syndrome (IBS) in adults where specialist assessment is being considered [1].

As a consequence of this guidance we have seen the workload for FCAL dramatically increase. Due to the nature and inherent inhomogeneity of this specimen and the increased workload, our laboratory evaluated the user-friendly CALEX® Cap Extraction device using the using the BÜHLMANN fCAL® turbo assay on the Abbott ARCHITECT platform.

BÜHLMANN fCAL® Turbo Assay Verification

Within-batch imprecision

Two patient samples (one at ~cut-off point and one positive) were run 10 times on the same day for the turbo assay on the Abbott ARCTITECT platform. Results were obtained from patient samples and extractions performed by two users.

Table 1. Within-batch imprecision

	fCAL® turbo assay	
	Low level	High level
Mean	45.03	379.84
SD	3.99	41.67
%CV	8.85	10.97

Between-batch imprecision

Due to instability of patient faecal samples, internal quality control (IQC) material was used to generate between batch imprecision data. Results collected over a 10 day period and included IQC lot change (lots 4901 and 1502 used).

Table 2. Between -batch imprecision

	fCAL® turbo assay	
	Low level	High level
Mean	74.63	259.27
SD	3.02	10.71
%CV	4.04	4.13

Variability between users

In order to determine the variability of results based of different staff using the CALEX® Cap Extraction devices, three users all prepared five extracts from the same sample. Results in Table 3 demonstrate data from all users gave similar imprecision data as calculated above (see table 1). There was no statistical significant different between users ($p<0.5$).

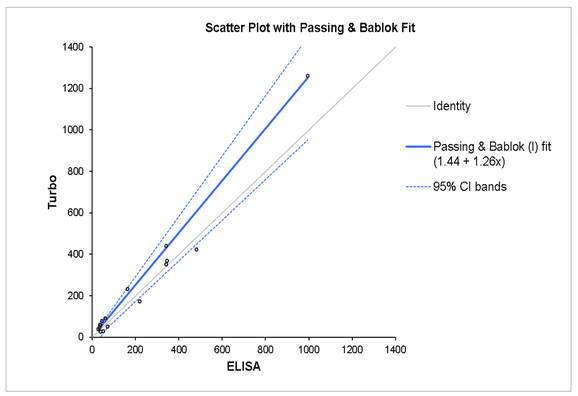
Table 3. User variability using the CALEX® Cap Extraction Devices

	User 1	User 2	User 3
Mean	45.4	44.3	41.8
SD	2.6	4.0	4.5
%CV	5.7	9.0	10.7

Comparison of Turbo vs. ELISA BÜHLMANN methods

Extracted faecal samples (a combination of EQA and patients) using the fCAL® turbo and ELISA methods were compared.

a)



b)

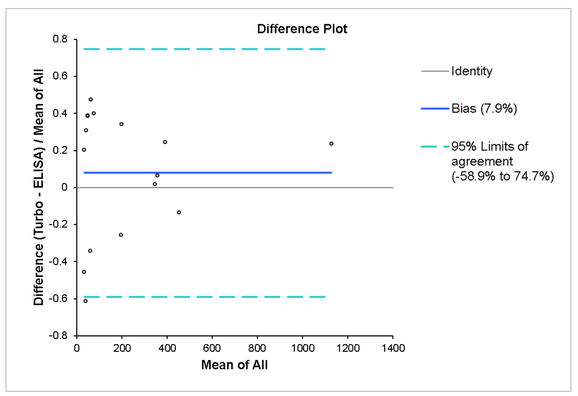


Figure 1. Scatter (a) and difference plot (b) comparing the BÜHLMANN ELISA (reference) and BÜHLMANN Turbo (new) assay.

Passing and Bablok regression was y = 1.44 + 1.26 and Altman-Bland difference plot demonstrated a +7.9% positive bias against the ELISA method.

Sample stability

The kit insert states that the stability of faecal samples at 2-8ºC is 6 days and the stability of extracts at 2-8ºC is 6 days and at -20ºC is >6 months. This was assessed using three different patient samples, over 7 days, stored at different temperatures.

Table 4. Stool stability at room temperature, stored in the fridge and freezer.

	Room Temperture					
	Day 0			Day 3		
Sample	Rep 1	Rep 2	Average	Rep 1	Rep 2	Average
1	853.7	848.7	851.2	557.7	580.2	568.95
2	681.5	668.2	674.85	518.6	523.4	521
3	647.6	649.9	648.75	267.7	289.6	278.65
				P value vs. day 0		0.0505

	Fridge (2-8°C)								
	Day 0			Day 3			Day 7		
Sample	Rep 1	Rep 2	Average	Rep 1	Rep 2	Average	Rep 1	Rep 2	Average
1	853.7	848.7	851.2	768.7	759.1	763.9	743	732.1	737.55
2	681.5	668.2	674.85	495.8	491.8	493.8	473	443.3	458.15
3	647.6	649.9	648.75	483.7	490.2	486.95	420.8	431.4	426.1
				P value vs. day 0		0.0375	P value vs. day 0		0.0349

