patient chemotherapy room using the port system and a portable pump.

Regimens

The modified FOLFOX6 regimen included leucovorin 400mg/m² intravenous (IV) on day 1, FU 400mg/m² IV on day 1 followed by 2,400mg/m² IV over 46 hours, and oxaliplatin 85mg/m² IV on day 1. All therapy was administered every 2 weeks for a total of 10-13 doses. Treatment was administered until progression of disease or unmanageable toxic effects occurred. After the eighth to tenth FOLFOX cycle, tri-weekly mFOLFOX6 maintenance therapy was initiated. In the case of disease progression, the FOLFIRI (5) regimen could be introduced.

Adverse event reporting

Adverse events were reported using the National Cancer Institute Common Terminology Criteria for Adverse Events version 3.0. Routine reporting was required for all events ≥ grade 3, every 6 weeks during active treatment.

Dose modifications

Oxaliplatin and FU were reduced for grade 3 and 4 adverse events except for diarrhea and stomatitis, wherein only FU was reduced for less than grade 4 toxicity, or for neurologic toxicities, wherein only oxaliplatin was modified for grades 3 and 2, provided the toxicity exceeded 7 days in duration.

RESULTS

Patient characteristics

Twenty-nine patients with metastatic CRC were treated by the modified FOLFOX6 regimen from May 2005 to December 2009. The patient characteristics are shown in **Table 1**. The patient ages ranged from 39 to 77 years (median 62 years). Nineteen (65.5%) had liver metastases, and 11 (37.9%) had multiple metastatic sites. A total of 446 cycles of treatment were administered with a median of 16 cycles per patient (range 13 to 23 cycles). The median number of tri-weekly mFOLFOX6 cycles administered was 7 (range 5-14). The median cumulative dose of oxaliplatin administered was 1,360mg/m² (range 1,105-1,955mg/m²). After the study, all patients received the FOLFIRI regimen as the second-line chemotherapy in most cases. At a median follow-up of 23 months (**Table 2**), 15 patients were deceased. The median progression-free survival time was 9.4 months.

Toxicity

All patients had peripheral neuropathy during treatment, but grade 3 neurotoxicity was observed in only 2 patients (6.9%). The frequencies of hematologic and non-hematologic toxicity were very low.

DISCUSSION

The current standard first-line chemotherapy for metastatic CRC is the FOLFOX regimen (3,4). Cumulative sensory neurotoxicity is a well-known dose-limiting factor of oxaliplatin (3). Alternative strategies of oxaliplatin-based therapy with decreased cumulative neurologic toxicity, such as the OPTIMOX regimen, have also been examined (6). A high rate of hematological or nonhematological toxicity is observed with the use of the FOLFOX regimen at the conventional biweekly schedule. Prevention or cure is one option for which carbamazepine, gabapentin, calcium, and magnesium have already been investigated (7). However, neuromodulatory agents have shown rather disappointing activity in the prevention of oxaliplatin-induced neurotoxicity (8,9).

We developed a new treatment with the mFOLFOX6 regimen administered biweekly for 10-13 consecutive cycles followed by a 3-week rest period, after which treatment was resumed with cycles of tri-weekly mFOLFOX6 administration at standard doses. The present study showed that the standard mFOLFOX6 treatment, followed by tri-weekly mFOLFOX6 maintenance therapy, was associated with a very low incidence of hematological and non-hematological toxicities, including severe neurotoxicity (3.1%), in 32 evaluable metastatic CRC patients. Furthermore, the median RD (9.2 months) and median PFS (8.6 months) were comparable with those usually observed with the mFOLFOX6 regimen (7). The low incidence of severe neurotoxicity observed in our study was clearly related to the planned tri-weekly treatment schedule. The tri-weekly mFOLFOX regimen is not only a way to decrease neurotoxicity but also a new strategy to administer chemotherapy with advantages in costs and quality of life without a deterioration of survival rates.

CONCLUSIONS

The tri-weekly mFOLFOX6 maintenance chemotherapy was associated with a very low incidence of grade 3 neurotoxicity. RD and PFS were comparable to those usually reported in the treatment of metastatic CRC patients. The tri-weekly maintenance strategy may reduce toxicity and maintain efficacy.

TABLE 1. Patient Characteristics.

Characteristics	No. of Patients (%)
Age Median, years	62
Range, years	39-77
Gender	
Male	14 (48.3)
Female	15 (51.7)
Primary tumor Colon	14 (48.3)
Site Rectum	15 (51.7)

TABLE 2. Response to treatment (n=29).

Complete Response	0 (0%)
Partial Response	17 (58.6%)
Stable Disease	10 (34.5%)
Progressive Disease	2 (6.9%)
Response Rate	58.6%
Time to progression	9.4 months (95% CI 8.2-10.5)
Median survival time	23 months (95% CI 19.6-27.8)

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Tumor Response and Negative Distal Resection Margins of Rectal Cancer after Hyperthermochemoradiation Therapy

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Abstract. *Background: The safety of regional hyperthermia has been tested in locally advanced rectal cancer. The aim of this study was to assess the effects of shorter distal margins on local control and survival in rectal cancer patients who were treated with preoperative hyperthermochemoradiation therapy (HCRT) and underwent rectal resection by using the total mesorectal excision (TME) method. Patients and Methods: Ninety-three patients with rectal adenocarcinoma who received neoadjuvant HCRT (total radiation: 50 Gy) were included in this study. Surgery was performed 8 weeks after HCRT, and each resected specimen was evaluated histologically. Length of distal surgical margins, status of circumferential margins, pathological response, and tumor node metastasis stage were examined for their effects on recurrence and survival. Results: Fifty-eight (62.4%) patients had tumor regression, and 20 (21.5%) had a pathological complete response. Distal margin length ranged from 1 to 55 mm (median, 21 mm) and did not correlate with local recurrence (p=0.57) or survival (p=0.75) by univariate analysis. Kaplan-Meier estimates of recurrence-free survival and local recurrence for the <10 mm versus ≥10 mm groups were not significantly different. Positive circumferential margins and failure of tumors to respond were unfavorable factors in survival. Conclusion: Distal resection margins that are shorter than 10 mm but are not positive appear to be equivalent to longer margins in patients who undergo HCRT followed by rectal resection with TME. To improve the down-staging rate, additional studies are needed.*

Cancer of the lung and bronchus, prostate, and colorectum in men and of the lung and bronchus, breast, and colorectum in women continue to be the most common fatal type of cancer (1). Colorectal cancer alone is expected to account for 9% (26,580) of all male and 9% (24,790) of all female cancer deaths in 2010. More than one-third of colorectal carcinomas occur in the rectum. An important concern in rectal cancer is a high local recurrence rate, as opposed to that in colon cancer. Current guidelines from the National Comprehensive Cancer Network (2) recommend that all patients with clinical stage II/III rectal cancer should be treated with preoperative chemoradiation followed by total mesorectal excision (TME). In locally advanced rectal cancer, the addition of 5-fluorouracil (5-FU) to preoperative radiotherapy has been shown to improve the pathological complete response rate, tumor down-staging, and locoregional control compared with radiotherapy alone (3). Hyperthermia is a procedure that involves heating tissues to a high temperature ranging from 41 to 43°C. This therapy has been combined with radiotherapy and/or chemotherapy for many years, with remarkable success in treating advanced and recurrent cancer. Hyperthermia affects cells in the S phase, inhibits sub-lethal damage repair, and improves oxygenation, making it an attractive therapy to combine with radiation and/or chemotherapy in the hopes of synergy (4). A previous study reported the additional effect of hyperthermia over preoperative radiation alone without any increase in adverse effects (5). Local hyperthermic therapy in combination with radiation has been shown to be less invasive; therefore, the use of local hyperthermia with radiation for local advanced rectal cancer has been recommended as a preoperative therapy.

Sphincter-preserving ultra-low anterior resection (LAR) is preferred to abdominoperitoneal resection (APR) with permanent colostomy for tumors located at least 2 cm proximal of the anal sphincter complex. Previous studies have shown that close distal margins are associated with an increased risk of mucosal recurrence and overall cancer recurrence (6). We have been conducting a clinical trial of regional hyperthermia in

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Key Words: Rectal cancer, hyperthermo-chemo-radiation therapy, HCRT, distal margins, tumor response.

combination with chemoradiotherapy (hyperthermochemo-radiation therapy; HCRT) by using chronochemotherapy (7) for locally advanced rectal cancer (8). The advantages of preoperative HCRT include tumor down-staging, improved resectability, overall survival, and increased anal sphincter preservation (9). The aim of this study was to assess the effects of shorter distal margins on local control and survival in rectal cancer patients who were treated with preoperative HCRT and underwent rectal resection by using the TME method.

Patients and Methods

Patients and therapeutic strategy. Between January 2004 and March 2011, 93 patients with proven rectal adenocarcinoma who underwent HCRT followed by surgery were included in this study. During the diagnostic work-up, all patients underwent staging for distant metastases with computed tomography of the abdomen and thorax. T Stage was determined by magnetic resonance imaging, especially T2-weighted imaging before and after HCRT. The extent and location of the tumor were classified according to the TNM (10).

Preoperative HCRT. All patients in this study underwent preoperative HCRT at the Department of Radiology and Radiation Oncology, Gunma University Hospital. The radiation treatment was delivered by 10-MV x-rays through a three-field box technique. The clinical target volume encompassed the primary tumor and the entire mesorectal tissue. The total radiation dose was 50 Gy, with daily fractions of 2.0 Gy on 5 consecutive days per week. Chemotherapy consisted of 5-FU (250 mg/m^2 per day) and levofolinate calcium (25 mg/m^2 per day) administered by continuous infusion at night for 5 days a week in the 1st, 3rd, and 5th weeks of radiation. Two to five hyperthermia sessions were performed once a week with 8 MHz radiofrequency capacitive heating equipment (Thermotron-RF 8, Yamamoto Vinita Co., Ltd., Japan).

