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Plasma D-Dimer Level in Patients with Colorectal Cancer: Its Role as a Tumor Marker

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Abstract: The purpose of this study was to explore the relationship between the preoperative plasma D-dimer (DD) levels and the tumor pathology of colorectal cancer. The plasma DD levels were measured preoperatively in 108 patients with colorectal cancer, and then were correlated with the tumor pathology and stage. The diagnostic value of the DD levels for the tumor stage was then compared with that of the preoperative carcinoembryonic antigen (CEA) levels. The preoperative DD levels were higher in patients with either a large-sized tumor or a tumor showing deep wall penetration. Lymph-node metastasis, lymphatic invasion, hepatic metastasis, and peritoneal dissemination were all associated with higher DD levels. A stepwise increase in the median DD level was found with the tumor stage. The preoperative DD levels also significantly correlated with CEA levels. When a cutoff value of 0.6 μg/ml was used in the DD assay, the sensitivity and specificity for Dukes C or D cancer were 67.2% and 64.0%, and those for Dukes D cancer were 91.3% and 57.6%, respectively. Although the DD assay was less specific, its diagnostic value in the preoperative staging of colorectal cancer was comparable to that of the CEA assay. The measurement of the preoperative DD level is thus considered to be useful for the preoperative staging of colorectal cancer.

Key Words: D-dimer, colorectal cancer, carcinoembryonic antigen

Introdution

Malignancy is frequently accompanied by the systemic activation of hemostasis and fibrinolysis.[1,2] Thromboembolic diseases such as arterial and venous thrombosis, migratory thrombophlebitis, and pulmonary embolism are important complications of cancer. In addition, intravascular coagulation with fibrinolysis also commonly occurs in cancer patients, and often leads to a subclinical state of disseminated intravascular coagulation.

Evidence of the tumor-associated activation of both coagulation and fibrinolysis can be provided by the measurement of such fibrin split products as D-dimer (DD) in the patient's plasma. DD is a stable end-product of cross-linked fibrin degradation by plasmin. A recent hemoagglutination assay has made the measurement of the plasma DD level routinely available in clinical laboratories.[3]

The elevation of the plasma DD level has been reported to be associated with ovarian malignancy,[4,5] cervical cancer of the uterus,[4,6] lung cancer,[7,8] and prostate cancer.[9] Regarding colorectal cancer, Edwards et al.[10] reported that the preoperative plasma DD level was elevated in patients with Dukes C cancer. However, they studied only a small number of patients, and no study has yet been conducted on a large number of patients.

We measured the preoperative plasma DD level in patients undergoing laparotomy for colorectal cancer, and explored the relationship between the preoperative DD levels and the pathological findings, including the stage of the tumors. The diagnostic value of the DD levels for the tumor stage was thus compared with that of the plasma carcinoembryonic antigen (CEA) level.

Patients and Methods

This study was done on a total of 108 patients who underwent laparotomy for the purpose of resecting a primary colorectal cancer at the Department of Surgery, Koshigaya Hospital, Dokkyo University School of Medicine, between March 1995 and August 1996. The patient group comprised 76 men and 32 women. The

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mean (range) of the patients' ages was 62.2 (27–86) years. Any patients who had undergone other operations within 3 months, those with liver cirrhosis, those demonstrating hematological disorders, and those associated with either intestinal obstruction or peritonitis were excluded from the study, because their plasma levels of DD were likely to be higher even without colorectal cancer. In patients having multiple primary colorectal cancers, the histologically most advanced lesion was used in the analysis. The location of the tumor was the cecum in 7, the ascending colon in 8, the transverse colon in 11, the descending colon in 6, the sigmoid colon in 32, and the rectum in 44 patients. The sizes of the primary tumors were expressed by their greatest diameter. Tumor pathology, such as differentiation, and the extent of wall penetration, lymph-node metastasis, lymphatic invasion, and venous invasion was microscopically diagnosed from hematoxilin–eosin staining of the resected specimens in 106 patients whose tumor was resected. In two patients whose tumor was unresectable, only the size, the extent of the wall penetration, and lymph-node metastasis were recorded from macroscopic findings at laparotomy. The presence of synchronous hepatic metastasis was diagnosed from the findings at laparotomy, including intraoperative ultrasonography.

Tumor stage was determined according to a modification of Dukes' classification applied in Japan: 18 Dukes A, a cancer with neither penetration through the muscularis nor lymph-node metastasis; 32 Dukes B, a cancer with penetration through the muscularis but without lymph-node metastasis; 35 Dukes C, a cancer with lymph-node metastasis; and 23 Dukes D, a cancer with distant metastasis such as hepatic metastasis and peritoneal dissemination.

