correlations between adiponectin/leptin and BMI were weaker in the SCCE patients than in the controls in agreement with the above-mentioned previous reports and with our previous report for gastric cancer (Nakajima et al. 2009).

Resistin is expressed mainly in the bone marrow, peripheral mononuclear cells, lungs, placental tissue and pancreatic β cells; however, its biological effects have remained unclear to date. Three case–control studies for the risk of MDS, multiple myeloma or colorectal cancer have been reported (Dalamaga et al. 2008, 2009; Kumor et al. 2009). Dalamaga et al. demonstrated the decreased level of resistin in MDS patients, and speculated that it was due to a compensatory response to the upregulation of other inflammatory factors etiologically linked to myelodysplasia. They also reported the decreased level of resistin in patients with multiple myeloma. Kumor et al. reported a decreased resistin level in colorectal cancer patients. Comparatively, we previously reported that the elevated resistin level may be a biomarker of gastric cancer (Nakajima et al. 2009). Here, resistin in SCCE patients was significantly higher than those in controls, and the correlation between BMI and resistin was demonstrated weakly only in the controls. It implicates that resistin may be a biomarker of SCCE and it also gradually increased with tumor stage progression. This may imply that resistin is a biomarker for the progression of SCCE, rather than a risk factor in SCCE carcinogenesis.

Visfatin is also a new adipocytokine found to be identical to a previously known pre-B cell colony enhancing factor expressed by lymphocytes. The clinical correlations of visfatin with cancer have been rarely reported. In the present study, visfatin level showed no difference between SCCE patients and controls. Previous studies have investigated the correlation between the risk of cancer and obesity in reference to insulin resistance involving insulin, C-peptide, IGFBP and IGF (Giovannucci 2001). Insulin resistance has also been investigated in terms of its correlation with cancer-induced cachexia (Kerem et al. 2008). In this study, C-peptide level showed no difference between SCCE patients and controls. Moreover, its correlation with BMI was demonstrated weakly only in the controls.

The results of the present study suggest that resistin may be a biomarker of SCCE, as well as of stage progression of SCCE. In addition, the impaired responses to body weight loss of adiponectin and leptin in SCCE were suggested. A prospective cohort study is needed to completely elucidate the importance of adipocytokines as biomarkers of SCCE and the causative association between cancer-induced cachexia and the changes in adipocytokines levels. Furthermore, an in vitro study will also be necessary to evaluate the direct function of these adipocytokines on cancer cells.

Acknowledgment This work was supported by the Ministry of Health, Labor and Welfare of Japan.

References

Arita Y, Kihara S, Ouchi N, Takahashi M, Maeda K, Miyagawa J, Hotta K, Shimomura I, Nakamura T, Miyaoka K, Kuriyama H, Nishida M, Yamashita S, Okubo K, Matsubara K, Muraguchi M, Ohmoto Y, Funahashi T, Matsuzawa Y (1999) Paradoxical decrease of an adipose-specific protein, adiponectin, in obesity. Biochem Biophys Res Commun 257:79–83

Calle EE, Thun MJ (2004) Obesity and cancer. Oncogene 23:6365–6378

Chia VM, Newcomb PA, Lampe JW, White E, Mandelson MT, McTiernan A, Potter JD (2007) Leptin concentrations, leptin receptor polymorphisms, and colorectal adenoma risk. Cancer Epidemiol Biomarkers Prev 16:2697–2703

Dalamaga M, Karmaniolas K, Nikolaidou A, Chamberland J, Hsi A, Dionyssiou-Asteriou A, Mantzoros CS (2008) Adiponectin and resistin are associated with risk for myelodysplastic syndrome, independently from the insulin-like growth factor-I (IGF-I) system. Eur J Cancer 44:1744–1753

Dalamaga M, Karmaniolas K, Panagiotou A, Hsi A, Chamberland J, Dimas C, Lekka A, Mantzoros CS (2009) Low circulating adiponectin and resistin, but not leptin, levels are associated with multiple myeloma risk: a case–control study. Cancer Causes Control 20:193–199

Fantuzzi G (2005) Adipose tissue, adipokines, and inflammation. J Allergy Clin Immunol 115:911–919 quiz 920

Giovannucci E (2001) Insulin, insulin-like growth factors and colon cancer: a review of the evidence. J Nutr 131:3109S–3120S

Ishikawa M, Kitayama J, Kazama S, Hiramatsu T, Hatano K, Nagawa H (2005) Plasma adiponectin and gastric cancer. Clin Cancer Res 11:466–472

Jamieson NB, Brown DJ, Michael Wallace A, McMillan DC (2004) Adiponectin and the systemic inflammatory response in weight-losing patients with non-small cell lung cancer. Cytokine 27:90–92

Kerem M, Ferahkose Z, Yilmaz UT, Pasaoglu H, Ofluoglu E, Bedirli A, Salman B, Sahin TT, Akin M (2008) Adipokines and ghrelin in gastric cancer cachexia. World J Gastroenterol 14:3633–3641

Kumada M, Kihara S, Ouchi N, Kobayashi H, Okamoto Y, Ohashi K, Maeda K, Nagaretani H, Kishida K, Maeda N, Nagasawa A, Funahashi T, Matsuzawa Y (2004) Adiponectin specifically increased tissue inhibitor of metalloproteinase-1 through interleukin-10 expression in human macrophages. Circulation 109:2046–2049

Kumor A, Daniel P, Pietruczuk M, Malecka-Panas E (2009) Serum leptin, adiponectin, and resistin concentration in colorectal adenoma and carcinoma (CC) patients. Int J Colorectal Dis 24:275–281

Nakajima TE, Yamada Y, Hamano T, Furuta K, Gotoda T, Katai H, Kato K, Hamaguchi T, Shimada Y (2009) Adipocytokine levels in gastric cancer patients: resistin and visfatin as biomarkers of gastric cancer. J Gastroenterol 44:685–690

Pannacciulli N, Vettor R, Milan G, Granzotto M, Catucci A, Federspil G, De Giacomo P, Giorgino R, De Pergola G (2003) Anorexia nervosa is characterized by increased adiponectin plasma levels and reduced nonoxidative glucose metabolism. J Clin Endocrinol Metab 88:1748–1752

Rubenstein JH, Dahlkemper A, Kao JY, Zhang M, Morgenstern H, McMahon L, Inadomi JM (2008) A pilot study of the association of low plasma adiponectin and Barrett's esophagus. Am J Gastroenterol 103:1358–1364

Stattin P, Lukanova A, Biessy C, Soderberg S, Palmqvist R, Kaaks R, Olsson T, Jellum E (2004) Obesity and colon cancer: does leptin provide a link? Int J Cancer 109:149–152

Wang Y, Lam KS, Xu JY, Lu G, Xu LY, Cooper GJ, Xu A (2005) Adiponectin inhibits cell proliferation by interacting with several growth factors in an oligomerization-dependent manner. J Biol Chem 280:18341–18347

Wei EK, Giovannucci E, Fuchs CS, Willett WC, Mantzoros CS (2005) Low plasma adiponectin levels and risk of colorectal cancer in men: a prospective study. J Natl Cancer Inst 97:1688–1694

Wolf I, Sadetzki S, Kanety H, Kundel Y, Pariente C, Epstein N, Oberman B, Catane R, Kaufman B, Shimon I (2006) Adiponectin, ghrelin, and leptin in cancer cachexia in breast and colon cancer patients. Cancer 106:966–973

Wolfe BE, Jimerson DC, Orlova C, Mantzoros CS (2004) Effect of dieting on plasma leptin, soluble leptin receptor, adiponectin and resistin levels in healthy volunteers. Clin Endocrinol (Oxf) 61:332–338

Wolk A, Gridley G, Svensson M, Nyren O, McLaughlin JK, Fraumeni JF, Adam HO (2001) A prospective study of obesity and cancer risk (Sweden). Cancer Causes Control 12:13–21

Yang WS, Lee WJ, Funahashi T, Tanaka S, Matsuzawa Y, Chao CL, Chen CL, Tai TY, Chuang LM (2001) Weight reduction increases plasma levels of an adipose-derived anti-inflammatory protein, adiponectin. J Clin Endocrinol Metab 86:3815–3819

Yokota T, Oritani K, Takahashi I, Ishikawa J, Matsuyama A, Ouchi N, Kihara S, Funahashi T, Tenner AJ, Tomiyama Y, Matsuzawa Y (2000) Adiponectin, a new member of the family of soluble defense collagens, negatively regulates the growth of myelomonocytic progenitors and the functions of macrophages. Blood 96:1723–1732

JJCO Japanese Journal of Clinical Oncology

Jpn J Clin Oncol 2010;40(5)404–411
doi:10.1093/jjco/hyp192
Advance Access Publication 4 February 2010

Phase I and Pharmacokinetic Study of ABI-007, Albumin-bound Paclitaxel, Administered Every 3 Weeks in Japanese Patients with Solid Tumors

Kazuhiko Yamada[1], Noboru Yamamoto[1], Yasuhide Yamada[1], Toru Mukohara[2,3], Hironobu Minami[2,3] and Tomohide Tamura[1*]

[1]Department of Internal Medicine, National Cancer Center Hospital, Tokyo and [2]Department of Oncology/ Hematology, National Cancer Center Hospital East, Kashiwa, Japan

[3]Present address: Medical Oncology, Department of Medicine, Kobe University Hospital, Kobe, Japan.

*For reprints and all correspondence: Tomohide Tamura, Department of Internal Medicine, National Cancer Center Hospital, 5-1-1 Tsukiji, Chuo-ku, Tokyo 104-0045, Japan. E-mail: ttamura@ncc.go.jp

Received August 25, 2009; accepted December 20, 2009

Objective: ABI-007 is a novel Cremophor® EL-free nanoparticle albumin-bound paclitaxel. This Phase I study was designed to evaluate tolerability and determine recommended dose for Japanese patients when ABI-007 was administered in every-3-week schedule. Pharmacokinetics of paclitaxel was also assessed.

Methods: Patients with advanced solid tumors refractory to standard therapy received a 30 min intravenous infusion of ABI-007 every 3 weeks without pre-medications at 200, 260 or 300 mg/m^2, respectively. Tolerability and recommended dose were determined by the standard '3 + 3' rule.

Results: No dose-limiting toxicity was observed, despite the dose escalation. In another cohort, 260 mg/m^2 was re-evaluated and resulted in no dose-limiting toxicity. Grade 3 or 4 neutropenia was reported for the majority of patients ($n = 8$) but no incidence of febrile neutropenia. Non-hematological toxicities were generally mild except for Grade 3 sensory neuropathy ($n = 3$). Pharmacokinetic study demonstrated the area under the curve of paclitaxel increased with increasing the dosage, and comparable pharmacokinetic parameters to the western population. Partial response was observed in three non-small cell lung cancer patients. Two of whom had received docetaxel-containing chemotherapy prior to the study.

Conclusions: ABI-007 administered in every-3-week schedule was well tolerated up to 300 mg/m^2, and recommended dose was determined at 260 mg/m^2 in consideration of efficacy, toxicities and similarity of pharmacokinetic profile in western studies. Additional studies of single-agent ABI-007 as well as platinum-based combinations, particularly in patients with non-small cell lung cancer, are warranted.

Key words: nanoparticle albumin-bound paclitaxel – ABI-007 – Phase I – pharmacokinetic – Japanese

INTRODUCTION

ABI-007 (Abraxane®; Abraxis Bioscience, Los Angels, CA, USA) is a novel Cremophor® EL (polyoxyethylated castor oil)-free albumin-bound nanoparticle formulation of paclitaxel. This formulation allows for a higher paclitaxel concentration in the suspension, serving to reduce the administration volume and time. No pre-medication to prevent the Cremophor® EL-induced hypersensitivity reaction is needed. In addition, non-polyvinyl chloride infusion system and in-line filtration are not necessarily applied given no leaching of plasticizers (1,2).

In the Phase I study of every-3-week (Q3W) schedule conducted in the USA, the dose of ABI-007 was escalated from 135 to 375 mg/m^2, and maximum tolerated dose (MTD) and recommended dose (RD) were established at 300 mg/m^2. It was exceedingly higher than that of solvent-based paclitaxel (Taxol®; Bristol–Myers Squibb, Princeton, NJ, USA), 175 mg/m^2 (1). Dose-limiting toxicities (DLTs) were keratitis, blurred vision, sensory neuropathy, stomatitis and neutropenia. Maximum concentration (C_{max}) and the area under the curve from time zero to infinity (AUC_{inf}) of paclitaxel increased linearly over the ABI-007 dose range of 135–300 mg/m^2 administered over 30 min. Volume of distribution of ABI-007 is characterized by the larger distribution than solvent-based paclitaxel, indicating extensive extravascular distribution of the drug (3). C_{max} and AUC_{inf} values for individual patients correlated well with toxicities.

