

In Vitro–Chemosensitivity Test Using the Collagen Gel Droplet Embedded Culture Drug Test (CD-DST) for Malignant Pleural Mesothelioma: Possibility of Clinical Application

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Purpose: An in vitro–chemosensitivity test using the collagen gel droplet embedded culture drug test (CD-DST), established by Kobayashi et al. (*Jpn J Cancer Res* 2001; 92: 203–10), has been widely used on various tumors. This study retrospectively evaluated its possibility of clinical application to patients with malignant pleural mesothelioma (MPM).

Patients and Methods: CD-DST using 26 fresh specimens obtained by biopsy or surgery on MPM patients investigated in vitro responses to cisplatin (CDDP), carboplatin (CBDCA), doxorubicin (ADR), etoposide (VP-16), 5-fluoruracil (5-FU), gemcitabine (GEM), vinorelbine (VNR), irinotecan (SN-38), and docetaxel (TXT). Correlations between CD-DST data and clinical effects were then assessed for some MPM patients undergoing chemotherapy.

Results: The rate of in vitro sensitivity to each chemoagent (N = tested number) was 35% for CDDP (N = 23), 14% for CBDCA (N = 21), 7% for ADR (N = 15), 15% for VP-16 (N = 13), 0% for 5-FU (N = 15), 45% for GEM (N = 11), 25% for VNR (N = 8), 40% for SN-38 (N = 5), and 44% for TXT (N = 9). No difference was observed between CD-DST data of each chemoagent and histological type. Of these MPM patients, 14 clinical effects on 13 patients who underwent chemotherapy for primary or recurrent disease were reviewed in comparison with CD-DST data of each chemoagent. Among 10 chemotherapies including in vitro–sensitive chemoagents, 3 led to partial response (PR), and 7 resulted in four stable diseases (SDs) and 3 to progressive diseases (PDs). In contrast, among 4 chemotherapies using in vitro–resistant chemoagents, SD and PD were observed in 1 and 3, respectively. In regard to the clinical response rate, CD-DST sensitivity, specificity, and accuracy in the 14 examined chemotherapies were respectively 100%, 36%, and 50%, and in regard to the disease control rate, they were 88%, 60%, and 71%. CD-DST data for the chemoagents were to a limited extent significantly correlated with the disease control status of chemotherapy (p = 0.052).

Conclusion: Although the number of tested MPM specimens was small, CD-DST data obtained by biopsy or surgical-fresh specimens of MPM marginally correlated to the disease control effect of chemotherapy for this disease. Therefore CD-DST may possibly be applied to selecting the chemotherapy regimen for MPM. To determine the possibility of a clinical application of this test to MPM, a prospective clinical study of a greater number of patients will be necessary. (*Ann Thorac Cardiovasc Surg* 2008; 14: 355–362)

Key words: malignant pleural mesothelioma, collagen gel droplet embedded culture drug test, in vitro–chemosensitivity test, surgical specimen

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Introduction

Malignant pleural mesothelioma (MPM) is usually an aggressive tumor that is resistant to conventional modes of treatment, such as chemotherapy, radiotherapy, and surgery. As a result, it is well known that overall prognosis is extremely poor even today, with median survival periods of only 6 to 12 months. Since most patients with MPM suffer from the advanced stage, chemotherapy has played an important role in the treatment of this disease. In fact, chemotherapy has been tested systemically on patients with MPM, but the reported level of activity of chemoagents was at one time generally unsatisfactory. However, several promising active chemoagents have been developed in this decade, and recent meta-analysis has confirmed that chemotherapy use on this disease can have small but modest survival benefits and that it can also improve tumor response rates.¹⁻⁴⁾ In consideration of these recent problems, chemotherapy might be made more effective by individualizing the treatment of each patient based on drug sensitivity data obtained from specimens of the patient's tumor. From this point of view, the clinical application of the *in vitro*-chemosensitivity test is a promising strategy for chemotherapy.

