UK NEQAS

International Quality Expertise

Hepatitis B serology scheme results

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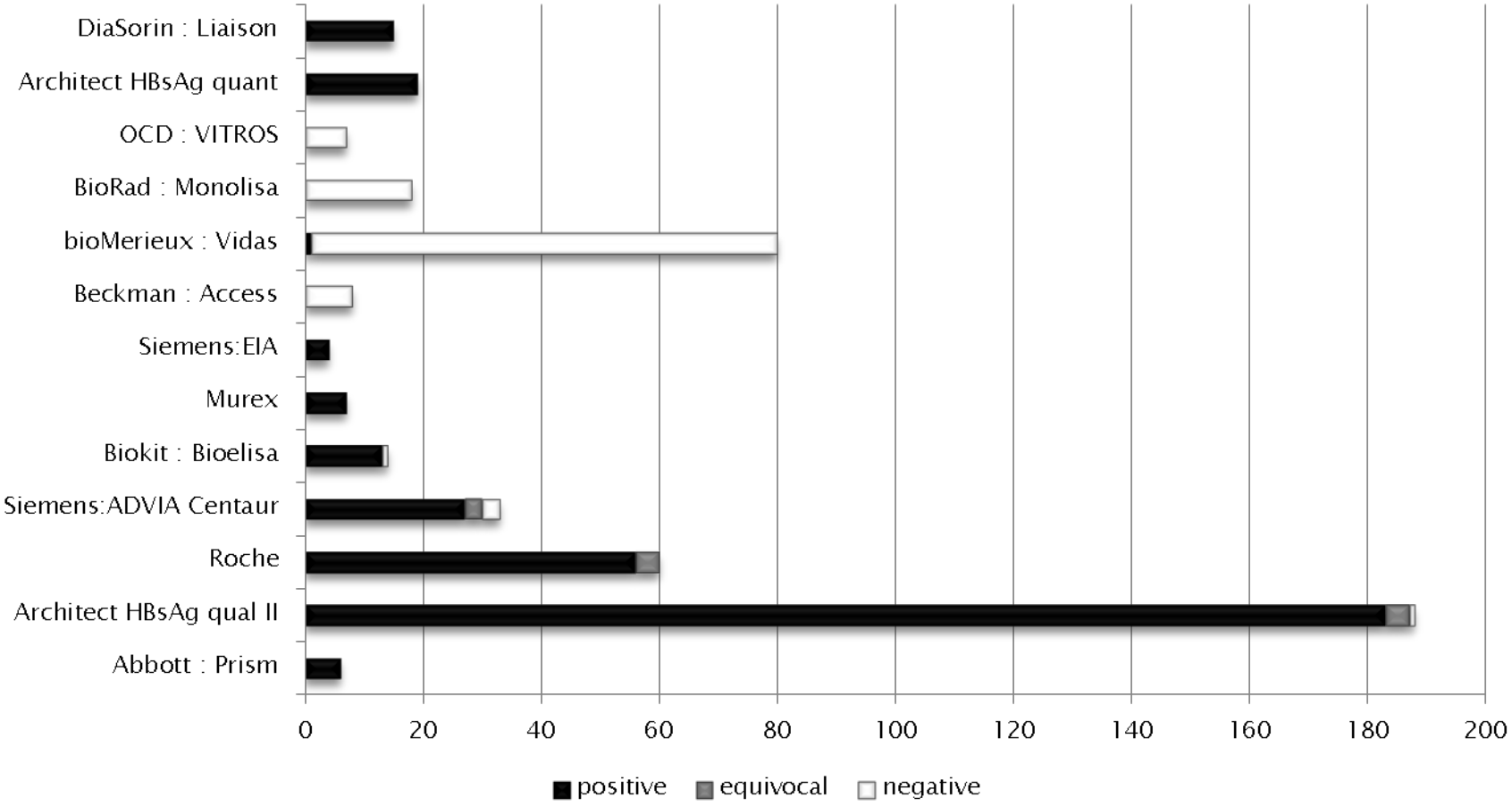
Hepatitis B serology scheme

