Prognostic relevance of *SAMSN1* expression in gastric cancer

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Abstract. The prognosis for patients with advanced gastric cancer (GC) remains poor. The identification of biomarkers relevant to the recurrence and metastasis of GC is advantageous for stratifying patients and proposing novel molecular targets. In the present study the oncological roles of SAM domain, SH3 domain and nuclear localization signals 1 (*SAMSN1*), a mediator of B-cell function, were elucidated in GC. The expression and methylation status of *SAMSN1* were investigated in a panel of 11 GC cell lines. Immunohistochemical staining was performed to determine the pattern of SAMSN1 protein expression in gastric tissues. The prognostic impact of *SAMSN1* expression was determined by analyzing 175 pairs of surgically resected gastric tissues. A marked decrease in the level of *SAMSN1* mRNA was detected in 8/11 GC cell lines as compared with that in a non-transformed intestinal epithelium cell line (FHs 74) without promoter methylation. The mean expression level of *SAMSN1* mRNA was reduced in GC tissues compared with normal adjacent tissues, an observation that was independent of tumor differentiation. The pattern of SAMSN1 protein expression was significantly correlated with that of *SAMSN1* mRNA. Low *SAMSN1* mRNA expression was significantly associated with tumor size (>60 mm; P=0.026) and shorter overall survival times (P=0.004). Multivariate analysis identified low *SAMSN1* mRNA expression as an independent prognostic factor for poor overall survival (hazard ratio, 1.80; 95% confidence interval, 1.07–3.05; P=0.025). The difference in survival between the low and high *SAMSN1* expression groups was more marked in patients with stage II/III GC compared to those with stage IV GC. In patients with stage II/III GC who underwent curative surgery, low *SAMSN1* expression was associated with reduced disease free survival times. The results of the present study indicate that downregulation of *SAMSN1* transcription may affect the progression and recurrence of GC, and therefore may represent a novel biomarker of GC.

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Introduction

The high incidence of gastric cancer (GC) and its associated mortality pose severe threats to human health (1,2). Although curative gastrectomy followed by adjuvant therapy has been demonstrated to prolong the survival of patients with stage II/III GC, certain patients develop locoregional or distant recurrence (3-5). Patients with stage IV GC almost always possess a poor prognosis (6,7). Identifying biomarkers relevant to the recurrence and metastasis of GC may assist clinicians in tailoring therapies by identifying high-risk patients and proposing novel molecular targets for the treatment of GC.

Recent analysis of gene and protein expression profiles, as well as oncogenic signaling pathways, suggests the existence of molecular subtypes of GC (8-10). This molecular diversity leads to clinical heterogeneity (8). Although GCs represent a biologically heterogeneous group of diseases, treatment strategy is generally determined by clinical stage alone, with no consideration of the molecular characteristics of the cancer (2). Detailed molecular characterization of a patient's tumor may enable tailored therapies that improve the likelihood of a positive outcome and decrease toxicity.

SAM domain, SH3 domain and nuclear localization signals 1 (*SAMSN1*) encodes one of a family of SH3-domain containing cytoplasmic adaptor proteins expressed in lymphocytes (11,12). *SAMSN1* is mainly expressed by hematopoietic cells and mediates B-cell activation and differentiation. The *SAMSN1* gene is located on chromosome 21q11-21, within a region associated with heterozygous deletions that is frequently present in lung cancer cells, suggesting that *SAMSN1* acts as a tumor suppressor (13,14). This possibility is supported by the study of Noll *et al* (15), which revealed that *SAMSN1* is a suppressor of multiple myeloma (15). To date, the precise role of *SAMSN1* in oncogenesis remains to be fully elucidated, particularly in cancer of the digestive tract, including GC. The present study hypothesized that the dysregulation or absence of *SAMSN1* expression contributes to the initiation and progression of GC. The aims of the present study were to investigate the clinical significance of *SAMSN1* expression, define the mechanism of *SAMSN1* transcriptional regulation, establish whether *SAMSN1* contributes to tumorigenesis and assess the clinical utility of *SAMSN1* as a potential prognostic marker and as a target for therapy in GC.

