

## 22ABOT2

### Exercise instructions

### Distributed on 11/04/2022 – Closing on 25/04/2022

#### Material Provided

- Please refer to the COSHH sheet provided for all Health and Safety aspects of the samples. <https://www.ukneqash.org/download/177/UKNEQASBTCOSHHGeneralInstructionsPDF>
- 1 group O and 2 group B samples for anti-A titration, prepared from filtered fresh frozen plasma\*. These samples should be centrifuged prior to testing.
- 1 group A rr red cell sample (approximately 30% in modified Alsever's solution). These cells should be prepared for use in the appropriate diluent.
- Three group A 'patient' red cell samples suspended to approximately 10% in modified Alsever's solution, for A subtyping and labelled Patient W, Y, and Z. These cells should be prepared for use in the appropriate diluent.

\* Those who undertake renal (or other solid organ) transplant, please treat these as samples from patients awaiting an ABO incompatible transplant from a group A living donor.

#### Testing

- Samples should be treated as per clinical samples.
- Using the red cells provided, undertake titration of anti-A on the 3 plasma samples using your in-house techniques, and if possible the equivalent standard techniques using Bio-Rad technology as described below.
- If you perform group A subtyping in clinical practice, then undertake A<sub>1</sub> subtyping on the three 'patient' red cell samples.

#### Standard techniques (Bio-Rad)

- Prepare dilutions of plasma in saline (PBS or NaCl) using a doubling dilution method. Make the dilutions with a minimum volume of 200µl, using an automatic pipette. Use a new tip to dispense each dilution.
- Prepare a 0.8 - 1% red cell suspension in CellStab (use ID-diluent 2 if CellStab is not available).
- Read the endpoint of the titration as the last **weak** reaction.

#### LISS indirect antiglobulin test (IAT) using IgG or polyspecific cards

- a) Add 50ul of cells suspended in CellStab or ID-diluent 2 to each microtube
- b) Add 25ul of each plasma dilution to the corresponding microtube
- c) Incubate at 37°C for 15'
- d) Centrifuge 10' in DiaMed centrifuge

#### Direct agglutination at room temperature (DRT) using NaCl cards

- a) Add 50ul of cells suspended in CellStab or ID-diluent 2 to each microtube
- b) Add 50ul of each plasma dilution to the corresponding microtube
- c) Incubate at RT for 15'
- d) Centrifuge 10' in DiaMed centrifuge

#### Return of results

Record results of the titrations with your in-house and/or standard techniques, via the UKNEQAS website ([www.ukneqasbtlp.org/participants/login](http://www.ukneqasbtlp.org/participants/login)). Instructions for data entry can be found at <https://www.ukneqash.org/downloads/ABOT%20data%20entry%20instructions.pdf>

If you have any problems with the material or any queries regarding the questionnaire or results please contact the BTLT team.

**With thanks for your participation**