Surgery and postoperative therapy. Rectal resection was performed using the principles of TME 8 weeks after the completion of HCRT. A complete 6-month course of adjuvant 5-FU-based chemotherapy was typically recommended for all medically fit patients completing HCRT and curative surgery. The majority of the patients received oral 5-FU/leucovorin.

Pathology. Each resected specimen was examined for histological changes after HCRT according to the histological criteria of the Japanese Classification of Colorectal Carcinoma (11). Grades were assigned according to the amount of necrosis, degeneration, and lytic change of the tumor in the estimated total amount of the lesion (12). Grading of the histopathological response was performed by pathologists. The distal margin was defined as the gross distance between the distal edge of the tumor or post-treatment fibrosis, if present, and the distal mucosal resection margin.

Results

Patients' characteristics. Ninety-three patients with lower third rectal cancer were included in this study; the median age was 64 years. Sixty-eight patients were males, and 25 patients were females, with a male-to-female ratio of 2.7: 1. Patient characteristics are shown in Table I. All patients tolerated this regimen without hematological toxicity. The non-hematological toxicities observed were diarrhea in one patient and anorexia in one patient, both with grade 3 cancer. When the clinical pretreatment stage was compared with the pathologic results, down-staging of the T and N stages was possible in 51 (45.2%) and 54 patients (58.1%), respectively. The overall down-staging rate, including both the T and N stages, was 63.4% (59/93). Significant down-staging estimated in the primary lesion, in which tumors were undetectable by MRI and colonoscopy and negative results were obtained from biopsies, occurred in 47 patients (50.5%). Anterior resection was performed with colorectal anastomosis using a double-stapler device in 55 (59.1%) patients and with coloanal anastomosis using the hand-sewn technique in 21 (22.6%) patients. Abdominoperineal resection was performed in 17 (18.3%) patients. The overall sphincter preservation rate in the present study was 81.7% (76 out of 93 patients).

Table I. *Patients' characteristics.*

Characteristic	No. of cases (%)
Age, years	
Median (range)	64 (43-85)
Gender	
Male	68 (73.1)
Female	25 (26.9)
Stage	
II	39 (41.9)
III	50 (53.8)
IV	4 (4.3)
Surgical technique	
APR	17 (18.3)
LAR	76 (81.7)
Sphincter-preserving rate	81.7%

APR, Abdominoperitoneal resection; LAR, low anterior resection.

The pathological diagnoses obtained from surgical specimens are shown in Table II. Pathological complete response (pCR) of the primary tumor (Figure 1) and lymph nodes on the pathological specimen was observed in 20 patients (21.5%), and one patient showed pCR of the primary tumor but had residual tumor cells in the regional node. All patients had pathologically negative distal resection margins. Patients with pCR after HCRT had no better outcome than did those without pCR (Table II). pCR after HCRT might not be indicative of a prognostically favorable biological tumor profile. For the LAR specimen, the distal resection margins ranged from 1 to 55 mm (median 21 mm), not including the anastomotic staple rings. Fourteen (15.1%) patients who underwent LAR had distal resection margins of <10 mm. Seven (7.5%) patients were found to have positive circumferential margins with the pelvic side wall.

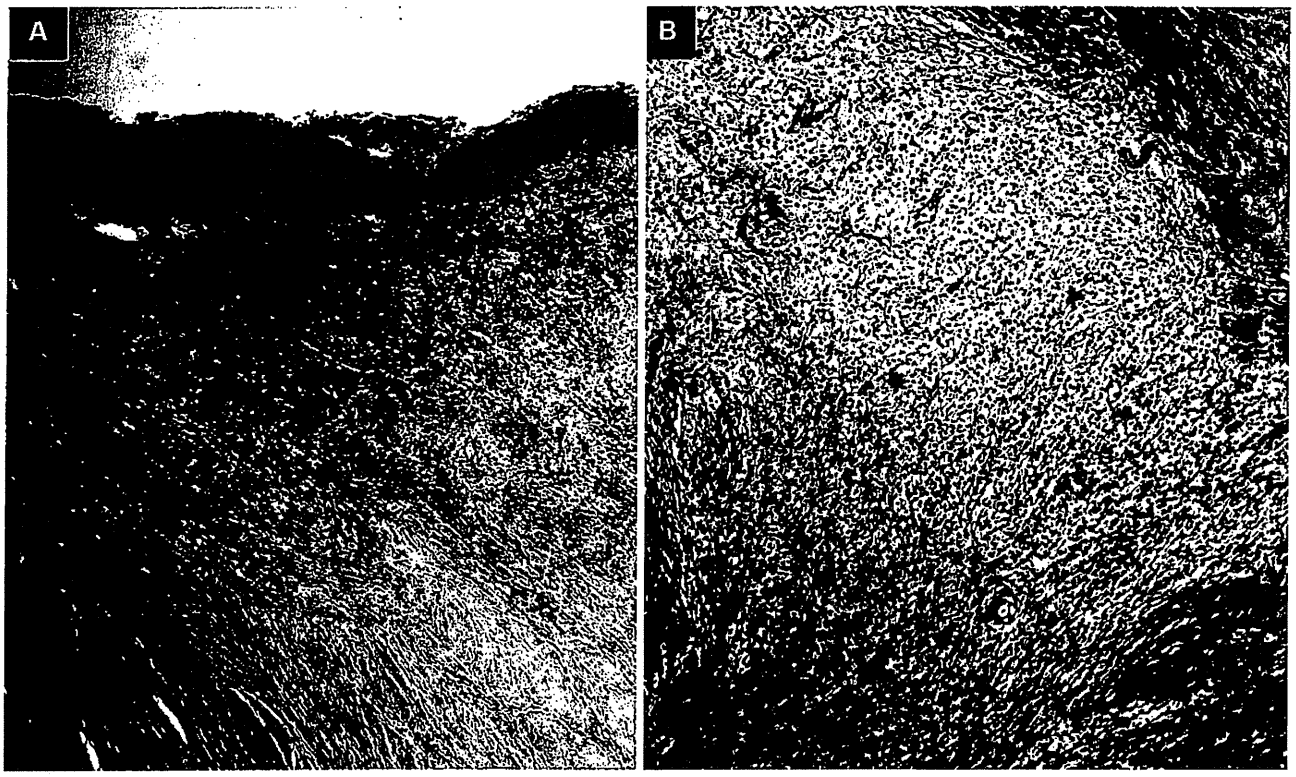

Figure 1. *Microscopic evaluation of tumor response. A: Low magnification (×25) image showing the ulcer at the tumor bed site. B: Higher magnification (×100) image deep in the wall of the fibrosis shows a high number of lymphocytes and fibroblasts scattered throughout the scar tissue.*

The median follow-up was 37 months (range 4-81 months) for all patients, and 84 patients were still alive at the time of writing. A distal margin length shorter than 10 mm did not correlate with local recurrence (p=0.57) or survival (p=0.73) by univariate analysis. Kaplan-Meier estimates of recurrence-free survival and local recurrence for the <10 mm *versus* >10 mm groups were not significantly different (Figure 2). However, positive circumferential margin and down-staging were related to overall survival (Figures 3 and 4).

Discussion

Whether LAR or APR is performed, ensuring that the intact rectum and mesorectum are removed with clear surgical margins is immensely important to prevent the local recurrence of rectal cancer. Preoperative chemoradiation can potentially increase the feasibility of sphincter-preserving resections by reducing the tumor volume and by defending against local tumor extensions (13). Given the additional effects of hyperthermia on chemoradiation therapy, HCRT may compensate for the narrow circumference and distal resection margins. The rate of

Table II. *Comparison of clinicopathologic variables.*

Distal resection margin	<10 mm n=14	≥10 mm n=79	p-value
Local recurrence	1 (7.1%)	3 (3.8%)	0.5745
Survival	13 (92.9%)	71 (89.9%)	0.7313
Pathologic circumferential resection margin	**– n=86**	**+ n=7**	**p-value**
Local recurrence	3 (3.5%)	1 (14.3%)	0.1795
Survival	79 (91.9%)	5 (71.4%)	0.029
Pathologic complete response	**pCR (–) n=73**	**pCR (+) n=20**	**p-value**
Local recurrence	4 (2.3%)	0 (0%)	0.2896
Survival	70 (91.9%)	20 (71.4%)	0.1006
Down-staging	**– n=34**	**+ n=59**	**p-value**
Local recurrence	3 (8.8%)	1 (1.7%)	0.1049
Survival	27 (79.4%)	57 (96.6%)	0.0187

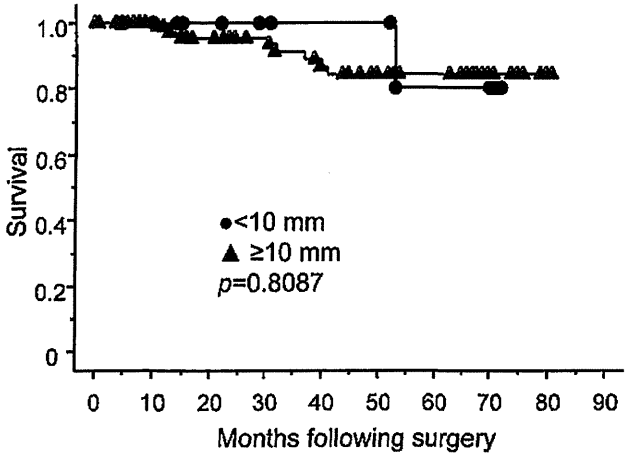

Figure 2. *Kaplan-Meier distribution of overall survival for patients with <10 mm distal margins versus those with ≥10 mm distal margins.*

