ADJUVANT IMMUNOTHERAPY OF LUNG CANCER WITH BCG CELL WALL SKELETON (BCG-CWS)

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Four hundred fifty-five patients with lung cancer were treated with oil-attached cell-wall skeleton of bacillus Calmette-Guérin (BCG-CWS) as adjuvant immunotherapy following initial conventional therapy. The overall survival period of the patients was prolonged significantly as compared with that of 380 patients in a historical control group receiving initial conventional therapy alone ($p < 0.0001$). The prolongation of the survival period of the patients was also statistically significant when classified according to clinical stages and histological cell types. The therapeutic effect was remarkable in patients combined with malignant pleurisy. Intrapleural injection of BCG-CWS resulted in not only prevention of accumulation of pleural effusion and abrogation of tumor cells but also in prolongation of survival period ($p = 0.016$). No serious side effects due to BCG-CWS were experienced. From the previous experimental studies and clinical experiences with human tumors, it can be concluded that adjuvant immunotherapy with BCG-CWS is a useful therapeutic modality for lung cancer, especially in cases combined with malignant pleurisy.

***Cancer* 43:1314–1319, 1979.**

THE CLINICAL BENEFITS of immunotherapy with bacillus Calmette-Guérin (BCG) for human cancer have been confirmed in prolonging the survival period of the patients as well as duration of remission of the disease.[3,5,8,9,13,14,15,19] However, many investigators have also noticed various kinds of serious side effects during the therapy.[3,11,12,20,21,23] In previous studies, we have shown that cell-wall skeleton (CWS), having a principal structure of mycolic acid-arabinogalactan-mucopeptide complex, is a biologically active component in immunopotentiation.[25,26,27] Recent studies have demonstrated that the material has a potent immunotherapeutic effect on human tumors as well as animal tumors when given in oil-attached form.[2,16,28,29,30,31,32]

This report expands our initial cooperative clinical study with the use of BCG-CWS for immunotherapy of lung cancer patients in 11 collaborative hospitals.

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Accepted for publication May 22, 1978.

PATIENTS AND METHODS

Patients

A total of 455 patients with primary lung cancer treated in our collaborative group consisting of 11 hospitals were entered in this study between April 1974 and December 1976. All patients were classified to either squamous cell carcinoma, adenocarcinoma, small cell carcinoma or large cell carcinoma by histological examination of the sputum, pleural fluid and/or biopsy material.

Clinical staging was done according to the classification by Mountain.[17] All patients received initially conventional therapy such as surgery, irradiation and/or chemotherapy.

Oil-attached BCG-CWS was prepared by the same method as used previously.[28,29,30]

Procedure of Immunotherapy with BCG-CWS

In principle, treatment with BCG-CWS was introduced following the initial conventional therapy, and in some cases during the therapy. After skin test with 10 μg of BCG-CWS, in most cases 200 to 400 μg of oil-attached BCG-CWS was administered weekly or biweekly into the skin of bilateral upper arms as

0008–543X/79/0400/1314 $0.80

induction of the immunotherapy. In some cases, the same dose of BCG-CWS was injected directly into the intrathoracic unresectable tumor at surgery, intrabronchial tumor with bronchofiberscope, or cutaneous metastatic tumor. In 55 patients combined with malignant pleurisy histologically certified, BCG-CWS was injected repeatedly into the affected pleural cavity at one or two weeks intervals.

Following several injections of BCG-CWS as mentioned above, patients were referred to maintenance therapy, in which 100 or 200 μg of BCG-CWS was injected monthly into the skin of bilateral upper arms. The termination of the therapy was not fixed.

When patients showed hyperreaction at initial skin test with BCG-CWS or during the treatment with BCG-CWS, the dose given was adequately reduced.

Evaluation of Immunotherapy

Three hundred eighty lung cancer patients in the same hospital, who had conventional therapy alone between April 1971 and March 1974 were used as historical controls.[7] Not included in the control group are patients whose cell types were out of the four cell types mentioned above. The distribution of the major clinical features which have been known to affect a prognosis of the disease is shown in Table 1.[22] The sex of the patients and the modality of the initial conventional treatment including irradiation and operation were a little different in distribution between the both groups.

The effect of the immunotherapy was evaluated in terms of survival period from admission.[4] Survival rate was calculated with life table method and the difference between both groups was evaluated statistically by the generalized Wilcoxon test according to Gehan.[6]

TABLE 1. Major Clinical Features of Patients

Clinical features	BCG-CWS treated (455 cases)	Control (380 cases)	Chi² test
Cell types			
Squamous cell carcinoma	38.2% (174)	44.7% (170)	
Adeno-carcinoma	42.6 (194)	38.4 (146)	
Small cell carcinoma	14.3 (65)	10.8 (41)	
Large cell carcinoma	4.9 (22)	6.1 (23)	NS
Clinical stages			
I	11.9% (54)	10.5% (40)	
II	12.5 (57)	13.2 (50)	
III	75.6 (344)	76.3 (290)	NS
Sex			
Male	73.8% (366)	80.5% (306)	
Female	26.2 (119)	19.5 (74)	$p < 0.05$
Age (years)	60 ± 10	62 ± 10	NS
Treatments			
Operation	34.7% (158)	23.9% (91)	$p < 0.05$
Irradiation	56.3 (256)	66.1 (251)	$p < 0.05$
Chemotherapy	69.9 (318)	65.8 (250)	NS
Conservative	2.6 (12)	3.9 (15)	NS
Patients entry	1974–1976	1971–1974	

Number of patients is indicated in parentheses.
NS: not significant.

