

FETAL RED CELL DETECTION KIT

Product Code 05-050

TEST PRINCIPLE Haemoglobin A is readily eluted from adult red cells at a low pH while cord red cells or red cells from the newborn which contain haemoglobin F are more resistant. A suitable reagent will fully elute the adult haemoglobin A from a thin blood film without eluting the red cells containing haemoglobin F. The resistant haemoglobin can then be stained red with an appropriate counterstain.

SAMPLES Test samples for the investigation of the presence of fetal red cells in the maternal circulation should be collected into EDTA, citrated or heparinised tubes. Clotted samples are **not** suitable.

REAGENTS PROVIDED

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| 1. | Eluting reagent | 3 x 100mls |
| 2. | Counterstain | 3 x 100mls |

Reagents are ready for use.

REAGENTS NOT PROVIDED

1. Fixative solution

Use absolute ethyl alcohol or methanol and dilute with tap water (80mls alcohol + 20mls tap water). This fixing reagent should be stored at room temperature and changed every 7 days or before. See 'Notable Points' below.

Inverclyde manufacture 'FIXOL' a stable fixative solution made specifically for this purpose. It is recommended that FIXOL be used as the fixing reagent (order code 05-051).

STORAGE

Store the reagents at room temperature.

RECOMMENDED METHOD

Allow samples to come to room temperature.

Fixing- Mix the sample to be investigated thoroughly. Prepare a **thin** blood film and allow to air dry for 20 minutes.

Fix in 80% ethyl alcohol for 5 minutes.

Wash thoroughly in tap water and allow to air dry.

NB. If Inverclyde 'Fixol' is used as the fixative reagent the films can be air dried for two minutes and fixed over 3 minutes.

Fixed films can be stored and used for up to 72 hours.

NB Over fixation can partly inhibit elution.

Elution Flood the fixed dried film with the eluting fluid and allow 30 seconds for elution to occur.

Wash thoroughly in running tap water. Do not allow to dry.

Counterstaining Flood the film with the counterstain and allow 2 minutes for adequate staining.

Wash thoroughly in running tap water and allow to dry.

MICROSCOPIC EXAMINATION

Examine the eluted/counterstained film under the x 40 dry objective. It is helpful to rack the condenser down and to increase the light intensity. Fetal cells or cells containing HbF stain bright red while normal adult cells (HbA) appear as ghosts or stain very pale pink.

Leucocytes stain grey/blue.

CONTROLS

Positive and negative controls must be included in every batch of tests.

Positive control- Add an appropriate aliquot of fresh ABO group O cord red cells to a fresh ABO group O adult sample. (5 - 10 μ to 5mls respectively).

Ensure that the adult sample does not contain any other irregular blood group antibodies.

Negative control- Any fresh adult sample which does not contain HbF.

NOTABLE POINTS

1. The control samples should be prepared regularly.

2. Keep the eluting reagent away from heat at all times. Do not

smoke when using this reagent.

3. A precipitate may develop in the eluting reagent after some months. This will not interfere with its eluting properties.

4. Poor quality staining (adult cells staining pink or red due to improper elution) is likely to be a result of improper fixing. Care must be taken to ensure that the fixing agent is accurately diluted with water and changed every week or before.

5. Fixol is a stable fixative solution with a 2 year expiry from the date of manufacture. It is strongly recommended that this reagent be used with the FCD kit.