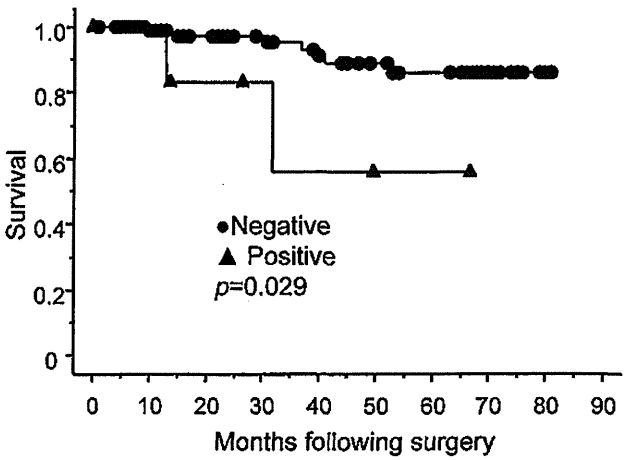

Figure 3. *Kaplan-Meier distribution of overall survival for patients with negative circumferential margins versus those with positive circumferential margin.*

pCR after neoadjuvant chemoradiation therapy ranged from 10 to 16% in various series examined in a review (14). Recent efforts incorporating newer cytotoxic and molecularly targeted agents into chemoradiation therapy regimens have been reported. Large randomized trials showed that the addition of weekly oxaliplatin to fluoropyrimidine-based chemoradiation led to an increase in grade 3/4 toxicity but no difference in pCR rates. Early phase trials evaluating the anti-epidermal growth factor receptor antibody cetuximab in combination with chemoradiation reported modest pCR rates of 5 to 12%. In this study, the pCR rate was 21.5%, which compares favorably with that observed in the other reports. Moreover, our study showed a greater reduction of adverse effects through the use of chronochemotherapy (8).

Tumor distance from the anal verge was significantly greater after HCRT, thus it was possible to carry out sphincter-preserving surgery in a larger proportion of the patients. The close distal resection margin, even if it was shorter than 10 mm, was not related to local recurrence and survival in this study. A positive circumferential margin was also not related to local failure (Table II). Obtaining negative distal and circumferential margins remains a goal of rectal cancer surgery after HCRT. The malignant potential and behavior of tumor after HCRT might be different from the pretreatment status. Further investigations are needed in order to archive better individual oncologic results.

Down-staging in this study was a good predictor of outcome. Down-staging oncological treatment has not been viewed as an additional therapy, and there is no evidence-based protocol to follow if the tumor fails to regress or increases in size after HCRT. Trials of chemotherapy with

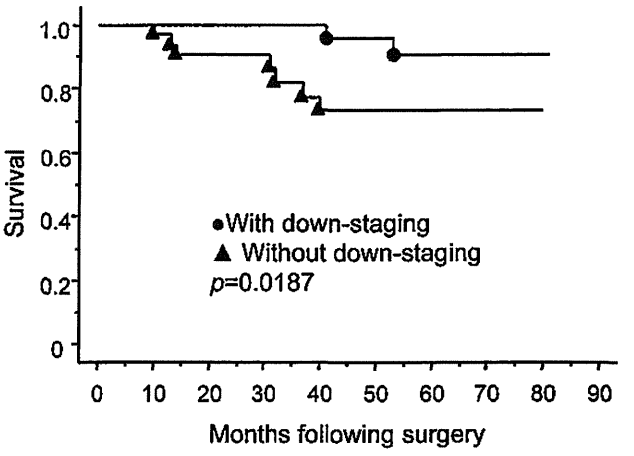

Figure 4. *Kaplan-Meier distribution of overall survival for patients with down-staging versus those without down-staging.*

new biologic agents in a preoperative setting for patients whose disease fails to down-stage after HCRT are needed. New techniques for rectal cancer surgery also need to be investigated. However, a prospective randomized study that inspects the adequacy and safety of the distal and circumferential resection margins would be difficult to set up. The limitations of our study include the small number of patients and the short follow-up period. The results of our study are encouraging in terms of the rate of down-staging, pCR, and sphincter-preserving surgery. The favorable results of our study might be due to additional hyperthermia with chemoradiation therapy. Further investigations to improve the down-staging rate of HCRT for rectal cancer are required.

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Received August 4, 2011
Revised September 24, 2011
Accepted September 27, 2011

日本消化器外科学会雑誌. 2011;44(9):1105-1110

メシル酸イマチニブによる術前化学療法を施行しpCRが得られた胃原発GIST局所再発の1切除例

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尾辻　英彦[1)]　前田　隆雄[1)]　田中　寛[1)]　吉田カツ江[2)]
待木　雄一[1)]　桑野　博行[4)]

症例は78歳の男性で，2004年11月，胃gastrointestinal stromal tumor（以下，GISTと略記）に対して噴門側胃切除術を施行した．免疫組織学的検査でKITおよびCD34が陽性，核分裂像が高度であり高リスクのGISTと診断されたため，術後より1年9か月の間，イマチニブ(400mg/日)の投与を行った．2008年11月の腹部造影CTで残胃に局所再発を認めたため，イマチニブの投与を再開した．投与3か月後の腹部造影CTでは腫瘍の縮小を認め，再手術を施行した．切除標本の病理組織学的検査では，腫瘍細胞は認めず，血管腫様の組織を認めるのみでpathological complete response（以下，pCRと略記）と判断した．横隔膜と肝外側区への浸潤を伴う再発胃GISTに対して，イマチニブによる術前化学療法を施行後に切除し，pCRが得られた希少な症例を経験したため報告する．

はじめに

GISTは消化管に発生する間葉系腫瘍のうち，平滑筋細胞，神経細胞への分化を示さず，免疫染色検査でKIT陽性を示すものとされる[1)]．

これまで治療抵抗性であったGISTの再発例，転移例に対して，現在はメシル酸イマチニブ(以下，イマチニブと略記)の投与が第一選択となっている[2)]．イマチニブは高い奏効率が報告されているが，単独での治癒は困難であり，長期投与による耐性や休薬後の腫瘍増大などの問題も指摘されている．そのため，切除可能な場合には外科的切除が必要と考えられ，外科的治療の介入によって，治療効果の向上が期待される．今回，我々は局所再発を来たした胃GISTに対してイマチニブを投与後に切除し，pCRが得られた症例を経験したので報告する．

症　例

患者：78歳，男性

主訴：特になし

既往歴：大腸癌．

現病歴：2004年11月，穹隆部の胃GISTに対して噴門側胃切除術（食道残胃吻合）を施行した．腫瘍径は13cmで，病理組織学的検査では紡錘形細胞の増殖を認め，免疫組織化学的検査ではKITおよび

＜2011年2月16日受理＞別刷請求先：木村　明春　〒376-0024　桐生市織姫町6—3　桐生厚生総合病院外科

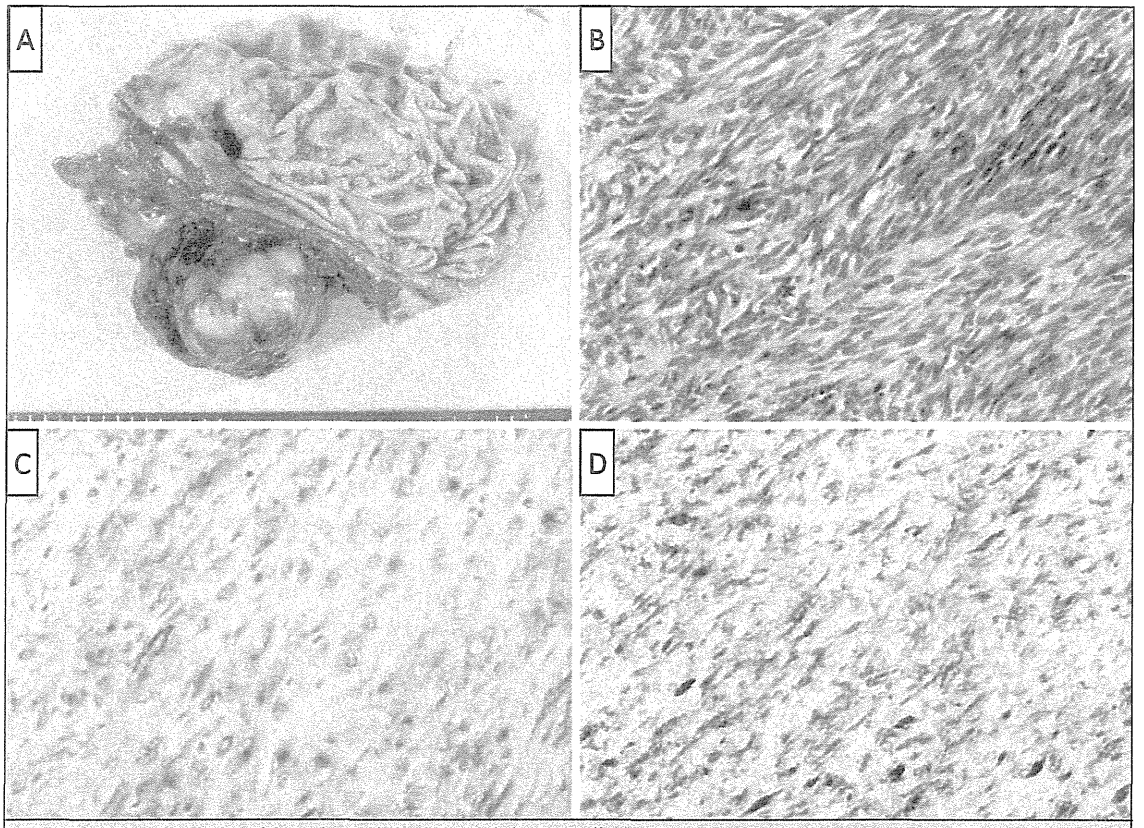

Fig. 1 Macroscopic and histological findings of the initially resected tumor of the stomach. A: The tumor measured 13 cm in diameter. B: Proliferation of spindle cell. C and D: The tumor cells were positive for c-kit (C) and CD34 (D).

