

AM05879PU-N**Monoclonal Antibody to Rituximab - Purified**

Alternate names:	MabThera, Rituxan
Quantity:	0.2 mg
Concentration:	1.0 mg/ml
Background:	Rituximab is a therapeutic reagent directed against the human CD20 cell surface antigen.
Host / Isotype:	Rat / IgG2a
Recommended Isotype Controls:	SM15P, SM15PX
Clone:	MB2 A4
Immunogen:	F(ab)2 fragment of Rituximab. Spleen cells from immunised rats were fused with cells of the NS-1 mouse myeloma cell line
Format:	State: Liquid purified IgG fraction from Tissue Culture Supernatant Purification: Affinity Chromatography on Protein G Buffer System: TRIS buffered saline pH 8.0 Preservatives: 0.09% Sodium Azide
Applications:	ELISA: 5 µg/ml as coating antibody. Flow Cytometry: Use 10 µl of 50 µg/ml diluted antibody to label 10 ⁶ cells in 100 µl. Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Specificity:	This antibody specifically recognises the idiotypic determinants expressed by the Rituximab humanised monoclonal antibody. It does <u>not</u> recognise other CD20 antibodies. This antibody has been used in ELISA assays to monitor the levels of Rituximab in patient serum following therapy. It has been used to detect Rituximab bound to the surface of the Raji B cell line, however detection of Rituximab bound ' <i>in vivo</i> ' to B-CLL cells has not been demonstrated. It is possible that complement deposition on Rituximab opsonised cells inhibits binding of the anti-Rituximab antibody to cell bound Rituximab (3).
Storage:	Store undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer. Avoid repeated freezing and thawing. Shelf life: one year from despatch.
General Readings:	1. Bayne, M. et al. (2003) Phase I/II study of fractionated radioimmunotherapy in relapsed low grade non Hodgkin lymphoma. Br. J. Cancer. 88 : S38. 2. Cragg, M. S. et al. (2004) Apparent modulation of CD20 by rituximab: an alternative explanation. Blood. 103: 3989-3990. 3. Beum, P. V. et al. (2004) Complement activation and C3b deposition on rituximab-opsonized cells substantially blocks binding of phycoerythrin-labeled anti-mouse IgG probes to rituximab. J. Immunol. Methods. 294:

37-42.

4. Hampson, G. et al. (2010) Validation of an ELISA for the determination of rituximab pharmacokinetics in clinical trials subjects. *J. Immunol. Methods* 360: 30-38.

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6. Daydé, D. et al. (2009) Tumor burden influences exposure and response to rituximab: pharmacokinetic-pharmacodynamic modeling using a syngeneic bioluminescent murine model expressing human CD20. *Blood*. 113: 3765-72.

7. Aung, T. et al. (2011) Exosomal evasion of humoral immunotherapy in aggressive B-cell lymphoma modulated by ATP-binding cassette transporter A3. *Proc Natl Acad Sci U S A*. 108: 15336-41.

8. Schmidt, E. et al. (2009) Immunogenicity of rituximab in patients with severe pemphigus. *Clin Immunol*. 132: 334-41.

9. McDonald, V. et al. (2010) Rituximab pharmacokinetics during the management of acute idiopathic thrombotic thrombocytopenic purpura. *J Thromb Haemost*. 8: 1201-8.

10. Kagan, L. et al. (2011) Subcutaneous Absorption of Monoclonal Antibodies: Role of Dose, Site of Injection, and Injection Volume on Rituximab Pharmacokinetics in Rats. *Pharm Res*. Sep 2. [Epub ahead of print].

11. Kagan, L. and Mager, D.E. (2013) Mechanisms of subcutaneous absorption of rituximab in rats. *Drug Metab Dispos*. 41: 248-55.

12. Cragg, M. S. et al. (2004) A new anti-idiotypic antibody capable of binding rituximab on the surface of lymphoma cells. *Blood*. 104:2540-2.

13. Kagan, L. et al. (2014) Interspecies pharmacokinetic modeling of subcutaneous absorption of rituximab in mice and rats. *Pharm Res*. 31: 3265-73.

14. Blasco, H. et al. (2009) Pharmacokinetics of rituximab associated with CHOP chemotherapy in B-cell non-Hodgkin lymphoma. *Fundam Clin Pharmacol*. 23: 601-8.

15. Pers, J.O. et al. (2007) BAFF-modulated repopulation of B lymphocytes in the blood and salivary glands of rituximab-treated patients with Sjögren's syndrome. *Arthritis Rheum*. 56: 1464-77.

Pictures:

Detection of Rituximab spiked into 10% human serum by sandwich ELISA. Human anti Rituximab was used as the coating antibody and detection was performed by adding HRP conjugated anti-Rituximab antibody AM05879PU-N. *Data are shown as the mean of three independent measurements.*

