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Added Value - Use of EQA data

Gareth Davies

Summary Of Talk

- Determining Sensitivity and Specificity
  - Pregnancy Testing EQA Programme – Pregnancy Sensitivity Study
- Pre-analytical EQA
  - Serum Indices Programme
- Post-analytical EQA
  - Porphyrin EQA Programme - PBG Clinical Cases

An investigation to assess the sensitivity limits for Pregnancy Testing kits and devices on the Weqas Urine Pregnancy EQA Programme

Pool Code	Distribution Code	Spiked hCG conc. (IU/L)
1	W107, sample 4	5
2	W106, sample 4	10
3	W107, sample 3	15
4	W106, sample 3	20
5	W107, sample 2	25
6	W106, sample 2	30
7	W107, sample 1	40
8	W106, sample 1	50
9	W105, sample 2	Non – spiked

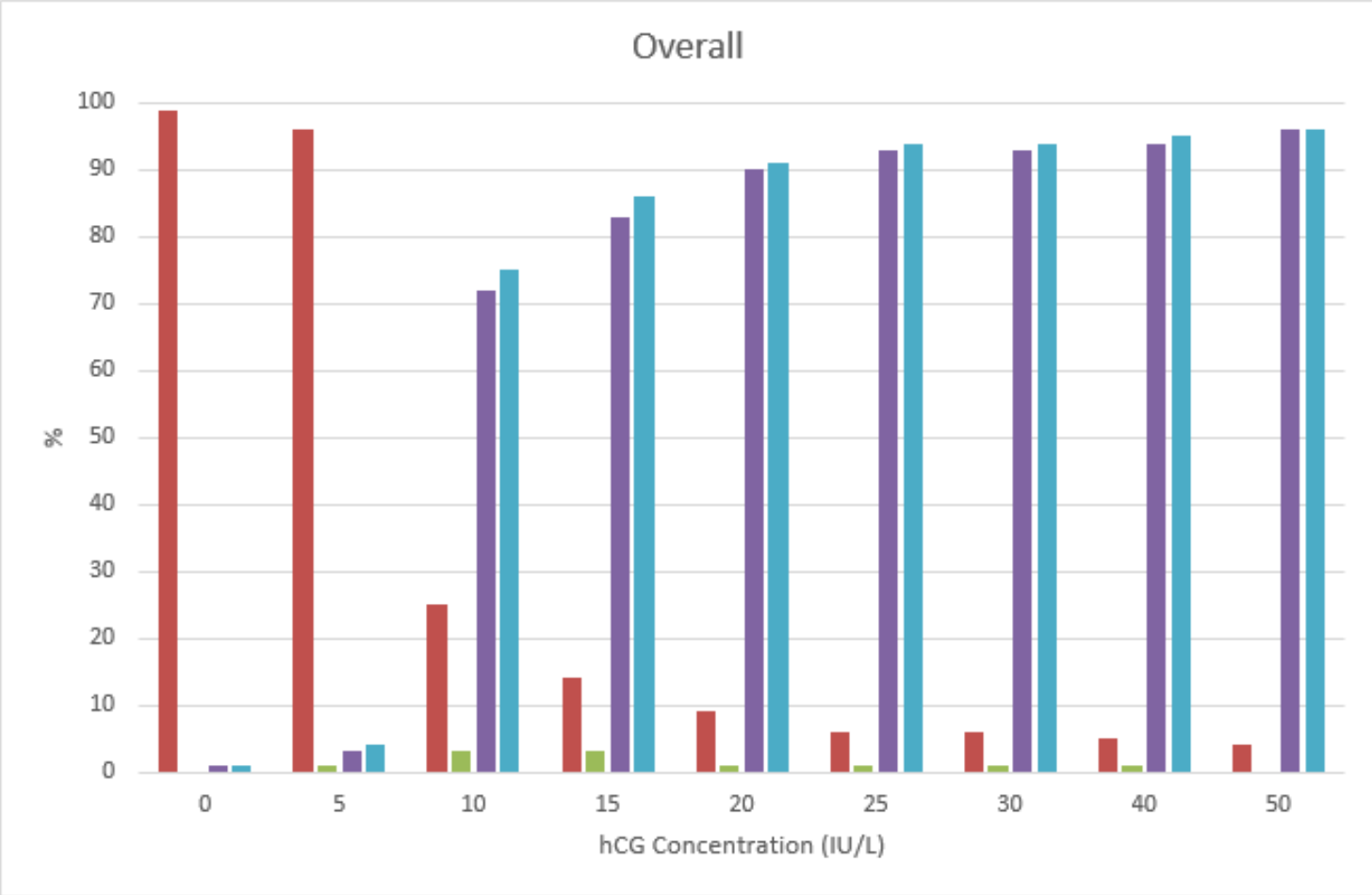
Number of Reported Results

Device / Kit	Number of reported results								
	Distribution W106 (Pools 2, 4, 6 & 8)				Distribution W107 (Pools 1, 3, 5 & 7)				Distribution W105 (Pool 9)
	2	4	6	8	1	3	5	7	9
AIDE Diag One Step Device	13	13	12	13	12	12	12	12	11
Alere Easy hCG	108	111	111	111	94	94	94	94	113
Alere hCG	155	155	155	155	138	138	138	137	170
SureStep One Step hCG	23	24	24	24	22	22	22	22	23
Checkmate	21	21	21	21	22	22	22	22	21
BioNexia	40	40	40	40	42	42	42	42	33
BioSign hCG DXpress	69	74	74	74	63	63	64	63	67
BioSign hCG Visual	36	37	37	37	43	43	43	43	48
One Step hCG	51	51	51	51	48	48	48	48	43
Clinitek Status	421	444	444	446	411	434	439	435	416
Qupid One Step	13	13	13	13	13	13	13	13	11
SureScreen hCG GHCGC	88	89	89	89	66	66	66	66	68
Total	**1038**	**1072**	**1071**	**1074**	**974**	**997**	**1003**	**997**	**1024**