For the measurement of plasma DD and CEA levels, peripheral blood was obtained from the cubital vein of each patient within 3 days before operation. The plasma levels of DD were measured using a latex agglutination assay (LPIA-Ace, Diaiatron, Tokyo Japan). Because of the sensitivity of the DD assay, the plasma levels of DD less than 0.06 μg/ml were considered to be 0.06 μg/ml. The plasma levels of CEA were measured with an enzyme immunoassay (Glaozyme, Wako, Osaka, Japan) using the same blood samples.

The Mann-Whitney U-test or Kruskal-Wallis test was used for the comparisons of variables between the patient groups. Spearman's rank correlation analysis was carried out to examine the correlations between the two numerical variables.

Table 1. Comparisons of preoperative plasma D-dimer levels according to tumor stage and pathological findings

	n	Median	IQR	*P*
Stage				
Dukes A	18	0.21	(0.07–0.44)	
Dukes B	32	0.57	(0.25–0.82)	<0.0001
Dukes C	35	0.60	(0.27–1.19)	
Dukes D	23	1.05	(0.78–2.35)	
Site				
colon	64	0.67	(0.29–1.52)	0.2058
rectum	44	0.56	(0.22–0.84)	
Size				
≤20 mm	14	0.14	(0.06–0.32)	0.0006
20 <≤ 50 mm	54	0.62	(0.27–1.05)	
50 mm<	35	0.80	(0.41–1.53)	
Differentiation				
well	79	0.53	(0.22–1.04)	0.1967
moderate	19	0.96	(0.62–1.80)	
mucinous	8	0.62	(0.25–1.67)	
Penetration through the muscularis				
negative	20	0.22	(0.07–0.44)	0.0013
positive	86	0.68	(0.36–1.47)	
Lymph-node metastasis				
negative	55	0.48	(0.19–0.86)	0.0273
positive	53	0.78	(0.40–1.61)	
Lymphatic invasion				
negative	23	0.22	(0.06–0.58)	0.0013
positive	83	0.68	(0.37–1.46)	
Venous invasion				
negative	79	0.56	(0.22–0.98)	0.0402
positive	27	0.86	(0.48–1.80)	
Hepatic metastasis				
negative	17	0.53	(0.21–0.94)	<0.0001
positive	91	1.60	(0.80–2.47)	
Peritoneal dissemination				
negative	99	0.58	(0.23–1.10)	0.060
positive	9	1.05	(0.62–2.26)	

Values show the number of the patients, medians, and interquartile ranges (IQR)

Results

In Fig. 1, the distribution of the plasma DD level is plotted according to the tumor stage. The plasma DD level increased significantly with the advance of the tumor stage (Table 1).

Comparisons of the plasma DD levels according to the site, the size, the differentiation, and the presence or absence of the bowel wall penetration, lymph-node metastasis, lymphatic invasion, venous invasion, hepatic metastasis, and peritoneal dissemination are shown in Table 1.

The plasma DD levels did not significantly differ by either the site or the differentiation of the tumor. However, the larger the tumor, or the more deeply the tumor penetrated the bowel wall, the higher the plasma DD level. The plasma DD levels were also higher in patients demonstrating lymph-node metastasis than in those without lymph-node metastasis. Lymphatic invasion of the tumor was associated with a higher plasma DD level. Plasma DD levels were also higher in the patients

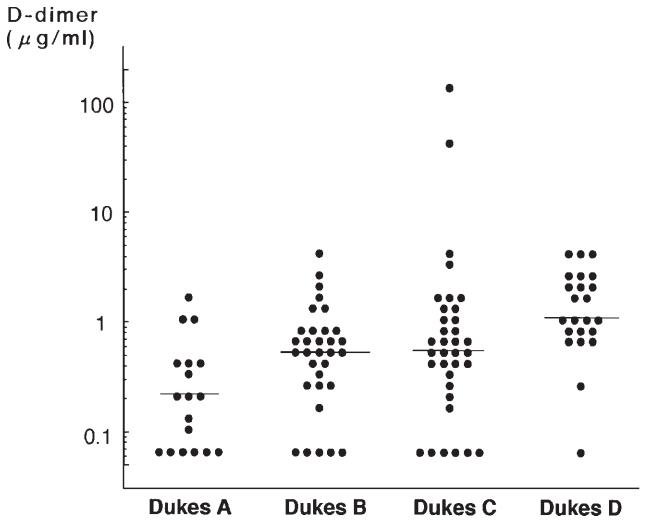

Fig. 1. Preoperative plasma D-dimer levels in patients with colorectal cancer according to Dukes' stages. *Bars* show the medians