In the Phase III pivotal study of 454 patients with metastatic breast cancer, Q3W schedule of ABI-007 260 mg/m^2 produced the superior outcome to the same schedule of solvent-based paclitaxel, 175 mg/m^2: significantly higher response rate and prolonged time to progression [33% vs. 19% ($P < 0.001$) and 23.0 vs. 16.9 weeks ($P = 0.006$), respectively] and significantly lower incidence of Grade 4 neutropenia, despite a 49% higher paclitaxel dose [9% vs. 22% ($P < 0.001$)] (4). The dosage and schedule used in this Phase III study lead to the approved labeling worldwide.

According to the clinical utility and study data reported overseas, ABI-007 seems to be an effective treatment. This Phase I study aimed to evaluate tolerability, DLT and RD in Japanese patients with solid tumors when administered in Q3W schedule. Efficacy, toxicity and pharmacokinetics (PK) were also evaluated as secondary objectives, followed by the evaluation on ethnic difference in PK.

PATIENTS AND METHODS

Patient Eligibility

Patients aged 20–74 years with histologically or cytologically diagnosed malignant solid tumors refractory to standard therapies or for which there was no effective treatment were eligible. They had to have an Eastern Cooperative Oncology Group (ECOG) performance status (PS) of 0–2, and a life expectancy of ≥60 days. Eligibility criteria also included adequate renal, liver and bone marrow function, defined as serum creatinine (Cr) ≤1.5 mg/dl, serum total bilirubin (TB) ≤1.5 mg/dl, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) <100 IU/l, respectively, serum albumin ≥3.0 g/dl, white blood cell count ≤12 000/mm^3, absolute neutrophil count ≥2000/mm^3, platelets ≥100 000/mm^3 and hemoglobin ≥9.0 g/dl. Patients with prior exposure to taxanes were eligible for the study. Key exclusion criteria included the following: (i) surgery within 4 weeks; (ii) chemotherapy within 3 weeks; (iii) radiotherapy within 3 weeks; (iv) history of radiation to more than 30% of hematopoietic marrow; (v) pre-existing sensory neuropathy ≥Grade 2; (vi) pleural effusion and ascites that required drainage; (vii) brain metastasis showing symptoms or requiring treatment; (viii) hepatitis B or C virus or human immunodeficiency virus infection; (ix) chronic steroid treatment; (x) pregnancy, lactation, suspicion of being pregnant; (xi) serious pre-existing medical conditions such as uncontrolled infections, pulmonary fibrosis, diabetes, severe heart disease and psychogenic disorders.

This study was approved by the Institutional Review Board at the National Cancer Center and conducted according to Japanese Good Clinical Practice guidelines. All patients provided written informed consent prior to study entry.

Study Design and Treatment

This Phase I, open label, dose-finding study was conducted at National Cancer Center and National Cancer Center East.

ABI-007 was supplied by TAIHO Pharmaceutical Co., Ltd, Tokyo Japan. Each vial contained 100 mg of paclitaxel and ~900 mg of frozen-dried Albumin Human (United States Pharmacopeia). The prescribed dose of ABI-007 was prepared in 5 mg (paclitaxel)/ml of physiological saline as a suspension. The drug was administered via 30 min i.v. without pre-medication and in-line filtration.

Evaluated dose levels were 200, 260 or 300 mg/m^2, as shown in Table 1, repeated every 3 weeks. The rationale for selected dose range was the following: the upper level, 300 mg/m^2—MTD determined in a US Phase I study; the middle level, 260 mg/m^2—the approved dose in the US/EU, and the lower level, 200 mg/m^2—one dose level below MTD examined in the foregoing Phase I study. The dose range also factored in PK: linear PK of ABI-007 over the dose range 80–300 mg/m^2 and the same level and activity of CYP2C8 and CYP3A4 between Japanese and Caucasians (5). Dose escalation was capped at 300 mg/m^2. In the event that MTD exceeded the cap, further steps in investigation would be discussed among study sponsor, principal investigator and medical experts.

The dose escalation followed the standard '3 + 3' rule. Three patients were evaluated at the first dose level, and in the absence of DLTs, three additional patients were entered at the next dose level. If one of the three patients encountered a DLT, another cohort was to be added at the same dose level. The MTD was defined as the dose level at which two out of three to six patients experienced DLT. The RD

Table 1. Dose levels

Level	Dose (mg/m^2)	No. of patients entered	No. of courses
1	200	3	9
2	260	6	23
3	300	3	14

was defined as the dose level that is one level below MTD, and consequently, a total of six patients were to be treated at RD to further evaluate the safety profile.

DLTs were pre-defined as any of the following drug-related toxicities that had occurred during the first course: (i) Grade 4 thrombocytopenia; (ii) Grade 3 thrombocytopenia requiring platelet transfusion; (iii) Grade 4 neutropenia over 4 days; (iv) Grade 3 or 4 febrile neutropenia; and (v) Grade 3 or 4 non-hematologic toxicity. Dose was reduced by one level when DLT occurred in the first course, and reduction was allowed when the toxicities corresponding to DLT or Grade 2 neuropathy occurred in the second course or later.

Patient Evaluation

Pre-treatment evaluation included a complete history and physical examination, a complete blood count with differential, serum chemistry profile, urinalysis including pregnancy test, chest X-ray and electrocardiogram. Serum chemistry profile included electrolytes, Cr, urea nitrogen, TB, AST, ALT, lactic dehydrogenase, alkaline phosphatase, total protein, albumin and C-reactive protein. Baseline imaging studies and serum tumor marker levels were obtained at the discretion of treating physician. Toxicity assessment, physical examination and all blood tests except serum tumor markers were repeated on a weekly basis.

Toxicities were graded according to Common Terminology Criteria for Adverse Events (CTCAE), version 3.0. Patients were considered evaluable for toxicity if they received at least one dose of the study drug. Objective response to therapy was assessed every 4–6 weeks according to Response Evaluation Criteria in Solid Tumors (RECIST), version 1.0 (6).

Blood Sampling and PK Analysis

Whole blood samples of 7 ml each were collected in 6 ml of heparinized tube and 1 ml of K3-EDTA tube to determine the PK of ABI-007 at time points: 0, 0.25, 0.5 (end of infusion), 0.75, 1, 1.5, 2, 4, 10, 24, 48 and 72 h. Heparinized samples were immediately centrifuged at 1000 *g* for 15 min in 4°C and resultant plasma was stored in aliquot, whereas K3-EDTA samples were softly mixed in normal temperature. These samples were stored at less than or equal to −20°C until analyzed. The sample was analyzed for paclitaxel using liquid chromatography/tandem mass spectrometry in Alta Analytical Laboratory (El Dorado Hills, CA, USA). The limit of quantification for paclitaxel in plasma and whole blood was 1.00 and 5.00 ng/ml, respectively, and the range of reliable response in these samples was 1.00–500 and 5.00–5000 ng/ml, respectively.

PK parameters were determined from each patient's whole blood/plasma paclitaxel concentration profile. They were evaluated by non-compartmental analysis using the WinNonlin software package (Ver4.1, Pharsight Corp., CA, USA). The C_{max} of paclitaxel was obtained directly from experimental data. The elimination constant (λz) was obtained by log-linear regression analysis of the terminal phase of the whole blood/plasma concentration vs. time profile. The elimination half-life ($t_{1/2}$) was determined by taking the ratio of natural log of 2 and λz. The AUC_{inf} was estimated by summing the areas from time zero to the last measured concentration–time point (AUC_{0-t}), calculated using the linear-logarithmic trapezoidal method, and the extrapolated area. The dose–area relationship (i.e. total ABI-007 dose divided by AUC_{inf}) was used to determine total body clearance (CL). The volume of distribution (Vz) was determined by taking the ratio between CL and λz.

Table 2. Patient characteristics

Characteristics	No. of patients
Total no. of patients	12
Male/female	10/2
Age (years)	
Median	61
Range	45–69
ECOG performance status	
0	3
1	9
Tumor type	
NSCLC	6
Parotid gland	1
Ovary	1
Bladder	1
Pharyngeal and esophageal	1
Colon	1
Thymoma	1
Prior treatment	
Surgery	9
Radiotherapy	3
Chemotherapy	12
No. of prior chemotherapy	
1	1
2	4
≥3	7
Prior taxane therapy	
Yes	
Solvent-based paclitaxel	1
Docetaxel	5
Solvent-based paclitaxel and docetaxel	2
No	4

ECOG, Eastern Cooperative Oncology Group; NSCLC, non-small cell lung cancer.

Table 3. Hematologic toxicities (all courses)

Dose levels	Level 1 (200 mg/m^2)			Level 2 (260 mg/m^2)			Level 3 (300 mg/m^2)			All		
No. of patients (no. of courses)	$n = 3$ (9)			$n = 6$ (23)			$n = 3$ (14)			$n = 12$ (46)		
CTCAE grade	1–2	3	4	1–2	3	4	1–2	3	4	1–2	3	4
Leucopenia	2	0	0	3	2	0	3	0	0	8	2	0
Neutropenia	1	1	0	1	3	1	1	2	0	2	6	2
Anemia	1	0	0	2	0	0	1	0	0	4	0	0
Thrombocytopenia	0	0	0	1	0	0	1	0	0	2	0	0

CTCAE, Common Terminology Criteria for Adverse Events.

Descriptive statistics were used for baseline characteristics, safety assessment, and PK variables. Regression analysis of individual C_{max}, and AUC_{inf} vs. dose was performed to gain an appreciation of PK linearity. The SAS software package (ver8.2, SAS Institute, Inc., NC, USA) was used for statistical analysis.

RESULTS

Patients and Treatment

Between August 2006 and June 2007, 12 patients were enrolled and treated in this study at two participating centers in Japan. Patient characteristics are summarized in Table 2. Most patients were male (83%) with a median age of 61 (range, 45–69) years and all patients were ECOG PS 0–1. The predominant type of tumor was non-small cell lung cancer (NSCLC). Nine patients had surgery for primary tumors, seven had received more than three prior chemotherapy regimens and eight had received prior taxane-containing chemotherapy.

The patients were treated at the following dose levels: 200 mg/m^2 (Level 1, $n = 3$), 260 mg/m^2 (Level 2, $n = 6$) and 300 mg/m^2 (Level 3, $n = 3$). All were evaluable for safety and PK, and 11 for efficacy (one had no adequate measurable lesions for RECIST criteria).

DLT, Tolerability and RD

No DLTs were observed through the dose escalation to the highest Level 3; therefore, the MTD was not reached methodologically. To decide on the potential RD, study sponsor, medical advisor and principal investigators jointly reviewed the reference data in the foreign studies (1,4,7) and favored 260 mg/m^2 from tolerability and safety perspectives, particularly the development of cumulative neurotoxicity. Additional three patients were then accrued to 260 mg/m^2 cohort to repeat the assessment. None of DLTs being experienced by the additional patients, 260 mg mg/m^2, was established as RD.

Safety

A total of 46 courses of ABI-007 was administered, and the median number of courses administered per patient was 3 (range, 1–11). No acute hypersensitivity reactions were observed during the infusion period. The most common toxicities were neutropenia, leucopenia, lymphopenia, alopecia and sensory neuropathy. The incidences of hematologic toxicities by dose level are shown in Table 3. Grade 3 or 4 neutropenia was often experienced in more than half of patients throughout the study; however, no febrile neutropenia was observed. The median time to onset of Grade 3 or 4 neutropenia was 15.0 (range, 8–34) days, and the median time to recovery to $<$Grade 2 was 6.5 (range, 3–14) days. There were no episodes of $\geq$Grade 2 or greater thrombocytopenia, and anemia was mostly mild. Frequent non-hematological toxicities were sensory neuropathy, alopecia, arthralgia/myalgia and rash (Table 4). The sensory neuropathy was manifested by paresthesia in a symmetric, stocking/glove distribution, and the median time to the first indication or exacerbation from the baseline was 7 days. The severity of non-hematologic toxicities was generally mild except for three cases of Grade 3 sensory neuropathy at Level 2 ($n = 1$) and Level 3 ($n = 2$), which cumulatively exacerbated from Grade 1 observed in the first week of the first course (range, 3–6 days from the administration) to Grade 3 during the third or later course (range, 3–11 courses from the administration). Among the three patients who experienced Grade 3 sensory neuropathy, one patient had received taxane-containing chemotherapy prior to the study. A variety of ocular toxicities including superficial keratopathy reported in the initial Phase I study of USA were not observed in this study. Treatment delay occurred in one patient at each Levels 2 and 3 due to the neurotoxicity, dose reduction occurred in two patients at each Levels 2 and 3 due to the neurotoxicity, and treatment was discontinued in three patients at each Levels 2 and 3, comprising five patients with treatment-related neurotoxicity and one patient with unrelated neutropenia.