Percentage Positive results for each kit / device at each different measured hCG concentration

% of Positive Results for Specified Concentration (IU/L)	0 IU/L	5 IU/L	10 IU/L	15 IU/L	20 IU/L	25 IU/L	30 IU/L	40 IU/L	50 IU/L
Alere Easy hCG	0	2	37	48	86	90	94	97	99
Sure Screen hCG GHCGC	0	2	39	65	74	82	81	85	84
BioSign hCG Visual	0	2	44	79	34	86	92	86	97
Qupid One Step	0	0	46	85	62	92	85	92	92
Alere hCG	1	4	57	78	87	92	90	93	92
One Step hCG	2	10	59	75	69	81	73	81	86
Sure Step One Step hCG	0	0	70	73	96	86	96	86	100
Bio Nexia	0	5	73	90	88	95	95	95	98
AIDE Diag One Step Device	0	0	84	75	84	75	83	75	84
Checkmate	5	0	86	86	100	86	95	95	100
Clinitek Status	1	2	91	93	99	98	100	99	100
BioSign hCG DXpress	0	3	96	97	99	98	100	100	100

Percentage of negative results (red), equivocal results (green), positive results (purple) and positive plus equivocal (non-negative results) (light blue) for each pregnancy testing kit / device.



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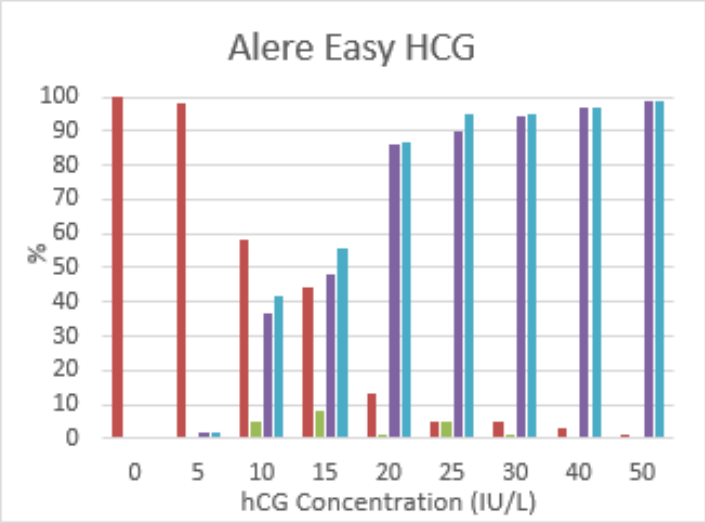