- Three distributions/ year
- 6 specimens/ distribution
- Liquid human serum
- Report on HBsAg status (and Hepatitis B markers for anti-HBc, anti-HBc IgM, HBeAg, anti-HBe, if tested)

Distribution 3787, spec. 2931



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Distribution 3787, spec. 2931

Table 1: Assays detecting HBsAg in sample

	Abbott : Prism	Architect HBsAg qual II	Roche	Siemens:ADVIA Centaur	Biokit : Bioelisa	Murex	Siemens:EIA
median	69.535	7.62	3.39	12.37	0.1395	0.589	0.285
mean	66.515	7.425	3.35	12.08	0.151	0.611	0.915
SD	24.66	2.58	0.81	5.98	0.027	0.177	1.325
min	38.99	1.53	0.1	0.05	0.132	0.338	0.191
max	105.82	13.91	5.61	29.9	0.185	0.891	2.901
cut off	Index=1	Index=1	Index=1	Index=1	OD range: 0.06 – 0.10	OD range: 0.1 – 0.173	OD range: 0.058 – 0.082
n	6	188	60	33	14	7	4

Table 2: Assays failed to detect HBsAg in sample

	Beckman : Access	OCD : VITROS	bioMerieux : Vidas	Bio-Rad : Monolisa
median	0.375	0.09	0.03	0.019
mean	0.376	0.090	0.038	0.045
SD	0.041	0.023	0.021	0.12
min	0.3	0.06	0	0.011
max	0.43	0.13	0.15	0.38
cut off	Index=1	Index=1	TV=0.13	OD range: 0.06 – 0.08
n	8	7	80	18

Distribution 3787, spec. 2931

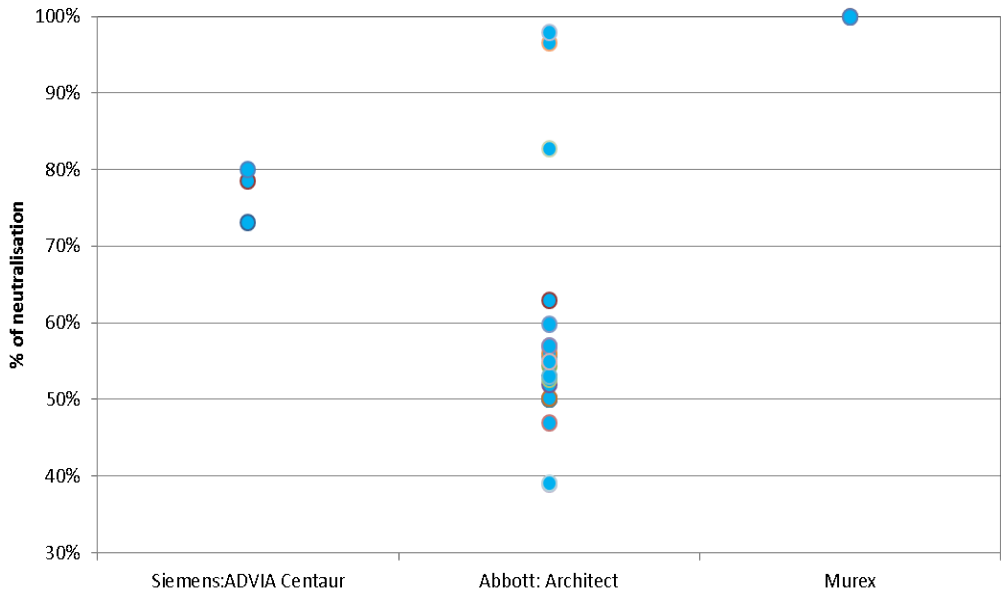
- Quantitative result: 100% positive

	Architect HBsAg quant	DiaSorin : Liaison
median	0.95	3.3
mean	0.956	2.897
SD	0.43	1.12
min	0.3	0.75
max	2	4.3
cut off	0.05 IU/mL	0.05 IU/ml
n	19	15

