

ab204911 Antibody Concentration Kit (Gold/Magnetic labelling)

**A product of Expedeon, an
Abcam company**

Applicable to Expedeon product codes: 262-0010

For concentrating and cleaning up antibodies.

This product is for research use only and is not intended for diagnostic use.

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1. Overview

Antibodies are sometimes only available at concentrations that are too low for antibody conjugation or other antibody applications. The Antibody Concentration Kit (Gold/Magnetic labelling) uses a simple spin column to easily and quickly remove excess buffer from the antibody thereby providing a more concentrated antibody solution. The kit can also be used to reduce the concentration of additives often found in antibody formulations, such as salt, citrate and azide, that may interfere in subsequent conjugation reactions.

The Antibody Concentration Kit (Gold/Magnetic labelling) also allows the researcher to perform a simple buffer exchange to transfer the antibody into a more favourable buffer for conjugation.

Antibodies purified using the Antibody Concentration Kit (Gold/Magnetic labelling) are fully compatible with our Gold and Magnetic conjugation kits. Please note that the Antibody Concentration and Clean Up Kit for Latex & Europium ([ab269965](#)) should be used if the antibody is to be used with the Latex and Europium conjugation kits. Where both a buffer exchange and a concentration step are required, these can be performed together by adjusting the volume used to recover the antibody accordingly.

2. Materials Supplied and Storage

Store kit at +4°C immediately on receipt.

Item	Quantity	Storage temperature
Spin cartridge	3X Spin cartridges (up to 0.5 mL)	4°C
Conjugation Buffer	1X Vial	4°C

Reagents are ready to use as supplied.

3. Assay Procedure

For buffer exchange, proceed directly to section 3.2.

3.1 Concentration of antibody solution:

1. Add up to 0.5 mL antibody to spin cartridge
2. Spin for 1 to 3 minutes in a microfuge at a recommended maximum speed of 15,000 x g to reduce the buffer volume in the spin cartridge to between 50 and 100 µL.
3. Repeat steps 1 and 2 as many times as is necessary to process the entire antibody to the desired concentration. It may be necessary to discard the excess buffer collected in the collection tube between spins. Avoid spinning the antibody dry as reconstitution of the antibody will be difficult and significant antibody loss may occur.
4. Recover the concentrated antibody from the spin cartridge.

Δ Note: It is advisable not to spin the antibody dry as reconstitution of the antibody will be difficult and significant antibody loss and degradation may occur. Minimum volume advised for recovery is 50µl.

Δ Note: Please note other proteins present in the buffer such as BSA will also be concentrated using this method.

*Δ **Note:** Spin times will vary depending on buffer composition and volume as well as centrifuge speed.*

3.2 Buffer exchange using spin column assembly:

1. Add up to 0.5 mL antibody to spin cartridge.
2. Spin for 1 to 3 minutes in a micro centrifuge at a recommended speed of 15,000 x g to reduce buffer volume to 100 µL.
3. Discard the excess liquid in a collection tube.
4. Add 400 µL conjugation buffer to the antibody in the spin cartridge
5. Spin for 1 to 3 minutes in a microfuge at a recommended maximum speed of 15,000 x g to reduce buffer volume to 100 µL.
6. Discard the excess liquid in collection tube.
7. Repeat steps 4 to 6 at least 5 times to exchange antibody buffer.
8. Recover antibody from the spin cartridge.

*Δ **Note:** Each cycle leads to a reduction in the concentration of low molecular weight substances (smaller than 10 kDa). However, the concentration of proteins such as BSA will be unchanged.*

*Δ **Note:** If the antibody requires a buffer exchange as well as being concentrated, it can be recovered in less volume at the end of the buffer exchange step. Minimum volume advised for recovery is 50 µL.*

*Δ **Note:** The exchange process is more efficient if the volume is reduced to 50 µL instead of 100 µL at each cycle.*

*Δ **Note:** Spin times will vary depending on buffer composition and volume as well as centrifuge speed.*

4. Test for Protein

Wherever possible protein values should be determined using an absorbance at 280 nm. For an IgG using a 1 cm light path an OD₂₈₀ of 1.0 is equivalent to an antibody concentration of 0.714 mg/mL.

When using Bradford-type reagents it is important to use an IgG standard curve. The absorbance generated by this type of reagent is dependent on the protein used. For example, using a BSA standard curve to determine the protein concentration of an IgG solution will result in a 2.3-fold under-estimate of the IgG concentration.

5. Antibody storage:

Store at 4°C. Other storage conditions (e.g. frozen at -70°C) may also be satisfactory. The sensitivity of any antibody to freeze thaw should be determined by experimentation on small aliquots.

Technical Support

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