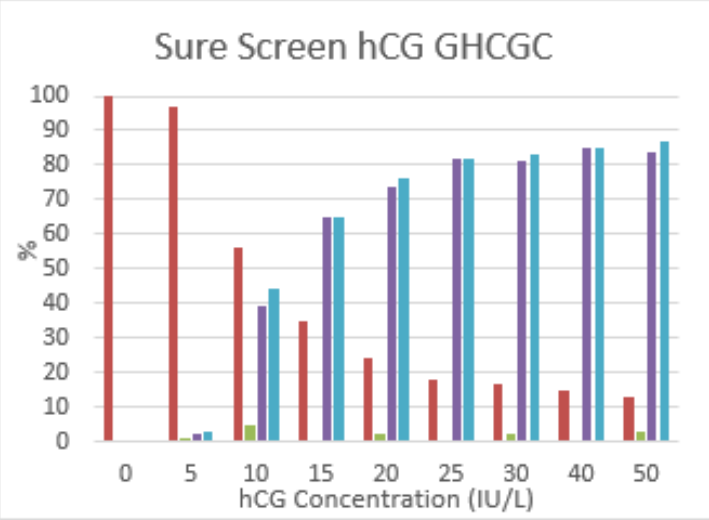

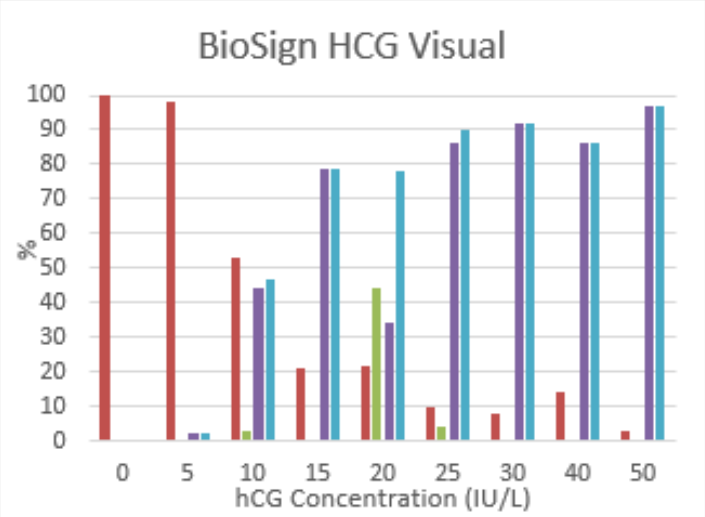

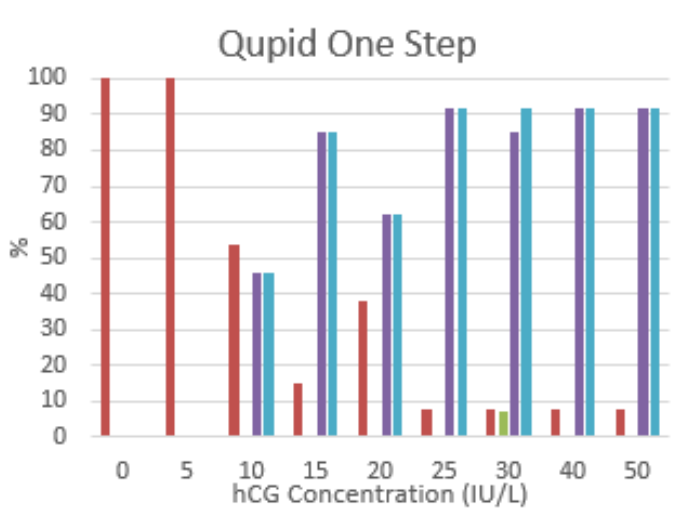

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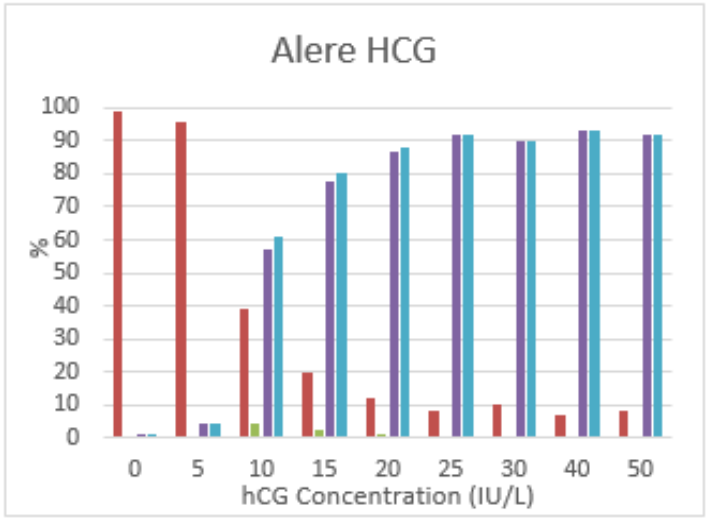