Recently, Kobayashi et al.⁵⁻⁷⁾ developed a novel test of this kind, the collagen gel droplet embedded culture drug sensitivity test (CD-DST), using various types of malignant neoplasms. This technique has been safely and widely applied in Japan, and recent clinical and experimental data may support its usefulness in practical medicine,⁵⁻¹³⁾ but to date, the *in vitro*-chemosensitivity data of MPM remain unknown. This present study was then performed to evaluate retrospectively the association between CD-DST results, which had been obtained from biopsy or surgically resected MPM tumors, and the clinical effect of chemotherapy using some representative agents.

Patients and Methods

From 1990 to 2005, the Department of Thoracic Surgery of our organization diagnosed MPM in 45 patients. Of this total, 26 were enrolled for a CD-DST analysis using biopsy or surgical specimens, under the informed consent of each patient. CD-DST for cisplatin (CDDP), carboplatin (CBDCA), doxorubicin (ADR), etoposide (VP-16), 5-fluorouracil (5-FU), gemcitabine (GEM), vinorelbine (VNR), irinotecan (SN-38), and docetaxel

(TXT) were performed, and some *in vitro*-sensitivity data were obtained in 23 samples. Three samples were uninformative for reasons of technical failure: low growth rate in 2 and no viable malignant cells in 1.

CD-DST was performed as described previously by Kobayashi et al.⁵⁻⁷⁾ Briefly, biopsied or surgically resected MPM specimens were digested in dispersion collagenase enzyme, and the dispersed cancer cells were incubated in a collagen gel-coated flask. The viable cells alone adhering to the collagen gel layer were then collected and added to reconstructed Type I collagen solution (Cellmatrix Type CDTM; Nitta Gelatin Inc., Yao, Japan). Three drops of these mixtures were placed in each well of a 6-well multiplate, and CDDP (0.2 ug/ml), CBDCA (2.0 ug/ml), ADR (0.02 ug/ml), VP-16 (1.0 ug/ml), 5-FU (1.0 ug/ml), GEM (0.08 ug/ml), VNR (0.05 ug/ml), SN-38 (0.03 ug/ml), or TXT (0.1 ug/ml) were then added to each well, and the plate was incubated for 24 h. After removal of the medium containing each chemoagent, each well was incubated with PCM-2 medium (Primaster, Nitta Gelatin Inc., Yao, Japan) for 7 days. Neural red was then added to stain colonies in the collagen gel droplets, which were finally fixed with formalin. The *in vitro*-chemosensitivity effect of each chemoagent was expressed as a ratio of the total colony volume (T) of the treated cells to that of the untreated cells (C). Originally, a sample with a ratio of T to C of 50% or less, greater than 60%, and from 51% to 60% was defined as *in vitro* sensitive, resistant, and borderline, respectively, but in the present study, the cutoff ratio was regarded to be 60%, namely, samples with a ratio of T to C of 60% or less being considered as *in vitro* sensitive.

Patient characteristics are summarized in Table 1. Nineteen men and 7 women whose age at biopsy or surgery ranged from 41 to 83 years (mean, 58) were studied. The past history of asbestos exposure was demonstrated in 15 patients (58%). Tumor histology was the epithelioid type in 13 patients (50%), the biphasic type in 8 (31%), and the sarcomatoid type in 5 (19%). The tumor's International Mesothelioma Interest Group (IMIG) stage¹⁴⁾ at biopsy or surgery was stage II in 6 patients (23%), stage III in 13 patients (50%), and stage IV in 7 patients (27%). The sample resection mode for CD-DST was biopsy in 7 patients (27%), and surgical specimens were applied in 19 patients (81%). Two samples were obtained from neck metastatic lymph node in 1 patient and from brain metastasis in another; the remaining samples were from intrathoracic dissemi-

Table 1. Characteristics of patients (N = 26)