Table 4. Non-hematologic toxicities (all courses)

Dose levels	Level 1 (200 mg/m^2)		Level 2 (260 mg/m^2)		Level 3 (300 mg/m^2)		All	
No. of patients (no. of courses)	$n = 3$ (9)		$n = 6$ (23)		$n = 3$ (14)		$n = 12$ (46)	
CTCAE grade	1–2	3	1–2	3	1–2	3	1–2	3
Sensory neuropathy	1	0	5	1	1	2	7	3
Alopecia	3	0	4	0	3	0	10	0
Myalgia	0	0	6	0	3	0	9	0
Rash	2	0	4	0	1	0	7	0
Arthralgia	1	0	4	0	2	0	7	0
Asthenia	2	0	2	0	2	0	6	0
Motor neuropathy	0	0	3	0	2	0	5	0
Nausea	2	0	1	0	1	0	4	0
Anorexia	3	0	1	0	0	0	4	0
Vomiting	1	0	2	0	0	0	3	0
Diarrhea	2	0	0	0	0	0	2	0
Stomatitis	0	0	0	0	2	0	2	0

Grade 4 toxicities were not observed.

Table 5. Anti-tumor response

	Tumor type	Prior taxane therapy	Response
Level 1 (200 mg/m^2)	NSCLC	+	PD
	NSCLC	+	PR
	Parotid gland	+	PD
Level 2 (260 mg/m^2)	NSCLC	+	PD
	NSCLC	−	PR
	Ovary	+	PD
	NSCLC	+	PR
	Colon	−	PD
	Thymoma	−	SD
Level 3 (300 mg/m^2)	Bladder	−	SD
	NSCLC	+	NE
	Pharyngeal and esophageal	+	SD

PD, progressive disease; PR, partial response; SD, stable disease; NE, not evaluable.

Response

Eleven of 12 patients were evaluable for anti-tumor response (Table 5). Partial responses were observed in three NSCLC patients. Of them, two of whom had received docetaxel-containing chemotherapy prior to the study. The first patient, entered at Level 1, had received 6 courses of ABI-007, and the second and third patients, entered at Level 2, 11 and 6 courses, respectively. The both responders in Level 2 attained disease control until the treatment discontinuation due to the sensory neuropathy.

Pharmacokinetics

Blood samples for PK analysis were available from all of 12 patients following the first course of treatment. A semi-log plot of the mean values of paclitaxel concentration for each dose level vs. time is shown in Fig. 1. After 30 min infusion of ABI-007, the concentration of paclitaxel began to decrease immediately upon cessation of the infusion with $t_{1/2}$ of 17.3–27.3 h in the whole blood, which is nearly comparable with that of standard dose of solvent-based paclitaxel (6), and the decline of paclitaxel concentration from maximum was multiphasic.

The mean PK parameters of paclitaxel are summarized in Table 6. C_{max}, AUC_{0-t} and AUC_{inf} of paclitaxel when administered as a 30 min infusion of ABI-007 increased with increasing dosage. CL and Vz of the blood sample showed the small inter-patient variability, and the mean ± SD values (CV%) for CL and Vz at the dose level of 260 mg/m^2 were 18.1 ± 2.33 (12.9 CV%) (l/h/m^2) and 510 ± 96.8 (19.0 CV%) (l/m^2), respectively. These values slightly decreased with increased dosage. It was considered that there was no remarkable difference in calculated values of PK parameters between whole blood and plasma. Regression analysis suggested the dose-proportionality of ABI-007 within the dose range in this study (R^2 of $C_{max} = 0.4470$, R^2 of

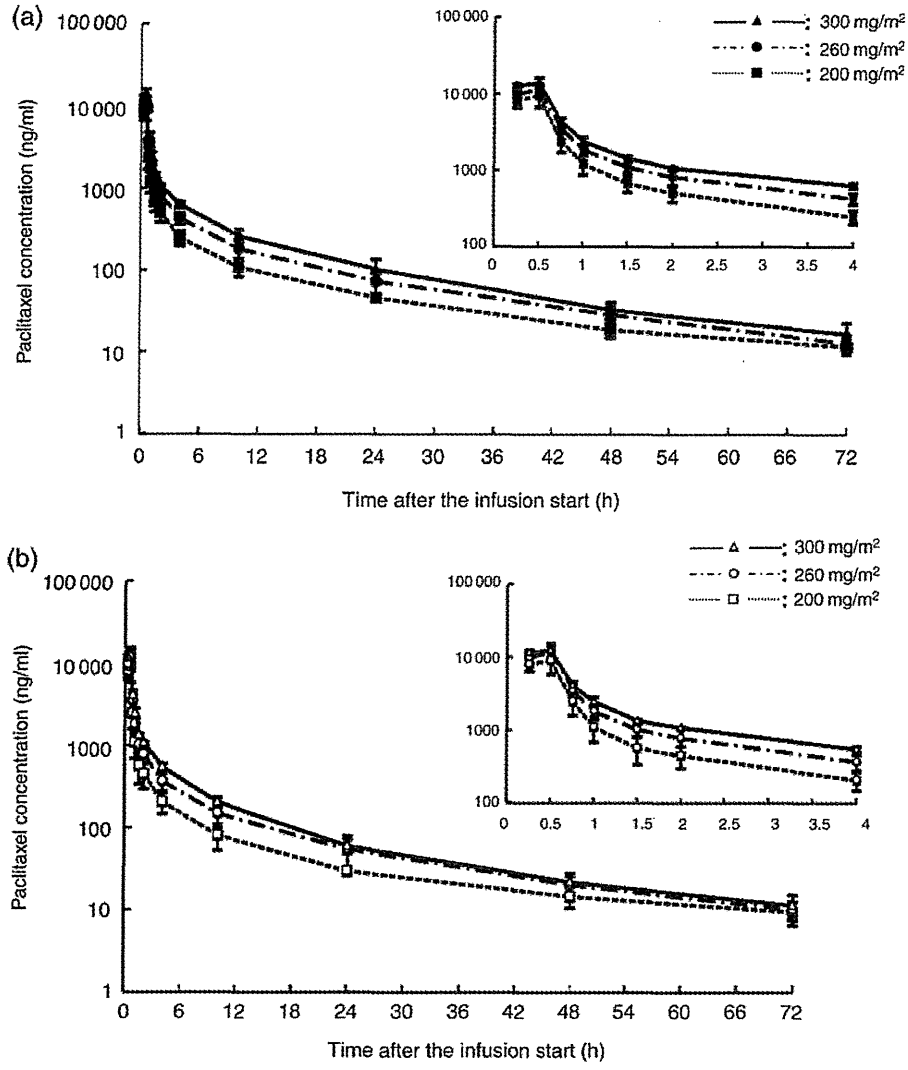

Figure 1. (a) Mean whole blood concentration–time profiles of paclitaxel. (b) Mean plasma concentration–time profiles of paclitaxel.

$AUC_{inf} = 0.7177$); however, it was difficult to establish the linearity due to those narrow dose range and small data size.

DISCUSSION

In the Phase I study where ABI-007 was administered in Q3W schedule in Japanese patients, no DLT occurred at any dose level of 200, 260 and 300 mg/m^2. Because MTD was not reached by the 3 + 3 rule, selection of RD was attributed to the consideration of reasonable tolerability, toxicities and PK profile. Since paclitaxel treatment was characterized for the cumulative neurotoxicity, dose selection also took into account the development of sensory neuropathy throughout the study. Consequently, 260 mg/m^2 was reassessed as potential RD and established as RD in the absence of applicable DLT. Outcome of sensory neuropathy in all treatment courses also provided the justification for the feasibility of 260 mg/m^2 (Table 7). Among 260 and 300 mg/m^2 cohorts, every patient experienced neuropathic events, in which Grade 3 or 4 event was more frequent in 300 mg/m^2 (two out of three patients) than in 260 mg/m^2 cohorts (one out of six patients). Moreover, all the three patients in 300 mg/m^2 cohort discontinued the treatment due to neuropathic events as opposed to two out of six patients in 260 mg/m^2 cohort.

In terms of treatment-related toxicities, Grade 3 or 4 neutropenia was experienced in 15 of 46 treatment courses (32%). Nonetheless, no febrile neutropenia was observed. Median duration of recovery from Grade 3 or 4 to $<$Grade 2 was 6.5 days (range, 3–14). No treatment delay was caused by neutropenia. In addition, platelet decrease $\geq$Grade 2 was not observed throughout the study. On the whole, hematological toxicities were mild. In regard to sensory neuropathy, the median time to the first indication or exacerbation from the baseline was 7 days, which

was relatively early to that of solvent-based paclitaxel. Especially for Grade 3 sensory neuropathy, the indication or exacerbation fell within the first week of the first course, ranging from 3 to 6 days; the time to improve from Grade 3 to Grade 2 or 1 was 21, 26 and 46 days in the respective cases. Although the improvement tended to delay when compared with median 22 days in a previous Phase III study (4), it still remains controversial because of the great difference in the sample sizes between the two studies. Meanwhile, other non-hematological toxicities including mucositis—the DLT of the US Phase I study—were generally mild to moderate up to 300 mg/m^2.

PK profiles of ABI-007 have revealed the small inter-patient variability, and the AUC and C_{max} of paclitaxel increased with increasing the dosage. In whole blood samples, there was a significant correlation between the doses and PK parameters. The linearity was uncertain in the face of wide confidence interval (CI) with small sample size, however, presumable from the other reported data showing the linearity over a wide dose range: 80–300 mg/m^2 (2) and PK equality between Japanese and western population (3).

Anti-tumor response was demonstrated in the patients with NSCLC including the patients who had received prior taxane-containing therapy.

Multiple previous studies of ABI-007 also reported the promising data in the patients with NSCLC. In a Phase II trial, 260 mg/m^2 of ABI-007 was administered alone in the same Q3W schedule as our study in the first-line setting, overall response rate was 16.3% (95% CI, 5.24–27.31%) and the disease control rate was 48.8% (95% CI, 33.90–63.78%) (8). More recently, weekly (QW) schedule of ABI-007 was also reported: 125 mg/m^2 of ABI-007 was administered in monotherapy on days 1, 8 and 15 every 4 weeks, the response rate was 30% ((95% CI, 16–44%) and the disease control rate was 50% (95% CI, 35–66%) (9). Despite the higher incidence of ≥Grade 3 neutropenia and sensory neuropathy relative to the Q3W schedule, QW schedule was well tolerated and active.

In conclusion, no DLT observed at any dose levels, and ABI-007 was well tolerated up to 300 mg/m^2 in Japanese

Table 6. PK parameters of paclitaxel

	200 mg/m^2 ($n = 3$)		260 mg/m^2 ($n = 6$)		300 mg/m^2 ($n = 3$)	
	Mean	CV (%)	Mean	CV (%)	Mean	CV (%)
Whole blood						
C_{max} (ng/ml)	9430	28.3	11 635	13.0	13 833	15.3
AUC_{inf} (ng h/ml)	10 360	22.0	14 593	13.7	19 138	12.2
$t_{1/2}$ (h)	24.3	10.9	19.5	7.9	18.3	1.9
CL (l/h/m^2)	19.9	21.6	18.1	12.9	15.8	11.2
Vz (l/m^2)	689	15.3	510	19.0	417	9.7
Plasma						
C_{max} (ng/ml)	9040	34.0	12 000	17.6	12 700	20.5
AUC_{inf} (ng h/ml)	9146	29.6	13 330	20.7	16 271	11.2
$t_{1/2}$ (h)	29.0	17.7	20.8	19.5	19.8	9.8
CL (l/h/m^2)	23.1	26.4	20.2	21.5	18.6	10.6
Vz (l/m^2)	935	11.7	620	36.9	527	7.0

PK, pharmacokinetic; CV, coefficient of variation; C_{max}, maximum concentration; AUC_{inf}, area under the concentration–time curve up to ∞ hours; $t_{1/2}$, terminal elimination half-life; CL, clearance; Vz, volume of distribution based on terminal phase.

