

## 25ABOT3

### Exercise instructions

### Distributed on 14/07/2025 – Closing on 28/07/2025

#### Material Provided

Please refer to the COSHH sheet provided for all Health and Safety aspects of the samples.

[https://haembtbp.ipassportqms.com/document\\_download/M2ZIYWMzMTetNTYwNi00YjEzLTg3YTItYTNmMzAzN2FiMGY5](https://haembtbp.ipassportqms.com/document_download/M2ZIYWMzMTetNTYwNi00YjEzLTg3YTItYTNmMzAzN2FiMGY5)

- 3 vials of plasma, to be used for anti-A titration, prepared from filtered fresh frozen plasma.
  - Patient 1 – containing group B plasma - to be used vs. the A<sub>1</sub> cells provided
  - Patient 2 – containing group B plasma - to be used vs. the A<sub>1</sub> cells provided
  - Patient 3 – containing group O plasma - to be used vs. the A<sub>1</sub> cells provided
- 1 vial of group A<sub>1</sub> rr red cells suspended in 20-30% modified Alsever's solution
  - A<sub>1</sub> rr red cells - to be used as a reagent red cell for titration vs Patients 1, 2, and 3
- 3 vials of group A 'patient' red cells suspended in 20-30% modified Alsever's solution
  - Patient W – for A subtyping
  - Patient Y – for A subtyping
  - Patient Z – for A subtyping

#### Testing

- Samples should be treated as per clinical samples.
- Plasma samples should be centrifuged prior to testing.
- Red cell samples should be prepared for use in the appropriate diluent.
- Treat the plasma samples for Patients 1, 2 and 3 as being from patients awaiting an ABO incompatible transplant from a group A living donor.
- Use the 30% red cells (group A<sub>1</sub>) provided to undertake titration of the plasma samples against the cells indicated above using your in-house techniques, and if possible, the equivalent standard techniques using Bio-Rad technology as described below.
- Do not use local cells for titration, use the cells provided.
- If A subtyping is performed in clinical practice, undertake A 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 50µl of cells suspended in CellStab or ID-diluent 2 to each microtube
- b) Add 25µl 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 50µl of cells suspended in CellStab or ID-diluent 2 to each microtube
- b) Add 50µl 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://haembtbp.ipassportqms.com/document\\_download/ZmNINTI3OTEtY2RhZC00NDA4LTlhZjktMDA0YzlyMG15Zm15](https://haembtbp.ipassportqms.com/document_download/ZmNINTI3OTEtY2RhZC00NDA4LTlhZjktMDA0YzlyMG15Zm15)

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

**With thanks for your participation**

**Additional information**

**Paper instructions will no longer be sent with samples, the COSHH document will still always be sent, and a link for the instruction will be included on the COSHH document. You can find exercise instructions online in the data entry page, or by going to or by going to the link below and replacing XXXX with the exercise code.**  
**<https://www.ukneqash.org/downloads/XXXXInstructions.pdf>**