		Number of patients (%)
Gender	Male	19 (73)
	Female	7 (27)
Age	Mean [range]	58 [41–83]
Asbestos exposure	Yes	15 (58)
	No or unknown	11 (42)
Smoking	Current	12 (46)
	Ex	5 (19)
	Never	9 (35)
Histology	Epithelioid	13 (50)
	Biphasic	8 (31)
	Sarcomatoid	5 (19)
Stage (IMIG)	II	6 (23)
	III	13 (50)
	IV	7 (27)
Resection mode	Biopsy	7 (27)
	Surgery	19 (81)
Sample site	Intrathoracic	24 (73)
	Others	2 [neck node, brain] (8)
Chemotherapy history	No	23 (88)
	Yes	3 (12)
CD-DST	Success	23 (88)
	Failure	3 (12)

ex, ex-smoker; IMIG, International Mesothelioma Interest Group; CD-DST, collagen gel droplet embedded culture drug test.

nated lesions. Chemotherapy had been performed before CD-DST analysis in 3 patients (12%) as induction therapy.

Although chemotherapy performed in the present series was of diverse regimens, three types were in principal categorized: CDDP-based, single agent (GEM or VNR), and others. CDDP-based combination chemotherapy (CDDP plus GEM) was administered according to Byrne et al.¹⁵⁾ with at least two courses. A single agent using GEM or VNR was performed with 4 to 8 cycles according to Kindler et al.¹⁶⁾ and Steele et al.,¹⁷⁾ respectively. ADR was administered with a minor change by Sørensen et al.,¹⁸⁾ and VP-16 was also administered according to Sahnoud et al.,¹⁹⁾ with minor modification. CBDCA was administered at area under the blood concentration time curve (AUC) 4–5 dose, in single agent according to Raghavan et al.,²⁰⁾ and VP-16 combination chemotherapy according to Tueni et al.,²¹⁾ with minor change.

The clinical effect of chemotherapy was principally evaluated after each cycle according to both radiological and clinical findings. The local effect on intrathoracic primary or recurrent disease was assessed mainly by chest computed tomography (CT) on the modified

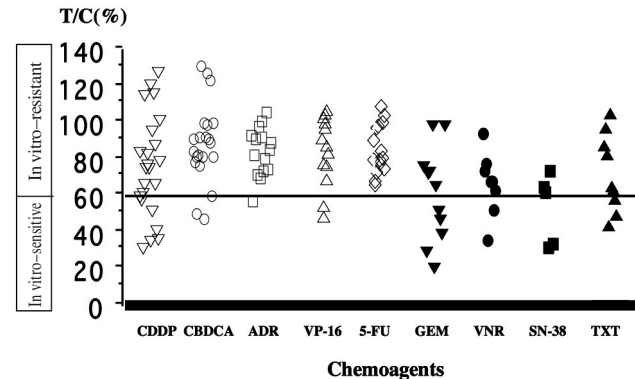


Fig. 1. In vitro-chemosensitivity profile of each chemoagent against MPM.

This figure shows a distribution of the CD-DST T/C ratio (%) of each chemoagent. T/C ratio of 60% or less is judged as in vitro sensitive, and that of more than 60% as in vitro resistant. T/C, total colony of the treated cells / total colony of the untreated cells; CDDP, cisplatin; CBDCA, carboplatin; ADR, doxorubicin; VP-16, etoposide; GEM, gemcitabine; VNR, vinorelbine; SN-38, irinotecan; TXT, docetaxel.

response evaluation criteria in solid tumours (RECIST) criteria.²²⁾ Complete remission (CR) was defined as the disappearance of all target measurable lesions for at least 4 weeks with no evidence of tumor elsewhere. Partial response (PR) was defined as a reduction of 30% or during a period of at least 4 weeks without the development of new metastatic lesions. Progressive disease (PD) was defined as an increase of 20% or more in the total tumor measurement over the nadir measurement or the appearance of new lesions. Stable disease (SD) was defined as meeting neither the criteria of PR nor of PD. Response cases included CR and PR cases, and disease control cases included response cases plus SD cases.

The statistical analysis was performed using Fisher's exact probability test or unpaired comparison test software (StatView-J, Version 5.0; SAS Institute Inc., Cary, NC, USA).