RESULTS

Survival Period

One hundred sixty-three patients in the immunotherapy group and 39 patients in the control group are still alive as of April 1977. The survival rates of all patients in the both groups are shown in Fig. 1. The 50% survival rates of all patients were 13.5 and 8.5 months in the immunotherapy and control group. The difference was statistically significant ($p < 0.0001$). Table 2 shows 50% survival periods of the patients according to histological cell type and clinical stage. The patients with small cell carcinoma at various clinical stages were evaluated inclusively in one group, because they were too small in number to be classified according to clinical stages. The prolongation of survival period in immunotherapy group was also significant ($p = 0.0008$).

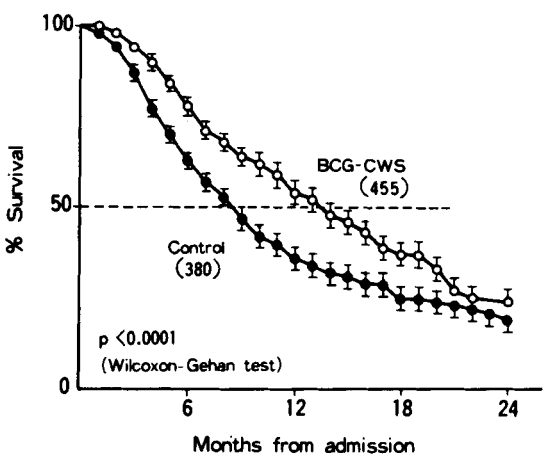

FIG. 1. The survival of all patients. The survival rates are depicted in SE.

TABLE 2. Fifty percent Survival According to Histological Cell Type and Clinical Staging

Histological cell types	Clinical stages	50% survival months: BCG-CWS group	50% survival months: Control group	Statistical significance (Wilcoxon-Gehan)
Small cell carcinoma	I, II, III	11.0 (65)	5.5 (41)	p = 0.0008
Large cell carcinoma	I, II, III	11.0 (22)	7.5 (23)	NS
Squamous cell carcinoma and adenocarcinoma	I, II	>23.0* (90)	23.0 (74)	NS
Squamous cell carcinoma	III	12.0 (123)	8.0 (141)	p = 0.0005
Adenocarcinoma	III	12.0 (155)	6.5 (101)	p < 0.0001

* The patients are still alive at 70%. NS: not significant.
Number of patients is indicated in parentheses.

In large cell carcinoma, the 50% survival periods of the patients were 11 months in the immunotherapy group and 7.5 months in the control group. However, the number of the patients was considerably limited and the difference in survival period hardly deserved a statistical evaluation.

The 50% survival period of the patients with squamous cell carcinoma and adenocarcinoma in stage I and II was 23 months in the control group, while 70% of the patients in the immunotherapy group were still alive at the same time. The 50% survival periods of 123 patients with squamous cell carcinoma and 155 patients with adenocarcinoma in stage III were 12 months in the immunotherapy groups, whereas those of 141 and 101 respective control patients were 8 and 6.5 months. The both differences were statistically significant (p = 0.0005 in squamous cell carcinoma, p < 0.0001 in adenocarcinoma).

Fifty-five patients with malignant pleurisy receiving BCG-CWS immunotherapy survived much longer than controls (Fig. 2). In this group, all major clinical features of the patients statistically corresponded to those of controls as indicated in Fig. 2.

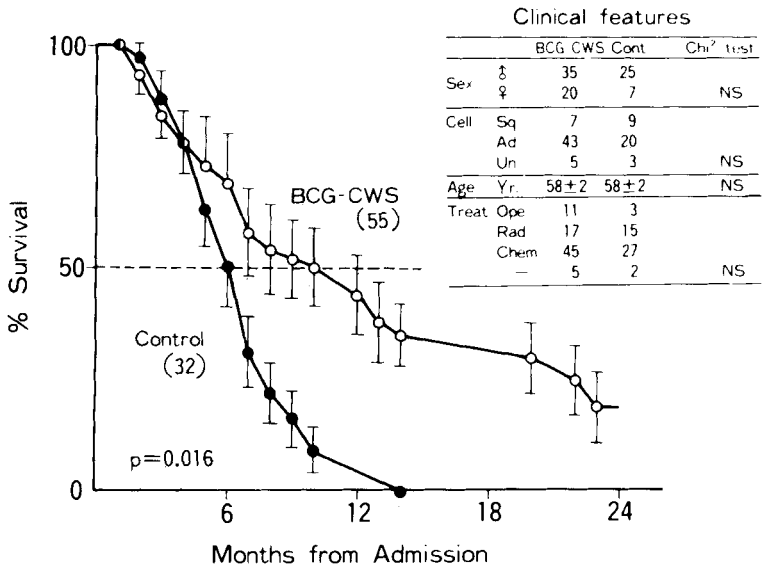

FIG. 2. The survival of patients with malignant pleural fluid receiving the intrapleural BCG-CWS immunotherapy and conventional therapy alone.

