Amersham AMDEX sheep anti-mouse IgG-HRP

Product Specification Sheet

Introduction

Product code

RPN4201

About

Sheep anti-mouse IgG conjugated to multiple HRP molecules on a dextran backbone. This conjugate is specific for rat and mouse IgG.

Important

Read these instructions carefully before using the products.

Intended use

The products are intended for research use only, and shall not be used in any clinical or *in vitro* procedures for diagnostic purposes.

The product may not be used to perform or offer commercial services of any kind and may not be incorporated into a diagnostic kit of any type intended for resale.

Safety

For use and handling of the products in a safe way, refer to the Safety Data Sheets.

Storage

Store, in the form supplied, at 2–8°C.

Expiry

See outer packaging.

Components

Buffer 500 µL	Stabilized in 50% TRIS Buffer and 50% HRP Conjugate Stabilizer
Conjugate concentration	Approximately 1 mg HRP/mL
Dilution	For ELISA, it is recommended that the conjugate should be diluted in the range 1:1 000 to 1:4 000.

Use

The conjugate is supplied for use in ELISA systems.

It can be used in direct substitution for a standard anti-mouse HRP conjugate, without the need to alter existing protocols.

For optimum results it is necessary to:

- Dilute the conjugate to a suitable titre.

  It is important to optimise the dilution by setting up a range test. In our own experiments we have found a dilution range of 1:1000 to 1:4000 to be suitable for ELISA applications. This range is given for guidance only and should be determined by the user.

- Dilute the conjugate in a suitable buffer.

  It is essential that the conjugate is diluted in a suitable buffer.

  For optimum results the following buffer is suggested:

  Starting buffer: PBS pH 7.2, 0.1% BSA, 0.1% Tween™.

  Optimise depending on application and signal: noise ratio to PBS pH 7.2, up to 3% BSA, up to 1% Tween.

For optimum results the following buffer is suggested:

Starting buffer: PBS pH 7.2, 0.1% BSA, 0.1% Tween.

Optimise depending on application and signal: noise ratio to PBS pH 7.2, up to 3% BSA, up to 1% Tween.

Troubleshooting

Table 1. Symptom: No signal.

Diagnosis	Remedy
One or more reagents omitted.	Re-run the assay, taking care over additions.
One or more reagents used too dilute.	Re-run the assay using higher concentrations.

Table 2. Symptom: High non-specific binding (NSB).

Diagnosis	Remedy
Improperly washed plate.	Re-run the assay, taking special care over the wash steps (Thorough rinsing with a washbottle is preferable to using an automatic plate-washer. It is important that plates are banged firmly to remove all liquid from wells after each step).
One or more reagents used too concentrated.	Re-run the assay using lower concentrations.
Insufficient blocking agents present.	Re-run the assay in buffers with higher concentrations of Tween and BSA.

Table 3. Symptom: Optical density (OD) readings too high.

Diagnosis	Remedy
If standard readings within expected range, samples are too concentrated.	Re-run the assay after diluting the samples in a suitable buffer.
If all readings too high, substrate incubation too long.	Re-run the assay, allowing less time between additions of substrate and stop solutions.
Improperly washed plate.	Re-run the assay (see *Table 2, on page 1*).
Amdex conjugate too strong.	Dilute the conjugate further.

Table 4. Symptom: OD readings too low.

Diagnosis	Remedy
Samples too dilute to read on standard curve.	Samples outside assay range. Re-assay after concentrating down.
All readings (including standards) too low; substrate incubation too short.	Re-run the assay, allowing more time between additions of substrate and stop solutions.
Amdex conjugate too dilute.	Re-run the assay with the conjugate at a high concentration.

Table 5. Symptom: Poor standard curve shape.

Diagnosis	Remedy
Errors in diluting standards.	If the problem hampers reading of the assay, re-run with fresh standards.
Improperly washed plate.	Re-run the assay (see *Table 2, on page 1*).

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