



Title	Antibody-Induced Redistribution of Marek's Disease Tumor-Associated Surface Antigen (MATSA) on Lymphoblastoid Line (MSB-1) Cells Derived from Marek's Disease Lymphoma
Author(s)	Matsuda, Haruo; Ikuta, Kazuyoshi; Miyamoto, Hiroyuki et al.
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PRELIMINARY REPORT

ANTIBODY-INDUCED REDISTRIBUTION OF MAREK'S DISEASE TUMOR-ASSOCIATED SURFACE ANTIGEN (MATSA) ON LYMPHOBLASTOID LINE (MSB-1) CELLS DERIVED FROM MAREK'S DISEASE LYMPHOMA

HARUO MATSUDA, KAZUYOSHI IKUTA, HIROYUKI MIYAMOTO
and SHIRO KATO

Department of Pathology, Research Institute for Microbial Diseases, Osaka University,
Yamada-kami, Suita, Osaka

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A "cap" formation due to temperature dependent and ligand-induced redistribution of various receptors on lymphocyte membrane was first described by Smith and Hollers (1970) using lymphocytes treated with concanavalin A labeled with fluorescein isothiocyanate (FITC). Taylor et al. (1971) also demonstrated a "cap" formation due to the redistribution of immunoglobulin (Ig) molecules on B lymphocytes treated with anti-Ig antibody labeled with FITC. The phenomenon has been extensively studied by de Petris and Raff (1972) and Unanue et al (1972). Receptors forming a "cap" are generally destined to be interiorized due to pinocytosis of the cell (Taylor et al., 1971).

Using sera of rabbits and chickens immunized with Marek's disease (MD) lymphoblastoid MSB-1 line cells (Akiyama and Kato, 1974), we have demonstrated MD tumor-associated surface antigen (MATSA) reported by Witter et al. (1975) on most cells of all six MD lymphoblastoid cell lines by indirect fluorescent antibody (FA) technique (Matsuda et al., 1976). All the procedures for FA technique was carried out at 4 C as described by Witter et al. (1975). Although the appearance of MATSA demon-

strated by FA technique is annular along the surface of the cells, the stain was not even, but patchy.

In this preliminary paper, we reported the temperature-dependent polarization, cap-formation and cap-exclusion phenomenon of MATSA determinants on the surface of MSB-1 cells as detected by reaction with rabbit anti-MSB-1 serum.

Unfixed living MSB-1 cells were washed three times with cold phosphate buffered saline (PBS), resuspended into one drop (about 0.05 ml) of unconjugated rabbit anti-MSB-1 serum, as previously described (Matsuda et al., 1976), and reacted for 30 min at 4 C. After washing with three changes of a large volume of cold PBS, the cells were resuspended in one drop of fluorescein-conjugated goat anti-rabbit Ig and reacted for 20-30 min at 4 C. After a final wash in cold PBS or TC medium 199, the cells were further incubated for 5-120 min at 37 C. Samples were taken at various times and immediately examined with an Olympus fluorescence microscope.

It was observed that MATSA were distributed in patches on the surface of almost all MSB-1 cells stained in the cold (Fig. 1).

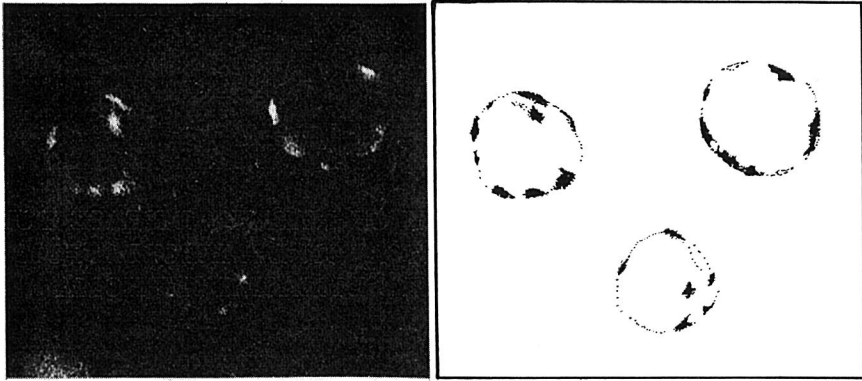


FIGURE 1. Fluorescent spots in addition to linear surface staining.