Results

As also shown in methods, whereas CD-DST was successfully performed in 23 MPM specimens (88%), 3 offered no CD-DST data because of a low growth rate in 2 and no viable malignant cells in 1. As for the resection mode in such CD-DST-failure specimens, surgery was performed on 1, and a biopsy on another. Histologically, 2 were epithelioid types and 1 was a sarcomatoid type. No previous chemotherapy was performed on

Table 2. In vitro–chemosensitivity of each chemoagent against MPM

Chemoagent	In vitro–sensitive No.	Tested No.	%
Cisplatin (CDDP)	8	23	35
Carboplatin (CBDCA)	3	21	14
Doxorubicin (ADR)	1	15	7
Etoposide (VP-16)	2	13	15
5-fluorouracil (5-FU)	0	15	0
Gemcitabine (GEM)	5	11	45
Vinorelbine (VNR)	2	8	25
Irinotecan (SN-38)	2	5	40
Docetaxel (TXT)	4	9	44

MPM, malignant pleural mesothelioma.

these specimens.

CD-DST data of each chemoagent are shown in Fig. 1 and Table 2. Figure 1 shows the distribution of the CD-DST T/C value (%) of each chemoagent, and Table 2 shows a summary of in vitro–sensitive case rates. The rate was 35% in CDDP, 14% in CBDCA, 7% in ADR, 15% in VP-16, 0% in 5-FU, 45% in GEM, 25% in VNR, 40% in SN-38, and 44% in TXT. The correlation between the CD-DST data of each chemoagent and clinicopathological factor (i.e., histology and stage) was also analyzed, but no significant difference was observed because of the small number of tested cases. Figure 2 shows a representative distribution of CD-DST T/C value (%) of CDDP and GEM according to histological type, epithelioid versus nonepithelioid, indicating no statistical difference between in vitro–sensitivity status and histology (CDDP: $p = 0.458$; GEM: $p = 0.111$).

Of the 7 patients with CD-DST data obtained by biopsy, 4 underwent first-line chemotherapy against unresectable MPM disease (Table 3-1). Five chemotherapy courses were evaluated to examine an association between the CD-DST data and the clinical effect. For 1 patient (Pt. No. 2), two regimens of first- and second-line chemotherapy were analyzed. The clinical effect of chemotherapy including at least one CD-DST–sensitive chemoagent was PR in two regimens (Pt. No. 2) and PD in one (Pt. No. 1).

Table 3-2 shows an association between CD-DST data and the chemotherapeutic effect on postoperative recurrent disease. Of the 6 patients, 5 underwent GEM-based chemotherapy, and unfortunately no PR was obtained. However, of 4 chemotherapies, including at least 1 CD-DST–sensitive chemoagent, the clinical effect was SD in three regimens (Pt. Nos. 1, 3, and 6).

On the other hand, the 2 chemotherapies, including CD-DST–resistant chemoagents, showed PD.

Table 3-3 shows an association between CD-DST data and the induction of the chemotherapeutic effect on surgically resected primary lesions. Unlike other CD-DST data, these three sets of data were analyzed using the posttreated samples. All patients underwent chemotherapy, including at least CD-DST–sensitive chemoagents, and the clinical response was PR in 1 patient and SD in 2.

Table 4 shows a summary of CD-DST data and the clinical effect on MPM patients undergoing three types of chemotherapy described above. Chemotherapy, including at least one CD-DST–sensitive chemoagent, showed PR in three regimens, SD in five, and PD in two. On the other hand, chemotherapy including CD-DST–resistant chemoagent showed SD only in one regimen and, unfortunately, PD in three. From the point of view of CD-DST data in regard to clinical response (including PR), we found sensitivity, specificity, positive prediction value (PPV), negative prediction value (NPV), and accuracy to be 100%, 36%, 30%, 100%, and 50%, respectively. Also, in regard to disease control (including PR and SD), sensitivity, specificity, PPV, NPV, and accuracy were 88%, 60%, 80%, 75%, and 71%, respectively. In vitro–sensitive data according to CD-DST of the chemoagents were to a limited extent significantly correlated with the disease control status of chemotherapy ($p = 0.052$).