Table 7. Grade change in sensory neuropathy (all courses)

Level	Case	Before administration	Course no.									
			1	2	3	4	5	6	7	8	9	10	11
Level 1	1-2	0	0	0	1	1	1	1[a]	—	—	—	—	—
Level 2	2-1	1	2	—	—	—	—	—	—	—	—	—	—
	2-2	0	1	1	1	1	1	1	1	1	2	2	3[a]
	2-3	0	0	1	—	—	—	—	—	—	—	—	—
Level 3	3-1	0	1	1	2	2	2	3[a]	—	—	—	—	—
	3-2	0	1	1	1	2	2[a]	—	—	—	—	—	—
	3-3	0	2	2	3[a]	—	—	—	—	—	—	—	—
Level 2	2-4	0	1	1	1	2	2	2[a]	—	—	—	—	—
	2-5	0	1	—	—	—	—	—	—	—	—	—	—
	2-6	0	1	1	—	—	—	—	—	—	—	—	—

—, end of study.
[a]Study-off due to sensory neuropathy.

patients. RD in this schedule was determined to be 260 mg/m^2 in consideration of efficacy, toxicities and similarity of PK profile in the western studies. Additional studies of single-agent ABI-007 and platinum-based combinations are warranted.

Acknowledgements

The preliminary results of this study were presented in part at the AACR-NCI-EORTC International Conference, San Francisco, CA, October 2007.

Funding

This trial was funded by Taiho Pharmaceutical (Tokyo, Japan).

Conflict of interest statement

Hironobu Minami and Tomohide Tamura receive remuneration for the lectures from Taiho Pharmaceutical (Tokyo, Japan).

References

1. Ibrahim NK, Desai N, Legha S, Soon-Shiong P, Theriault RL, Rivera E, et al. Phase I and pharmacokinetic study of ABI-007, a Cremophor-free, protein-stabilized, nanoparticle formulation of paclitaxel. *Clin Cancer Res* 2002;8:1038–44.
2. Nyman DW, Campbell KJ, Hersh E, Long K, Richardson K, Trieu V, et al. Phase I and pharmacokinetics trial of ABI-007, a novel nanoparticle formulation of paclitaxel in patients with advanced nonhematologic malignancies. *J Clin Oncol* 2005;23:7785–93.
3. Sparreboom A, Scripture CD, Trieu V, Williams PJ, De T, Yang A, et al. Comparative preclinical and clinical pharmacokinetics of a cremophor-free, nanoparticle albumin-bound paclitaxel (ABI-007) and paclitaxel formulated in Cremophor (Taxol). *Clin Cancer Res* 2005;11:4136–43.
4. Gradishar WJ, Tjulandin S, Davidson N, Shaw H, Desai N, Bhar P, et al. Phase III trial of nanoparticle albumin-bound paclitaxel compared with polyethylated castor oil-based paclitaxel in women with breast cancer. *J Clin Oncol* 2005;23:7794–803.
5. Shimada T, Yamazaki H, Mimura M, Inui Y, Guengerich FP. Interindividual variations in human liver cytochrome P-450 enzymes involved in the oxidation of drugs, carcinogens and toxic chemicals: studies with liver microsomes of 30 Japanese and 30 Caucasians. *J Pharmacol Exp Ther* 1994;270:414–23.
6. Therasse P, Arbuck SG, Eisenhauer EA, Wanders J, Kaplan RS, Rubinstein L, et al. New guidelines to evaluate the response to treatment in solid tumors. European Organization for Research and Treatment of Cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. *J Natl Cancer Inst* 2000;92:205–16.
7. Ibrahim NK, Samuels B, Page R, Doval D, Patel KM, Rao SC, et al. Multicenter phase II trial of ABI-007, an albumin-bound paclitaxel, in women with metastatic breast cancer. *J Clin Oncol* 2005;23:6019–26.
8. Green MR, Manikhas GM, Orlov S, Afanasyev B, Makhson AM, Bhar P, et al. Abraxane, a novel Cremophor-free, albumin-bound particle form of paclitaxel for the treatment of advanced non-small-cell lung cancer. *Ann Oncol* 2006;17:1263–8.
9. Rizvi NA, Riely GJ, Azzoli CG, Miller VA, Ng KK, Fiore J, et al. Phase I/II trial of weekly intravenous 130-nm albumin-bound paclitaxel as initial chemotherapy in patients with stage IV non-small-cell lung cancer. *J Clin Oncol* 2008;26:639–43.

original article

Annals of Oncology 21: 1001–1005, 2010
doi:10.1093/annonc/mdp464
Published online 29 October 2009

Phase II study of oxaliplatin plus S-1 as first-line treatment for advanced gastric cancer (G-SOX study)

W. Koizumi[1]*, H. Takiuchi[2], Y. Yamada[3], N. Boku[4], N. Fuse[5], K. Muro[6], Y. Komatsu[7] & A. Tsuburaya[8]

[1]*Department of Gastroenterology/Gastrointestinal Oncology, Kitasato University School of Medicine, Sagamihara;* [2]*Department of Gastroenterology, Osaka Medical College, Takatsuki;* [3]*Gastrointestinal Medical Oncology Division, National Cancer Center Hospital, Tokyo;* [4]*Division of Gastrointestinal Oncology, Shizuoka Cancer Center, Sunto-gun;* [5]*Division of Gastrointestinal Oncology and Digestive Endoscopy, National Cancer Center Hospital East, Kashiwa;* [6]*Department of Clinical Oncology, Aichi Cancer Center Hospital, Nagoya;* [7]*Department of Cancer Chemotherapy, Hokkaido University Hospital Cancer Center, Sapporo and* [8]*Department of Gastrointestinal Surgery, Kanagawa Cancer Center, Yokohama, Japan*

Received 8 August 2009; accepted 17 August 2009

Background: The efficacy and safety of oxaliplatin combined with S-1 (SOX regimen) for unresectable advanced or recurrent gastric cancer were investigated.

Patients and methods: Oxaliplatin was administered i.v. (100 mg/m^2) on day 1, while S-1 was administered orally (80 mg/m^2/day, b.i.d.) for 14 days followed by a 7-day rest. This schedule was repeated every 3 weeks.

Results: Among 55 patients enrolled, one patient received oxaliplatin for the other study, and three patients were considered unsuitable against the inclusion criteria. Accordingly, 51 patients were assessable for efficacy. The response rate was 59%, and the disease control rate was 84%. The median progression-free survival time was 6.5 months, the 1-year survival rate was 71%, and the median survival time was 16.5 months. In 54 patients assessed for safety, the major grade 3/4 toxic effects were neutropenia (22%), thrombocytopenia (13%), anemia (9%), anorexia (6%), fatigue (6%), and sensory neuropathy (4%).

Conclusion: These findings indicate that SOX regimen with oxaliplatin at a dose of 100 mg/m^2 is feasible and shows promising efficacy against advanced gastric cancer.

Key words: advanced gastric cancer, oxaliplatin, phase II, S-1, SOX

introduction

Chemotherapy for advanced gastric cancer was proven to be superior to best supportive care in terms of survival and quality of life [1–3]. Phase III studies have been carried out to compare epirubicin/cisplatin/5-fluorouracil (5-FU) with 5-FU/doxorubicin/methotrexate, cisplatin/5-FU with docetaxel/cisplatin/5-FU, and 5-FU/cisplatin with capecitabine/cisplatin [4–6]. On the basis of the results of these studies, advanced gastric cancer is mainly treated with combination chemotherapy that includes fluoropyrimidine derivatives and platinum compounds.

Oxaliplatin is a third-generation platinum compound that was developed to improve tolerability and ease of administration compared with cisplatin [7]. The non-inferiority of oxaliplatin-based regimens to cisplatin-based regimens was demonstrated in the Revised European-American Lymphoma (REAL)-2 phase III study [8]. In addition, the result of phase III study comparing 5-FU/leucovorin/cisplatin with 5-FU/leucovorin/oxaliplatin showed that oxaliplatin was at least as effective as cisplatin [9].

S-1 is an orally active prodrug of 5-FU that contains tegafur (which is continuously metabolized to 5-FU) blended with two modulators, gimeracil and potassium oxonate [10]. In Japan, advanced gastric cancer is mainly treated with S-1 alone or S-1 combined with other drugs. The SPIRITS phase III study demonstrated the superiority of S-1 plus cisplatin to S-1 alone [11]. The S-1 plus cisplatin regimen was also investigated by the FLAGS phase III study carried out in Western countries, which demonstrated that S-1 plus cisplatin was at least as effective as 5-FU plus cisplatin and less toxic [12].

We conducted a multicenter phase II study to evaluate the efficacy and safety of the combination regimen of S-1 and oxaliplatin (SOX regimen) in advanced gastric cancer as first-line therapy.

patients and methods

patients' eligibility

The following criteria were used to enroll patients for the present study. All patients had unresectable advanced or recurrent gastric cancer excluding the esophagus and gastroesophageal junction, confirmed by histological or

*Correspondence to: Dr W. Koizumi, Department of Gastroenterology/Gastrointestinal Oncology, Kitasato University School of Medicine, 2-1-1 Asamizodai, Sagamihara, Kanagawa 228-8520, Japan. Tel: +81-42-748-9111; Fax: +81-42-748-5120; E-mail: koizumi@med.kitasato-u.ac.jp

cytological examination. They had survived at least 4 weeks if extended or standard surgery had been carried out (or at least 2 weeks after minor surgery) and were able to take oral drugs. They were aged ≥20 years, had an Eastern Cooperative Oncology Group performance status (PS) of zero to two, and were expected to survive for at least 2 months. In general, they had not received prior chemotherapy, but those who had completed postoperative adjuvant therapy at least 180 days before enrollment were eligible. They had at least one measurable lesion according to RECIST guidelines [13]. They also had adequate bone marrow function (hemoglobin level ≥ 80 g/l, white blood cell count of 3–12 × 10^9/l, neutrophil count ≥ 1.5 × 10^9/l, and platelet count ≥ 100 × 10^9/l), liver function (total bilirubin ≤ 1.5 × the institutional upper limit of normal, aspartate aminotransferase/alanine aminotransferase ≤ 2.5 × the institutional upper limit of normal, and alkaline phosphatase ≤ 2.5 × the institutional upper limit of normal), and renal function (serum creatinine level ≤ 1.5 mg/dl and creatinine clearance ≥ 50 ml/min). All patients provided written informed consent.

This study was carried out in accordance with the Helsinki declaration and Good Clinical Practice guidelines and was approved by the institutional review boards of all participating medical institutions.

treatment plan

Oxaliplatin was administered i.v. at a dose of 100 mg/m^2 on day 1. S-1 was administered orally at a dose of 80 mg/m^2/day b.i.d. for 14 days (from the evening on day 1 until the morning on day 15), followed by a 7-day rest period in the 3-weekly schedule. Treatment was repeated until there was disease progression, unacceptable toxicity, or withdrawal of consent.

In the event of grade 4 neutropenia or febrile neutropenia or grade 3 diarrhea or stomatitis, the doses of oxaliplatin and S-1 were reduced by one dose level from the next cycle. If grade 2 sensory neuropathy not recovering by the end of the cycle or grade 3 sensory neuropathy occurred, the dose of oxaliplatin was reduced by one dose level from the next cycle after recovering to grade 2 or less. If grade 2 thrombocytopenia continued ≥8 days after the scheduled day for starting the next cycle or if platelet transfusion was required, oxaliplatin was reduced by one dose level from the next cycle. Oxaliplatin and S-1 could be reduced by two dose levels, but treatment was discontinued if subsequent reduction was indicated. The doses of oxaliplatin and S-1 could be reduced by 25 mg/m^2 and 10–30 mg/day, respectively, for each level. Treatment was discontinued if grade 4 diarrhea, stomatitis, or sensory neuropathy occurred, if grade 3 sensory neuropathy failed to recover by the time when the next cycle was scheduled, if grade 2 thrombocytopenia continued ≥15 days after the scheduled day for starting the next cycle, or if the rest period of S-1 was over 21 days.

evaluation

The data on the patients' characteristics, a 12-lead electrocardiogram, computed tomography (CT) scans, and tumor marker levels (CA19-9 and carcinoembryonic antigen) were obtained within 14 days of enrollment, while hematology tests, biochemistry tests, and assessment of symptoms and signs were carried out within 7 days before enrollment. During the study, hematology tests, biochemistry tests, and assessment of symptoms and signs were carried out every week until the end of the fourth cycle and subsequently every 3 weeks. CT scans were carried out and tumor markers were measured every 6 weeks (every 2 months after the best overall response was achieved).