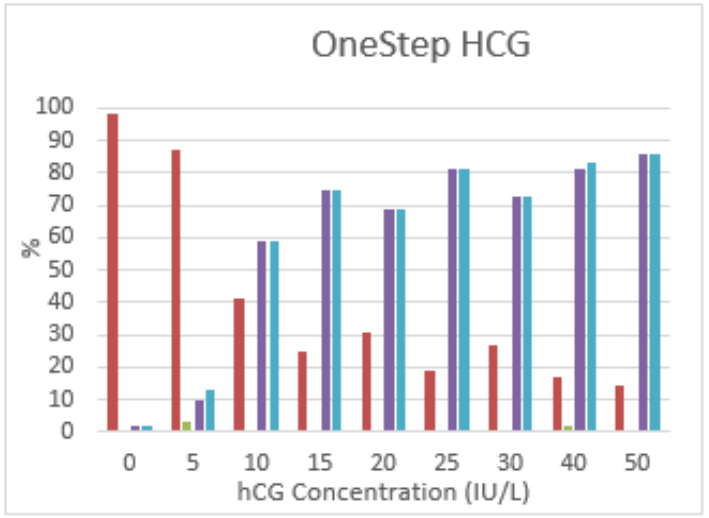

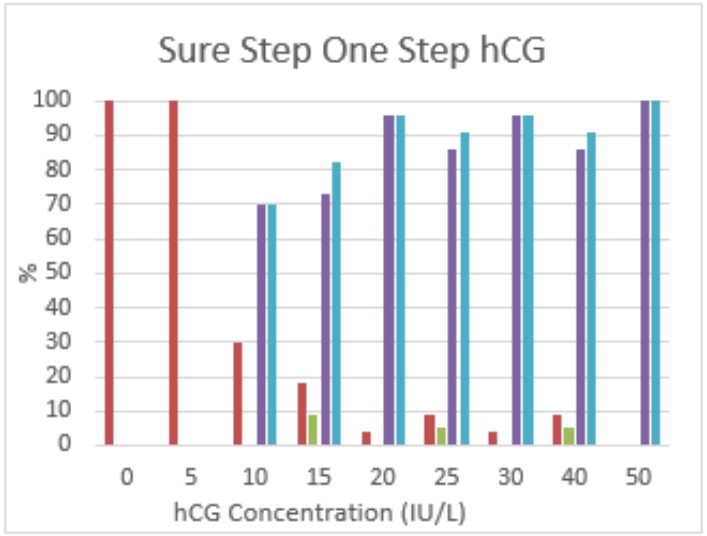

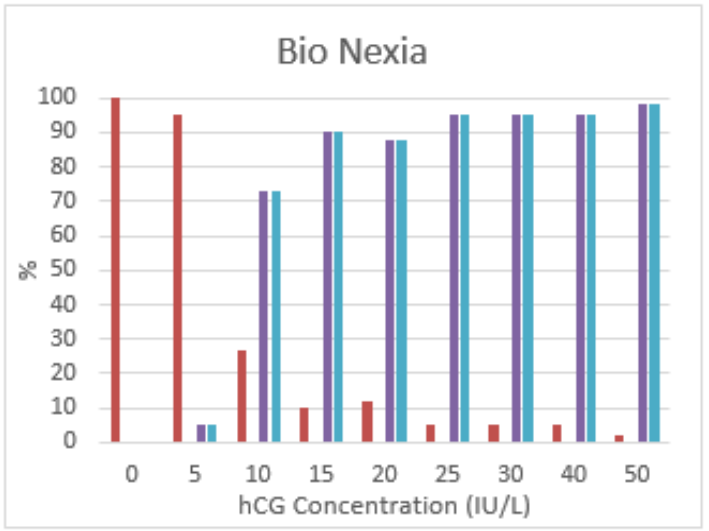

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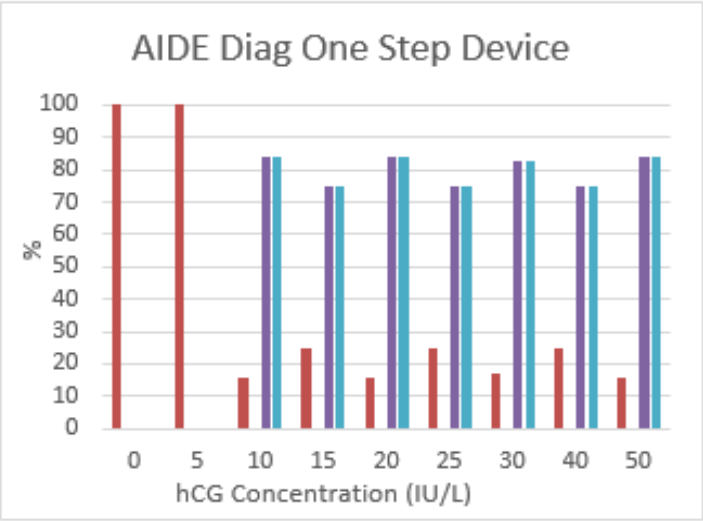

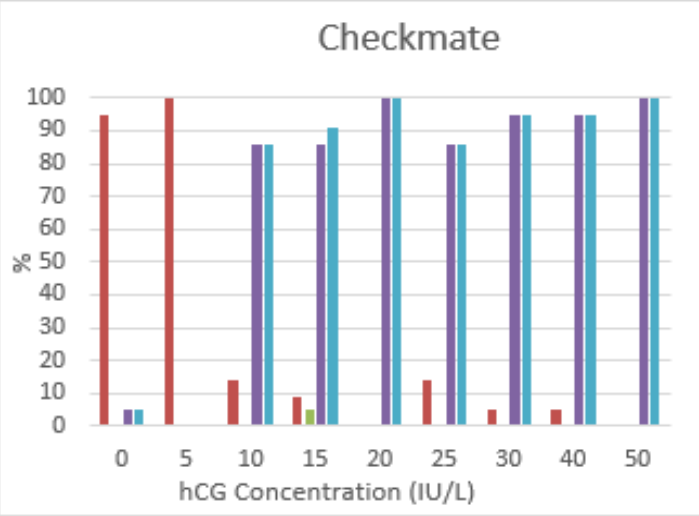

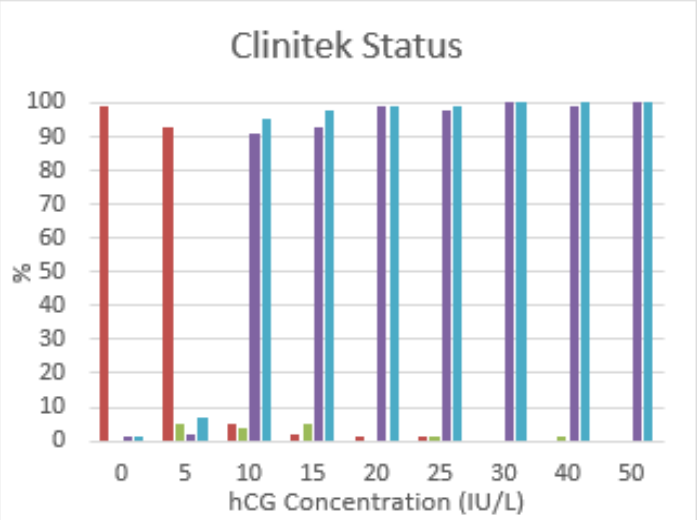

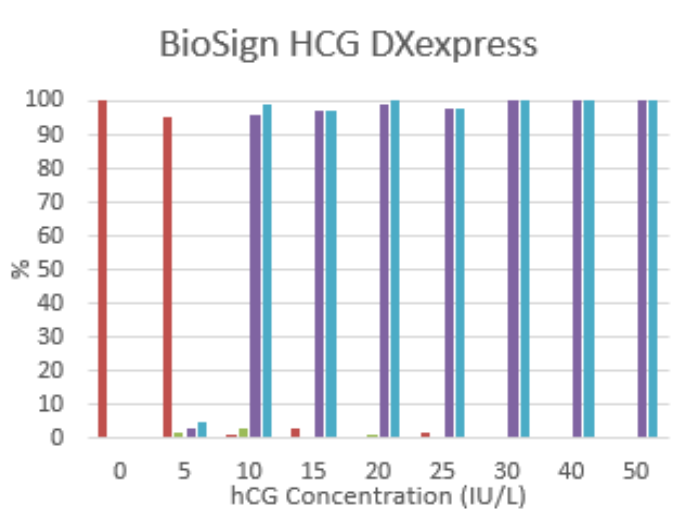

Overall Results

- The majority of users correctly identified the true negative (unadulterated urine from healthy non-pregnant female donors), with just 0.7% of users (7 out of 1024) reporting a positive result.
- At a hCG concentration of 5 IU/L, 96% of users reported a negative result for this sample. The One Step hCG users reported the highest number of non-negative results for this sample (13%) with 5 users reporting positive and 1 equivocal result.
- At a concentration of 10 IU/L, well below the stated cut-off of most methods, 71% of users reported a positive result, with 8 of the 12 methods reporting a positive rate of > 50%. The 2 machine read devices, the Clinitek Status and the BioSign hCG DXpress had the highest analytical sensitivity with 95% and 99% reporting non- negative results (positive plus equivocal) respectively.
- At a hCG concentration of 15 IU/L, all methods, except for the Alere Easy hCG, reported > 50% positive results. Overall, 83% of all users reported a positive result at this concentration.
- At a hCG concentration of 20 IU/L, all methods reported > 50% positive results for this sample with 90% of all users reporting a positive result.
- At a hCG concentration of 25 IU/L, 93% of all users reporting a positive result with a positivity rate of 98% for the two machine read devices.
- Overall positive rates at hCG concentrations of 30, 40 and 50 IU/L were 94%, 95% and 96% respectively.