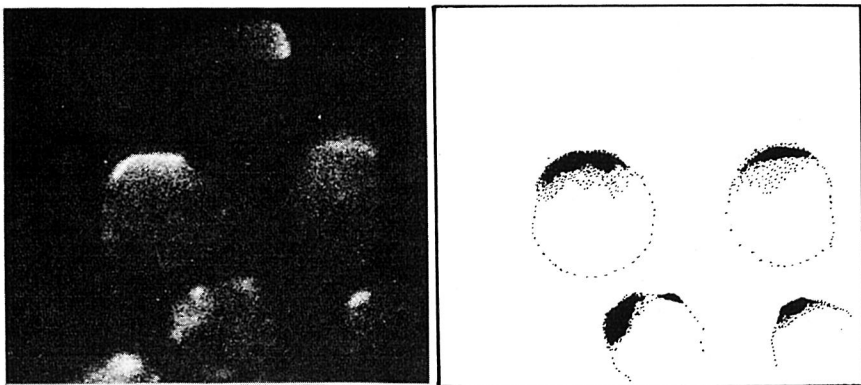


FIGURE 2. Asymmetrical distribution of fluorescent spots, forming a "cap".

However, within 30 min of incubation at 37 C, the fluorescent spots from the patchy area moved over the surface of about 50% of the cells to one pole, agglutinated and formed a "cap" (Fig. 2). Within 120 min, most of the caps which had been formed on the most cells, were pushed out to the top of the cytoplasmic projection (Fig. 3) and finally excluded and set free from the cell surface (Fig. 4).

On the other hand, when the temperature was maintained at 4 C, cap-formation was scarcely observed at time periods of up to 120 min. The pushing out of the cap and the detachment of the small fluorescent sphere from the surface of MSB-1 cells seem to be a phenomenon different from pinocytosis. The

movement of MATSA molecules on or in the membrane is of interest not only because of its relevance to our understanding of the movement of the membrane antigen but also because of its possible importance in the relationship of the membrane tumor-specific antigen and of their immune expression. Further studies on the biological significance of these phenomena are in progress.

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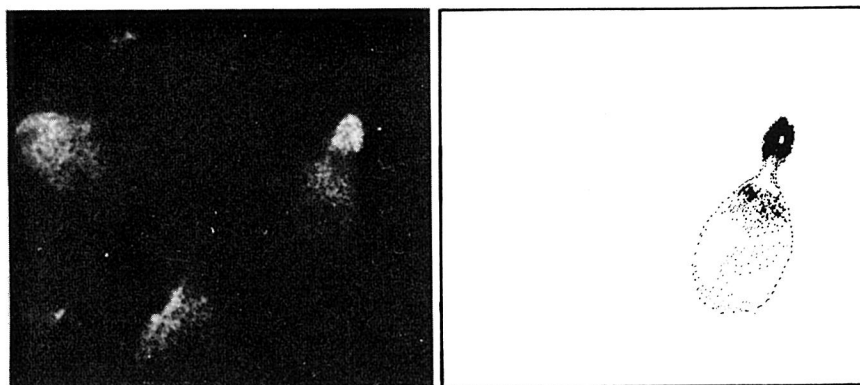


FIGURE 3. Cytoplasmic projection of "cap" from one pole of the cell surface.

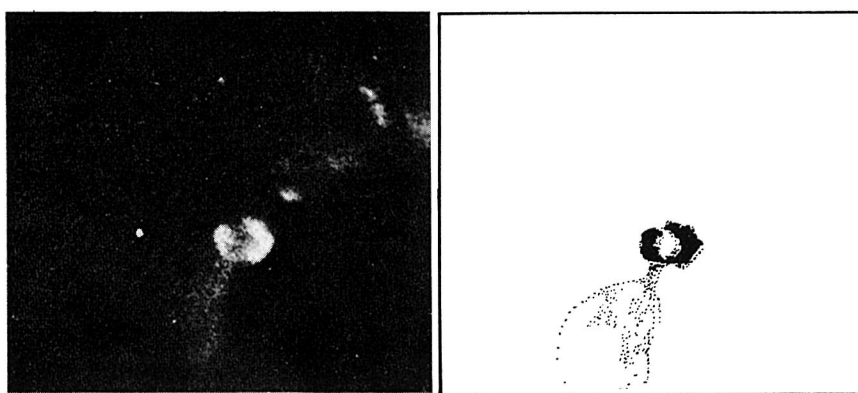


FIGURE 4. Detachment of a small fluorescent sphere from the cell surface.

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