Responses were evaluated according to the RECIST guidelines. To confirm partial response (PR) (30% or greater decrease in the sum of the longest diameter of target lesions, referenced against the baseline sum of the longest diameter of target lesions together with stabilization or decrease in size of nontarget lesions) or complete response (CR) (disappearance of all target and nontarget lesions together with normalization of tumor marker levels), tumor measurements were repeated no <4 weeks after objective response was firstly obtained. Responses were assessed by the independent review committee. Overall survival (OS) was defined as the time from treatment initiation to death from any cause. Progression-free survival (PFS) was the time from treatment initiation to first documentation of disease progression detected by the review committee or death from any cause (censored at second-line chemotherapy). Time-to-treatment failure (TTF) was the time from treatment initiation to discontinuation of treatment, first documentation of disease progression by the review committee, or death from any cause. Toxic effects were evaluated according to the Common Terminology Criteria for Adverse Events V3.0.

statistical analysis

The primary end point was the response rate (RR), while the secondary end points were OS, PFS, TTF, and safety. The required sample size was calculated to be at least 49 patients on the null hypothesis of the RR of ≤40% versus the alternative hypothesis of the RR of >60%, power 80%, and α 2.5% (one sided). The 95% confidence interval (CI) was calculated for the RR, PFS, and TTF. OS, PFS, and TTF were calculated by the Kaplan–Meier method. Safety was analyzed in all patients who received at least one dose of study medication.

The cut-off date for RR, PFS, TTF, and safety was 27 May 2008, while that for OS was 13 July 2009.

results

patients' characteristics

Fifty-five patients were enrolled from April to December in 2007. Among them, one patient who received oxaliplatin for the other study by mistake was excluded from all analyses. Three other patients were excluded from efficacy analysis because of prior chemotherapy (methotrexate), severe interstitial pneumonia, or absence of measurable lesions (one patient each). Accordingly, 51 patients formed the efficacy analysis set (Table 1), while 54 patients were analyzed for safety. The median age of the 51 patients was 63 years (range 30–77 years) and the PS was zero or one in 50 patients. Prior adjuvant chemotherapy with S-1 had been carried out in one patient, while 50 patients had received no prior chemotherapy.

treatment

At the data cut-off date, treatment was ongoing in eight patients. The major reasons for discontinuation of treatment in 46 patients were disease progression (63%), adverse events (28%), and withdrawal of consent (2%).

The median number of treatment cycles was 6.0 (range 1–16+). The median dose intensity was 88 mg/m^2/3 weeks for oxaliplatin and 867 mg/m^2/3 weeks for S-1, and the median relative dose intensity was 87.5% and 85.7%, respectively. The median total dose was 600 mg/m^2 for oxaliplatin and 5966 mg/m^2 for S-1.

efficacy

The response was assessed as PR, stable disease (SD) (less than a 30% reduction and less than a 20% increase in the sum of the longest diameter of target lesions, referenced against the baseline sum of the longest diameter of target lesions together with stabilization or decrease in size of nontarget lesions), and progressive disease (PD) in 30, 13, and 5, respectively, of the 51

Table 1. Patients' profile ($n = 51$)

Characteristic	No. of patients	%
Median age, years (range)	63 (30–77)	
Sex		
Male	34	67
Female	17	33
ECOG PS		
0	32	63
1	18	35
2	1	2
Disease status		
Advanced	47	92
Recurrent	4	8
Primary tumor		
No	12	24
Yes	39	77
Prior adjuvant chemotherapy		
No	50	98
Yes	1	2
Histology		
Diffuse	35	69
Intestinal	16	31
Sites of metastasis		
Lymph nodes	41	80
Liver	23	45
Lung	9	18
Peritoneum	7	14
Other	9	18
No. of metastases		
1	22	43
≥2	29	57

ECOG PS, Eastern Cooperative Oncology Group performance status.

Table 2. Objective response to treatment ($n = 51$)

Response	No. of patients	% (95% CI)
CR	0	0
PR	30	59
SD	13	26
PD	5	10
Not evaluable	3	6
Overall response rate	30	59 (44.2–72.4)
Disease control rate (CR + PR + SD)	43	84 (71.4–93.0)

CI, confidence interval; CR, complete response; PR, partial response; SD, stable disease; PD, progressive disease.

patients in the efficacy analysis set (three were not assessable). The RR was 59% (95% CI 44.2% to 72.4%) and the disease control rate (CR + PR + SD) was 84% (95% CI 71.4% to 93.0%) (Table 2).

The median follow-up period was 16.5 months as of 13 July 2009. The median survival time (MST) was 16.5 months (95% CI 13.2–22.3 months) (Figure 1), median PFS was 6.5 months (95% CI 4.8–11.2 months) (Figure 2), and median TTF was 4.8 months (95% CI 4.0–5.6 months). The patients who received

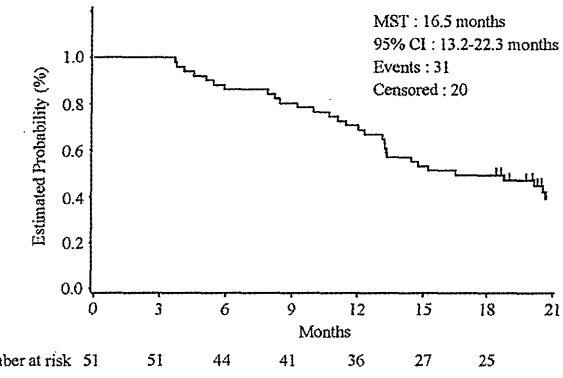

Figure 1. Kaplan–Meier estimates of overall survival ($n = 51$).

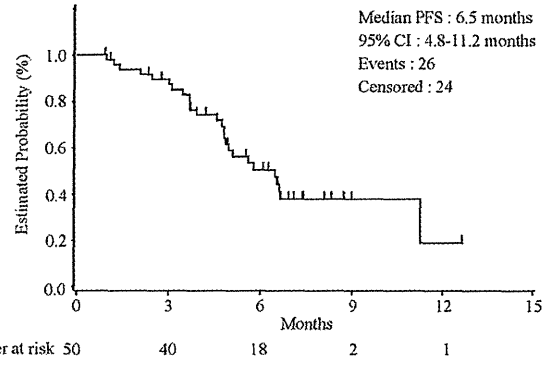

Figure 2. Kaplan–Meier estimates of progression-free survival ($n = 50$).

the second-line chemotherapy without PD were censored at the date of image examination immediately before the second-line chemotherapy in PFS analysis. The 1-year survival rate was 70.6% (95% CI 58.1% to 83.1%).

Forty-one of the 46 patients (89%) who discontinued treatment received second-line chemotherapy. One patient (2%) with PR underwent surgery and pathological CR was observed.

safety assessment

Grade 3/4 toxicity occurred in 33 of the 54 patients (61%) in the safety analysis set. Grade 3/4 leukopenia, neutropenia, thrombocytopenia, anemia, anorexia, and fatigue were noted in 2 (4%), 12 (22%), 7 (13%), 5 (9%), 3 (6%), and 3 patients (6%), respectively (Table 3). The median onset of thrombocytopenia in all grades was after 42 days and the nadir platelet count was seen at 113 days. The median time from the nadir to grade 0 or platelet count of treatment initiation was 15 days and the duration of thrombocytopenia in all grades was 21 days. Sensory neuropathy was observed in 48 patients (89%), but grade 3/4 neuropathy occurred only in two patients (4%). The median cumulative dose of oxaliplatin associated with sensory neuropathy of any grade was 150 mg/m^2 (grade 1: 150 mg/m^2, grade 2: 900 mg/m^2). There were no treatment-related deaths.

doi:10.1093/annonc/mdp464

Table 3. Toxicity of therapy ($n = 54$)

Toxicity (CTCAE)	No. of patients (%)					
	Grade 1	Grade 2	Grade 3	Grade 4	All grades	Grade 3/4
Hematological						
Leukopenia	15 (28)	16 (30)	2 (4)	0	33 (61)	2 (4)
Neutropenia	3 (6)	15 (28)	12 (22)	0	30 (56)	12 (22)
Thrombocytopenia	25 (46)	9 (17)	7 (13)	0	41 (76)	7 (13)
Anemia	14 (26)	14 (26)	4 (7)	1 (2)	33 (61)	5 (9)
Non-hematological						
Nausea	27 (50)	10 (19)	1 (2)	0	38 (70)	1 (2)
Vomiting	15 (28)	4 (7)	0	0	19 (35)	0
Diarrhea	17 (32)	4 (7)	1 (2)	0	22 (41)	1 (2)
Anorexia	21 (39)	16 (30)	2 (4)	1 (2)	40 (74)	3 (6)
Fatigue	24 (44)	14 (26)	2 (4)	1 (2)	41 (76)	3 (6)
Rash	13 (24)	2 (4)	0	0	15 (28)	0
Pigmentation	20 (37)	2 (4)	0	0	22 (41)	0
Hand–foot syndrome	12 (22)	2 (4)	0	0	14 (26)	0
Stomatitis	20 (37)	1 (2)	0	0	21 (39)	0
Increased creatinine	3 (6)	0	0	0	3 (6)	0
Febrile neutropenia	0	0	1 (2)	0	1 (2)	1 (2)
Sensory neuropathy	35 (65)	11 (20)	2 (4)	0	48 (89)	2 (4)

CTCAE, Common Terminology Criteria for Adverse Events V3.0.

discussion

Advanced gastric cancer is usually treated by combination chemotherapy with fluoropyrimidine derivatives and platinum compounds. Several recent large-scale phase III studies have shown that the RR ranges from 25% to 54%, median PFS from 2.9 to 7 months, and MST from 8.6 to 13 months [5, 6, 8, 9, 11, 14]. Unfortunately, these results are not satisfactory. In Japan, S-1 plus cisplatin is considered to be the standard treatment for advanced gastric cancer on the basis of the results of two phase III studies: the JCOG9912 study demonstrated non-inferiority of S-1 to i.v. infusion of 5-FU [14] and the SPIRITS study showed that S-1 plus cisplatin was superior to S-1 alone [11]. In the SPIRITS study, the RR, median PFS, and MST achieved with S-1 plus cisplatin were 54%, 6.0 months, and 13 months, respectively. However, more frequent incidences of grade 3/4 adverse events were reported as compared with S-1-alone group, and the combination regimens with improved safety are expected.

With the present SOX regimen, the RR was 59%, median PFS was 6.5 months, 1-year survival was 70.6%, and MST was 16.5 months, indicating similar efficacy to that of S-1 plus cisplatin. The excellent result of our SOX regimen in MST may be explicable by good PFS and feasible safety profile, which enabled patients to receive the second-line chemotherapy in the high proportion (89%). The efficacy of SOX regimen was also comparable with epirubicin and oxaliplatin plus capecitabine in the REAL-2 study (1-year survival rate of 47% and MST of 11.2 months) [8], which demonstrated that oxaliplatin was as effective as cisplatin combined with epirubicin and 5-FU or capecitabine.

Comparison of safety between the present SOX regimen and S-1 plus cisplatin that were reported previously [11] indicates a lower incidence of grade 3/4 toxicity with SOX regimen than S-1 plus cisplatin for leucopenia (4% versus 11%), neutropenia (22% versus 40%), anemia (9% versus 26%), anorexia (6% versus 30%), and nausea (2% versus 11%). The incidence of grade 3/4 thrombocytopenia was higher with SOX regimen (13% versus 5%). Sensory neuropathy is a characteristic toxicity of oxaliplatin, and 89% of the patients receiving SOX regimen had neuropathy, but only 4% had severe (grade 3/4) neuropathy. These results indicate that SOX regimen is more tolerable and tends to be superior to S-1 plus cisplatin in terms of safety.

