

Note added in proof. Bauer *et al.*¹⁵ have independently applied the lactoperoxidase technique to isolate immunoglobulin from murine splenic lymphocytes.

JOHN J. MARCHALONIS
JOHN L. ATWELL*
ROBERT E. CONE†

Walter and Eliza Hall Institute of
Medical Research, Parkville, Victoria 3052

Received July 13; revised September 20, 1971.

* Australian Commonwealth research scholar.
† Postdoctoral fellow of the Damon Runyon Memorial Fund.

- ¹ Dwyer, J. M., and Mackay, I. R., *Lancet*, i, 1189 (1970).
- ² Ehrlich, P., *Pro. Roy. Soc.*, B, 66, 424 (1900).
- ³ Marchalonis, J. J., Cone, R. E., and Samter, V., *Biochem. J.*, 124, 921 (1971).
- ⁴ Marchalonis, J. J., *Biochem. J.*, 113, 299 (1969).
- ⁵ Eisen, H. N., Kern, M., Newton, W. T., and Helmreich, E., *J. Exp. Med.*, 110, 187 (1959).
- ⁶ Perper, R. J., Zee, T. W., and Mickelson, M. M., *J. Lab. Clin. Med.*, 72, 842 (1968).
- ⁷ Marchalonis, J. J., and Schonfeld, S. A., *Biochim. Biophys. Acta*, 221, 604 (1970).
- ⁸ Marchalonis, J. J., *Devel. Biol.*, 25, 479 (1971).
- ⁹ Edelman, G. M., and Marchalonis, J. J., in *Methods in Immunology and Immunochemistry* (edit. by Williams, C. A., and Chase, M. W.), 1, 405 (Academic Press, New York, 1967).
- ¹⁰ Parish, C., and Marchalonis, J., *Anal. Biochem.*, 34, 436 (1970).
- ¹¹ Bankhurst, A. D., Warner, N. L., and Sprent, J., *J. Exp. Med.*, 134, 1005 (1971).
- ¹² Greaves, M. F., and Hogg, N. M., *Proc. Third Sigrid Julius Symp. Cell Co-operation in the Immune Response* (edit. by Cross, A., Kosunen, T., and Makela, O.), 145 (Academic Press, New York, 1971).
- ¹³ Cone, R. E., Marchalonis, J. J., and Rolley, R. T., *J. Exp. Med.*, 134, 1373 (1971).
- ¹⁴ Marchalonis, J. J., Cone, R. E., and Atwell, J. L., *J. Exp. Med.* (in the press).
- ¹⁵ Bauer, S., Vitetta, E. S., Sherr, C. J., Schenkein, T., and Uhr, J. W., *J. Immunol.*, 166, 1133 (1971).