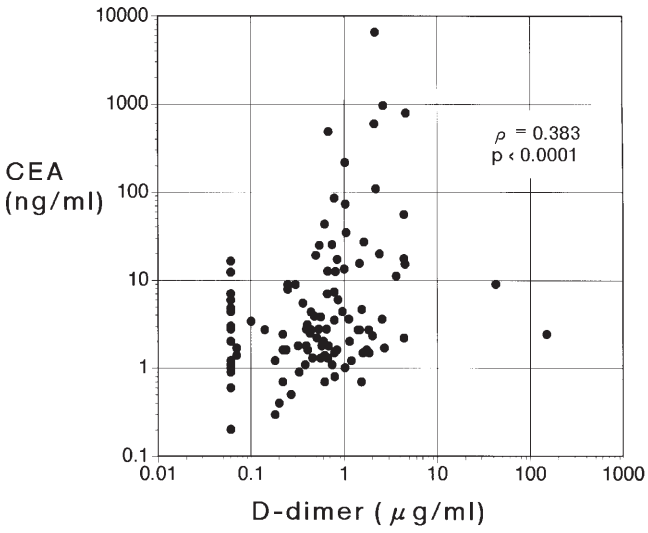

Fig. 2. Correlation between the preoperative plasma D-dimer level and the carcinoembryonic antigen (*CEA*) level in patients with colorectal cancer

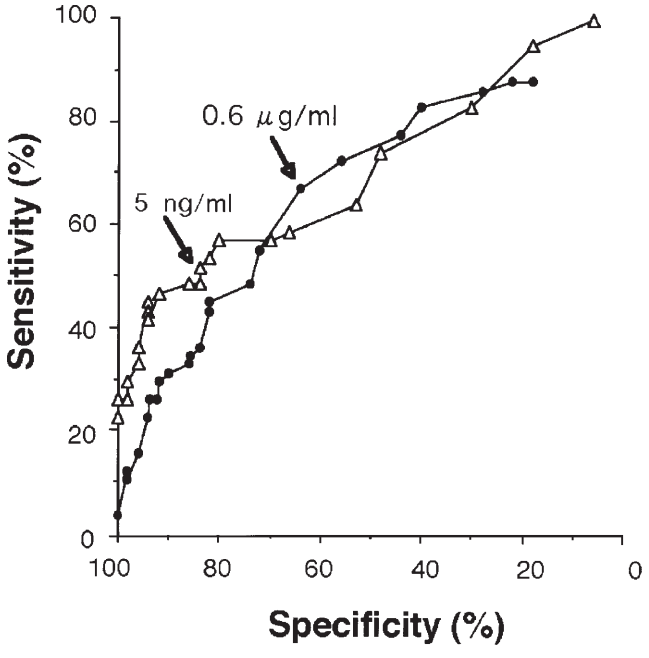

Fig. 3. Receiver operator characteristic curves of the preoperative D-dimer (*circles*) level and the CEA level (*triangles*) for the diagnosis of Dukes C or D colorectal cancer

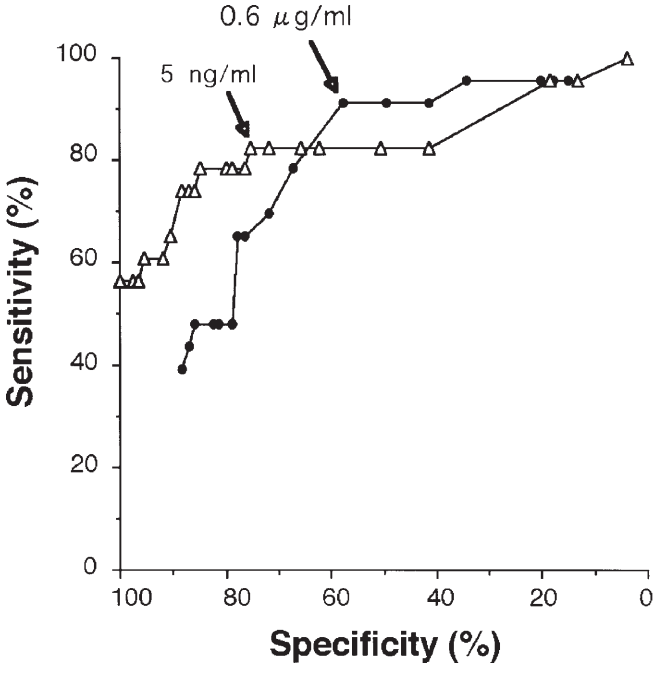

Fig. 4. Receiver operator characteristic curves of the preoperative D-dimer (*circles*) level and the CEA (*triangles*) level for the diagnosis of Dukes D colorectal cancer

with hepatic metastasis than in those not demonstrating hepatic metastasis. The patients with peritoneal dissemination also had higher plasma DD levels than those without peritoneal dissemination (Table 1).