Yamada et al. [15] reported that the treatment was discontinued at high frequency (28%) due to prolonged thrombocytopenia when metastatic colorectal cancer patients were treated with S-1 plus 130 mg/m^2 of oxaliplatin. This discontinuation was supposed to be caused by the geniality of dose reduction criteria which allowed the reduction of oxaliplatin only in case of occurrence of grade 3 or more toxicity in terms of thrombocytopenia. The incidence of thrombocytopenia was 93% in all grades and 28% in grade 3/4, resulting in low median relative dose intensity of S-1 74.6% and that of oxaliplatin 82.8%. Zang et al. [16] also reported the study of SOX regimen with 130 mg/m^2 of oxaliplatin in patients with metastatic colorectal cancer. In their study, the treatment was interrupted in cases of grade 2 or higher toxicity until the recovery to grade 0 or 1, and the doses of oxaliplatin and S-1 were reduced after a second occurrence of grade 2 toxicity. As a result, the incidence of thrombocytopenia was 13% in grade 3/4, and the median relative dose intensity of oxaliplatin and S-1 was 82% and 82%, respectively. In this study, we used 100 mg/m^2 dose of oxaliplatin as SOX regimen for advanced gastric cancer to decrease the incidence of thrombocytopenia considering the possible bleeding from the primary tumor and to maintain the dose intensity of S-1, which have been demonstrated to a key drug against advanced gastric cancer as a single agent. In this new regimen, the incidence of

thrombocytopenia was 13% in grade 3/4 without reducing the antitumor activity. The median relative dose intensity of oxaliplatin and S-1 was 87.5% and 85.7%, respectively, indicating that the treatment was carried out as scheduled in most of patients in this study.

In conclusion, SOX regimen with oxaliplatin at a dose of 100 mg/m^2 was effective and well tolerated in patients with advanced gastric cancer. SOX regimen has the potential to replace current regimens such as S-1 plus cisplatin or 5-FU plus cisplatin because of similar efficacy with less toxicity and more convenient treatment. Further investigation of this SOX regimen is expected.

funding

Yakult Honsha Co., Ltd.

acknowledgements

We are grateful to I. Hyodo, Y. Sakata, N. Masuda, and F. Nagamura for their kind advice and to A. Sato, K. Yoshikawa, and K. Miyagawa who carried out the independent review committee. We also thank S. Sugiura and S. Takahashi for their helpful advice. This study has been presented at the Annual Conference of the American Society of Clinical Oncology, Orlando, FL, 2009. This study was registered with Japan Pharmaceutical Information Center Clinical Trials Information (no. 070374).

disclosure

All authors declared no conflicts of interest.

references

1. Murad AM, Santiago FF, Petroianu A et al. Modified therapy with 5-fluorouracil, doxorubicin, and methotrexate in advanced gastric cancer. Cancer 1993; 72: 37–41.
2. Glimelius B, Hoffman K, Haglund U et al. Initial or delayed chemotherapy with best supportive care in advanced gastric cancer. Ann Oncol 1994; 5: 189–190.
3. Pyrhönen S, Kuitunen T, Nyandoto P, Kouri M. Randomised comparison of fluorouracil, epidoxorubicin and methotrexate (FEMTX) plus supportive care with supportive care alone in patients with non-resectable gastric cancer. Br J Cancer 1995; 71: 587–591.
4. Waters JS, Norman A, Cunningham D et al. Long-term survival after epirubicin, cisplatin and fluorouracil for gastric cancer: results of a randomized trial. Br J Cancer 1999; 80: 269–272.
5. Van Cutsem E, Moiseyenko VM, Tjulandin S et al. Phase III study of docetaxel and cisplatin plus fluorouracil compared with cisplatin and fluorouracil as first-line therapy for advanced gastric cancer: a report of the V325 Study Group. J Clin Oncol 2006; 24: 4991–4997.
6. Kang YK, Kang WK, Shin DK et al. Capecitabine/cisplatin versus 5-fluorouracil/cisplatin as first-line therapy in patients with advanced gastric cancer: a randomized phase III noninferiority trial. Ann Oncol 2009; 20: 666–673.
7. Kidani Y, Inagaki K, Tsukagoshi S. Examination of antitumor activities of platinum complexes of 1,2-diaminocyclohexane isomers and their related complexes. Gann 1976; 67: 921–922.
8. Cunningham D, Starling N, Rao S et al. Capecitabine and oxaliplatin for advanced esophagogastric cancer. N Engl J Med 2008; 358: 36–46.
9. Al-Batran SE, Hartmann JT, Probst S et al. Phase III trial in metastatic gastroesophageal adenocarcinoma with fluorouracil, leucovorin plus either oxaliplatin or cisplatin: a study of the Arbeitsgemeinschaft Internistische Onkologie. J Clin Oncol 2008; 26: 1435–1442.
10. Kato T, Shimamoto Y, Uchida J et al. Possible regulation of 5-fluorouracil-induced neuro- and oral toxicities by two biochemical modulators consisting of S-1, a new oral formulation of 5-fluorouracil. Anticancer Res 2001; 21: 1705–1712.
11. Koizumi W, Narahara H, Hara T et al. S-1 plus cisplatin versus S-1 alone for first-line treatment of advanced gastric cancer (SPIRITS trial): a phase III trial. Lancet Oncol 2008; 9: 215–221.
12. Ajani JA, Rodriguez W, Bodoky G et al. Multicenter phase III comparison of cisplatin/S-1 (CS) with cisplatin/5-FU (CF) as first-line therapy in patients with advanced gastric cancer (FLAGS). Gastrointestinal Cancers Symposium. 2009 Abstr 8.
13. Therasse P, Arbuck SG, Eisenhauer EA et al. New guidelines to evaluate the response to treatment in solid tumors. European Organization for Research and Treatment of Cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. J Natl Cancer Inst 2000; 92: 205–216.
14. Boku N, Yamamoto S, Shirao K et al. Randomized phase III study of 5-fluorouracil (5-FU) alone versus combination of irinotecan and cisplatin (CP) versus S-1 alone in advanced gastric cancer (JCOG9912). J Clin Oncol *(Meeting Abstracts)* 2007; 25: (Abstr LBA4513).
15. Yamada Y, Tahara M, Miya T et al. Phase I/II study of oxaliplatin with oral S-1 as first-line therapy for patients with metastatic colorectal cancer. Br J Cancer 2008; 98: 1034–1038.
16. Zang DY, Lee BH, Park HC et al. Phase II study with oxaliplatin and S-1 for patients with metastatic colorectal cancer. Ann Oncol 2009; 20: 892–896.

Cancer Science
The official journal of the Japanese Cancer Association

Adipocytokines as new promising markers of colorectal tumors: Adiponectin for colorectal adenoma, and resistin and visfatin for colorectal cancer

Takako Eguchi Nakajima,[1] Yasuhide Yamada,[1,6] Tetsutaro Hamano,[2] Koh Furuta,[3] Takahisa Matsuda,[4] Shin Fujita,[5] Ken Kato,[1] Tetsuya Hamaguchi[1] and Yasuhiro Shimada[1]

[1]Division of Gastrointestinal Oncology, National Cancer Center Hospital, Tokyo; [2]Hamano Statistical Analysis Ltd., Tokyo; Divisions of [3]Clinical Laboratory, [4]Endoscopy and [5]Colon Surgery, National Cancer Center Hospital, Tokyo, Japan

(Received November 11, 2009/Revised January 20, 2010/Accepted January 22, 2010/Online publication March 15, 2010)

Adipocytokines are adipocyte-secreted hormones associated with some malignancies such as colorectal, breast, and prostate cancer. We hypothesized that changes in the levels of adipocytokines may indicate the carcinogenesis and progression of colorectal cancer and adenoma, and investigated the association of the blood levels of several adipocytokines through a case-control study. Blood levels of adiponectin, leptin, resistin, visfatin, and C-peptide at diagnosis were measured in 115 colorectal cancer patients and 115 age-, sex-, and body mass index-matched controls. The same analysis was performed in 72 colorectal adenoma patients and 72 controls. Logistic regression models were used for estimating odds ratios and 95% confidence intervals, and one-way ANOVA was performed to determine the prevalence of each variable between two or more groups. Resistin and visfatin levels in cancer patients were significantly higher than those of controls on multivariate analysis ($P = 0.03$ and $P < 0.01$, respectively). Stage progression significantly correlated with resistin and visfatin levels ($P < 0.01$ for both). The adiponectin level in adenoma patients was significantly lower than that of controls on multivariate analysis ($P = 0.04$). Its level was inversely correlated with the number of adenoma ($P = 0.02$), but not correlated with the size of adenoma. Resistin and visfatin may be good biomarkers of colorectal malignant potential and stage progression. Adiponectin level may be a good biomarker of colorectal adenoma. (*Cancer Sci* 2010; 101: 1286–1291)

Adipocytokines, such as adiponectin, leptin, resistin, visfatin, tumour necrosis factor (TNF)-α, and interleukin (IL)-6 are cytokines secreted by visceral adipose tissue, and they have recently been suggested to be associated with obesity-related diseases.[1,2] Many epidemiologic studies have shown a positive correlation between obesity and increased risk of colorectal cancer and adenoma as well as other cancers at various sites (e.g. breast, prostate gland, and endometrium).[3–5]

In obesity mouse models, severe macrophage invasion was observed in the vascular/stromal compartment of adipose tissue, suggesting that excess adiposity is associated with chronic inflammation.[6,7] Other reports have shown that prostaglandin E2 stimulates leptin secretion from cultured human adipose tissue cells and that cyclooxygenase 2 inhibitors prevent an increase in leptin production.[8] In inflammation-associated colorectal cancers, such as those associated with inflammatory bowel diseases, non-genetic stimuli such as overexpression of IL-6 also enhance the survival and proliferation of preneoplastic cells.[9] Leptin was also reported to induce IL-6 production by $Apc^{Min/+}$ colon epithelial cells which leads to autocrine/paracrine trans IL-6 receptor signaling.[10] This results in the promotion and survival proliferation of preneoplastic cells. On the other hand, adiponectin reportedly inhibits inflammation and angiogenesis while leptin induces tumor angiogenesis.[11,12]

These findings in epidemiological and basic research suggest that adipocytokines may well contribute to the induction of carcinogenesis and tumor progression. Therefore, we hypothesized that changes in the levels of adipocytokines may indicate the carcinogenesis and progression of colorectal cancer and adenoma. To evaluate whether adipocytokines are stronger biomarkers of colorectal cancer and adenoma than body mass index (BMI), we performed a BMI-matched case-control study and investigated the association between the blood levels of several adipocytokines and colorectal cancer and adenoma.

Materials and Methods

Study population. After approval of the study protocol by the Institutional Review Board of the National Cancer Center, patients who underwent upper total colonoscopy at the hospital from February 1999 to February 2007, who were considered to have no active malignancies except colorectal cancer and no inflammatory bowel diseases, and whose blood samples at diagnosis before any treatments for colorectal cancer or adenoma could be obtained, were identified and invited to participate in the study. Patients who had been newly and pathologically diagnosed with colorectal cancer by biopsy using colonoscopy and treated at our hospital were identified as colorectal cancer patients among the enrolled patients. Age-, sex-, and BMI-matched controls (1:1) were identified among patients who had been diagnosed as free from colorectal cancer or adenoma by colonoscopy. Among the enrolled patients, we identified those patients who had been newly undergone hot-biopsy, polypectomy, or endoscopic mucosal resection and were pathologically diagnosed with colorectal adenoma at our hospital as colorectal adenoma patients. Age-, sex- and BMI-matched controls (1:1) were identified among patients who had been diagnosed as free from colorectal cancer or adenoma by colonoscopy. BMI at diagnosis was calculated based on the data in medical records as follows: weight (kg)/height $(m)^2$. All subjects (patients and controls) provided informed consent prior to the collection and analysis of blood samples. Clinical and pathological information for both groups was obtained from medical records.