Discussion

MPM is a tumor of mesothelial cells in the plural cavity, occurring mostly after asbestos exposure of very long latency periods (30–40 years). In consideration of the history of the asbestos industry in Japan, the incidence of MPM is unhappily increasing and is predicted to continue rising markedly into the next decade, as it is in other countries.^{23,24} Unfortunately, MPM is generally a refractory tumor for which chemotherapeutic regimens have been far from satisfactory in achieving clinical responses, and most patients have a clinically advanced stage at diagnosis. Therefore the position of this disease in the league table of cancer-related deaths is rising remarkably. Because diagnosis can be difficult and can offer little hope for a cure, and because the disease has the potential for extremely unpleasant symptoms, its therapeutic strategy should be immediately established.

Table 3-1. Association between in vitro-sensitivity data and chemotherapeutic effect (biopsy specimen)

Pts No.	Specimen	Histology	Chemoagent	In vitro-data		Chemotherapy	Clinical effect
				T/C value (%)	(S or R)		
1	Biopsy	Sarco	CDDP	58	(S)	CDDP + GEM	PD
			GEM	28	(S)		
2	Biopsy (brain)	Biphasic	CDDP	74	(R)	1) CDDP + GEM	PR
			GEM	19	(S)		
3	Biopsy (lymph node)	Biphasic	VNR	34	(S)	2) VNR	PR
			ADR	83	(R)	ADR	PD
4	Biopsy	Biphasic	CDDP	87	(R)	CDDP + VP-16 + ADR	SD
			VP-16	112	(R)		
			ADR	101	(R)		

pts, patients; T/C, total colony of the treated cells / total colony of the untreated cells; S, sensitive; R, resistant; sarco, sarcomatoid type; CDDP, cisplatin; GEM, gemcitabine; VNR, vinorelbine; ADR, doxorubicin; VP-16, etoposide; PD, progressive disease; PR, partial response; SD, stable disease.

Table 3-2. Association between in vitro-sensitivity data and chemotherapeutic effect on recurrent disease (surgical specimen)

Pts No.	Specimen	Histology	Chemoagent	In vitro-data		Chemotherapy	Clinical effect
				T/C value (%)	(S or R)		
1	Surgery	Epi	CDDP	40	(S)	CDDP + GEM	SD
			GEM			Unknown	
2	Surgery	Biphasic	GEM	71	(R)	GEM	PD
3	Surgery	Sarco	GEM	16	(S)	GEM	SD
4	Surgery	Epi	GEM	38	(S)	GEM	PD
5	Surgery	Epi	GEM	97	(R)	GEM	PD
6	Surgery	Epi	CBDCA	60	(S)	CBDCA	SD

pts, patients; T/C, total colony of the treated cells / total colony of the untreated cells; S, sensitive; R, resistant; epi, epithelioid type; sarco, sarcomatoid type; CDDP, cisplatin; GEM, gemcitabine; CBDCA, carboplatin; SD, stable disease; PD, progressive disease.

Table 3-3. Association between in vitro-sensitivity data and induction chemotherapeutic effect on primary disease (surgical specimen)

Pts No.	Specimen	Histology	Chemoagent	In vitro-data		Chemotherapy	Clinical effect
				T/C value (%)	(S or R)		
1	Surgery	Epi	CDDP	60	(S)	CDDP + GEM	PR
			GEM	72	(R)		
2	Surgery	Epi	CDDP	74	(R)	CDDP + GEM	SD
			GEM	51	(S)		
3	Surgery	Biphasic	CBDCA	51	(S)	CBDCA + VP-16	SD
			VP-16	46	(S)		

T/C, total colony of the treated cells / total colony of the untreated cells; S, sensitive; R, resistant; epi, epithelioid type; CDDP, cisplatin; GEM, gemcitabine; CBDCA, carboplatin; VP-16, etoposide; PR, partial response; SD, stable disease.

Table 4. Summary of the association between in vitro-sensitivity data and chemotherapeutic effect

	Chemotherapeutic effect		
	PR	SD	PD
In vitro-chemosensitivity data			
Sensitive	3	5	2
Resistant	0	1	3

PR, partial response; SD, stable disease; PD, progressive disease.