Conclusions

- All kits / devices reported positive results at concentrations well below their stated kit cut-off.
- The 2 instruments (Clinitek Status and BioSign hCG DXpress) reported a higher % positive rate at 10 IU/L than the visually-read kits/devices.
- The 2 instruments (Clinitek Status and BioSign hCG DXpress) reported a higher % positive rate for all samples above 25 IU/L than the visually-read kits/devices.

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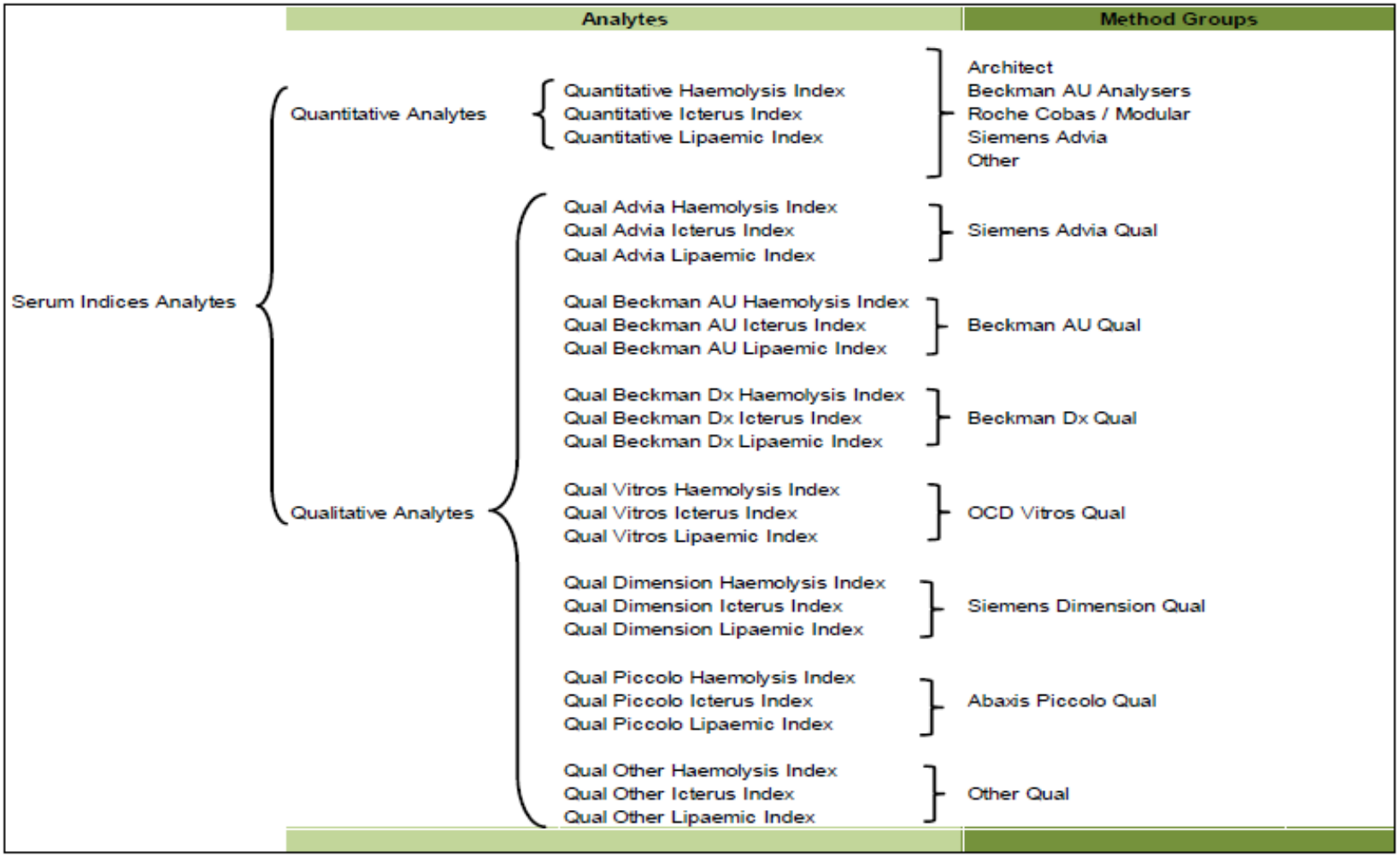
- Pre-analytical EQA
  - Serum Indices Programme
- Post-analytical EQA
  - Porphyrin EQA Programme - PBG Clinical Cases

Pre-analytical EQA - Serum Indices Programme

Table 1 - Units of Reporting			
Platform	**Units**		
	Haemolysis	**Icterus**	**Lipaemia**
Siemens	Ordinal , mg/dL	Ordinal , mg/dL	Ordinal , mg/dL
Abbott	g/L	µmol/L	mmol/L
Beckman	Ordinal , mg/dL	Ordinal , mg/dL	Ordinal , mg/dL
Roche	mg/dL , mAbs, µmol/L	mg/dL, mAbs , µmol/L	mg/dL, mAbs
Vitros	mg/dL	Ordinal	Ordinal, mg/dL

Pre-analytical EQA - Serum Indices Programme



Pre-analytical EQA - Serum Indices Programme

Haemolysis

Tgt Value	Qual Tgt
0.5	+
1.0	++
1.4	++
2.7	+++
3.1	++++

Icterus

Tgt Value	Qual Tgt
60	+
100	+/++
160	++
200	++
260	+++
342	+++
360	+++
460	++++
480	++++

Lipaemia

Tgt Value	Qual Tgt
2.5	++
2.97	++
4.0	++
7.0	++
9.4	+++
12.14	+++

Pre-analytical EQA – Qualitative Results

Haemolysis

From Quantitative Haemolysis Index Results				
Sample Number	1	2	3	4
Mean	0	2.8	0	0
SD	0.01	0.13	0.01	0.01
CV %	130%	5%	93%	104%