	Freezer (below -18°C)								
	Day 0			Day 3			Day 7		
Sample	Rep 1	Rep 2	Average	Rep 1	Rep 2	Average	Rep 1	Rep 2	Average
1	853.7	848.7	851.2	687.5	710.4	698.95	784.6	720.5	752.55
2	681.5	668.2	674.85	573.2	780.5	676.85	679.5	667.4	673.45
3	647.6	649.9	648.75	619.4	622.5	620.95	552.5	566.3	559.4
				P value vs. day 0		0.3359	P value vs. day 0		0.1785

Table 5. Extract stability at room temperature, stored in the fridge and freezer.

	Room Temperature	
Sample	Day 0	Day 3
1	851.2	849.4
2	674.85	513.7
3	648.75	714.5
P value vs. day 0		0.6776

	Fridge (2-8°C)		
Sample	Day 0	Day 3	Day 7
1	851.2	802.5	808.7
2	674.85	794.9	693.5
3	648.75	700	721.6
P value vs day 0		0.4919	0.6725

	Freezer (below -18°C)		
Sample	Day 0	Day 3	Day 7
1	851.2	793.1	852.3
2	674.85	678.8	651.8
3	648.75	619.4	651.4
P value vs day 0		0.2607	0.5203

Significant differences between day 0 and indicated time periods (p values highlighted in red) for samples stored at room temperate and in the fridge. For stool stability, only samples stored in the freezer (below -18ºC) were stable for up to 7 days. Once extracted, samples were stable in the fridge (2-8ºC) or in the freezer (below -18ºC) for up to 7 days.

Implementation to Primary Care

In order to provide the best possible service, there was close collaboration with our gastroenterology team to provide a new local standardised pathway for primary care to follow.