The plasma DD levels correlated significantly with the plasma CEA levels ($\rho = 0.383$, $P < 0.0001$, Fig. 2). Higher plasma DD levels were also associated with higher plasma CEA levels.

The receiver operator characteristic (ROC) curves for the DD assay and the CEA assay in the diagnosis of Dukes C or D cancer, and those in the diagnosis of Dukes D cancer are shown in Figs. 3 and 4, respectively.

Table 2. Sensitivity, specificity, positive predictive value, negative predictive value and diagnostic accuracy of the D-dimer assay, carcinoembryonic antigen (CEA) assay, and combinations of D-dimer assay and CEA assay for Dukes C or D cancer, and those for Dukes D cancer

	Sensitivity	Specificity	PP	NP	Accuracy
Dukes C or D tumors					
D-Dimer alone	67.2	64.0	68.4	62.7	65.7
CEA alone	51.7	84.0	78.9	60.0	66.6
D-Dimer or CEA	77.6	56.0	67.1	68.3	67.6
D-Dimer and CEA	41.3	92.0	85.7	57.5	64.8
Dukes D tumors					
D-Dimer alone	91.3	57.6	36.8	96.1	64.8
CEA alone	78.2	76.5	47.3	92.9	76.8
D-Dimer or CEA	95.7	47.1	32.8	97.6	57.4
D-Dimer and CEA	73.9	87.1	60.7	92.5	84.3

Values show the percentages
PP, positive predictive value; NP, negative predictive value

The ROC curve is a diagram with the sensitivity (%) plotted on the vertical axis with the false-positive rate [100 − specificity (%)] plotted on the horizontal axis. The possible combinations of sensitivity and specificity are found by varying the cutoff value, and they are then plotted as a curve. Since the top left corner shows the ideal combination of 100% sensitivity and 100% specificity, a cutoff value which gives the point closest to the top left corner on the ROC curve is considered to be the most useful in the assay. In addition, in a comparison of the two assays, the assay with the ROC curve furthest into the top left corner is thus suggested to be better than the other.

The cutoff values of the DD assay was determined to be 0.6 μg/ml from the ROC curve. The ROC curves of the CEA assay also showed the generally accepted cutoff value of 5 ng/ml to be a reasonable one. A comparison of the ROC curves between the DD assay and the CEA assay showed that the two assays were almost equal for Dukes C or D tumors, whereas the DD assay was more sensitive but less specific than the CEA assay for Dukes D tumors. Sensitivity, specificity, positive predictive value, negative predictive value, and diagnostic accuracy of the DD assay (cutoff 0.6 μg/ml), the CEA assay (cutoff 5 ng/ml), and combined assays of DD and CEA assay (either DD elevation or CEA elevation, and both DD elevation and CEA elevation) for Dukes C or D tumor and those for Dukes D tumor are shown in Table 2.

Discussion

The activation of the hemostasis system has been detected in various malignancies, including gastrointestinal malignancies.[1,2] DD is a stable end-product of fibrin degradation, and the plasma level of DD can be used as a marker of fibrin formation and degradation.[3]

Edwards et al.[10] found higher plasma DD levels in the patients with Dukes C colorectal cancer than in those with Dukes A,B cancer. They thus suggested that the plasma DD level might be a useful tumor marker of colorectal cancer.[10] However, the number of patients in their study was small.

We studied 108 patients with primary colorectal cancer, and the plasma DD level was thus found to correlate with the tumor pathology and stage. Our study confirmed that the plasma DD levels also correlated significantly with the pathological findings and the stage of the tumor. The plasma DD levels were higher in the patients having either a large-size tumor or one with a deep penetration. A higher plasma DD level was associated with lymphatic invasion and distant metastasis. Consequently, patients with an advanced-stage tumor had higher preoperative DD levels.

The mechanism of the hypercoagulability in patients with colorectal cancer has yet to be fully clarified. Wojtukiewicz et al.[11] used immunohistochemical techniques with fresh frozen specimens of colon cancer to study the pathways of thrombin formation and fibrinolysis. Because they could not identify some coagulation factors such as factor VII, factor X, or fibrin in tumor cells, they thus concluded that coagulation activation in colon cancer may be triggered by a soluble product that exerts its effect at sites distant from the tumor. Pineo et al.[12] demonstrated that mucin entering into the circulation caused hypercoagulability, although it remained speculative as to how such mucin gained access to the circulation. Dover et al.[13] found significantly more factor X-activating activity in colorectal

carcinoma tissue than in the adjacent nonmalignant mucosa.