Analyte: Qual Advia Haemolysis Index

	Sample Number			
	1	2	3	4
Interpretation	Neg,-	+++	Neg,-	Neg,-

Number of Responses	Sample 1	Sample 2	Sample 3	Sample 4
Neg,-	30	-	30	30
+	-	-	-	-
++	-	-	-	-
+++	-	30	-	-
++++	-	-	-	-
TOTAL	30	30	30	30

Analyte: Qual Beckman AU Haemolysis Index

	Sample Number			
	1	2	3	4
Interpretation	Neg,-	+++	Neg,-	Neg,-

Number of Responses	Sample 1	Sample 2	Sample 3	Sample 4
Neg,-	35	-	35	35
+	-	-	-	-
++	-	-	-	-
+++	-	10	-	-
++++	-	24	-	-
+++++	-	1	-	-
TOTAL	35	35	35	35

Icterus

From Quantitative Icterus Index Results				
Sample Number	1	2	3	4
Mean	1.6	1.6	1.8	171.8
SD	2.30	1.99	2.24	9.49
CV %	144%	127%	123%	6%

Analyte: Qual Advia Icterus Index

	Sample Number			
	1	2	3	4
Interpretation	Neg,-	Neg,-	Neg,-	++

Number of Responses	Sample 1	Sample 2	Sample 3	Sample 4
Neg,-	23	23	23	-
+	-	-	-	3
++	-	-	-	20
+++	-	-	-	-
++++	-	-	-	-
TOTAL	23	23	23	23

Analyte: Qual Beckman AU Icterus Index

	Sample Number			
	1	2	3	4
Interpretation	Neg,-	Neg,-	Neg,-	++

Number of Responses	Sample 1	Sample 2	Sample 3	Sample 4
Neg,-	26	26	26	-
+	-	-	-	-
++	-	-	-	26
+++	-	-	-	-
++++	-	-	-	-
+++++	-	-	-	-
TOTAL	26	26	26	26

Lipaemia

From Quantitative Lipaemic Index Results				
Sample Number	1	2	3	4
Mean	0.2	1.9	0.2	0.2
SD	0.07	0.38	0.07	0.07
CV %	34%	20%	36%	35%

Analyte: Qual Advia Lipaemia Index

	Sample Number			
	1	2	3	4
Interpretation	Neg,-	++	Neg,-	Neg,-

Number of Responses	Sample 1	Sample 2	Sample 3	Sample 4
Neg,-	23	1	23	23
+	-	22	-	-
++	-	-	-	-
+++	-	-	-	-
++++	-	-	-	-
TOTAL	23	23	23	23

Analyte: Qual Beckman AU Lipaemic Index

	Sample Number			
	1	2	3	4
Interpretation	Neg,-	++	Neg,-	Neg,-

Number of Responses	Sample 1	Sample 2	Sample 3	Sample 4
Neg,-	26	-	26	26
+	-	-	-	-
++	-	26	-	-
+++	-	-	-	-
++++	-	-	-	-
+++++	-	-	-	-
TOTAL	26	26	26	26

Pre-analytical EQA – Quantitative Results

Distribution:		IRL Base	IRL Base+H&L	IRO Base	IRO Base+I	IRR Base	IRR Base+L
Semi-Quant Haemolysis Index (g/L)							
Instrument							
	Overall Mean	0.01	0.52	0.01	0.01	0.01	0.01
	Overall SD	0.01	0.08	0.01	0.01	0.01	0.01
	Overall Number	131	131	136	137	134	135
	Reference Value	0	+	0	0	0	0
Architect	Instrument Mean	0.0	0.51	0.0	0.0	0.0	0
	Instrument SD	0.0	0.03	0.0	0.0	0.0	0.01
	Number	28	27	31	32	34	34
Cobas	Instrument Mean	0.01	0.54	0.01	0.01	0.01	0.01
	Instrument SD	0.01	0.04	0.01	0.01	0.01	0.01
	Number	82	86	96	96	93	94

Pre-analytical EQA – Quantitative Results

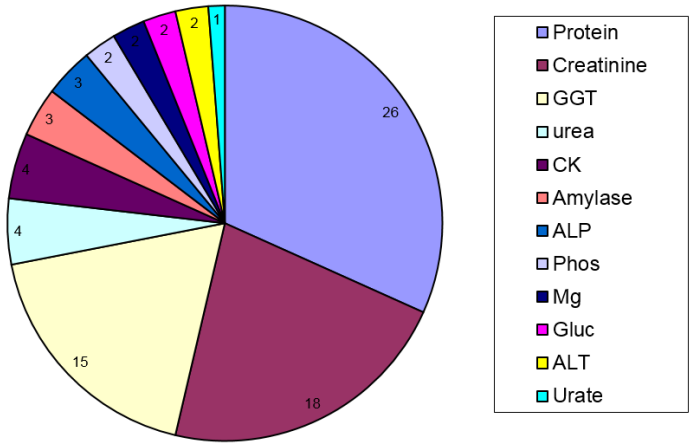
Distribution:		IRL Base	IRL Base+H&L	IRO Base	IRO Base+I	IRR Base	IRR Base+L
Semi-Quant Icterus Index (umol/L)							
Instrument							
	Overall Mean	1.89	0.06	1.82	171.8	0.91	0.03
	Overall SD	2.61	0.02	2.24	9.49	1.39	0.18
	Overall Number	136	125	152	152	139	135
	Reference Value	0	0	0	180	0	0
Architect	Instrument Mean	4.9	0.07	4.2	172.5	2.39	0.05
	Instrument SD	1.89	0.21	0.83	7.96	1.45	0.27
	Number	25		31	32	35	33
Cobas	Instrument Mean	0.0	0.0	0.0	171.2	0.0	0.0
	Instrument SD	0.0	0.0	0.0	10.76	0.0	0.0
	Number	82	82	85	85	85	85