Adipocytokines and C-peptide measurements. All blood samples were stored at −20°C until use. None of the samples were previously thawed. Blood levels of adiponectin, resistin,

[6]To whom correspondence should be addressed.
E-mail: yayamada@ncc.go.jp

doi: 10.1111/j.1349-7006.2010.01518.x

Table 1. Clinical characteristics of patients with colorectal cancer and controls

	Patients (n = 115)	Controls (n = 115)	P-values
Age (years)	63.7 ± 10.3	63.5 ± 10.5	0.99
Sex			
Female (%)	46 (40.0)	46 (40.0)	
Male (%)	69 (60.0)	69 (60.0)	1.00
Body mass index	22.9 ± 2.9	23.1 ± 2.7	0.897
Stage*			
0	23	–	–
I	23	–	–
II	19	–	–
III	23	–	–
IV	27	–	–
Location			
Right colon	55	–	–
Left colon	7	–	–
Rectum	53	–	–
Macroscopic type*			
0 – Ip	5	–	–
0 – Isp	6	–	–
0 – Is	10	–	–
0 – IIa	17	–	–
0 – IIb	0	–	–
0 – IIc	0	–	–
0 – III	0	–	–
1	1	–	–
2	73	–	–
3	1	–	–
4	1	–	–
5	1	–	–
Histological type*			
Well-differentiated adenocarcinoma	86	–	–
Moderately differentiated adenocarcinoma	21	–	–
Poorly differentiated adenocarcinoma	7	–	–
Mucinous adenocarcinoma	1	–	–

Data are presented as mean ± SD. *Japanese Classification of Colorectal Carcinoma 6th edition.

visfatin, and C-peptide at diagnosis were measured by SRL (Tokyo, Japan). Adiponectin was determined by enzyme-linked immunosorbent assay (ELISA) (Otsuka Pharmaceutical, Tokyo, Japan) with a sensitivity of 1.9 μg/mL, an intra-assay coefficient of variation of 3.5–5.1%, and an inter-assay coefficient of variation of 6.0–8.7%. Resistin was determined by ELISA (BioVender Laboratory Medicine, Brno, Czech Republic) with a sensitivity of 1.1 ng/mL, an intra-assay coefficient of variation of 2.8–3.4%, and an inter-assay coefficient of variation of 5.1–6.9%. Leptin was measured using radioimmunoassay kits (Linco Research, St. Charles, MO, USA) with a sensitivity of 0.5 ng/mL, an intra-assay coefficient of variation of 3.4–8.3%, and an inter-assay coefficient of variation of 3.0–6.2%. Visfatin was determined by ELISA (Adipo Gen, Seoul, Korea) with a sensitivity of 0.13 ng/mL, an intra-assay coefficient of variation of 4.4–10.4%, and an inter-assay coefficient of variation of 6.4–9.9%. C-peptide was determined by ELISA (Fujirebio, Tokyo, Japan) with a sensitivity of 0.04 ng/mL, an intra-assay coefficient of variation of 1.96–2.97%, and an inter-assay coefficient of variation of 1.06–2.60%. Duplicate measurements were performed in a single experiment.

Statistical analysis. The results of the comparison of clinical characteristics between patients and controls was evaluated by the χ^2-test for categorical variables and two-sample t-test for continuous variables. Conditional logistic regression models were used for estimating odds ratios and 95% confidence intervals to evaluate the association of each variable with colorectal cancer or adenoma. One-way ANOVA was performed to examine the prevalence of each variable between tumor stage groups. Log transformations were conducted on variables prior to analysis to achieve normal distribution. Differences with a P-value <0.05 were considered significant. All statistical analyses were carried out using the SAS system (version 9.1.3; SAS Institute, Cary, NC, USA).

Results

Adipocytokines and C-peptide, and colorectal cancer. The clinical characteristics and adipocytokine and C-peptide levels of the 115 colorectal cancer patients and 115 controls are shown in Tables 1 and 2. There was no significant difference in age, sex, and BMI between the two groups. Results of the univariate and multivariate logistic regression analyses are shown in Table 3. Resistin and visfatin levels were significantly higher in the colorectal cancer patients than in the controls on multivariate analysis (P = 0.03 and P < 0.01, respectively). Linear contrast analysis was conducted to evaluate the correlation between each variable and tumor stage defined by the Japanese Classification of Colorectal Carcinoma 6th edition (Table 4). Resistin and visfatin levels gradually increased with tumor stage progression (P < 0.01 and P < 0.01, respectively).

Adipocytokines and C-peptide, and colorectal adenoma. The clinical characteristics and adipocytokine and C-peptide levels of the 72 colorectal adenoma patients and 72 controls are shown in Tables 5 and 6. There was no significant difference in age, sex, and BMI between the two groups. Results of the univariate and multivariate logistic regression analyses are shown in Table 7. Multivariate analysis showed that adiponectin levels were significantly lower in the colorectal adenoma patients than in the control patients (P = 0.04). Linear contrast analysis was conducted to evaluate the correlation between each variable and the number of adenomas (Table 8a). Adiponectin level inversely correlated with the number of adenomas (P = 0.02). The size of the largest adenoma among all the adenomas of a patient showed no significant correlation with any variables (Table 8b).

Table 2. Blood adipocytokine levels in patients with colorectal cancer and controls

	Patients				Controls			
	n	Median value	25th quartile value	75th quartile value	n	Median value	25th quartile value	75th quartile value
Adiponectin (μg/mL)	115	8.9	6.6	13	115	8.9	5.7	12.9
Resistin (ng/mL)	115	4.5	3.1	6.4	115	3.1	2.2	4.7
Leptin (ng/mL)	115	3.7	2.4	5.7	114	4.2	2.3	6
Visfatin (ng/mL)	115	3.9	2.1	7.9	115	1.4	0.8	2.6
C-peptide (ng/mL)	114	0.2	0.1	0.4	111	0.3	0.1	0.6

Table 3. Univariate and multivariate analysis of patients with colorectal cancer and controls

	Univariate analysis		Multivariate analysis	
	Odds ratios (95% confidence intervals)	*P*-values	Odds ratios (95% confidence intervals)	*P*-values
Adiponectin*	1.227 (0.653–2.307)	0.52	0.802 (0.321–2.003)	0.64
Resistin*	2.850 (1.700–4.777)	<0.01	2.067 (1.053–4.055)	0.03
Leptin*	0.799 (0.458–1.393)	0.43	1.057 (0.477–2.342)	0.89
Visfatin*	3.142 (2.064–4.783)	<0.01	2.985 (1.862–4.787)	<0.01
C-peptide*	0.711 (0.550–0.920)	0.01	0.983 (0.663–1.458)	0.93

*Log-transformed.

Table 4. Association between adipocytokine levels and stage progression of colorectal cancer

	Control		Stage 0		Stage 1		Stage 2		Stage 3		Stage 4		*P*-values
	n	mean ± SD	*n*	mean ± SD	*n*	mean ± SD	*n*	mean ± SD	*n*	mean ± SD	*n*	mean ± SD	
Adiponectin*	115	2.3 ± 0.5	23	2.3 ± 0.4	23	2.2 ± 0.6	19	2.3 ± 0.5	23	2.1 ± 0.5	27	2.3 ± 0.4	0.94
Resistin*	115	1.2 ± 0.5	23	1.3 ± 0.5	23	1.6 ± 0.5	19	1.5 ± 0.5	23	1.5 ± 0.6	27	1.7 ± 0.5	<0.01
Leptin*	114	1.4 ± 0.7	23	1.4 ± 0.7	23	1.4 ± 0.7	19	1.5 ± 0.8	23	1.3 ± 0.5	27	1.1 ± 0.6	0.11
Visfatin*	115	0.2 ± 1.1	23	0.8 ± 1.2	23	1.3 ± 1.1	19	1.0 ± 0.9	23	1.5 ± 1.0	27	1.8 ± 0.9	<0.01
C-peptide*	111	−1.4 ± 1.2	23	−1.6 ± 1.2	23	−1.6 ± 1.1	19	−1.9 ± 1.2	22	−1.8 ± 1.1	27	−1.6 ± 1.0	0.17

*Log-transformed. Data are presented as mean ± SD.

Linear contrast analysis was also conducted to evaluate the correlation between adiponectin and the adenoma-carcinoma sequence, and the result was not significant (data not shown).

Table 5. Clinical characteristics of patients with colorectal adenoma and controls

	Patients (*n* = 72)	Controls (*n* = 72)	*P*-values
Age (years)	66.8 ± 7.3	66.7 ± 7.1	0.99
Sex			
Female (%)	22 (30.6)	22 (30.6)	
Male (%)	50 (69.4)	50 (69.4)	1.00
Body mass index	23.0 ± 2.8	22.8 ± 2.8	0.74
Number of adenomas			
2>	44	–	–
≥3	28	–	–
Location			
Right colon	33	–	–
Left colon	27	–	–
Rectum	12	–	–
Macroscopic type*			
0 – Ip	4	–	–
0 – Isp	13	–	–
0 – Is	24	–	–
0 – IIa	31	–	–
0 – IIb	0	–	–
0 – IIc	0	–	–
0 – III	0	–	–
Histological atypia			
Moderate atypia	64	–	–
Severe atypia	78	–	–
Maximum size			
−5 mm	14	–	–
6–10 mm	24	–	–
11–20 mm	17	–	–
>20 mm	17	–	–

Data are presented as mean ± SD. *Japanese Classification of Colorectal Carcinoma 6th edition.

Discussion

The results of this case-control study suggest that resistin and visfatin may be good biomarkers of colorectal malignant potential independently from BMI, and also of stage progression of colorectal cancer. Adiponectin may be a good biomarker of colorectal adenoma independently from BMI. For gastric cancer, we have reported similar results, namely, resistin and visfatin levels in gastric cancer patients were significantly higher than those in controls, and gradually increased with tumor stage progression. Furthermore, adiponectin levels tended to be lower in early stage gastric cancer patients than in controls.[13]

Obesity is recognized as a strong risk factor for the development of several cancers.[3–5] However, many experimental and case-control studies have suggested that BMI is not the best and only marker for elucidating the physiology of obesity. Recently, adipocytokines produced by adipose tissue have been the subject of intense investigation as novel risk markers not only of metabolic syndrome but also of cancers, particularly those indicating a correlation between their risk of development and obesity such as colorectal cancer and adenoma.[14–20] To the best of our knowledge, however, the present study is the first report to evaluate a difference in visfatin level between colorectal cancer patients and controls, and the only one report has been reported for a difference in resistin level so far.[21]

Adiponectin suppresses the secretion of inflammatory cytokines such as TNF-α, and induces the secretion of anti-inflammatory cytokines such as IL-10 in the atherogenic process.[22–24] Furthermore, it has been reported to inhibit tumor growth by suppressing angiogenesis *in vitro* and *in vivo*.[25] In case-control studies, the correlation between adiponectin level and colorectal cancer remains controversial[19,26]. An inverse correlation between adiponectin level and colorectal adenoma has been also reported.[27] Our results showed an inverse correlation between adiponectin and colorectal adenoma. However, we had no information regarding body weight changes in the patients and controls before the sampling, and thus it was not possible to determine whether the decrease in adiponectin levels in the patients was caused by obesity before the sampling. It was also difficult to determine when the adiponectin level decreased, either before or after colorectal adenoma development. Instead

doi: 10.1111/j.1349-7006.2010.01518.x

Table 6. Blood adipocytokine levels in patients with colorectal adenoma and controls

	Patients				Controls			
	n	Median value	25th quartile value	75th quartile value	*n*	Median value	25th quartile value	75th quartile value
Adiponectin (μg/mL)	72	7.5	5.4	10.3	72	8.8	6.3	13.6
Resistin (ng/mL)	72	3.1	2.4	4.8	72	2.8	1.9	3.9
Leptin (ng/mL)	71	3.3	2.4	5.4	72	3.3	1.8	5.4
Visfatin (ng/mL)	72	1	0.6	2.8	72	1.6	0.7	2.8
C-peptide (ng/mL)	71	0.3	0.1	0.7	69	0.2	0.1	0.5

Table 7. Univariate and multivariate analysis of patients with colorectal adenoma and controls

	Univariate analysis		Multivariate analysis	
	Odds ratios (95% confidence intervals)	*P*-values	Odds ratios (95% confidence intervals)	*P*-values
Adiponectin*	0.363 (0.169–0.780)	0.01	0.422 (0.189–0.946)	0.04
Resistin*	1.293 (0.706–2.368)	0.41	1.200 (0.595–2.420)	0.61
Leptin*	1.497 (0.772–2.901)	0.23	1.331 (0.662–2.677)	0.42
Visfatin*	0.883 (0.661–1.180)	0.40	0.872 (0.604–1.260)	0.47
C-peptide*	1.208 (0.893–1.634)	0.22	1.023 (0.704–1.484)	0.91

*Log-transformed.