Materials and methods

Cell lines and tissue samples. The GC cell lines MKN1, MKN45, MKN74, NUGC2, NUGC3, NUGC4 and SC-6-JCK were obtained from the Japanese Collection of Research Bioresources Cell Bank (Osaka, Japan). The AGS, KATOIII and N87 cell lines were acquired from the American Type Culture Collection (Manassas, VA, USA). The GCIY was obtained from Tohoku University, Sendai, Japan. A control, non-tumorigenic epithelial cell line (FHs 74) was purchased from the American Type Culture Collection. Cells were cultured in RPMI-1640 medium (Thermo Fisher Scientific, Inc., Waltham, MA, USA) supplemented with 10% fetal bovine serum (Thermo Fisher Scientific, Inc.) and maintained in a 5% CO_2 atmosphere at 37˚C. For FHs 74 cells, the medium was additionally supplemented with 30 ng/ml epidermal growth factor (Sigma-Aldrich; EMD Millipore, Billerica, MA, USA). Total RNA was extracted using an RNeasy Mini kit (Qiagen GmbH, Hilden, Germany) and used as a template for the generation of complementary DNA as described previously (16,17). Primary GC tissues and corresponding normal adjacent tissues were collected from 175 patients who underwent gastric resection for GC without neoadjuvant therapy at Nagoya University Hospital (Nagoya, Japan) between November 2001 and December 2012. Patients who received neoadjuvant therapy were excluded, as it was difficult to obtain cancer cells from scarred tissues. Following collection, tissue samples were immediately frozen in liquid nitrogen and stored at -80˚C until the time of RNA extraction. Corresponding normal adjacent gastric mucosa samples were obtained from each patient and were collected from a region no less than 5 cm from the tumor edge. To determine whether the expression status of *SAMSN1* differed according to tumor histology, patients were categorized into two histological subtypes: Differentiated (papillary, well differentiated and moderately differentiated adenocarcinoma) and undifferentiated (poorly differentiated adenocarcinoma, signet ring cell carcinoma and mucinous carcinoma) (18). Since 2006, adjuvant chemotherapy using S-1 (an oral fluorinated pyrimidine) has been administered to all Union for International Cancer Control (UICC) stage II/III GC patients (unless contraindicated by the patient's condition) (19,20). Patients were followed-up at least once every 3 months for 2 years following surgery, and then every 6 months for 5 years or until death. Physical examination, laboratory tests and enhanced computed tomography (chest and abdominal cavity) were performed at each visit (21). The chemotherapy regimen for patients with distant metastasis or recurrence was chosen at the physician's discretion. The present study conformed to the ethical guidelines of the World Medical Association Declaration of Helsinki: Ethical Principles for Medical Research Involving Human Subjects, and was approved by the Institutional Review Board of Nagoya University, Nagoya, Japan. Written informed consent for usage of clinical samples and data, as required by the institutional review board, was obtained from all patients (22).

SAMSN1 mRNA expression analysis. *SAMSN1* mRNA expression levels in 11 GC cell lines and 175 primary GC tissues and corresponding normal adjacent tissues were analyzed by reverse transcription-quantitative polymerase chain reaction (RT-qPCR) using an ABI StepOnePlus Real-Time PCR System (Applied Biosystems; Thermo Fisher Scientific, Inc.) in conjunction with the gene specific primers listed in Table I. Cycling conditions were as follows: One cycle at 95˚C for 10 min, 40 cycles at 95˚C for 5 sec and 60˚C for 60 sec To investigate the oncological role of *SAMSN1* in GC, correlation analysis was performed to evaluate the association between the pattern of *SAMSN1* mRNA expression and clinicopathological parameters, including patient survival following gastrectomy. Each of the 175 patients was assigned to one of two groups (low and high *SAMSN1* expression) according to their median level of *SAMSN1* mRNA expression in GC tissues. Additionally, the prognostic impact of *SAMSN1* mRNA expression on patients categorized according to the 7th UICC staging system was also evaluated (23).

Bisulfite sequence analysis. Genomic DNA from GC cell lines was treated with bisulfite using the EpiTect Bisulfate kits (Qiagen GmbH) and sequenced to determine the levels of DNA methylation according to previously published procedures (24).

Immunohistochemistry. The intensity and pattern of SAMSN1 protein expression was determined by immunohistochemical staining using 48 representative sections of well-preserved GC tissue as described previously (25). Sections were incubated for 1 h at room temperature with a rabbit polyclonal antibody raised against *SAMSN1* (catalog no., 13063-1-AP; ProteinTech Group, Inc., Chicago, IL, USA) diluted 1:400 in antibody diluent (Dako, Glostrup, Denmark). The samples were subsequently washed with phosphate buffered saline, followed by a 10 min incubation with biotinylated rabbit secondary antibody (Histofine SAB PO(R) kit; Nichirei Corporation, Tokyo, Japan) in a 1:1,000 dilution with ChemMateT antibody diluent (Dako). Sections were subsequently developed for 3 min using 3,3'- diaminobenzidine as the substrate (Nichirei Corporation). The patterns of *SAMSN1* staining in GC tissues and corresponding non-cancerous tissues were compared, and positive blood vessel staining provided an internal control for the immunolabeling procedure. Specimens were randomized and coded prior to analysis by two independent observers blinded to the status of the samples (26,27).