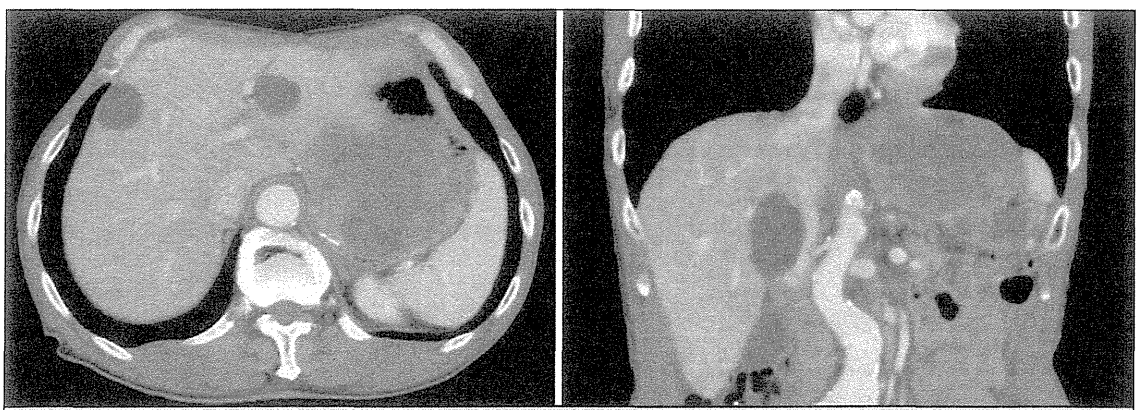

Fig. 2 Abdominal enhanced CT revealed local recurrence of GIST, invaded lateral segment of the liver and the diaphragm.

CD34 が陽性であった(**Fig. 1**). また核分裂像も高度であり, 高リスクの GIST であったため, 術後から 1 年 9 か月の間, イマチニブ (400mg/日) の投与を行った.

以後, 定期的に外来で経過観察を行い, 2008 年 3 月に撮影した CT では明らかな再発所見を認めなかった. 術後より約 4 年経過した 2008 年 11 月の腹部造影 CT で残胃に腫瘍性病変を認めた. 腫瘍は径 8.5×7.2cm で前回手術時の staple line を含め残胃を中心に存在しており, 局所再発と診断した. 腫瘍は肝外側区域および食道裂孔を含め左横隔膜へ広範に浸潤陽性と診断された (**Fig. 2**). 上部消化管内視鏡検査では胃体部を中心に粘膜下腫瘍様の隆起を認め, 胃内腔を圧排していた. 根治切除を行うには広範囲

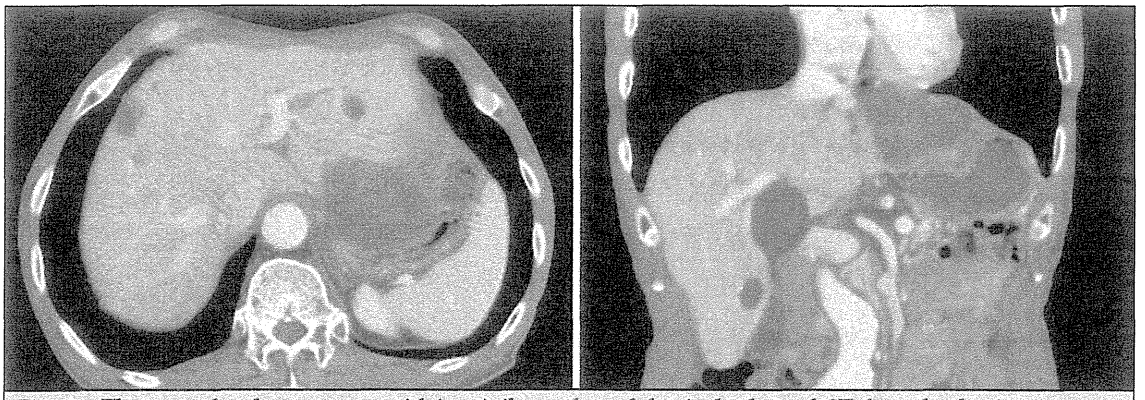

Fig. 3 Three months after treatment with imatinib mesylate, abdominal enhanced CT showed reduction in tumor size.

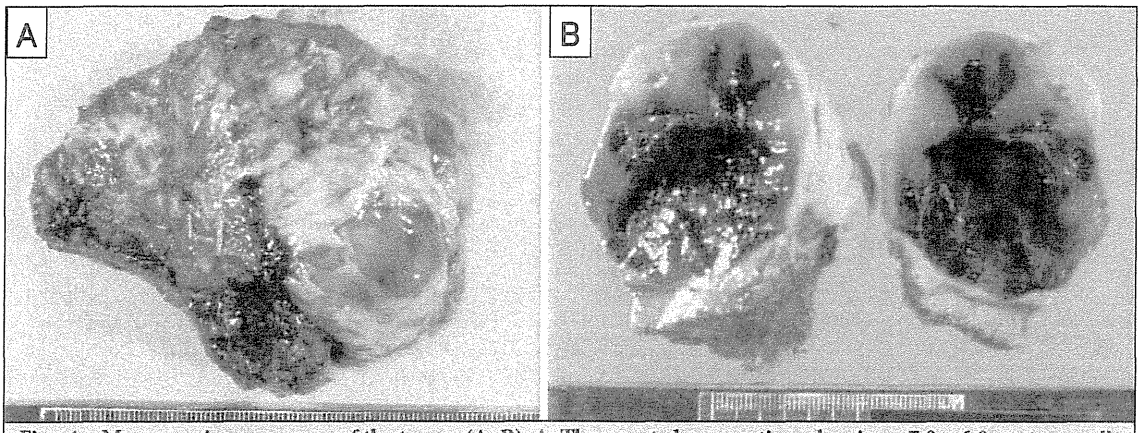

Fig. 4 Macroscopic appearance of the tumor (A. B). A: The resected preparation, showing a 7.0×6.0 cm tumor. B: The cut surface of the tumor is dark reddish like an old hematoma.

の横隔膜の合併切除が必要であると思われ，侵襲が大きいと考えられた．術前化学療法を行う方針とし，イマチニブ（400mg/day）を約 2 か月間投与した．2009 年 2 月の腹部造影 CT では腫瘍は縮小し，横隔膜への浸潤範囲も縮小していた（**Fig. 3**）．この時点で切除可能と判断し，手術目的に入院となった．

現症：体温 36.7℃，血圧 124/80mmHg，脈拍 64/分，腹部には腫瘤を触知せず，正中部に前回手術痕を認めた．

血液生化学検査：異常所見は認めず，腫瘍マーカーの上昇もみられなかった．

腹部造影 CT 所見：腫瘍は 7.5×6.0cm と縮小を認め，腫瘍内の造影効果も減弱していた（**Fig. 3**）．横隔膜への浸潤範囲も縮小していた．画像上 PR，切除可能と判断した．

イマチニブの効果は PR であったが，今後の投与継続による副作用や耐性の可能性も考えられたため切除する方針とした．

手術所見：腫瘍は肝外側区，食道胃吻合部付近を主座としていた．肉眼的に横隔膜への浸潤は認めなかった．残胃部分切除術，肝外側区域切除術を施行した．

切除標本肉眼所見：腫瘍は被膜を有する 7.0×6.0cm の充実性の腫瘤であった．割面では全体に暗赤色調を呈しており，典型的な GIST に見られるような黄白色調ではなかった（**Fig. 4**）．

病理組織学的検査所見：GIST 細胞は認めず，血管腫様組織が主体であった．viable cell は認めず，pCR と判定された（**Fig. 5**）．また遺伝子解析は行わなかった．

術後経過：術後にイマチニブの投与は行わず，外来にて経過観察中であるが，術後 1 年経過時点で再発は認めていない．

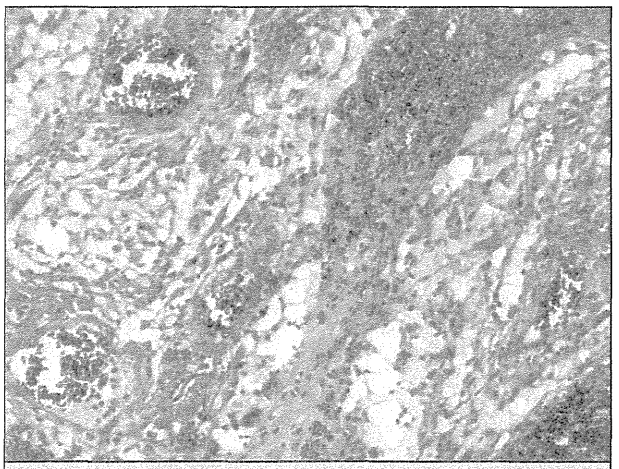

Fig. 5 Histopathological findings: Most of the tumor was replaced by angiomatoid tissue, and no viable tumor cells were detected.