Pre-analytical EQA – Quantitative Results

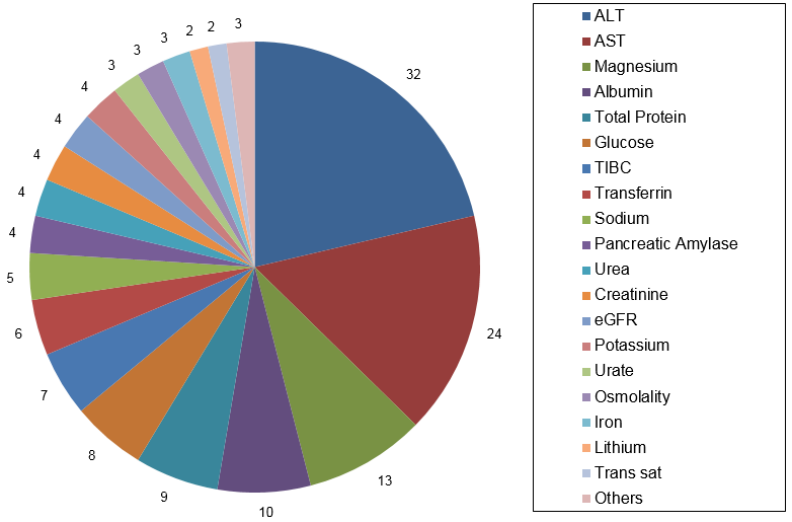
Distribution:		IRL Base	IRL Base+H&L	IRO Base	IRO Base+I	IRR Base	IRR Base+L
Semi-Quant Lipaemic Index (g/L)							
Instrument							
	Overall Mean	0.12	1.81	0.19	0.16	0.21	1.91
	Overall SD	0.06	0.29	0.06	0.06	0.07	0.38
	Overall Number	128	128	147	146	147	146
	Reference Value	0	2.0	0	0	0	2.0
Architect	Instrument Mean	0.02	1.39	0.08	0.04	0.1	1.37
	Instrument SD	0.02	0.09	0.02	0.03	0.01	0.13
	Number	28	27	29	33	32	35
Cobas	Instrument Mean	0.13	1.9	0.21	0.18	0.21	1.82
	Instrument SD	0.02	0.19	0.04	0.03	0.04	0.22
	Number	96	97	110	115	113	111

Pre-analytical EQA - Serum Indices Programme

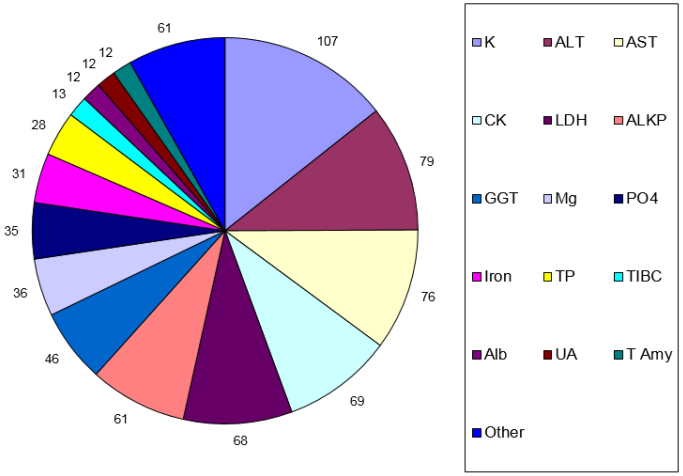
Icterus Sample, Bilirubin = 320 µmol/L



Lipaemic Sample, Triglyceride = 9 mmol/L



Haemolysed Sample, Hb = 1 g/L



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Post-analytical EQA – Porphyrin Clinical Cases

PBG Clinical Cases

Please note that for this distribution we have included an educational interpretative exercise. Clinical information has been provided below for each of the 3 PBG samples. Please enter your results for the 3 samples on this return sheet (as well as electronically on weqas.com). Alternatively you can send us a copy of your laboratory report including your comments.

Please treat all samples as you would a patient sample and comment on the results obtained, providing details of any further investigation you feel may be appropriate.

Lab Code: _____ *(please complete)*

PBG Sample 1

Result: .....µmol/l

Result:.....µmol/mmol Creatinine

Clinical Details:

A 35 year old known Variegate Porphyria (VP) patient, diagnosed on family screening, is admitted with acute abdominal pain, nausea and vomiting. A random urine sample is sent for porphobilinogen measurement.

Report Comment

Case 1

- *Clinical Details:* A 35 year old known Variegate Porphyria (VP) patient, diagnosed on family screening, is admitted with acute abdominal pain, nausea and vomiting. A random urine sample is sent for porphobilinogen measurement.
- *Results:*
- Quantitative PBG (µmol/L)
  - Overall Mean 112.53, SD 15.45, n=19
- Quant PBG/Creatinine Ratio (µmol/mmol)
  - Overall Mean 23.42, SD 3.21, n=14
- Creatinine (mmol/L)
  - Overall Mean 4.65, SD 0.17, n=14
- Qualitative PBG
  - 13 Positive
  - 7 Strong Positive

Case 1

- *Clinical Details:* A 35 year old known Variegate Porphyria (VP) patient, diagnosed on family screening, is admitted with acute abdominal pain, nausea and vomiting. A random urine sample is sent for porphobilinogen measurement.
- *Case Results (Expert Opinion):*
- This example was based on an actual case.
- In variegate porphyria (VP) urine PBG excretion is invariably normal during latency and increases only during an acute attack.
- In this case urine PBG excretion is increased 15-20 fold and therefore confirms an acute attack of porphyria as a cause of the symptoms.
- The patient is known to have VP so no other investigations are required.
- The result should be telephoned.
- Clinical advice (if sought) should be to provide symptomatic relief (opiates, IV fluids etc), to start haem-arginate therapy, establish possible precipitating factor (unsafe drugs, infection).
- We do not advocate monitoring of urine porphyrin and PBG following diagnosis and initiation of treatment for an acute attack.