Regarding the fibrinolytic activity, urokinase-type plasminogen activator (uPA) has been considered an important factor. uPA is the main plasminogen activator extracted from colorectal cancer tissue.[14] In addition, uPA has been detected in colorectal cancer tissue using an immunofluorescence study[15] or immunoperoxidase staining.[16] Furthermore, a study using a short-term organ culture showed that colorectal cancer secreted uPA.[17] The plasma level of uPA was also found to be elevated in patients with colorectal cancer.[18]

We thus speculate that the plasma DD levels are higher in patients with a more advanced-stage tumor because: firstly, larger tumors, which are usually more advanced, are likely to produce a greater quantity of substances activating coagulation and fibrinolysis; secondly, such substances thus gain more access to the circulation in tumors having lymphatic and venous invasion; thirdly, tumors with a higher fibrinolytic activity are likely to be more aggressive, because plasmin is known to activate collagenase IV which destroys the basement membrane in vitro;[19] and lastly, a large quantity of DD might thus be produced at the site of metastases. However, to confirm the above speculations, further biochemical and histopathological studies are required.

Preoperative diagnosis of the tumor stage is important in the selection of the surgical procedure. For Dukes A or B tumors, a local excision of the primary lesions might be theoretically curative, because the lesions do not have lymph-node metastasis. In contrast, for Dukes C tumors, an extended lymphadenectomy combined with a resection of the primary lesion is usually required. Moreover, for Dukes D tumors, treatment of both the primary and metastatic lesions is necessary. We thus explored the preoperative diagnostic value of the plasma DD level for Dukes C or D tumors and that for Dukes D tumors. In addition, we also compared the diagnostic value of the DD assay with that of the CEA assay.

The plasma CEA level has been the most commonly used tumor marker for colorectal cancer. Previous studies[20–22] have shown higher preoperative plasma CEA levels in patients having more advanced-stage tumors. However, because of the broad overlap in the ranges of values between the stages, the significance of the plasma CEA level in the preoperative diagnosis of tumor stage is thought to be limited.

The current study showed a significant correlation between the levels of preoperative DD and CEA. Broad areas of overlap between stages were also found in the plasma DD levels. The ROC curves show that the diagnostic value of the plasma DD level for tumor stage seems almost equal to that of the plasma CEA level. When the cutoff values of 0.6 μg/ml in the DD assay and 5 ng/ml in the CEA assay were applied, the DD assay was more sensitive but less specific than the CEA assay for Dukes C or D tumor. The diagnostic accuracy of the DD assay was 65.7%, while that of the CEA assay was 66.6%. Therefore, the DD assay or the CEA assay alone, or combinations of these two assays, may not be very useful to preoperatively determine whether the tumor stage is Dukes C or D.

For Dukes D tumor, however, the sensitivity and negative predictive values of the DD assay were 91.3% and 96.1%, respectively. In addition, the negative predictive value of a combination of the DD assay and the CEA assay (either DD elevation or CEA elevation) was 97.6%. Only 1 of 41 patients whose DD and CEA were both preoperatively less than the cutoff values had distant metastasis. Moreover, the positive predictive value of the other combination assay (both DD elevation and CEA elevation) was 60.7%. Seventeen of 28 patients, whose DD and CEA levels both were preoperatively elevated, also had distant metastasis. These results thus suggest that a combination of the DD assay and the CEA assay is useful in preoperatively identifying patients who are likely to have Dukes D tumors. In particular, if a patient preoperatively has elevated levels of both DD and CEA, detailed examinations for distant metastases are recommended.

In the current study, the prognostic significance of the preoperative plasma DD levels was not examined because of a short followup period. Studies on the CEA levels have shown that a higher preoperative CEA level is associated with a poor prognosis after a curative resection for Dukes B or C tumors,[20] or for Dukes C tumors.[21,22] The preoperative plasma DD levels might also have such prognostic significance, and this question needs to be clarified in a future study.

In conclusion, the current study demonstrated that the plasma DD level correlates with pathological findings and stages of colorectal cancer. Due to the broad areas of overlap in the ranges of values between stages, it is not possible to preoperatively diagnose the tumor stage based on the DD assay alone. Nevertheless, the DD assay combined with the CEA assay seems to be useful in identifying patients who are likely to have a Dukes D tumor.

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