考　察

本邦での GIST 診療ガイドライン[2]によると，再発・転移 GIST に対する治療の原則はイマチニブ投与である．再発 GIST に対しては，以前は再手術しか有効な治療法がなく，切除不能であった場合の予後は非常に不良であった．欧米で行われたイマチニブの第 II 相臨床試験[3]によると，奏効率で 53.7%，SD を含めた病勢コントロール率で 81.6% と，高い抗腫瘍効果が報告されている．また同試験の遺伝子解析では，イマチニブの抗腫瘍効果は c-kit 遺伝子変異部位により異なることが報告されており，エクソン 11 に変異がみられる症例では奏効率は 84% と最も高く，エクソン 9 の変異では 48% であった．

しかしながら，国内外の臨床試験の結果でもイマチニブ治療単独での CR 例はほとんどなく，イマチニブの投与により腫瘍のサイズが縮小しても，病理組織学的には viable な腫瘍細胞が一部に残存しているものと考えられている[4]．イマチニブの休薬により，早期の増悪が報告されており，病勢のコントロールにはイマチニブを可能なかぎり長期に継続することが必要である．その一方で，イマチニブ投与による副作用や二次耐性の出現により，イマチニブの継続が困難となる場合も少なくない．

近年では，再発 GIST に対しイマチニブを投与後に再切除を行い，良好な結果を得ている報告も散見される[4)~6)]．NCCN の GIST 治療ガイドライン[7]では，再発 GIST に対する外科的治療の原則として，イマチニブに反応のある GIST や，イマチニブで進行が停止し安定化した GIST，あるいは効果がなく進行する GIST で残存腫瘍が完全切除可能な場合には，外科的手術も適応となるとしている．

Rutkowski ら[8]や Gronchi ら[9]は，切除不能，転移再発 GIST に対するイマチニブ投与後の腫瘍切除について，イマチニブ奏効中の腫瘍切除が予後良好であり，腫瘍がイマチニブ耐性を獲得し病勢コントロールが得られない状態での手術では，切除率，生存率がともに低下し，予後不良になると報告している．

本症例も，局所再発を来した胃 GIST に対してイマチニブの投与を行い，病勢のコントロールを行ったうえで手術を施行し，残存腫瘍の完全切除を施行することができた．切除標本の病理組織学的検討では，腫瘍部には血管腫様の組織がみられるのみで，GIST の viable cell は認めず pCR と判断した．切除標本上 GIST 細胞は認められなかったが，イマチニブ投与により画像上縮小が得られたことから本症例は GIST の再発に矛盾しないと考えた．

イマチニブは腫瘍細胞の増殖を抑制し，アポトーシスを誘導するため，腫瘍細胞は嚢胞化や硝子様変性を起こすとされている[4]．本症例ではそのような変化は認めなかったが，血流の豊富な腫瘍細胞の中で血管が増生し，イマチニブの抗腫瘍効果により腫瘍細胞が消退，その結果栄養血管のみが残り，血管腫

Table 1 Reported cases of pathological complete response obtained after neoadjuvant chemotherapy by imatinib mesylate in Japan

Case	Author Year	Age Sex	Initial therapy	Recurrence	Tumor size before administration of IM (cm)	Response to treatment	Operation after administration of IM	Pathological findings
1	Abeshima[10] 2004	66 M	Administration of IM	None	Over fist size	PR	Total gastrectomy Left lobectomy of the liver	Hyaline degeneration
2	Nojiri[5] 2005	52 M	Partial gastrectomy	Local Liver	4 (Local), 7 (Liver)	SD	Proximal gastrectomy Partial hepatectomy	Hyaline degeneration
3	Koeda[6] 2007	56 M	Total gastrectomy	Spleen	4×4	PR	Splenectomy	Myxoid degeneration
4	Our case	78 M	Proximal gastrectomy	Local	8.5×7.2	PR	Partial gastrectomy Partial hepatectomy	Angiomatoid change

IM: imatinib mesylate

様の所見を呈したものと考えられた.

切除後の治療については, Rutkowski ら[8]や Gronchi ら[9]のいずれもイマチニブの投与継続が重要で, 術後できるだけ早期にイマチニブを再開することが必要であるとしている. 本症例は pCR が得られたため, 手術後のイマチニブ投与は行っていないが, 現在まで再発を認めていない.

胃 GIST に対して術前化学療法を行い切除した症例において pCR が得られることはまれである.「胃 GIST」,「メシル酸イマチニブ」をキーワードに医学中央雑誌刊行会 Web 版で 1983 年から 2010 年まで検索すると, 再発例も含めた胃 GIST に対して術前化学療法を施行後に手術を行った報告は 15 例であった. このうち切除標本で pCR を確認したのは 3 例[5][6][10]のみであった(会議録を除く). 表に示すように自験例を含めた 4 例の検討では, 安部島ら[10]が報告した 1 例は初回治療にイマチニブを使用しているが, その他の 3 例は手術後の再発例に対してイマチニブを使用している (Table 1). 本症例では初回切除後に 1 年 9 か月間イマチニブを投与しているが, 術後約 4 年で局所再発を来したため, イマチニブの投与を再開した. 本症例は再発時の腫瘍径も大きく, 横隔膜や肝外側区への浸潤も見られたが, イマチニブによる術前化学療法が奏効し手術可能となり, 切除標本で pCR が得られた希少な症例であると考えられた.

なお, 本論文の要旨は第 71 回日本臨床外科学会 (京都) で発表した.

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CASE REPORT

A Resected Case of Local Recurrence of GIST of the Stomach in which Pathological complete Response after Neoadjuvant Chemotherapy by Imatinib Mesylate

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A 78-year-old man underwent proximal gastrectomy for gastrointestinal stromal tumor (GIST) of the stomach in November 2004.The pathological diagnose was GIST, and it was positive for KIT and CD34 immunohistochemically, indicating it was in a high risk group. Administration of imatinib mesylate at a dose of 400 mg/day was given for 21 months after surgery as adjuvant chemotherapy. In November 2000, abdominal computed tomography (CT) revealed local recurrence of GIST invading the liver and diaphragm making curative resection difficult, so administration of imatinib mesylate was restarted. Three months after reinitiating imatinib mesylate treatment,abdominal CT showed reduction in tumor size. Therefore,we judged this lesion to be resectable and performed local resection. Histopathologically, the tumor was replaced by angiomatoid change,and no viable tumor cells were detected. Pathological complete response (pCR) was obtained. This was a rare case, in which local recurrence of GIST invading the liver and diaphragm was resected after neoadjuvant chemotherapy by imatinib mesylate, and pathological complete response was obtained.

Key Words: GIST, imatinib mesylate, neoadjuvant therapy

〔Jpn J Gastroenterol Surg. 2011;44(9):1105-1110〕

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Accepted: February 16, 2011

食道癌に対する化学療法後の手術

Esophagectomy after chemotherapy

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【ポイント】

◆ 切除不能食道癌は非常に予後不良である．

◆ 食道扁平上皮癌はほかの固形癌に比べて抗癌剤や放射線への感受性が高い．

◆ 切除困難症例を切除可能症例にするには補助療法が必要である．

臨外　67(1)：18～24，2012

はじめに

食道癌の治療成績は徐々に向上してきている．これは，診断や治療を含めた内視鏡技術の進歩や検診の普及などで比較的早期に癌が発見される割合が増えていることや，また，周術期管理の向上や頸部・胸部・腹部3領域の徹底したリンパ節郭清を中心とした手術手技の向上が背景にある．さらに，近年では術前または術後に補助化学療法を行うことで，さらに治療成績の向上をもたらしている[1,2]．一方，食道癌はほかの消化器癌に比べて悪性度が高く，他臓器浸潤や広範なリンパ節転移を伴った高度進行食道癌で発見されることもいまだ少なくない．腫瘍自体が他臓器浸潤のある症例（T4）や所属リンパ節を介した他臓器浸潤を認める場合は，相手臓器が容易に合併切除可能な臓器に限りT3症例に準じた手術適応を決定するが，気管・気管支や大血管への浸潤が認められる場合には一般に化学療法や放射線療法もしくは化学放射線療法が優先される．しかしながら，わが国の報告においてT4食道癌の治療成績は，手術症例において5年生存率が11.9%[3]，化学放射線療法の効果を検証した第Ⅱ相臨床試験（JCOG9516）においてもcomprete response（CR）率が15%，2年生存率が32%であり[4]，決して十分な治療成績とはいえない．そこで，局所進行食道癌においては，T4が解除できた時点で手術療法へ移行するか，根治的な非手術療法を継続するかの検討がなされる[5]．

本稿では，局所進行によって切除困難な食道癌の治療戦略について概説する．

局所進行食道癌に対する治療戦略

「食道癌診断・治療ガイドライン」（2007年4月版）[6]の治療アルゴリズムでは，進行食道癌stageⅢ（T4）・Ⅳaに対しては化学放射線療法もしくは放射線療法，遠隔臓器転移を伴うstageⅣbに対しては化学療法，放射線療法もしくは切除不能症例の治療と記載されており（図1），他臓器合併切除が可能な一部の症例を除いて初回治療として手術療法が選択されることはほとんどない．遠隔臓器転移がなく，腫瘍が気管・気管支や大動脈などの食道周囲臓器へ浸潤した症例（T4）や所属リンパ節転移を介した他臓器浸潤を認める場合は，癌が局所にとどまっていることから根治的化学放射線療法の適応となる．また，TNM分類でM1（LYM）に相当する可動性のない鎖骨上窩のリンパ節転移，もしくは腹腔動脈周囲リンパ節転移などが認められる症例も一般的に切除不能であり，かつ癌が局所（照射可能範囲）にとどまっていることから根治的化学放射線療法の適応となる．ただし，頸部食道癌では気管浸潤

