CLINICAL APPLICATIONS OF HUMAN MONOCLONAL ANTIBODIES


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ABSTRACT. Hybridomas producing human monoclonal antibodies (h-MoAbs) were generated by using lymphocytes of the regional lymph node from cancer patients or peripheral blood lymphocytes (PBL) from healthy volunteers which were immunized in vitro. The h-MoAbs thus obtained were applied to a wide range of clinical purposes, including the immunocytological detection of cancer cells in sputum, the radioimmunoimaging of lung cancer and the therapies of tetanus and cancer. Cancer cells in sputum can be simply detected by the immunostaining method using h-MoAb AE-6. Clear images of lung cancer xenografts in nude mice were obtained with 125I-labeled h-MoAb HB4C5. Tetanus was successfully cured by the use of a combination of h-MoAbs G2 and G6 in animal tests. Cancer xenografts in nude mice were regressed by the administration of h-MoAb S-97 conjugated with Pseudomonas exotoxin A (PE). These data indicate that h-MoAbs are clinically of potent value.

1. INTRODUCTION

For clinical applications of the MoAb, it is essential that the MoAb has a high specificity and an adequate antigen-binding activity. Since very few h-MoAbs have satisfactorily met these fundamental requirements as compared with those of mouse origin, mouse MoAbs have been employed as the major targeting vehicle thus far. However, the mouse MoAb administered to the human body inevitably gives rise to the human antibody to mouse MoAb (Tjandra et al. (1990)), which in the subsequent doses would cause adverse effects such as anaphylaxis and rapid clearance of the mouse MoAb from circulation (Seccamani et al. (1989)). Although humanized mouse MoAbs made by gene manipulation can reduce these adverse effects (Winter and Milstein (1991)), the use of human MoAbs is considered to be most preferable for clinical purposes (Borrebaeck et al. (1990) and Persson et al. (1991)).
We have generated a number of hybridomas which secrete h-MoAbs with high specificities and high antigen-binding affinities. In this proceedings, clinical applications of these h-MoAbs are shown.

2. MATERIALS AND METHODS

2.1. GENERATION OF HYBRIDOMAS SECRETING HUMAN MONOCLONAL ANTIBODIES

For the generation of hybridoma secreting h-MoAb AE-6, normal PBL were first treated with L-leucyl-L-leucine methyl ester and then immunized in vitro with the human lung cancer cell line A-549 in the culture medium containing MDP, IL-2 and IL-6 for 4 days. In this in vitro immunization system, PWM and LPS, on the contrary to our common sense, inhibited the immunization. The hyperimmunized PBL were fused with the fusion partner A4H12, a human leukemic T cell line. The fusion was carried out according to the method detailed for hybridoma HB4C5 (Murakami et al. (1985)). Hybridomas HB4C5 and S-97 were generated by fusing lymphocytes of the regional lymph nodes from cancer patients with the fusion partner line NAT-30 and RF-S1 (Kamei et al. (1990)), respectively, in 50% polyethylene glycol.

Hybridomas G2 and G6 were obtained by fusing PBL from hyper-vaccinated individuals with the RF-S1 line.

2.2. MONOCLONAL ANTIBODIES

All human MoAbs were produced by culturing the hybridomas at 37 C in the serum-free medium. MoAbs of IgM isotype were purified by the method described elsewhere (Yano et al. (1988)) and IgG isotype by affinity chromatography on protein A-Cellulofine (Seikagaku Kogyo Co., Ltd., Tokyo) as described previously (Kamei et al. (1990)).

2.3. IMMUNOCYTOLOGICAL DETECTION OF LUNG CANCER CELLS IN SPUTUM

Cytospin preparations of sputa from lung cancer patients and normal individuals were immunostained with h-MoAb AE-6. The immunoperoxidase staining was carried out as described previously (Hirose et al. (1991)) and the cytological detection of cancer cells was performed using a computer-aided image analyzer (Qube, Nexus Inc.).

2.4. RADIOIMMUNOIMAGING

Xenografted nude mice after the i.v. injection of $^{125}$I-labeled h-MoAb HB4C5 were subjected to the gamma-scintigraphy on day 5 post-injection using a gamma-scintillation camera (Model Sigma 410, Ohio Nuclear, U.S.A.) (Hashizume et al. (1990)).

3. RESULTS AND DISCUSSION

3.1. IMMUNOCYTOLOGICAL DIAGNOSIS OF LUNG CANCER WITH H-MOAB AE-6

The immunocytological detection of cancer cells in sputa can facilitate the early diagnosis of lung cancer. The conventional Papanicolaou's staining method is time-consuming and requires well-trained pathologists, and thus difficult to be applied to mass screening.

H-MoAb AE-6, highly reactive with lung cancer tissues with no reactivity to normal lung and bronchial tissues when examined by the avidin-biotin-peroxidase method, is considered to be highly specific