Most available chemotherapeutic agents have been tried on MPM, but none has consistently produced a response above 20% in the previous series.¹⁻⁴⁾ For example, the response rate of chemotherapy using such single agents as ADR, MMC, cyclophosphamide, ifosfamide, CDDP, CBDCA, and VP-16 was less than about 10%, even if we estimated it to be good. Combination chemotherapy, including agents of the old generation, has demonstrated similar or extremely few increased levels of response rates. In contrast, in the recent series using the new agents, such as GEM and VNR, the response rate has been reported to be more than 20% in a single regimen and more than 30% in a combined CDDP-based regimen.¹⁻⁴⁾

Thus to develop a more effective modality, chemotherapy guided by sensitivity and resistance assay may lead to rational treatment decisions for this disease. CD-DST, one of the in vitro-chemosensitivity tests using fresh tissues, has been widely applied in Japan for various neoplasms, and recent clinical and experimental data may support its usefulness in practical medicine.⁵⁻¹³⁾ Notably, this study may be the first to describe the in vitro-sensitivity status of fresh MPM tissues.

In the present study, in vitro-chemosensitivity profiles of conventional agents as well as the new agents were analyzed. It is interesting that the former agents, such as ADR, VP-16, and 5-FU, showed a low in vitro-sensitivity rate, ADR as 7%, VP-16 as 15%, and 5-FU as 0%, whereas the latter agents, such as GEM, VNR, SN-38, and TXT, showed slightly high in vitro-sensitivity rate, 45%, 25%, 40%, and 44%, respectively. These rates were almost compatible with those previously reported in the clinical course.¹⁻⁴⁾ Especially, GEM is widely considered to be a most promising chemotherapeutic agent against MPM in a single regimen as well as in combination with CDDP.¹⁵⁾ In fact, according to our data of GEM the sensitivity rate was also the highest (45%).

It is interesting that in comparison to the in vitro-sensitivity rate of the representative chemoagents to nonsmall cell lung cancer (NSCLC) in previous reports,^{9,25)} the present data of some agents to MPM were generally unfavorable for chemotherapy. For example, the in vitro-sensitivity rate of 5-FU to MPM was 0%, but to NSCLC it was 39%. Also, ADR was only 15% versus 38% to NSCLC. Thus in chemotherapy using single or combined agents of the old generation, our data support the clinically lower responder rate to this disease than that to NSCLC.

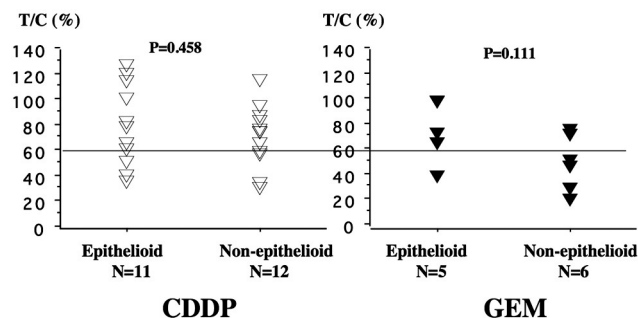


Fig. 2. Distribution of CD-DST T/C ratio of CDDP and GEM according to histological type.

The distribution of CD-DST T/C ratio of CDDP and GEM was not different between the epithelioid type and the nonepithelioid type of MPM.

T/C, total colony of the treated cells / total colony of the untreated cells; CDDP, cisplatin; GEM, gemcitabine; CD-DST, collagen gel droplet embedded culture drug sensitivity test; MPM, malignant pleural mesothelioma.

Ceresoli et al.²⁶⁾ described that the favorable effect of the treatment modality (mainly chemotherapy) for MPM patients was confirmed in the subset of patients with epithelial histology, whereas therapy had no impact on survival in patients with sarcomatoid MPM. Thus it seems that the epithelioid type of MPM has been regarded as being more chemosensitive than the sarcomatous type. In the present study, however, such a trend was not demonstrated (Fig. 2). Also, there was no association between in vitro sensitivity and the clinicopathological parameters examined in the study (data not shown). Since the number of tested samples was relatively small, such associations remain rather undetermined.