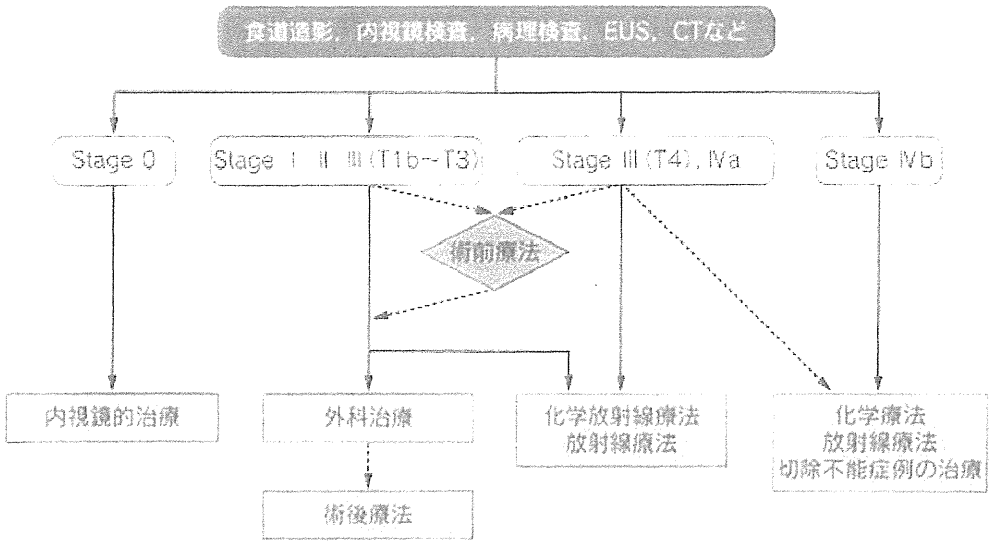

図1　食道癌治療アルゴリズム

（文献6より転載）

（T4）が認められた場合でも合併切除が可能であるため，手術療法が行われることもある．一方，腹部大動脈周囲リンパ節転移や遠隔臓器転移が認められる症例，通過障害が高度でなく頸部・胸部・腹部の3領域リンパ節転移が認められる症例に対しては全身化学療法が施行されることが一般的である．

切除困難（不能）食道癌に対する，手術を前提とした化学療法または化学放射線療法（induction therapy）後の食道切除の治療成績を検討した大規模試験は主に海外から報告されている．術前化学放射線療法＋手術と根治的化学放射線療法を比較するrandomized controlled trial（RCT）として，Stahlら[7]はcT3-4，N0-1，M0の食道扁平上皮癌患者を対象にinduction chemotherapy（5-FU＋ロイコボリン＋エトポシド＋シスプラチン）後に術前化学放射線治療（40 Gy）＋手術をする群と，根治的化学放射線治療（65 Gy以上）を施行する群の比較を行った．また，Bedenneら[8]はcT3，N0-1，M0の胸部食道癌患者を対象に化学放射線療法（5-FU＋シスプラチン＋照射30～46 Gy）後の奏効例に対して，手術群と根治照射まで化学放射線療法を継続する群とで比較試験を行っている．これらの試験の結果では，術前化学放射線が奏効した症例では化学放射線の継続が手術とほぼ同等の成績であるが，局所制御の点では術前化学放射線療法＋手術群のほうが優れているとの結果であった．

また，切除不能症例とは限らないが，術前に補助療法として化学療法あるいは化学放射線療法を行うことの有効性を検討した臨床試験が海外から報告されている[9]．術前化学療法による生存率改善効果を検討したRCTは9試験報告されており，そのうち生存率改善に寄与すると報告されたのは2編である．また，術前化学放射線療法による生存率改善効果を検討したRCTは7試験あり，そのうち生存率改善に寄与すると報告されたのは1編のみであった．しかしながら，これらの報告では手術単独での治療成績がわが国に比べて低く，頸部・胸部・腹部3領域リンパ節の徹底郭清が基本術式となっているわが国と，手術による局所制御を含めた治療効果の点で相違がみられる．また，これらのRCTでは扁平上皮癌と腺癌などの組織型の違いや，化学療法のレジメン，照射線量なども各試験で異なり，その検証に関しては慎重に判断する必要があると思われる．

一方，わが国においては，切除可能であるstage Ⅱ/Ⅲを対象とした臨床試験（JCOG9906）の結果において，根治的化学放射線療法（5-FU＋シスプラチン＋照射60 Gy）施行後の治療成績がCR率68%，3年生存率46%と報告されている[10]．この試験の結果は，手術関連死亡率が約10%とされるサルベージ手術を含んだ治療成績であり，単純にその成績を評価することはできないが，その高いCR率を考慮すると，切除困難症例に対するinduction therapyとして，照射線量の設定などの問題は含むが有用なプロトコールの1つとして挙げられると思われる．

Brain and Behavior

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Oxaliplatin-induced neurotoxicity involves TRPM8 in the mechanism of acute hypersensitivity to cold sensation

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Keywords
Menthol, neurotoxicity, oxaliplatin, transient receptor potential melastatin 8.

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Received: 20 September 2011;
Accepted: 14 December 2011

doi: 10.1002/brb3.34

Abstract

Oxaliplatin-induced peripheral neurotoxicity (OPN) is commonly associated with peripheral hypersensitivity to cold sensations (CS) but the mechanism is unknown. We hypothesized that the transient receptor potential melastatin 8 (TRPM8), a putative cold and menthol receptor, contributes to oxaliplatin cold hypersensitivity. To determine whether the TRPM8 is involved in acute OPN, varying concentrations of menthol were topically applied to the tongues of healthy subjects ($n = 40$) and colorectal cancer patients ($n = 36$) before and after oxaliplatin administration. The minimum concentration of menthol to evoke CS at the menthol application site was determined as the CS detection threshold (CDT). In healthy subjects, the mean CDT was 0.068. Sex and age differences were not found in the CDT. In advanced colorectal cancer patients, the mean CDT significantly decreased from 0.067% to 0.028% ($P = 0.0039$) after the first course of oxaliplatin infusions, and this marked CS occurred in patients who had grade 1 or less neurotoxicity, and grade 2 neurotoxicity, but not in those with grade 3 neurotoxicity. Further, the mean baseline CDT in oxaliplatin-treated patients was significantly higher than that of chemotherapy-naïve patients and healthy subjects (0.151% vs. 0.066%, $P = 0.0225$), suggesting that acute sensory changes may be concealed by progressive abnormalities in sensory axons in severe neurotoxicity, and that TRPM8 is subject to desensitization on repeat stimulation. Our study demonstrates the feasibility of undertaking CDT test in a clinical setting to facilitate the identification of early neurotoxicity. Moreover, our results indicate potential TRPM8 involvement in acute OPN.

Introduction

Oxaliplatin-induced peripheral neurotoxicity (OPN) is deleterious to patients both in terms of troublesome symptoms and the need to reduce or discontinue chemotherapy (Adelsberger et al. 2000). Oxaliplatin, a third-generation platinum analog, causes a unique spectrum of acute peripheral nerve hyperexcitability that has not been observed in patients receiving other platinum chemotherapeutic agents. Conversely, chronic oxaliplatin treatment induces an axonal neuropathy that is similar to that observed with other platinum-based compounds (Lehky et al. 2004). In clinical studies, approximately 90% of oxaliplatin-treated patients experienced unique acute OPN, particularly cold-induced paresthesia that is usually triggered by cold exposure and begins in the hands or feet but sometimes occurs around the mouth or in the throat (Raymond et al. 1998a; Raymond et al. 1998b; Grothey, 2003; Ali 2010;). It is an acute transient syndrome that may begin during drug infusion or within minutes, hours, or 1–2 days after administration but is usually self-limiting, often disappearing within a few days (Gamelin et al. 2002, 2006).

Recently, a wide repertoire of sensory transduction molecules that convert external environmental stimuli into neural activity has been identified (Basbaum et al. 2009). For example, the transient receptor potential (TRP) family of ion channels are the primary detectors of thermal stimuli (Jordt et al. 2003), and TRP melastatin 8 (TRPM8) determines whether temperatures are considered cool or cold

(McKemy et al. 2002; Peier et al. 2002; Daniels and McKemy 2007). However, to date, there is no evidence that TRPM8 is involved in the mechanisms of acute OPN.

Menthol, a potent TRPM8 agonist, has long been known to induce or intensify cold sensations by interacting with the peripheral cold receptor, TRPM8 (McKemy et al. 2002; Peier et al. 2002; Knowlton et al. 2010). The tongue is a well-characterized sensory organ, and TRPM8 is present in sensory lingual nerve fibers that mainly project from the trigeminal ganglion where they function as cold and menthol receptors on the tongue (Abe et al. 2005).

On the basis of these observations, we hypothesized that TRPM8 is involved in the mechanisms of acute OPN, especially marked sensitivity to cold. We tested this hypothesis by topically applying varying concentrations of menthol, a TRPM8 agonist, to the patients' tongue before and after oxaliplatin infusions to determine their sensitivity to cold sensation. The minimum concentration of menthol to evoke cold sensation (CS) at the menthol application site was determined as the cold sensation detection threshold (CDT).

The conventional clinical grading system was used to assess the severity of neurotoxicity in relation to CDT. Patients also completed self-report ratings of their sensitivity to cold sensation, and the results of these objective and subjective findings were compared.

Materials and Methods

Subjects and treatment regimen

A total of 76 subjects were enrolled in this study: 40 healthy subjects (24 women, 16 men; median age, 54 years; range, 22–85) and 36 patients (22 women, 14 men; median age, 57 years; range, 33–80) with advanced-stage colorectal cancer who received standard oxaliplatin in combination with infusional 5-fluorouracil/leucovorin (FOLFOX) as a first-line treatment. In the FOLFOX regimens, oxaliplatin (modified FOLFOX 6, 85 mg/m^2) was given intravenously over 2 h on day 1 in conjunction with leucovorin (200 mg/m^2) and followed by a 5-fluorouracil (5-FU) bolus injection (400 mg/m^2), repeated every 2 weeks. A continuous 24-h infusion of 5-FU (600 mg/m^2) was given over days 1 and 2. On day 2, leucovorin (200 mg/m^2; over 2 h) and 5-FU bolus (400 mg/m^2) were given intravenously.

