

- 6 Islam A. A new bone marrow biopsy needle with core securing device. *J Clin Pathol* 1982;35:359-64.
- 7 Ellman L. Bone marrow biopsy in the evaluation of lymphoma, carcinoma and granulomatous disorders. *Am J Med* 1976;60:1-6.
- 8 Burke JS. The value of the bone marrow biopsy in the diagnosis of hairy cell leukaemia. *Am J Clin Pathol* 1978;70:876-84.
- 9 Islam A, Catovsky D, Goldman JM, Galton DAG. Value of long core biopsy in detection of discrete bone marrow lesions. *Lancet* 1979;i:878.
- 10 Glauert AM, Glauert RH. Araldite as an embedding medium for electron microscopy. *J Biophys Biochem Cytol* 1958;4:191-4.
- 11 Luft JH. Improvements in epoxy resin embedding methods. *J Biophys Biochem Cytol* 1961;9:409-14.
- 12 Anderson WA, Andre J. The extraction of some cell components with pronase and pepsin from thin sections of tissue embedded in Epon-Araldite mixture. *J Microsc* 1968;7:343-9.
- 13 Spurr AR. A low viscosity epoxy resin embedding medium for electron microscopy. *J Ultrastruct Res* 1969;26:31-43.
- 14 Heyderman E, Monaghan P. Immunoperoxidase reactions in resin embedded sections. *Invest Cell Pathol* 1979;2:119-22.
- 15 Giddings J, Griffin RL, Maciver AG. Demonstration of immunoproteins in Araldite embedded tissues. *J Clin Pathol* 1982;35:111-14.
- 16 Islam A. Bone marrow structure in human leukaemias: A histological study by plastic embedding techniques. London: University of London (Thesis). 1982.
- 17 Stein H, Gatter K, Asbahr H, Mason DY. Methods in laboratory investigation: use of freeze-dried paraffin-embedded sections for immunohistologic staining with monoclonal antibodies. *Lab Invest* 1985;52:676-83.

Requests for reprints to: Dr A Islam, Department of Medical Oncology, Roswell Park Memorial Institute, 666 Elm Street, Buffalo, New York, 14263, USA.

Modified latex agglutination test for anticytomegalovirus, suitable for pretransfusion screening

J A J BARBARA, H MOULSDALE, S BROWN, P D GRIFFITHS,* M J BERRY,* M CONTRERAS *From the North London Blood Transfusion Centre, Deansbrook Road, Edgware, Middlesex, and the* Royal Free Hospital, London*

The increasing requirements of immunosuppressed patients for blood and blood components from donors uninfected with cytomegalovirus (CMV) led us to search for a sensitive, rapid, convenient and economical test for screening blood donations for antibodies against this virus.

Initially, we used a modification of a commercial haemagglutination (HA) assay (Cetus Corporation, Berkeley, California); this entailed 10-fold dilution of the red cells provided in the kit and assessment of agglutination in a microplate inclined at 70° after centrifugation at 387 g for one minute.¹ Although this test proved suitable for the selection of plasma with high titre anti-CMV for the manufacture of specific immunoglobulin, its sensitivity and specificity was unacceptably variable for selecting CMV free donations. This became apparent when 23% (28 of 124) donations found negative for anti-CMV by modified HA were found to be positive for CMV antibodies by radioimmunoassay. We therefore decided to use a modified latex agglutination as our routine screening test (CMV Scan, Becton and Dickinson, Baltimore, Maryland). We tested this assay in parallel with the modified HA test (MHA) using a sensitive radioimmunoassay described by Berry *et al*² as a standard for comparison.

Accepted for publication 15 July 1986

Methods

For the latex agglutination test 25 µl of test serum was pipetted on to black reaction cards and 15 µl of latex coated with CMV antigen was added and mixed, according to the manufacturer's recommendations. Positive and negative control sera were also tested. The card was placed on a humidified rotator for eight minutes and then read. Samples positive for anti-CMV showed clear agglutination, whereas negative ones remained cloudy. The test was made more economical by diluting two volumes of the antigen coated latex with one volume of the titration buffer provided with the kit. Furthermore, the volumes of reagents were reduced to 15 µl of test serum and 5 µl of diluted latex. Even with these reduced volumes, the reactions could be read easily.

Results

Before using the modified latex test for screening donor sera, we confirmed that the reduced volume modifications, with or without dilution of the latex, provided the same end point titres as the standard method when either sera or plasma with high titre or low titre anti-CMV were assessed. When 252 donor serum samples found to give negative CMV antibody results by modified haemagglutination screen tests were rescreened under routine testing conditions by the reduced volume latex agglutination test (method A), 58 (23%) were found to be positive by the second test. These 58 samples were then retested by various methods (table 1). A small number scored differently on this repeat testing.