Finally, in order to make clear the variation in patients' survival period from institution to institution, four representative hospitals were compared in which the number of patients were enough to be examined statistically. As shown in Table 3, BCG-CWS treated patients survived much longer than the control groups in every institution.

TABLE 3. Fifty Percent Survival Months of Treated and Control Patients by Institutions

Institution	BCG-CWS treated	Controls
Kinki Central Hospital for Chest Disease (Osaka)	9.0 (86)	5.5 (27)
Kyushu Cancer Center (Fukuoka)	19.0 (107)	9.5 (157)
Osaka Medical Center for Adult Disease (Osaka)	24.0 (44)	9.0 (12)
Tokyo Medical College	10.0 (128)	8.5 (120)

Four representative institutions, in which number of patients were enough to be compared statistically, were summarized in this table.
Number of patients is indicated in parentheses.

DISCUSSION

The present study revealed that additional immunotherapy with BCG-CWS to initial conventional treatment such as surgery irradiation and/or chemotherapy resulted in improvement of prognosis of the patients with lung cancer. Namely, the overall survival period of the patients in immunotherapy group was significantly longer than that of the control group. Between both groups, there was no significant difference in distribution of histological cell type, as well as clinical staging which has an important effect on prognosis. Accordingly, it can be concluded that the prolonged survival period in the immunotherapy group is not due to a difference in these clinical features between both groups.

This was further confirmed with analysis according to histological cell types of patients in stage III. In the immunotherapy groups, 50% survival periods of patients with squamous cell carcinoma and adenocarcinoma were prolonged, respectively, to 12 months with statistical significance as compared with 8 and 6.5 months in control groups. Particularly, the effect in patients with squamous cell carcinoma in stage III seems to be evaluable because other clinical features—sex and modality of initial conventional treatment—were also comparable.

In the present study, modality of initial conventional treatment was not quite comparable between the immunotherapy and control groups as indicated in Table 1. Although it has not been clear whether an annual change in percent of a mode of initial conventional therapy influences survival, in order to assess the influence on survival period further analysis of the survival period of all patients was carried out according to modality of initial therapy, irradiation and surgery. In each immunotherapy group, prolongation of survival period was significant as compared with controls ($p < 0.001$ in irradiation group, $p < 0.03$ in operation group). This indicates that the prolongation of the patients' survival period in BCG-CWS treated group was not originated from the increase in percent of the patients receiving operation.

The two year survival rate of patients with squamous cell carcinoma and adenocarcinoma in stage I and II were 50 and 70% in control and immunotherapy group, respectively. The difference is statistically insignificant, but it seems too early to be evaluated because many patients, most of whom had received operations, were still alive.

As previously reported, intrapleural injection of BCG-CWS into the affected pleural cavity of patients with malignant pleurisy resulted in prolongation of survival period. All clinical features such as indicated in Table 1 are not different in distribution in Chi^2 test between both groups. Although precise mechanism has remained undetermined, systemic immune potentiation and local effect on pleural effusion are possibly responsible for the clinical results.[18,29,30,31]

Previously, we have shown that BCG-CWS has an anti-tumor effect through the immunopotentiation at various levels when injected into the tumor-bearing hosts.[25,26,27,28,29,31] Some participants in this study have individually reported a close correlation between therapeutic effect and improvement of patient's cellular immune response during the therapy.[10,24,31] These results led us to suppose that immunotherapy with BCG-CWS can make the patient's immune mechanism able to react effectively against the tumor resulting in prolongation of the survival period.

The uneven effect of immunotherapy on 50% of the survival periods from institution to institution is probably related to the varied

distribution of cell types and clinical stages of patients in institutions.

In view of side effects, BCG-CWS was much less harmful as compared to living BCG, even when administered intrapleurally or intralesionally.[11,21,23] Ulcerations at the injected site of the skin were smaller and healed within 4 to 8 weeks. No case presented an enhancement of tumor growth due to the therapy.

In conclusion, the present study leads us to believe that adjuvant immunotherapy with BCG-CWS is a useful therapeutic modality in improving the prognosis of patients with lung cancer, even though considering that evaluation with historical control has a moot result.

APPENDIX

Participants in the trial were as follows:

Institution	*Responsible doctor*
Hiroshima University	Yukio Nishimoto
Kinki Central Hospital for Chest Disease (Osaka)	Kenji Sawamura
Kyoto University	Chiaki Nagase
Kyushu Cancer Center (Fukuoka)	Kosei Yasumoto
Niigata University	Yoshio Ito
Okayama University	Shozo Yamana
Osaka Medical Center for Adult Disease	Shoji Hattori
Osaka University	Yuichi Yamamura
Tokushima University	Eiro Tsubura
Tokyo Medical College	Yoshihiro Hayata
Toneyama National Hospital (Osaka)	Yoshihiro Yamamura

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