The subjects did not consume any spicy food 1 day prior to testing. They were also asked to refrain from eating, drinking, chewing gum, brushing their teeth, and using mouthwash for 2 h before testing, and we verified that the participants had observed these restrictions at the beginning of each session.

The present study was conducted in accordance with the Declaration of Helsinki for the care for human studies adopted by the Ethics Committee of Higashi-Asahikawa Hospital. All patients provided written informed consent.

Assessment of menthol in experiments 1 and 2

A solution of 5% L-menthol (from dry crystals; MERCK, Tokyo, Japan) was prepared in warm distilled water (41°C) at the time of application, and this solution was further diluted in warm distilled water to yield menthol solutions of 0.005%, 0.01%, 0.05%, 0.1%, 0.5%, and 1% (0.32 mM, 0.64 mM, 3.2 mM, 6.4 mM, 32 mM, and 64 mM, respectively). These solutions were topically applied with a cotton swab to the dorsal anterior tongue in two experiments (Fig. 1a). In experiment 1, the six different menthol solutions were administered to healthy subjects and patients with colon cancer prior to oxaliplatin exposure, and their subjective ratings of cold sensitivity were recorded. In experiment 2, patients were examined for alterations in the menthol-induced cold sensations before and 5–6 h after the patients receiving individual oxaliplatin infusions. The menthol concentrations used in this study were based on a previous human study (Albin et al. 2008). The vehicle control (warmed distilled water) was applied in the same manner.

Both experiments were performed in a room maintained at a constant temperature (22 ± 1°C) and a relative humidity of 55 ± 5%. The menthol testing was performed by two investigators (TK and MS) on all participants. Neither the individuals nor the investigator were aware of whether menthol or the vehicle was applied first because the substances were encoded by a technical assistant.

Cold sensations and Cold sensation detection threshold

The highest and lowest concentrations of the menthol solutions were set at 1% and 0.005%, respectively. Starting at the lowest and increasing to the highest menthol concentration, the solutions were applied with an interstimulus interval of 10 sec. For each stimulus, the subject was instructed to push a button as soon as he or she detected a CS (CDT). The CDT was considered the minimum menthol concentration. When no threshold was obtained, the highest concentration tested (1%) was entered as the threshold value.

Assessment of neurotoxicity

The National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE) version 3.0 was used to evaluate the severity of neurotoxicity: grade 1 (mild), loss of deep tendon reflexes or paresthesia not interfering with function; grade 2 (moderate), sensory alteration or paresthesia interfering with function but not activities of daily living; grade 3 (severe), sensory alteration or paresthesia interfering with activities of daily living; and grade 4, disabling (Trotti et al. 2003).

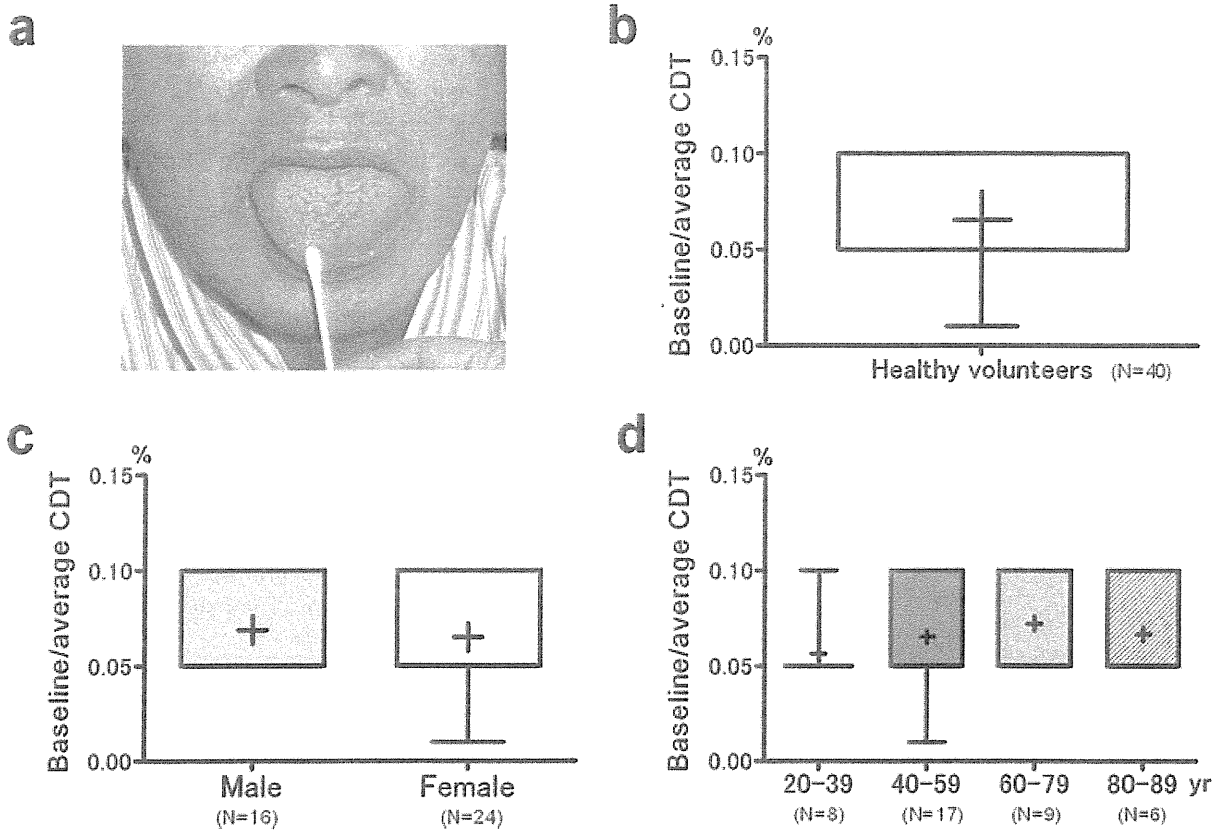

Figure 1. Effects of menthol on cold sensation and the detection threshold in healthy human subjects. (a) The menthol solution was topically applied with a cotton swab to the dorsal anterior tongue. (b) The mean baseline cold sensation detection thresholds (CDTs) in healthy human volunteers (n = 40) were 0.01% (1 of 40 subjects), 0.05% (26 of 40), and 0.1% (13 of 40). The overall mean CDT was 0.068 ± 0.026% (mean ± SD). (c) Significant sex difference in mean baseline CDT was not found. (d) Significant age difference in mean baseline CDT was not found. Cross is the mean of CDT.

Statistics

The effects of oxaliplatin were analyzed by the nonparametric Wilcoxon t-test for paired samples. In all of the statistical analyses, significance was determined using an alpha level of 0.05. All statistical procedures were performed using the IBM-SPSS software package version 18.0J for Windows (Tokyo, Japan) and the GraphPad Prism 4 statistics program (GraphPad Software, Inc., San Diego, CA).

Results

Effects of menthol on CS and CDT in healthy human subjects and patients with colon cancer (experiment 1)

All subjects noticed a significant feeling of coldness at the menthol application site. The CS occurred within the first 3 sec, reached an intensity plateau at approximately 5 sec and then disappeared within 10 sec. The intensity of the CS increased in a dose-dependent manner. None of the subjects experienced a CS when the vehicle control was applied. The mean baseline CDTs in healthy human volunteers were 0.01% (1 of 40 subjects), 0.05% (26 of 40), and 0.1% (13 of 40). The mean CDT was 0.068 ± 0.026% (SD) (Fig. 1b). To assess reproducibility, 40 healthy subjects were retested, and their CDTs were found not to differ significantly from the previous testing. Significant sex and age differences in mean baseline CDTs were not found as well (Fig. 1c and d). No serious adverse events occurred during the study and all doses of menthol were well tolerated.

The mean CDT in patients with colon cancer who had never received any chemotherapy was 0.067 ± 0.025% (n = 12). No significant difference in mean baseline CDT was observed between healthy subjects and patients with colon cancer. In addition, no serious adverse events occurred during the study and all doses of menthol were well tolerated.

Changes in the CDT before and after oxaliplatin administration (experiment 2)

Figure 2a shows the CDTs that were obtained before and after the first oxaliplatin administration in patients who had

