日本医放会誌：47（6），845—848，1987（昭62）

Evidence of Existence of Low Dose Radiation Induced Tumor Immunity

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Research Code No. : 405.9

Key Words : Tumor immunity, Tumor infiltrated lymphocyte, Monoclonal antibody, Lymphocyte subset, Radiotherapy

放射線治療によって誘発される腫瘍免疫の証明

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（昭和61年7月30日受付）
（昭和61年11月4日最終原稿受付）

放射線治療によって出現する腫瘍浸潤リンパ球の重要性については以前から指摘されてきたが，そのサブセットは未だ明らかにされていない．この研究では，新しい免疫組織化学的手法を用いて，放射線治療中の腫瘍浸潤リンパ球を解析した下咽頭癌の1症例において得られた典型的な結果について報告する．方法としては，種々のヒトリンパ球サブセットに対するモノクローナル抗体とビオチン・アビジン・ホースラディッシュペルオキシダーゼコンプレックス法を用いた．

治療前にはリンパ球浸潤は少ししかみられなかったが，1回2Gyにて2回照射時に早くも，癌細胞をとり囲んで著明なリンパ球浸潤がみとめられた．主に抗Leu-1陽性リンパ球から成り，その大部分は抗Leu-3a＋3b陽性を示した．さらに，HLA-DR陽性の腫瘍細胞やその間質細胞も著明にみられた．抗Leu-2a陽性リンパ球やBリンパ球の浸潤はわずかにみられたが，マクロファージやナチュラルキラー細胞の浸潤はみとめなかった．30Gy照射後に行なった手術での切除標本からは癌細胞は見出されなかった．

日常臨床における放射線治療では，同一の組織型のものでも，いわゆる放射線治療に効きやすい癌とそうでない癌があることはしばしば経験されるところである．放射線効果におけるその違いの最も重要な因子として，放射線治療によって誘導される腫瘍免疫の存在が考えられる．ここで報告した症例では，主に腫瘍に浸潤していたのは抗Leu-3a＋3b陽性リンパ球で，ヘルパー/インデューサーTリンパ球あるいは遅延型過敏症リンパ球（DTH細胞）であり，そのターゲットはHLA-DR抗原を発現した腫瘍細胞とその間質細胞であることが強く示唆される．それゆえ，放射線治療によって容易に腫瘍免疫が誘導される患者と，放射線の物理的作用のみによって治療せざるを得ない患者とを鑑別することが非常に重要であると思われる．

Introduction

Though the significance of tumor infiltrating lymphocytes induced by radiation therapy have been discussed[1], their subsets have not been clarified yet. In this study, we analysed tumor infiltrating lymphocytes induced by radiation therapy with a newly developed immunohistochemical method[2)3)].

Materials and Methods

The subsets of lymphocytes infiltrating in a tumor tissue from a patient with hypopharyngeal cancer (62 years old, male, squamous cell carcinoma, T3N1M0, Stage III) were analysed by the method of Biotin-Avidin-Horseradish peroxidase complex[2)3)] using a panel of 7 mouse monoclonal antibodies to human leukocyte antigens (Becton Dickinson Monoclonal Center, USA) (Table 1).

Results and Discussion

Before therapy, lymphocytes were only slightly infiltrated in the biopsy specimen (Fig. 1 left). At as early as 4 Gy doses of radiotherapy (2 Gy X 2), remarkable infiltration of lymphocytes was observed surrounding cancer cells. The immunohistochemically stained preparations of this specimen clarified the nature of the subsets of tumor infiltrating lymphocytes (Fig. 2). They were mainly composed of anti-Leu-1 positive lymphocytes (T lymphocytes), and most of them were anti-Leu-3a + 3b positive lymphocytes (helper/inducer T lymphocytes). They were both ## (remarkable) by Shimokawara's grading[4]. Moreover, HLA (Human leukocyte antigen)-DR positive tumor and interstitial cells were remarkably observed. Infiltration of anti-Leu-2a positive lymphocytes (cytotoxic/suppressor T lymphocytes) and anti-Leu-M3 positive lymphocytes (B lymphocytes) was + (slight). Anti-Leu-M3 positive cells (macrophages) and anti-Leu-11b positive lymphocytes (natural killer cells) were scarcely seen in this specimen. In the resected specimen after receiving 30 Gy doses, cancer cells had almost completely disappeared. Our clinical experiences with cancer therapy to cases of the same histopathological type disclosed that some patients respond well to radiotherapy while others do not. The most important factor concerning the difference in the response is supposed to be the existence of tumor immunity induced by radiotherapy in some of the patients. In the case reported here, anti-Leu-3a + 3b positive lymphocytes which infiltrated mainly into the tumor were helper/inducer T lymphocytes or delayed type hypersensitivity lymphocytes (DTH cells)[5]. Furthermore, their target cells were strongly suggested to be HLA-DR positive tumor cells and interstitial cells[6)7)]. On the other hand, anti-Leu-2a positive lymphocytes was reported to respond to tumor antigens in association with HLA-A, B and C[8]. Therefore, it should be very important to differentiate the patients to

Table 1 Monoclonal antibodies used in this study and their antigen distribution on immune cells

