What is a “Body Fluid”? 

- Fluids from the body other than samples commonly used for testing

- **NOT:** Plasma, serum, urine, CSF, whole blood, saliva, etc.
  - Where tests are **validated**

- **INCLUDES:** Pleural, peritoneal, pericardial, cyst, faeces, saliva, synovial, drain fluid, peritoneal dialysis fluid, CSF, urine, lymphatic, ocular
  - Where tests are **NOT validated**

https://en.wikipedia.org/wiki/Ascites
Body Fluids – a brief history

NATA/TGA publish new validation and registration requirements for in-house IVDs and for in-house modifications of TGA-approved methods

Dr Graham Jones: Routine method for CEA analysis was not manufacturer-validated for body fluid analysis
  • Poster at AACB ASM in Gold Coast validating Roche CEA for Fluid Analysis

The Victorian AACB Quality Assurance Group (VQAG) discussed the broader implications of Graham’s work, the NATA/TGA requirements re: the broad range of Body Fluid types/analytes measured
  • Acknowledged lack of validation for these

Acknowledgment: John Calleja and Fernando San Gil

Body Fluids – a brief history

October 2014
AACB + AACB Branch QAGs distributed a survey to the AU/NZ membership re: how fluid analysis requests are handled, measured and reported in their laboratories.

December 2016
John Calleja et al. report survey findings in *The Clinical Biochemist Reviews*

- Generally a non-uniform approach to test profiles analysed on Body Fluids
- Lack of uniformity in reporting of results e.g. RI and interpretive comments
- Clear lack of validation of in-house or manufacturer-based methods

2017
Changes came into effect for all NATA-accredited laboratories

- Commercial methods that were used outside the scope of the vendors’ specifications were targeted
- This was the case of nearly all Body Fluids testing!

Acknowledgement: John Calleja and Fernando San Gil
REQUIREMENTS FOR THE DEVELOPMENT AND USE OF IN-HOUSE IN VITRO DIAGNOSTIC MEDICAL DEVICES (IVDs)

(Fourth Edition 2018)
1. General Requirements

S1.4 If any modification is made to a commercially supplied IVD or an existing in-house IVD, it must be treated as a new in-house IVD, and the modification must be validated in accordance with these Requirements.

C1.4(i) This must include changes to intended use (including sample type) or indications for use.

C1.4(ii) Where an IVD has been modified, the validation steps required are determined by the nature of the modification. It must be demonstrated that the changes have been properly assessed and show that the assay continues to perform safely and effectively.
4. Particular Requirements – Analytical Performance

S4.4 The accuracy and imprecision of in-house IVDs must be determined by at least one of the following methods applicable to the relevant biological material:

(a) the use of certified reference material
(b) comparison with a definitive method or a reference method
(c) performance of recovery experiments
(d) use of validated in-house reference material
(e) performance in external proficiency-testing programs or laboratory sample exchange programs.
Creatinine

Cholesterol

LDH

Lipase

Sodium

Protein

Glucose

Lactate

OSmolality

pH

Triglycerides

AFP

CA 125

CA 15-3

CA 19-9

CEA

hCG

2018 Body Fluids Pilot Analytes

RCPA QAP
Results from the 2018 Body Fluids Pilot
Albumin

- 56 BCP
- 13 BCG

Non-Infectious Pleural Fluid
Parapneumonic Effusion
Malignant Ascites

Amylase

Non-Infectious Pleural Fluid
Parapneumonic Effusion
Malignant Ascites
Lactate Dehydrogenase

Non-Infectious Pleural Fluid
Parapneumonic Effusion
Malignant Ascites

Lipase

Non-Infectious Pleural Fluid
Parapneumonic Effusion
Malignant Ascites
Osmolality

- **Non-Infectious Pleural Fluid**
- **Parapneumonic Effusion**
- **Malignant Ascites**

### Osmolality Graphs

- **Specimen 1-01**
  - Undesirably Low: 429
  - Undesirably High: 527
  - Median Value: 456

- **Specimen 1-02**
  - Undesirably Low: 503
  - Undesirably High: 610
  - Median Value: 533

- **Specimen 1-03**
  - Undesirably Low: 473
  - Undesirably High: 473
  - Median Value: 572

Protein

- **Non-Infectious Pleural Fluid**
- **Parapneumonic Effusion**
- **Malignant Ascites**

### Protein Graphs

- **Specimen 1-01**
  - Undesirably Low: 7
  - Undesirably High: 29
  - Median Value: 19

- **Specimen 1-02**
  - Undesirably Low: 32
  - Undesirably High: 54
  - Median Value: 44

- **Specimen 1-03**
  - Undesirably Low: 40
  - Undesirably High: 56
  - Median Value: 50
Sodium

Non-Infectious Pleural Fluid

Parapneumonic Effusion

Malignant Ascites

Triglycerides

Non-Infectious Pleural Fluid

Parapneumonic Effusion

Malignant Ascites
2019 Body Fluids Program

RCPAQAP
The Royal College of Pathologists of Australasia
Quality Assurance Programs
Body Fluids Material

- Sample base is diluted human plasma
- Analyte spikes where required (e.g. CA-199)
- Fluid material—no reconstitution required
- Range of Chemistry and Tumour Marker Analytes tested
Body Fluids Program – Analytical

- 6 discrete samples
  - Each mimics a different body fluid type
- Run in duplicate over one year
  - 2 samples per run
  - 6 runs per year
Body Fluids Program – Clinical

- Paired serum results and clinical scenarios
  - Allow participants to identify the fluid type/fluid state
  - Encourage use of Light’s Criteria, Serum Ascites Albumin Gradient
Assessment Criteria Updates