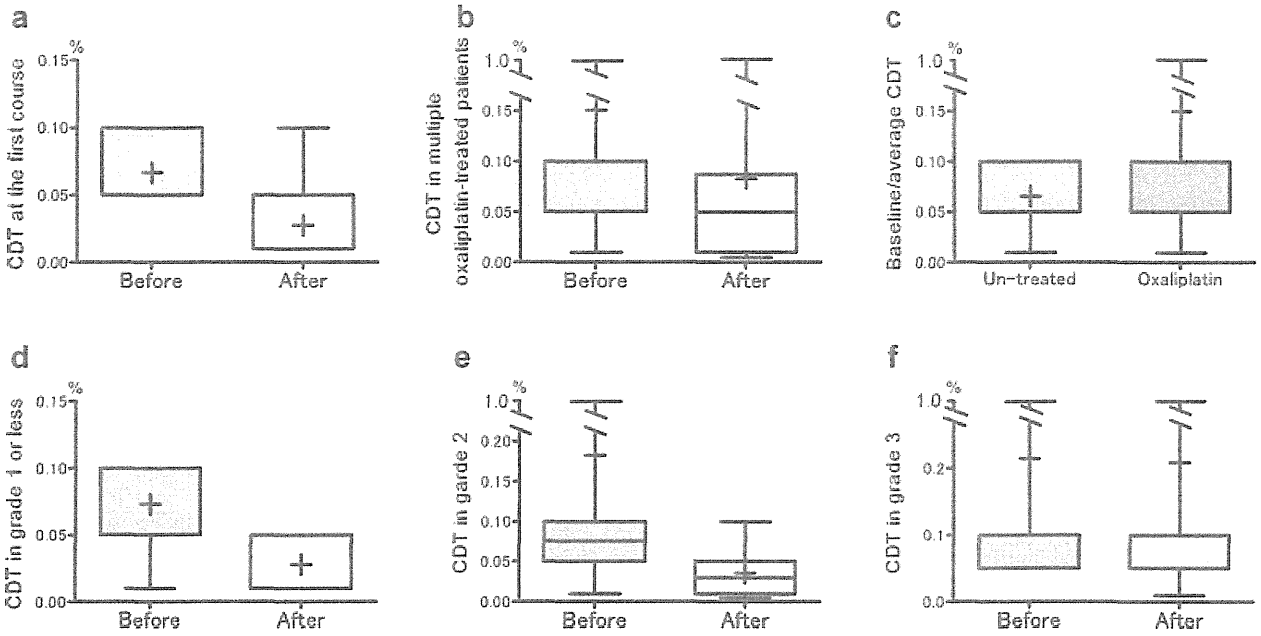

Figure 2. Changes in the cold sensation detection threshold (CDT) before and after oxaliplatin administration. (a) The CDT was determined by applying menthol before and after the first oxaliplatin administration. The CDT significantly decreased from 0.067 ± 0.025% (mean ± SD) to 0.028 ± 0.029% ($n = 12$, $P = 0.0025$). (b) Changes in the CDT before and after oxaliplatin administration in patients previously treated with oxaliplatin. The CDT significantly decreased from 0.151 ± 0.263% to 0.083 ± 0.198% ($n = 24$, $P = 0.0004$). (c) The mean baseline CDT was significantly higher in patients previously treated with oxaliplatin ($n = 24$) than in untreated subjects ($n = 52$) (0.151% vs. 0.066%, $P = 0.0225$). (d) The CDT was measured before and after oxaliplatin was administered to patients who had grade 1 or less neurotoxicity. The CDT significantly decreased from 0.073 ± 0.034% to 0.028 ± 0.021% ($n = 9$, $P = 0.0126$). (e) The CDT was measured before and after oxaliplatin was administered to patients who had grade 2 neurotoxicity. There was no significant difference in the CDTs ($n = 8$; before, 0.183 ± 0.332%; after, 0.036 ± 0.033%; $P = 0.022$). (f) The CDT was obtained before and after oxaliplatin was administered to patients who had grade 3 neurotoxicity. There was no significant difference in the CDTs ($n = 7$; before, 0.214 ± 0.347%; after, 0.209 ± 0.351%; $P = 1.0$). Cross is the mean of CDT.

never received chemotherapy. All but one patient were hypersensitive to menthol as indicated by a significant decrease in the CDT from 0.067 ± 0.025% to 0.028 ± 0.029% ($n = 12$, $P = 0.0025$). The CDTs were also measured before and after oxaliplatin administration in patients who had previously received oxaliplatin ($n = 24$, median, 330 mg/m^2; range, 85 – 2450 mg/m^2). Under these conditions, the CDT significantly decreased from 0.151 ± 0.263% to 0.083 ± 0.198% ($n = 24$, $P = 0.0004$) (Fig. 2b). Taken together, these findings show that the mean baseline CDT was significantly higher in patients previously treated with oxaliplatin ($n = 24$) than in untreated subjects ($n = 52$) (0.151% vs. 0.066%, $P = 0.0225$).

When the relationship between the CDTs and the CTCAE neurotoxicity ratings in oxaliplatin-treated patients was evaluated, the CDTs were found to be significantly decreased in patients who had grade 1 or less neurotoxicity (from 0.073% to 0.028%) ($n = 9$, $P = 0.0126$) (Fig. 2d), and grade 2 (from 0.183% to 0.036%) ($n = 8$, $P = 0.022$) (Fig. 2e), but not in those with grade 3 neurotoxicity (from 0.214% to 0.209%) ($n = 7$, $P = 1.0$) (Fig. 2f).

Discussion

Our results indicate a potential correlation between TRPM8 activity and OPN, especially in acute hypersensitivity to CS, and that acute changes in CDT may facilitate the identification of early OPN. In chemotherapy-naïve patients, significant sensitivity to topical menthol developed after the first oxaliplatin infusion, suggesting that oxaliplatin had indeed induced cold hypersensitivity. In contrast, patients with previous oxaliplatin exposure showed reduced cold hypersensitivity. With regard to the relationship between the CDT and neurotoxicity grade, we found that mild or moderate neurotoxicity was associated with significant changes in the CDT, while severe neurotoxicity was not associated with marked changes in the CDT. Whether the CDT remains unaltered in oxaliplatin-treated patients who do not develop OPN despite chronic oxaliplatin exposure requires further investigation. Nonetheless, these findings suggest that the CDT is a sensitive marker of early oxaliplatin-induced sensory disturbances.

Menthol activates the cold-transducing Ca^{2+} ion channel TRPM8 and increases cold-evoked currents (McKemy et al.

2002; Peier et al. 2002), and TRPM8 is naturally expressed sensory neurons (Reid et al. 2002; Abe et al. 2005; Kobayashi et al. 2005; Madrid et al. 2006). These TRPM8-expressing sensory neurons project into the superficial laminae of the spinal cord dorsal horn (Dhaka et al. 2008; Wrigley et al. 2009) that contains cold-sensitive neurons that project into the spinothalamic tract (Craig and Dostrovsky 2001). Thus, the cold-induced paresthesias after oxaliplatin administration that were accentuated by menthol might be mediated via the activation of TRPM8-expressing innocuous cold receptors, assuming that the receptors access central neurons. Although the precise mechanisms underpinning OPN are still uncertain, this study may serve as an entry point in furthering the mechanistic understanding of OPN. Oxaliplatin has also been shown to modify intracellular Ca^{2+} handling within the cell bodies of cultured neurons (Grolleau et al. 2001). A more recent study cited a possible mechanism for some of the oxaliplatin-induced effects that is related to the modification of surface charges around the ion channel: either due to extracellular Ca^{2+} chelation or binding of a charged biotransformation product of oxaliplatin to the channel (Broomand et al. 2009). In addition, the prospective CONcePT study confirmed that OPN could be strongly attenuated by pre- and post-treating patients with Ca^{2+} and Mg^{2+} infusions (Gamelin et al. 2008). These findings suggest a mode of action that involves a Ca^{2+}-dependent mechanism in OPN. Therefore, the Ca^{2+} ion channel TRPM8 appears to be a good candidate for understanding the Ca^{2+}-dependent mechanism in OPN.

The TRP ion channel family consists of approximately 28 mammalian cation channels (Gaudet, 2008; Talavera et al. 2008; Eid and Cortright, 2009) that are involved in a wide range of physiological and pathophysiological processes including taste, thermosensation, pain, and cell cycle regulation. The TRP ion channels present a novel mechanism for controlling Ca^{2+} transients in human neurons and represent potential targets for regulating neurite proliferation and outgrowth. Recent studies have shown that regulating TRPM8 ion channels may be a way of controlling Ca^{2+} transients in human neurons. We, therefore, hypothesized that oxaliplatin could alter calcium-sensitive voltage-gated Na channels through a pathway that involves Ca^{2+} ions that are likely mobilized by TRPM8.

Several limitations should be considered in light of our results. Firstly, we did not conduct additional follow-up of CDT after oxaliplatin infusion. Such data would provide a context for the length of time it takes for the CDTs to return to normal and would be very useful from a clinical translation standpoint to approximate the outcome of patients after oxaliplatin infusion. This approach will be incorporated into our next protocol. Second, we compared our CDT findings against the CTCAE grading system that is a gross general measure of neuropathy impairment (Trotti et al. 2003) that precludes specific measurement of cold allodynia symptoms. Hence, our menthol testing needs validation against a testing method that provides an objective evaluation of cold allodynia/parasthesia, preferably the gold standard of CS, such as quantitative sensory testing. The validation of the menthol testing using quantitative sensory tests will be one of the important future studies. In addition, although our healthy subjects and chemotherapy-naïve patients were similar in age, sex, and baseline CDTs, having colon cancer patients as controls rather than healthy volunteers would have established equivalency at baseline by accounting for the potential influence of cancer-specific changes on CDTs. Future studies would benefit from conducting additional evaluations of CDTs after oxaliplatin infusion, performing quantitative sensory testing, and using patients with colon cancer without OPN as controls.

The present data show that menthol may be used to determine and evaluate the neurotoxicity severity score, although the methodology using menthol has not been firmly established. Interestingly, patients with prior oxaliplatin exposure had significantly elevated CDT at baseline, and patients with grade 3 neurotoxicity did not show significant changes in the CDT before and after oxaliplatin administration. These findings suggest that TRM8 may be associated with the chronic stage of OPN. Unfortunately, in this study, these patients were not prospectively monitored for changes in the CDT during and after a long period of oxaliplatin treatment therefore, we could not confirm whether or not the CDT increased with OPN progression. A prospective, multicenter, randomized, double-blind study is needed to investigate the possibility of CDT as a diagnostic marker for OPN.

In conclusion, our findings indicate that OPN may be associated with TRPM8 in acute hypersensitivity to CS, and that additional studies on TRPM8 will enhance our understanding of the mechanisms of OPN. Further, our study demonstrates the feasibility of undertaking CDT test in a clinical setting to facilitate the identification of early neurotoxicity, although larger trials need to be conducted to confirm our findings.

Acknowledgments

Our heartfelt condolences and appreciation go to J. Iwamoto whose comments and suggestions were invaluable for our study. The assistance of K. Lee with manuscript preparation is gratefully acknowledged. The first two authors, T. Kono and M. Satomi, contributed equally to this work.

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