Table 8. Association between adipocytokine levels and clinical features of colorectal adenoma. (a) Association between adipocytokine levels and number of colorectal adenomas. (b) Association between adipocytokine levels and maximum size of colorectal adenomas

	Control		≤2		≥3		*P*-values
	n	mean ± SD	*n*	mean ± SD	*n*	mean ± SD	
(a)							
Adiponectin*	72	2.2 ± 0.5	44	2.0 ± 0.6	28	2.0 ± 0.4	0.02
Resistin*	72	1.1 ± 0.6	44	1.2 ± 0.5	28	1.1 ± 0.5	0.90
Leptin*	72	1.2 ± 0.6	43	1.2 ± 0.6	28	1.4 ± 0.5	0.15
Visfatin*	72	0.3 ± 1.2	44	0.2 ± 1.5	28	0.1 ± 1.1	0.40
C-peptide*	69	−1.5 ± 1.2	43	−1.2 ± 1.2	28	−1.2 ± 1.1	0.34

	Control		−5 mm		6–10 mm		11–20 mm		>20 mm		*P*-values
	n	mean ± SD	*n*	mean ± SD	*n*	mean ± SD	*n*	mean ± SD	*n*	mean ± SD	
(b)											
Adiponectin*	72	2.2 ± 0.5	14	1.9 ± 0.4	24	1.9 ± 0.4	17	1.9 ± 0.5	17	2.3 ± 0.6	0.48
Resistin*	72	1.1 ± 0.6	14	1.2 ± 0.4	24	1.2 ± 0.6	17	1.4 ± 0.5	17	1.0 ± 0.4	0.81
Leptin*	72	1.2 ± 0.6	13	1.6 ± 0.7	24	1.2 ± 0.5	17	1.1 ± 0.6	17	1.3 ± 0.6	0.53
Visfatin*	72	0.3 ± 1.2	14	0.0 ± 1.4	24	0.3 ± 1.2	17	0.6 ± 1.5	17	−0.4 ± 1.2	0.31
C-peptide*	69	−1.5 ± 1.2	13	−0.9 ± 0.8	24	−1.1 ± 1.2	17	−1.6 ± 1.2	17	−1.3 ± 1.2	0.64

*Log-transformed. Data are presented as mean ± SD.

of these limitations, we evaluated the correlation between the number of adenomas, the size of adenomas and adenoma-carcinoma sequence, and adiponectin to speculate the possibilities as "risk factors" for colorectal adenoma. The results showed that adiponectin level was inversely correlated with the number of adenoma. However, we could not elucidate why the adiponectin level was not correlated with the size of adenoma. If many more patients were enrolled in this study, a significant correlation between adiponectin levels and adenoma sizes may have been detected.

We have performed the above additional investigations into the relationship between adiponectin levels and colorectal carcinoma; however, our study has a few limitations. The BMI levels of the selected target group are very important and can affect the results of the study. The mean of BMI level of the patients in this study was 22.9, which was lower than that reported previously; this low BMI level may be attributed to the fact that all the patients were Japanese. Further, it is possible that variables other than those evaluated in this study may be correlated with adiposity and may influence the levels of adipocytokines. Therefore, the implications of our findings should be carefully evaluated considering these limitations.

Leptin primarily controls body fat stores and has also roles in promoting cellular proliferation, inhibiting cellular apoptosis, and inducing angiogenesis.[28] Over the years, the association between leptin levels and the risk of colorectal cancer or adenoma has remained controversial.[20,29] The expression of the leptin receptor in normal human colon mucosa, adenomas, and cancers suggests that a direct effect of leptin may be involved in carcinogenesis.[30] In the present study, however, the level of leptin was not significantly different between controls and patients with colorectal cancer or adenoma. In our previous

studies on the correlation between adipocytokines levels and gastric or esophageal cancer, we have shown that a strong correlation exists between leptin level and BMI. In this study, however, the BMI levels of patients and controls were similar; therefore, the value of leptin as a biomarker for colorectal could not be evaluated.[13,31]

Resistin has been demonstrated to be involved in inflammatory states corresponding to its predominant expression in mononuclear cells, particularly in atherosclerosis.[32,33] As for its correlation with cancer, three case-control studies on the risk of myelodysplastic syndrome, multiple myeloma, or colorectal cancer have been reported.[21,34,35] Dalamaga *et al.* demonstrated a decreased resistin level in myelodysplastic syndrome (MDS) patients, and speculated that it was due to a compensatory response to the up-regulation of other inflammatory factors etiologically linked to myelodysplasia. They also reported a decreased level of resistin in patients with multiple myeloma. Kumor *et al.* reported that the resistin levels in colorectal cancer patients are higher than those in controls and that the resistin levels in colorectal adenoma patients and controls were also significantly different. Our results showed that resistin levels, particularly in colorectal cancer patients, were significantly higher than those in controls independent of the BMI, and these levels gradually increased with progression in tumor stage. This may imply that resistin is a biomarker of colorectal malignant potential and stage progression.

Visfatin is a new insulinmimetic adipocytokine, which directly interacts with the insulin receptor but as the insulin-like growth factor receptor, and can subsequently promote cancer cell proliferation[36]. It is more highly expressed in primary colorectal cancer than in non-neoplastic mucosa.[37] Although the clinical correlations of visfatin with cancer have been rarely reported, we demonstrated here that it may be a novel and promising biomarker of colorectal cancer as well as resistin.

Taken together, the results suggest that resistin and visfatin may be good biomarkers of colorectal malignant potential independently of BMI, and also of stage progression of colorectal cancer. Adiponectin level may be a good biomarker of colorectal adenoma independently of BMI. Further investigations as to whether the changes in adipocytokine levels are the result and/or effects of colorectal cancer or adenoma development are needed, and the elucidation of this causative association will undoubtedly clarify the correlation between obesity and cancer. Histological studies on the expression of adipocytokines in cancer tissues also should be conducted to determine whether adipocytokines derived from cancer tissues or those derived from adipose tissues are important for carcinogenesis and tumor progression.

Acknowledgment

This work was supported by the Ministry of Health, Labor and Welfare of Japan.

Disclosure Statement

The authors have no conflict of interest.

References

1 Matsuzawa Y, Funahashi T, Kihara S, Shimomura I. Adiponectin and metabolic syndrome. *Arterioscler Thromb Vasc Biol* 2004; **24**: 29–33.
2 Rose DP, Komninou D, Stephenson GD. Obesity, adipocytokines, and insulin resistance in breast cancer. *Obes Rev* 2004; **5**: 153–65.
3 Calle EE, Thun MJ. Obesity and cancer. *Oncogene* 2004; **23**: 6365–78.
4 Nishii T, Kono S, Abe H *et al.* Glucose intolerance, plasma insulin levels, and colon adenomas in Japanese men. *Jpn J Cancer Res* 2001; **92**: 836–40.
5 Wolk A, Gridley G, Svensson M *et al.* A prospective study of obesity and cancer risk (Sweden). *Cancer Causes Control* 2001; **12**: 13–21.
6 Weisberg SP, McCann D, Desai M, Rosenbaum M, Leibel RL, Ferrante AW Jr. Obesity is associated with macrophage accumulation in adipose tissue. *J Clin Invest* 2003; **112**: 1796–808.
7 Xu H, Barnes GT, Yang Q *et al.* Chronic inflammation in fat plays a crucial role in the development of obesity-related insulin resistance. *J Clin Invest* 2003; **112**: 1821–30.
8 Fain JN, Leffler CW, Cowan GS Jr, Buffington C, Pouncey L, Bahouth SW. Stimulation of leptin release by arachidonic acid and prostaglandin E(2) in adipose tissue from obese humans. *Metabolism* 2001; **50**: 921–8.
9 Balkwill F, Coussens LM. Cancer: an inflammatory link. *Nature* 2004; **431**: 405–6.
10 Fenton JI, Hord NG, Lavigne JA, Perkins SN, Hursting SD. Leptin, insulin-like growth factor-1, and insulin-like growth factor-2 are mitogens in ApcMin/+ but not Apc+/+ colonic epithelial cell lines. *Cancer Epidemiol Biomarkers Prev* 2005; **14**: 1646–52.
11 Park HY, Kwon HM, Lim HJ *et al.* Potential role of leptin in angiogenesis: leptin induces endothelial cell proliferation and expression of matrix metalloproteinases in vivo and in vitro. *Exp Mol Med* 2001; **33**: 95–102.
12 Zhao X, Huang K, Zhu Z, Chen S, Hu R. Correlation between expression of leptin and clinicopathological features and prognosis in patients with gastric cancer. *J Gastroenterol Hepatol* 2007; **22**: 1317–21.
13 Nakajima TE, Yamada Y, Hamano T *et al.* Adipocytokine levels in gastric cancer patients: resistin and visfatin as biomarkers of gastric cancer. *J Gastroenterol* 2009; **44**: 685–90.
14 Petridou E, Mantzoros C, Dessypris N *et al.* Plasma adiponectin concentrations in relation to endometrial cancer: a case-control study in Greece. *J Clin Endocrinol Metab* 2003; **88**: 993–7.
15 Miyoshi Y, Funahashi T, Kihara S *et al.* Association of serum adiponectin levels with breast cancer risk. *Clin Cancer Res* 2003; **9**: 5699–704.
16 Petridou E, Papadiamantis Y, Markopoulos C, Spanos E, Dessypris N, Trichopoulos D. Leptin and insulin growth factor I in relation to breast cancer (Greece). *Cancer Causes Control* 2000; **11**: 383–8.
17 Goktas S, Yilmaz MI, Caglar K, Sonmez A, Kilic S, Bedir S. Prostate cancer and adiponectin. *Urology* 2005; **65**: 1168–72.
18 Hsing AW, Chua S Jr, Gao YT *et al.* Prostate cancer risk and serum levels of insulin and leptin: a population-based study. *J Natl Cancer Inst* 2001; **93**: 783–9.
19 Wei EK, Giovannucci E, Fuchs CS, Willett WC, Mantzoros CS. Low plasma adiponectin levels and risk of colorectal cancer in men: a prospective study. *J Natl Cancer Inst* 2005; **97**: 1688–94.
20 Stattin P, Lukanova A, Biessy C *et al.* Obesity and colon cancer: does leptin provide a link? *Int J Cancer* 2004; **109**: 149–52.
21 Kumor A, Daniel P, Pietruczuk M, Malecka-Panas E. Serum leptin, adiponectin, and resistin concentration in colorectal adenoma and carcinoma (CC) patients. *Int J Colorectal Dis* 2009; **24**: 275–81.
22 Fantuzzi G. Adipose tissue, adipokines, and inflammation. *J Allergy Clin Immunol* 2005; **115**: 911–9. quiz 20.
23 Kumada M, Kihara S, Ouchi N *et al.* Adiponectin specifically increased tissue inhibitor of metalloproteinase-1 through interleukin-10 expression in human macrophages. *Circulation* 2004; **109**: 2046–9.
24 Yokota T, Oritani K, Takahashi I *et al.* Adiponectin, a new member of the family of soluble defense collagens, negatively regulates the growth of myelomonocytic progenitors and the functions of macrophages. *Blood* 2000; **96**: 1723–32.
25 Wang Y, Lam KS, Xu JY *et al.* Adiponectin inhibits cell proliferation by interacting with several growth factors in an oligomerization-dependent manner. *J Biol Chem* 2005; **280**: 18341–7.
26 Ferroni P, Palmirotta R, Spila A *et al.* Prognostic significance of adiponectin levels in non-metastatic colorectal cancer. *Anticancer Res* 2007; **27**: 483–9.
27 Otake S, Takeda H, Suzuki Y *et al.* Association of visceral fat accumulation and plasma adiponectin with colorectal adenoma: evidence for participation of insulin resistance. *Clin Cancer Res* 2005; **11**: 3642–6.
28 Sierra-Honigmann MR, Nath AK, Murakami C *et al.* Biological action of leptin as an angiogenic factor. *Science* 1998; **281**: 1683–6.
29 Chia VM, Newcomb PA, Lampe JW *et al.* Leptin concentrations, leptin receptor polymorphisms, and colorectal adenoma risk. *Cancer Epidemiol Biomarkers Prev* 2007; **16**: 2697–703.
30 Hardwick JC, Van Den Brink GR, Offerhaus GJ, Van Deventer SJ, Peppelenbosch MP. Leptin is a growth factor for colonic epithelial cells. *Gastroenterology* 2001; **121**: 79–90.
31 Nakajima TE, Yamada Y, Hamano T *et al.* Adipocytokines and squamous cell carcinoma of the esophagus. *J Cancer Res Clin Oncol* 2010; **136**: 261–6.
32 Shetty GK, Economides PA, Horton ES, Mantzoros CS, Veves A. Circulating adiponectin and resistin levels in relation to metabolic factors, inflammatory

doi: 10.1111/j.1349-7006.2010.01518.x