In the present study, in vitro-chemosensitivity data were clinically applied in three patterns: the first was used for chemotherapy in nonsurgical cases (Table 3-1), the second in postoperative recurrent cases (Table 3-2), and the third in induction therapy cases (Table 3-3). In the first pattern, a CD-DST sample was usually concurrently obtained by a biopsy of intrathoracic or metastatic lesions for diagnosis and/or staging in advanced-stage disease. It is important that the samples in this pattern should be resected sufficiently for CD-DST analysis. In fact, in 1 patient CD-DST failure was a result of small and insufficient material. In the second pattern, the experimental sample was usually obtained by surgery, enough to analyze CD-DST. In contrast, the use of the third pattern of CD-DST in the present series

was fraught with some problems because CD-DST data were of postchemotherapeutic status. It was thought that these CD-DST data were clinically not so informative, but only confirmative for induction therapy. The first time we had scheduled that the biopsy material before induction therapy was used for CD-DST to select in vitro-sensitive chemoagents. Unfortunately, however, no appropriate case was enrolled because a sample collection for diagnosis in most cases had been already performed at another hospital. Further entry of such cases receiving induction therapy based on in vitro chemosensitivity is expected.

It was most important that there was a good association between CD-DST data and the chemotherapeutic response. Of 10 chemotherapies, including at least one CD-DST-sensitive chemoagent, 3 showed clinically PR, 5 showed SD, and 2 showed PD. Of 4 chemotherapies including CD-DST-resistant chemoagents, only 1 showed SD, and 3 showed PD. The sensitivity, specificity, PPV, NPV, and accuracy rate to CD-DST to clinical response (including PR) were 100%, 36%, 30%, 100%, and 50%, respectively, and to disease control (including PR and SD), sensitivity, specificity, PPV, NPV, and accuracy were 88%, 60%, 80%, 75%, and 71%, respectively. It is interesting that PR was at least achieved by chemotherapeutic regimens, including in vitro-sensitive chemoagents, and chemotherapy used in vitro-resistant chemoagents could have little expectation of effect. Thus CD-DST may provide clinically important information for selecting chemotherapy regimens for MPM.

It was noted that the present CD-DST data were principally based on a single-agent assay, because an in vitro-chemosensitive assay for combined chemoagents has been not established technically. In contrast, combination chemotherapy for MPM was often performed in clinical practice. According to the previous report,⁹⁾ there was no such major discrepancy for the assessment of combination chemotherapy based on single-agent assay, but since it is necessary to develop the combination assay of an in vitro-chemosensitivity test in the future, such projects are now aggressively ongoing.

Recently, pemetrexed (Alimta; Eli Lilly and Company, Indianapolis, IN),²⁷⁾ an antifolate agent, has been introduced with the hopes of it being a treatment for MPM. In particular, its combination with CDDP was shown to improve the median survival of MPM patients compared to CDDP single regimen by phase III study.²⁸⁾ The spread of this novel antimetabolic agent is definitely promising also in Japan. Therefore it should be

incorporated into the CD-DST system, if possible.

In vitro-chemosensitivity tests have been widely tried for clinical applications in various human malignancies.^{5–13)} Especially in the field of thoracic oncology, several preliminary reports have shown its hopeful usefulness clinically as well as experimentally, though almost all of these studies were retrospectively analyzed.^{9,13,25,29,30)} Therefore clinical application of such in vitro-chemosensitivity tests should be more aggressively promoted to select a better chemotherapy responder for MPM patients. Namely, individualized or order-made therapy based on the data of such in vitro-chemosensitivity tests may be proposed more even in the treatment of MPM in the future. Our results strongly support the possibility of clinical application.

In conclusion, CD-DST data obtained by biopsy or surgical-fresh specimens of MPM marginally correlated to the disease control effects of chemotherapy for it. To select the chemotherapy regimen and to expect a better response from each patient with MPM, the clinical application of this in vitro-sensitivity test should be prospectively driven under clinical study using more MPM patients.

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