Statistical analysis. Differences in the relative expression of *SAMSN1* mRNA (normalized to the level of glyceraldehyde-3-phosphate expression) between the two groups were analyzed using the Mann-Whitney U test. The χ^2 test was used to analyze the association between the expression status of

Table I. Primers and associated annealing temperatures.

Gene	Experiment	Direction	Sequence, 5'-3'	Product size, bp	Annealing temperature, ˚C
SAMSN1	RT-qPCR	Forward	TGCTCAAGAGAAAGCCATCC	97	60
		Reverse	TTATTCCGAAAACGATCGAAA		
	Bisulfite	Forward	TTGTTTTTATTTTGAGTTGTGTTTGT	416	62
	Sequencing 1	Reverse	ACTAAACTTCCTCCATTACTCTCTCTC		
	Bisulfite	Forward	AGTTATGTTTTTATTTATATTTAGAATGGG	257	64
	Sequencing 2	Reverse	TCACCCAAACTAAAATACAATAACA		
GAPDH	RT-qPCR	Forward	GAAGGTGAAGGTCGGAGTC	226	60
		Probe	CAAGCTTCCCGTTCTCAGCC		
		Reverse	GAAGATGGTGATGGGATTTC		

SAMSN1, SAM domain, SH3 domain and nuclear localization signals 1; *GAPDH*, glyceraldehyde-3-phosphate dehydrogenase; RT-qPCR, reverse transcription-quantitative polymerase chain reaction.

SAMSN1 and various clinicopathological parameters. A correlation between expression patterns of SAMSN1 protein and mRNA in gastric tissue specimens was also evaluated by the χ^2 test. Survival rates were calculated using the Kaplan-Meier method, and the difference in survival curves was analyzed using the log-rank test. Multivariate regression analysis was performed to detect prognostic factors using the Cox proportional hazards model, and variables with P<0.05 were entered into the final model. All statistical analysis was performed using JMP version 10 software (SAS Institute Inc., Cary, NC, USA). P<0.05 was considered to indicate a statistically significant difference.

Results

SAMSN1 expression and methylation status in GC cell lines. A marked decrease in the level of *SAMSN1* mRNA expression was detected in 8 (73%) of the 11 GC cell lines when compared with the FHs 74 control cell line. There was no marked difference in *SAMSN1* expression between cell lines derived from differentiated and undifferentiated GCs (Fig. 1A). No DNA methylation of the *SAMSN1* promoter was detected.

Patient characteristics. The patient population included 134 males and 41 females with an age range from 20-84 years (mean age, 64.7±11.8 years). Pathologically, 106 patients were diagnosed with undifferentiated GC and 69 with differentiated GC. A total of 39 patients were diagnosed with stage I disease, 29 with stage II, 51 with stage III and 56 with stage IV disease. A total of 119 patients with stage I-III disease underwent R0 resection. A total of 47/56 patients classified as UICC stage IV were assigned this diagnosis due to positive peritoneal lavage cytology, localized peritoneal metastasis or distant lymph node metastasis. A total of 6 of the patients with stage IV disease had synchronous liver metastasis and a single patient had lung metastasis, and these individuals underwent gastrectomy to control bleeding or obstruction to the passage of food.

SAMSN1 mRNA and protein expression in surgically resected tissues. The mean expression level of *SAMSN1* mRNA was reduced in GC tissues when compared with that in adjacent normal tissues (P<0.001). However, there was no significant difference in the expression of *SAMSN1* mRNA between patients with undifferentiated and differentiated GC (Fig. 1B; P=0.067). Immunohistochemical staining was subsequently performed to investigate the expression of SAMSN1 protein in those cases where the *SAMSN1* mRNA level in GC tissues was observed to be less or equivalent to that identified for corresponding non-cancerous tissues. Representative GC specimens with an increased, equivalent and reduced intensity of SAMSN1 protein staining in cancerous tissue compared with adjacent normal tissue are shown in Fig. 2A. In 48 of the patient samples examined, the pattern of SAMSN1 protein expression correlated significantly with that of the expression of *SAMSN1* mRNA (P=0.005; Fig. 2B).