Table 2 gives detailed results for nine negative or "discrepant" sera at repeat testing. Furthermore, when 50 serum samples from donors found to be negative for anti-CMV by modified haemagglutination and by the two latex test modifications, were tested by monoclonal radioimmunoassay, 48 were clearly negative, and two samples showed only traces of antibody to CMV. The figure shows the

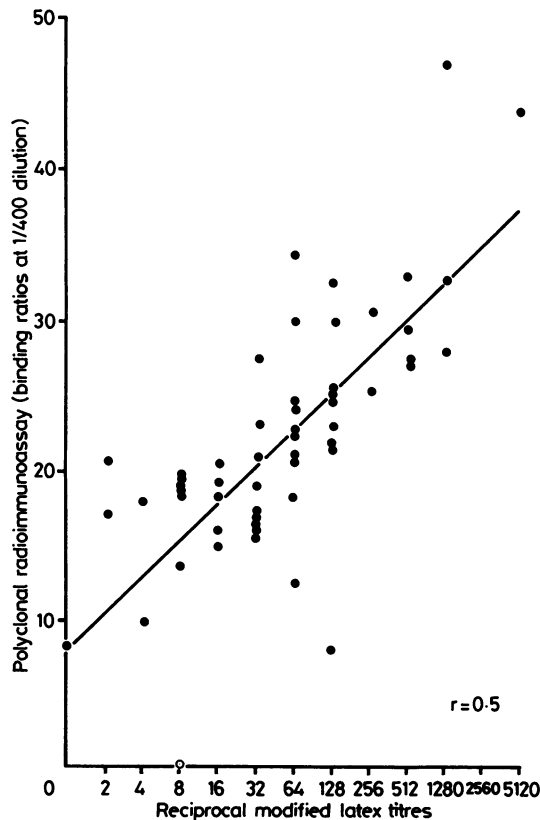
Table 1 Details of 58 sera, negative for anti-CMV by modified HA (but positive by latex screen) and retested by four methods

	Radioimmunoassay		Modified latex agglutination	
	Polyclonal antibody	Monoclonal antibody	Method A	Method B
No of anti-CMV positive (%)	54 (93)	53 (91)	55 (95)	53 (91)
No of anti-CMV negative (%)	4 (7)	5 (9)	3 (5)	5 (9)

A = volumes reduced by two thirds; B = volumes reduced by two thirds and latex diluted by one third.

Table 2 Detailed results of nine serum samples giving discordant results by different techniques

Sample identification No	Method used for retesting after original latex screen			
	Radioimmunoassay		Modified latex agglutination	
	Polyclonal	Monoclonal (anti-Fc IgG)	Titration (method A)	Screen (method B)
3	+	+	+ (1/32)	±
23	+	+	+ (1/32)	+
28	Negative	Negative	Negative	Negative
29	+	+	+ (1/128)	+
36	Negative	Negative	Negative	Negative
44	Negative	Negative	+ (1/8)	Negative
46	Negative	Negative	Negative	Negative
50	+	Negative	+ (1/4)	Negative
54	+	+	+ (1/4)	+



Correlation between modified latex (method A) and radioimmunoassay for anti-CMV assay.

correlation between modified latex and radioimmunoassay for anti-CMV assay.

Discussion

The results indicate that the modified latex tests, as well as being rapid and relatively economical (15 to 20p per test), are both reasonably sensitive and specific when compared with radioimmunoassay. At the North London Blood Transfusion Centre we find it a very practical and convenient test for fulfilling nearly 90% of the 3500 annual requests that we receive for platelet concentrates negative for CMV antibodies (de Silva *et al*, Proceedings of British Blood Transfusion Service third annual meeting, Oxford, September 4-7 1985). In agreement with Taswell *et al*,³ the latex test has become the method of choice at the Centre.

We acknowledge the generous gifts of reagents provided by Diamed Diagnostics and Becton and Dickinson.

References

- 1 Barbara JAJ. *Microbiology in blood transfusion*. Bristol: John Wright, 1983.
- 2 Berry NJ, Grundy JE, Griffiths PD. An improved radioimmunoassay method for the detection of IgG antibodies against cytomegalovirus. *J Virol Methods* 1986;13:343-500.
- 3 Taswell HF, Reisner RK, Rabe DE, Shelley CD, Smith TF. Comparison of three methods for detecting antibody to cytomegalovirus. *Transfusion* 1986;26:285-9.

Requests for reprints to: Dr J A J Barbara, North London Blood Transfusion Centre, Deansbrook Road, Edgware, Middlesex, England.