**Light’s Criteria (determines presence of exudate):**
- Lactate Dehydrogenase: tighten APS to equivalent in GC
- Total Protein: tighten APS to equivalent in GC

**Serum Albumin Ascites Gradient (indicates non peritoneal cause of ascites/portal hypertension):**
- Albumin: tighten APS to equivalent in GC

**Clinical Decision Limits:**
- Cholesterol: tighten APS to +/-0.3 to 2.5; then 12%
- Triglycerides: tighten APS to +/-0.3 to 1.2; then 25%

**Creatinine:**
- Target set using median of enzymatic methods

**Glucose and Osmolality:**
- Tighten APS to equivalent in General Chemistry
- No commutability issues expected
Changes Implemented

Tumour Markers:
• AFP, CA 125, CA 15-3 and hCG not included (cost prohibitive)
• CA 19-9 and CEA still available at higher levels than pilot

New analytes:
• Potassium
• Urate
• Urea

Improvements:
• Lipase
• pH
• Sodium
Results from Survey 1
**Malignant Pleural Effusion**

**Pleural Effusion**

### Albumin

- **Specimen 2-01**
  - Undesirably Low
  - Undesirably High

- **Specimen 2-02**
  - Undesirably Low
  - Undesirably High

### Amylase

- **Specimen 2-01**
  - Undesirably Low
  - Undesirably High

- **Specimen 2-02**
  - Undesirably Low
  - Undesirably High

Rizzi de Leon
Cholesterol

Glucose

Rizzi de Leon
Lactate Dehydrogenase

Pleural Effusion

Specimen 2-01

Specimen 2-02

Lipase

Undesirably Low

Undesirably High

No. of Laboratories

Undesirably Low

Undesirably High

No. of Laboratories

Undesirably Low

Undesirably High

No. of Laboratories

Rizzi de Leon
Osmolality

Protein
Sodium

Triglycerides
Malignant Pleural Effusion

Pleural Effusion

Urate

Specimen 2-01

Undesirably Low

Undesirably High

No. of Laboratories

0.005

0.065

>0.163

Median Value

Specimen 2-02

Undesirably Low

Undesirably High

No. of Laboratories

0.003

0.063

>0.161

Median Value

Urea

Specimen 2-01

Undesirably Low

Undesirably High

No. of Laboratories

0.2

1.2

>2.8

Median Value

Specimen 2-02

Undesirably Low

Undesirably High

No. of Laboratories

0.8

1.8

>3.4

Median Value

Rizzi de Leon
Creatinine

- Targets are set from the median of enzymatic creatinine results in Body Fluids program
- Correction factor used in rate blanked compensated Jaffe methods to allow for non-creatinine chromogens
- Jaffe may be prone to interferences in some fluids, e.g. peritoneal dialysis (high glucose)
- Not an issue clinically, but it shows the difference between serum method for fluid analysis

Enzymatic creatinine method measurements VS. Jaffe (alkaline picrate) methods in the Body Fluids & General Serum Chemistry programs
Lactate

- At equivocal concentrations, VITROS Lactate measures lower in the Body Fluids compared to VITROS Lactate in the General Chemistry
- Possibly combined with effect of using dry slide technology
pH

- pH of some body fluids can be used to guide clinical decisions
  - e.g. a pH of <7.20 is an indication of an infected pleural effusion
- Radiometer has an FDA approved application for pleural fluid pH analysis

Analytical Performance Specifications ±0.20
Potassium

- Listed as an “as found” analyte
  - High levels noted
- Continue to report potassium, as an EQA for potassium in body fluids
CA 19-9

Specimen 2.01

BF: Abbott ARCHITECT

TM: Abbott ARCHITECT

TM: OCD VITROS

TM: Roche cobas e

BF: OCD VITROS

BF: Roche cobas e
Roche instruments (cobas® E170/e601/e602) measuring lower in both Body Fluids and Tumour Markers programs

Further investigation still recommended
Clinical Evaluation

Question:
According to Light’s criteria, what is the most likely classification for this pleural fluid?

Options (please select one):
1. Transudative  2. Exudative

Commentary
According to Light’s criteria, a patient is considered to have an exudative effusion when any one of the following findings is present: a ratio of pleural fluid protein to serum protein higher than 0.5, a ratio of pleural fluid lactate dehydrogenase (LDH) level to serum LDH level higher than 0.6, or a pleural fluid LDH level higher than 200 U/L (or >67% of the upper limit of the normal range for serum LDH level).


Laboratories are not assessed on this feature.

Clinical Evaluation

This is a new feature intended for educational purposes. Clinical scenarios have been provided by the Working Party with an associated commentary and a suggested answer.
Summary

• Body Fluids Program: good after first survey results
• Not expecting large differences in performance in urine, serum and Body Fluids programs
• Some areas where further investigation may be warranted
• Work closely with the WP to review the program
Acknowledgements

AACB/RCPAQAP Body Fluids Working Party

• Dr Kay Weng Choy, Monash Pathology, Monash Medical Centre, Melbourne
• Dr Graham Jones, SydPath St Vincent’s Hospital, Sydney
• Mr John Calleja, Melbourne Pathology, Melbourne
• Dr Fernando San Gil, NSW Health Pathology South, The Wollongong Hospital, Wollongong
• Mr Peter Graham, RCPAQAP Blood Discipline Manager
• Ms Bernadette James, RCPAQAP Chemical Pathology Team

Royal North Shore Hospital (for sample pre-testing)
RCPAQAP Chemical Pathology Team