Prognostic implications of SAMSN1 mRNA expression. Patients were assigned to one of two groups according to their median *SAMSN1* mRNA expression level in GC tissues (high expression group, n=87; low expression group, n=88). Low *SAMSN1* mRNA expression was significantly associated with larger tumor size (>60 mm; P=0.026), but not tumor location or UICC stage (P=0.639) (Table II). Patients in the low *SAMSN1* expression group were more likely to have a shorter overall survival time than those in the high expression group (5-year survival rates were 43% and 66% for the high and low expression groups, respectively; P=0.004; Fig. 3A). In multivariate analysis for overall survival, low *SAMSN1* mRNA expression was identified to be an independent prognostic factor (hazard ratio, 1.80; 95% confidence interval, 1.07-3.05; P=0.025; Table III). When patients were categorized according to UICC stage, no significant differences in the mean expression level of *SAMSN1* mRNA was observed between groups (P>0.05, for each), suggesting that *SAMSN1* expression was independent of tumor stage (Fig. 3B).

Subsequently, a subgroup analysis of patients categorized according to UICC stage was performed. The survival difference between the low and high *SAMSN1* expression groups was more apparent in patients with stage II/III GC (P=0.025_ than those with stage IV GC (P=0.162) (Fig. 4A). Among 80 patients

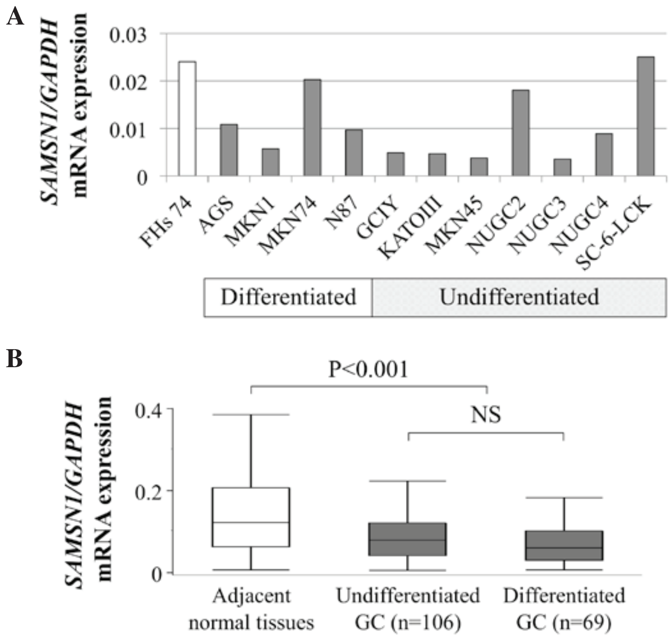
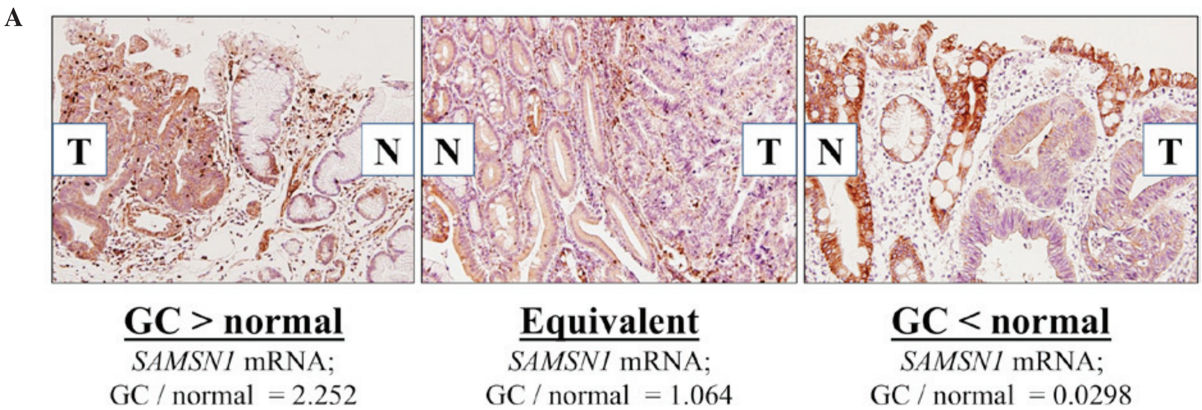


Figure 1. Expression status of *SAMSN1*. (A) A total of 8/11 GC cell lines had reduced *SAMSN1* mRNA expression compared with the FHs 74 cell line. (B) Quantification of *SAMSN1* mRNA expression in GC and adjacent normal tissues. The median level of *SAMSN1* mRNA expression was reduced in GC tissues compared with corresponding normal adjacent tissues, a finding that was independent of tumor differentiation status. Lines in the boxes indicate the median values. The upper and lower borders of the boxes indicate the quartile 4 and quartile 1 lines, respectively. The highest and lowest values are represented by horizontal lines. *SAMSN1*, SAM domain, SH3 domain and nuclear localization signals 1; GC, gastric cancer; NS, not significant.