Case 1

- *Clinical Details:* A 35 year old known Variegate Porphyria (VP) patient, diagnosed on family screening, is admitted with acute abdominal pain, nausea and vomiting. A random urine sample is sent for porphobilinogen measurement.
- *Participant Comments - Quantitative PBG Users:*
- 14 users provided comments
- 11 stated results consistent with acute attack
- 5 provided details of specialist porphyria services/referral labs for advice
- Others included – result to be phoned, ensure exclude other causes, look for precipitating factors
- *Participant Comments - Qualitative PBG Users:*
- 2 users provided comments – both stated results consistent with acute attack
- *Example comment:* The raised urinary porphobilinogen (PBG) concentration is consistent with an attack of acute porphyria. However, unfortunately there are no clear threshold values above which symptoms appear. Although typically PBG returns to normal within days of an acute attack of VP. Has the patient started haem arginate? It would be prudent to identify any precipitating factors including starvation, infection oral contraception, stress, alcohol and a wide range of common drugs.

Case 2

- *Clinical Details:* A 25 yr old female known to have Acute Intermittent Porphyria (AIP) presenting to A & E with abdominal pain.
- *Results:*
- Quantitative PBG (µmol/L)
  - Overall Mean 33.36, SD 4.45, n=19
- Quant PBG/Creatinine Ratio (µmol/mmol)
  - Overall Mean 5.46, SD 0.84, n=14
- Creatinine (mmol)
  - Overall Mean 6.194, SD 0.28, n=14
- Qualitative PBG
  - 1 Negative, 1 Equivocal (further investigation), 6 Weak Positive, 12 Positive

Case 2

- *Clinical Details:* A 25 year old female known to have Acute Intermittent Porphyria (AIP) presenting to A & E with abdominal pain.

- *Case Results (Expert Opinion):*
- The elevation in PBG is minimal and typical of values seen during remission or the latent phase of AIP.
- Unfortunately there is no clear threshold value above which symptoms appear but a current attack of acute porphyria is very unlikely and an alternative cause for her abdominal pain should be sought.
- Attacks of AIP are usually associated with gross elevations in PBG (typically 10 to 100 fold).
- There is a danger of an attack of porphyria developing if she is not managed appropriately so there is justification for repeating the test at intervals should her symptoms fail to resolve.

Case 2

- *Clinical Details:* A 25 year old female known to have Acute Intermittent Porphyria (AIP) presenting to A & E with abdominal pain.
- *Participant Comments - Quantitative PBG Users:*
- 14 users provided comments
- 10 stated PBG results can be elevated to this degree in between acute attacks
- 4 stated PBG results do not exclude or confirm an acute attack
- 5 provided details of specialist porphyria services/referral labs for advice
- 1 stated Raised PBG/creatinine ratio, consistent with an acute attack of AIP, although usually during an attack, urine PBG/creatinine ratio levels are at least five times the upper limit of the reference range
- 1 stated Elevated PBG, which *may* be consistent with an acute attack – suggest exclude other causes and contact NAPS for advice
- No Qualitative PBG users provided comments
- *Example comment:* Raised PBG/creatinine in a patient with known AIP. Diagnosis of an acute attack in a patient with known AIP is difficult to ascertain as it takes years for the levels to return to normal. Would need to exclude common causes of abdominal pain before diagnosing this as an acute attack. Detailed clinical history is important. Suggest liaison with National Acute Porphyria Service for advice.

Case 3

- *Clinical Details:* A GP sends a random urine sample from the 55 year old mother of a patient recently diagnosed with Acute Intermittent Porphyria (AIP).

- *Results:*
- Quantitative PBG (µmol/L)
  - Overall Mean 11.39, SD 1.42, n=19
- Quant PBG/Creatinine Ratio (µmol/mmol)
  - Overall Mean 0.77, SD 0.09, n=14
- Creatinine (mmol)
  - Overall Mean 13.97, SD 0.46, n=15
- Qualitative PBG
  - 13 Negative, 3 Equivocal (further investigation), 2 Weak Positive, 2 Positive

Case 3

- *Clinical Details:* A GP sends a random urine sample from the 55 year old mother of a patient recently diagnosed with Acute Intermittent Porphyria (AIP).
- *Case Results (Expert Opinion):*
- The normal PBG does not exclude latent porphyria in a family member of an affected patient.
- Urine PBG excretion can be increased in about 40% of pre-symptomatic AIP patients, although not before puberty.
- Further investigation is required using enzyme measurement or genetic testing.
- The gold standard test is genetics - Good practice would be to suggest referral of the patient to a clinical genetics service for counselling and to arrange genetic testing for this patient as well as testing for other family members.

Case 3

- *Clinical Details:* A GP sends a random urine sample from the 55 year old mother of a patient recently diagnosed with Acute Intermittent Porphyria (AIP).
- *Participant Comments - Quantitative PBG Users:*
- 14 users provided comments
- 5 stated results exclude current acute attack
- 14 stated that latent AIP could not be excluded
- 14 suggested further studies and /or discussion with NAPS
- *Participant Comments - Qualitative PBG Users:*
- No users provided comments
- *Example comment:* These results exclude a current attack of acute porphyria. During an attack both ALA and PBG are significantly raised. These results do not exclude a neurological porphyria in remission or the latent phase. This type of porphyria is inherited in an autosomal dominant manner so it is essential to carry out family studies in order to identify asymptomatic relatives who have inherited the defect and who will also be at risk of developing life-threatening porphyric attacks if exposed to precipitants. Further information on drugs can be found in the BNF or obtained from the Welsh Drug Information centre (Tel: 029 20724779). Suggest referral to the Porphyria Clinic

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Thank you

Any questions?