Anti-Leu-1	Pan T lymphocytes
	Activated T lymphocytes
Anti-Leu-2a	Cytotoxic/suppressor T lymphocytes
Anti-Leu-3a+3b	Helper/inducer T lymphocytes
	Delayed type hypersensitivity lymphocytes
Anti-Leu-HLA-DR	B lymphocytes
	Macrophages
	Activated T lymphocytes
	Tumor cells and their interstitial cells
Anti-Leu-M3	Macrophages
Anti-Leu 14	B lymphocytes
Anti-Leu 11b	Natural killer cells

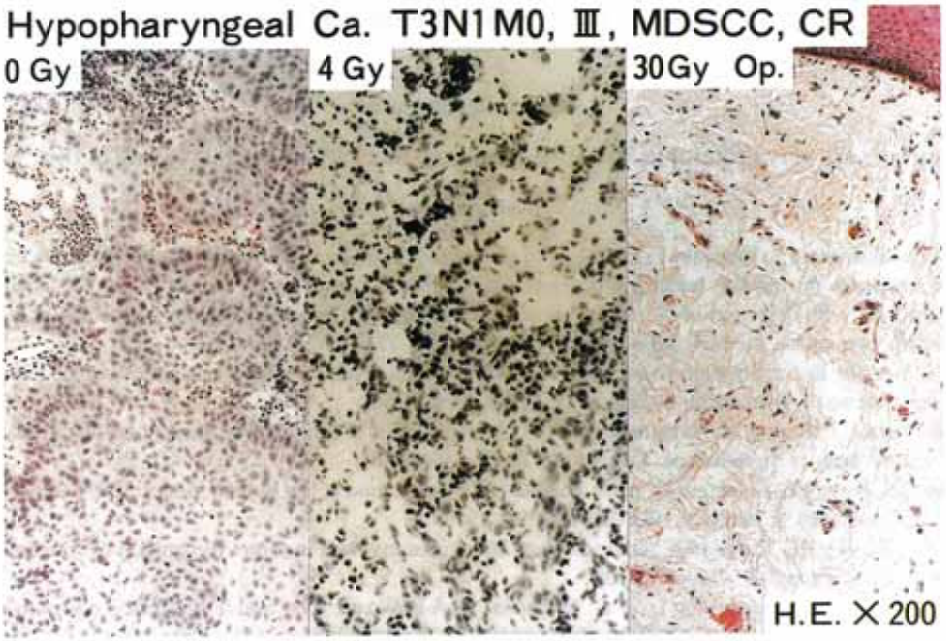

Fig. 1 Photomicrographs of the Hematoxylin-Eosin staining of hypopharyngeal cancer tissue from a patient (62 years old, male, squamous cell carcinoma, T3N1M0, Stage III) at pretreatment, after delivery of 4 Gy doses, and at operation after receiving 30 Gy doses.

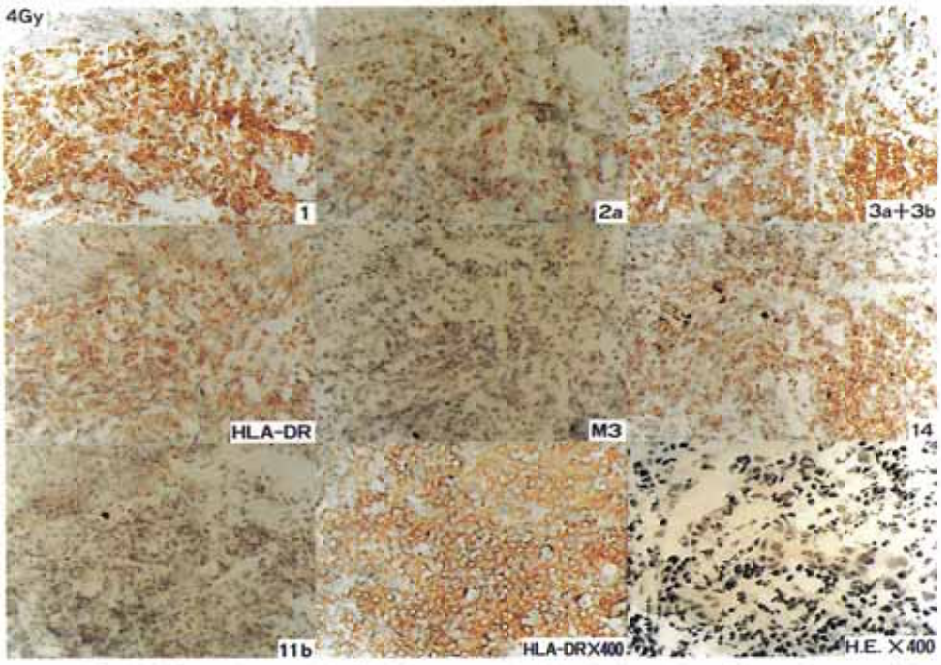

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Fig. 2 Photomicrographs of the immunohistochemical staining of the patient receiving 4 Gy doses of radiotherapy. The grade of infiltration of anti-Leu-3a + 3b positive lymphocytes and HLA-DR positive tumor and interstitial cells were both ## (remarkable).

whom tumor immunity can be easily induced by radiotherapy from those that can be treated only by the physical effect of radiotherapy.

Acknowledgement

The authors are very grateful to Miss Eva Garcia del Saz for her critical reading of this manuscript.

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