Distribution 3787, spec. 2931

- Neutralisation assays: 90% of results confirmed HBsAg

	Abbott: Architect	Siemens: ADVIA Centaur	Murex
median	53.7%	79%	100%
mean	57.6%	77.2%	100%
SD	0.15	0.04	0
min	39.0%	73.1%	100%
max	97.9%	80.0%	100%
n	24	3	3



Three results negative: 39%, 47%

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Process for investigation of suspected assay failure

…where an assay gives discrepant results in comparison to other assays results and for a significant number of participants.

- Report to the manufacturer
- Manufacturer: Investigations on the specimen and on any changes in the assay
- UK NEQAS & Manufacturer to establish the cause and whether this affects only EQA specimen or may affect clinical sample analysis as well
- Possible implications for clinical sample analysis: contact the MHRA (*MHRA: Medical & Healthcare products Regulatory Agency)*

UKNEQAS investigation

- Limit of detection for assays

	Beckman : Access	bioMerieux : Vidas	Bio-Rad : Monolisa	OCD : VITROS
WHO NIBSC 00/588	**0.056 IU/mL** (95% CI: 0.054–0.059)	**0.05 IU/mL** (L); 0.075 IU/mL (S)	**0.025 IU/ml** CI95 [0.019–0.037 IU/ml]	**0.080 IU/mL**

- Specimens characteristics:
  - HBsAg positive serum diluted 1:160 in HBsAg negative serum
  - Diluent serum negative for RF and IM
  - HBsAb in the sample:
    - HBsAg/HBsAb complexes
    - Free HBsAg detected with other assays

- HBsAg sequencing (HBsAg encoding region) in progress

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UKNEQAS investigation

- Clinical relevance: Prevalence of both HBsAg and HBs Ab is not frequent, but can be found in:
  - Patients with chronic hepatitis B
  - During late stage of infection (recovered infection)
  - Infection with different HBV subtypes
  - Mutations (presence of ‘a’ determinant mutant)

Manufacturers

- Results (anonymised) sent to manufacturers for their investigation
- Samples provided to manufacturers for their own investigation

- Manufacturers responses:
  - The test should be performed on undiluted serum or plasma (package insert)
  - Investigation and communication still on-going:
    - HBs Ab are probably the reason
    - Waiting for our report on HBV sequencing

Participants results (examples)

- Neutralisation 39%: participant not following their daily procedures with testing and interpretation:
  - We encourage participants to follow handling, testing (according to their algorithm) and interpretation of results the way they do with their daily samples in order to find deviations in the process
  - Investigating EQA failures, finding the root cause, putting in place preventive actions can be beneficial for daily routine testing not just for getting correct results in EQA schemes

Participants results (examples)

- Participants reporting more than one method:
  - Interpretation not followed by manufacturer instructions
  - Overall interpretation equivocal (individual test results negative and positive)

	Kit details	Kit change details	Batch number	Cut-off reading
HBsAg kit 1	Architect HBsAg qual II		56255LH00	1.00
HBsAg kit 2	bioMerieux : Vidas		160820-0	<0.13 =>0.13
HBsAg kit 3				

	Specimen : 2931
HBsAg report	○ Not Examined ○ Negative ◉ Equivocal ○ Positive
HBsAg kit 1	○ Not Examined ○ Negative ◉ Equivocal ○ Positive
HBsAg Test Reading kit 1	10.11
HBsAg kit 2	○ Not Examined ◉ Negative ○ Equivocal ○ Positive
HBsAg Test Reading kit 2	0.03
HBsAg kit 3	◉ Not Examined ○ Negative ○ Equivocal ○ Positive
HBsAg Test Reading kit 3	

Conclusions

- EQA providers – Participants – Manufacturers
  - We all aim to get the best service for the patients
  - With addressing and investigating the problems, we can improve our services