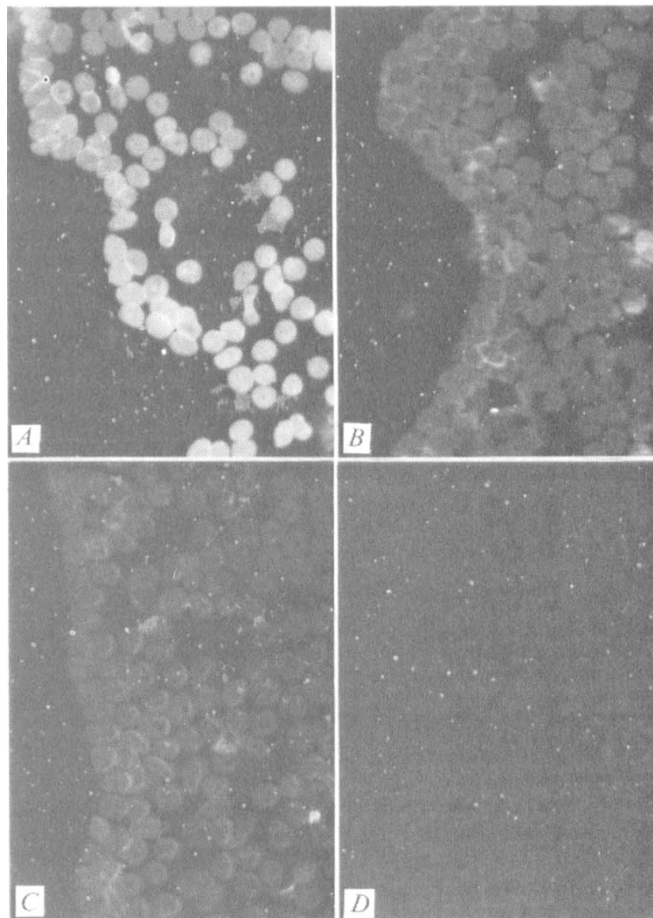


Fig. 1 Fluorescence photomicrograph of the smear of erythrocytes. A, Mouse; B, rat; C, golden hamster; D, sheep (× 560).

Erythrocyte Membrane-specific Antigen common to Several Species of Rodentia

THE rare existence of organ-specific antisera transcending species differences was pointed out by Coombs¹. Similar new organ, or "erythrocyte-specific", antisera reacting selectively to the erythrocytes of several species of Myomorpha are described here. The antisera, "anti-A", were prepared by the method of Adachi and Furusawa²: adult guinea-pigs were sensitized with the "antigen-A" (the protein portion of dd mouse erythrocyte membrane). The detailed procedure for preparing labelled antibody, the staining method and its specific reactivity to mouse erythrocytic cells have been reported before³. Circulating erythrocytes from various animals were stained with the fluorescein isothiocyanate-labelled "anti-A". Smear cell preparations were fixed with acetone at -20° C for 20 min and incubated with the labelled

antisera, which had been thoroughly absorbed with the emulsion of mouse liver and kidney to remove the cross-reacting antibodies, at 37° C for 90 min. A light microscope with a darkfield condenser was used for the fluorescent observations using Orsen's interference filter system. Clear fluorescent reactions, though of varied intensities, were observed with those animals classified as Myomorpha, that is, mouse, rat, golden hamster and Chinese hamster (Table 1, Fig. 1). Mammalian erythrocytes other than those of the guinea-pig used as the sensitized animal showed faint fluorescences. Nucleated erythrocytes of the chick, quail, turtle, toad and goldfish were negative to the reaction. The erythrocyte-specific reactivity of the labelled antisera was examined with the liver, kidney and thymus cells of the animals whose erythrocytes displayed intense fluorescences; they showed, however, no fluorescence.

It has been shown² that the Ouchterlony's immunodiffusion test revealed three specific precipitation lines in the "anti-A" and "antigen-A" reaction. In the experiment described here, the Ouchterlony test showed that both the antigenic fractions prepared from the erythrocyte membranes of mice ("antigen-A") and rat produced two common specific lines against the "anti-A"—surely, supporting the theory of transcendence of species differences of the "erythrocyte-specific" antisera.

MITSURU FURUSAWA
KEN-ICHI P. TAKAHASHI

Laboratory of Embryology,
Faculty of Science, Osaka City University,
Sugimoto-cho, Sumiyoshi-ku, Osaka

Received August 19, 1971.

¹ Coombs, R. R. A., *General Physiology of Cell Specialization* (edit. by Mazia, D., and Tyler, A.), 181 (McGraw-Hill, New York, 1963).

² Adachi, H., and Furusawa, M., *Exp. Cell Res.*, 50, 490 (1968).

³ Furusawa, M., and Adachi, H., *Exp. Cell Res.*, 50, 497 (1968).

Table 1 Distribution of the Erythrocyte Membrane-Specific Antigens in Various Animals

Animal tested	Fluorescent reaction in erythrocyte	Animal tested	Fluorescent reaction in erythrocyte
Goldfish	- *	Dog	+
African clawed toad	-	Rabbit	+
Turtle (<i>Clemmys</i>)	-	Squirrel	±
Chick	-	Guinea-pig	-
Quail	-	Chinese hamster	++
Sheep	±	Golden hamster	+++
Goat	±	Rat	+++
Cow	±	Mouse	++++
		Human (A, B, AB, O)	+

* + + + +, Intensive; + + + and + +, intermediate; +, very weak; ±, trace; -, negative fluorescence.