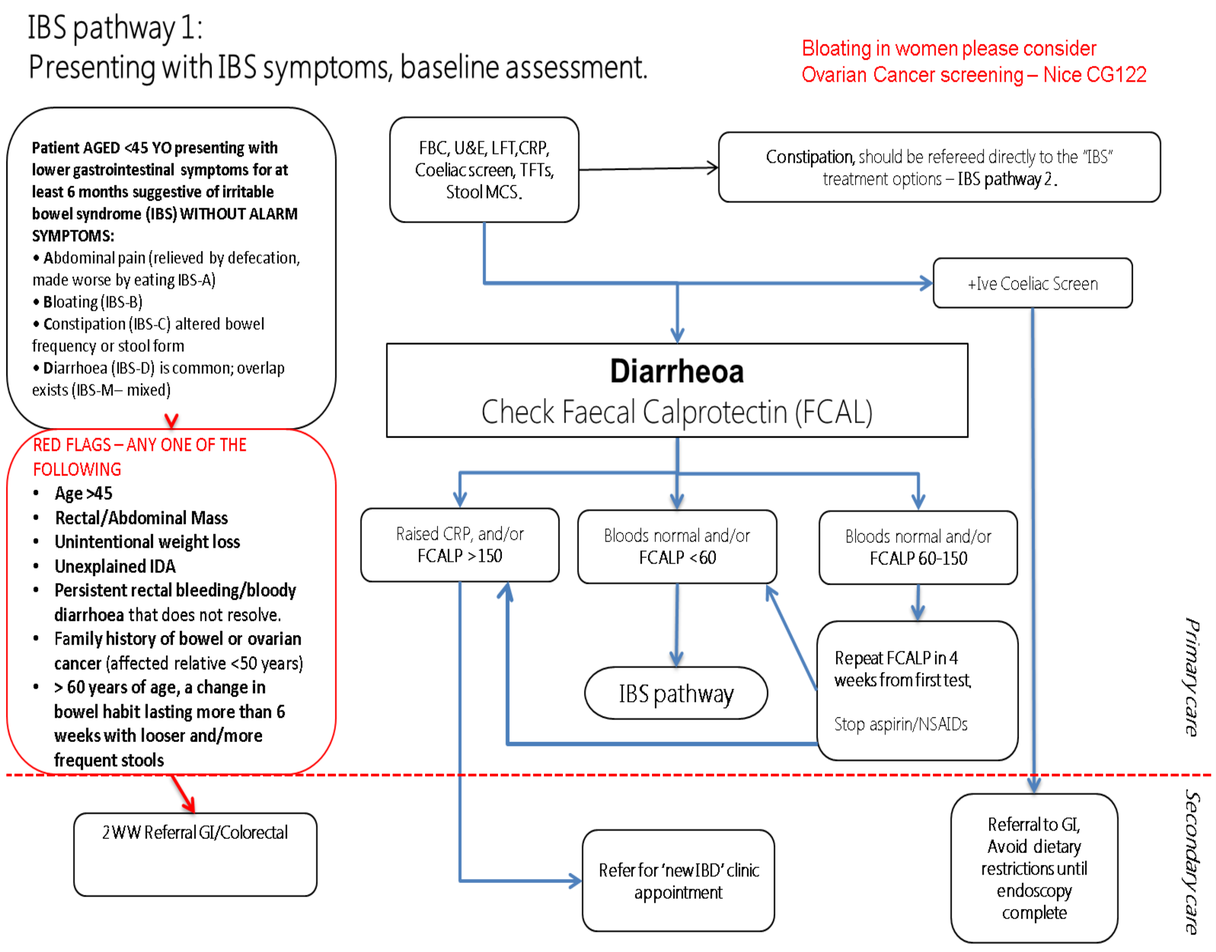


Figure 2. Primary care pathway for patients presenting with IBS symptoms.

The gastroenterology team delivered educational workshops to primary care and we provided further information and guidance via the Heart of England NHS Foundation Trust (HEFT) pathology website [2].

Post Implementation

In summer 2016, two of our local Clinical Commissioning Groups (CCGs) agreed to fund FCAL testing within the primary care setting. This additional funding was reflected in our FCAL workload figures with a dramatic increased in the number of GP requests from July 2016 onwards (see figure 3).

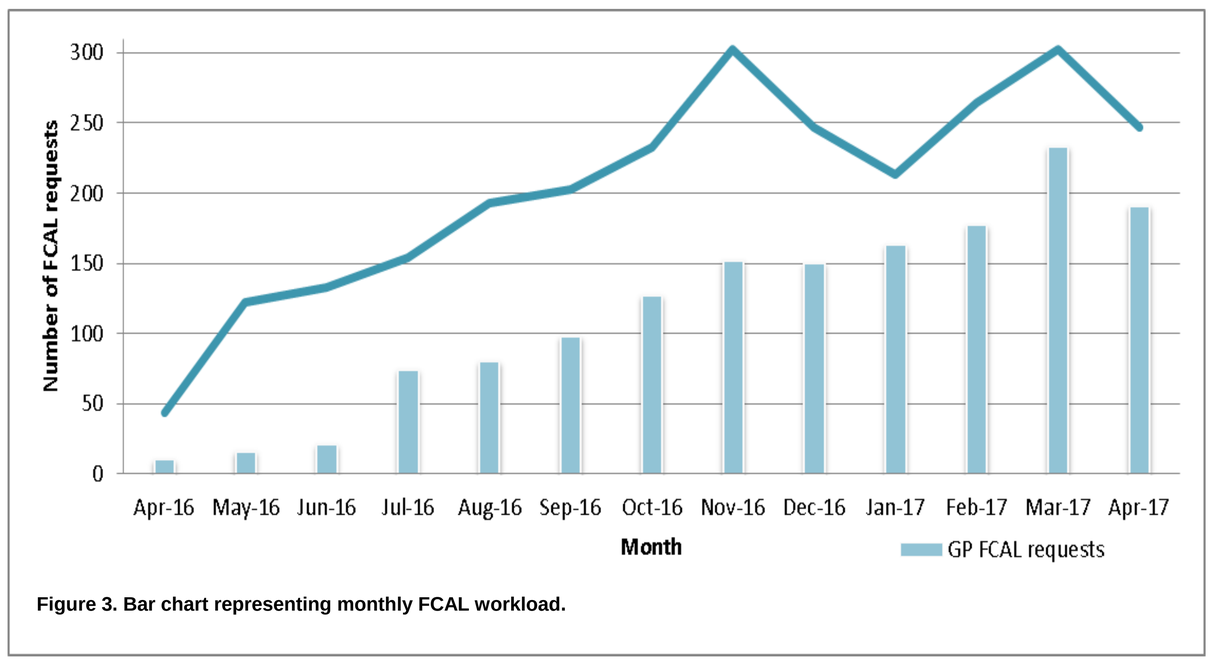


Figure 3. Bar chart representing monthly FCAL workload.

The FCAL workload has continued to rise thereafter, with slight drops observed during Christmas (December 2016) and Easter (April 2017).

The CALEX® Cap extraction device has allowed simplified sample handling, which has saved time and improved laboratory workflow especially as the devices can be loaded directly onto the Abbott ARCHITECT platform without the need to decant or dilute the sample.

Conclusions

Here we demonstrate the BÜHLMANN fCAL® turbo assay on the Abbott ARCHITECT platform is fit for purpose and we have recently received UKAS accreditation for this assay. Close collaboration with our gastroenterology team has enabled us to provide a new IBS local pathway for primary care to follow.

The CALEX® Cap Extraction devices have allowed the service to be maintained with the continuing increasing workload due to its ease of use and ability to be directly loaded onto the Abbott ARCHITECT platform.

References

[1] Faecal calprotectin diagnostic tests for inflammatory diseases of the bowel. NICE guidance [DG11]. October 2013.

[2] http://www.heftpathology.com/item/faecal-calprotectin-and-local-ibs-guidelines.html