B

		qPCR; GC/normal		
		>2	0.5 ~ 2	<0.5
Staining intensity	GC > normal	4	2	0
	Equivalent	2	20	5
	GC < normal	1	8	6

P=0.005

Figure 2. Immunohistochemical analysis. (A) Representative GC specimens with an increased, equivalent and reduced intensity of *SAMSN1* protein staining in cancerous tissue compared with adjacent normal tissue. Magnification, x100. The ratio of expression levels of *SAMSN1* mRNA between GC and corresponding normal adjacent tissue is shown below the figures. (B) A direct correlation was observed between SAMSN1 protein and mRNA expression in GC tissue specimens using the χ^2 test (P=0.005). GC, gastric cancer; *SAMSN1*, SAM domain, SH3 domain and nuclear localization signals 1; N, normal tissue; T, tumor tissue.

with stage II/III GC who underwent curative surgery, those who had a low level of *SAMSN1* mRNA expression in GC tissues were more likely to have shorter disease free survival times than those who had high *SAMSN1* mRNA expression (2-year survival rates were 50% and 81% for the low and high SAMSN1 expression groups, respectively; P=0.038; Fig. 4B).

Table II. Association between expression level of *SAMSN1* mRNA and clinicopathological parameters in 175 patients.

Variables	Low *SAMSN1* mRNA in GC tissue, n	High *SAMSN1* mRNA in GC tissue, n	P-value
Age, years			0.710
<65	38	40	
≥65	50	47	
Gender			0.891
Male	67	67	
Female	21	20	
Carcinoembryonic antigen, ng/ml			0.352
≤5	69	73	
>5	19	14	
Carbohydrate antigen 19-9, IU/ml			0.467
≤37	69	72	
>37	19	15	
Tumor location			0.719
Entire	6	8	
Upper third	17	19	
Middle third	31	24	
Lower third	34	36	
Tumor size, mm			0.026[a]
<60	43	57	
≥60	45	30	
Tumor depth, UICC classification			0.405
pT1-3	42	47	
pT4	46	40	
Differentiation			0.476
Differentiated	37	32	
Undifferentiated	51	55	
Lymphatic involvement			0.509
Absent	12	15	
Present	76	72	
Vessel invasion			0.708
Absent	40	42	
Present	48	45	
Infiltrative growth type			0.598
Invasive	31	34	
Expansive	57	53	
Lymph node metastasis			0.318
Absent	29	35	
Present	59	52	
Peritoneal lavage cytology			0.621
Negative	66	68	
Positive	22	19	
UICC stage			0.639
I	19	20	
II	14	15	
III	23	28	
IV	32	24	

[a]Statistically significant (P<0.05). UICC, Union for International Cancer Control; *SAMSN1*, SAM domain, SH3 domain and nuclear localization signals 1; GC, gastric cancer.

Table III. Univariate and multivariate analysis of prognostic factors for overall survival in 175 patients.

		Univariate			Multivariate		
Variables	n	Hazard ratio	95% CI	P-value	Hazard ratio	95% CI	P-value
Age, years (≥65)	97	1.00	0.63-1.60	0.991	1.08	0.64-1.85	0.782
Gender (female)	41	1.14	0.66-1.88	0.638	1.09	0.58-1.97	0.786
Carcinoembryonic antigen (>5 ng/ml)	33	1.66	0.93-2.79	0.083	1.13	0.61-2.01	0.688
Carbohydrate antigen 19-9 (>37 IU/ml)	34	2.16	1.25-3.60	0.007	1.58	0.89-2.79	0.133
Tumor location (lower third)	70	0.62	0.37-0.99	0.049	0.66	0.38-1.11	0.119
Tumor size (≥60 mm)	75	2.86	1.79-4.64	<0.001	1.53	0.91-2.61	0.106
Tumor depth (pT4, UICC classification)	86	3.92	2.39-6.65	<0.001	1.72	0.94-3.22	0.079
Tumor differentiation (undifferentiated)	106	1.75	1.08-2.92	0.023	1.22	0.68-2.23	0.507
Lymphatic involvement	148	5.93	2.21-24.3	<0.001	1.27	0.37-5.88	0.726
Vessel invasion	93	2.40	1.48-4.00	<0.001	1.70	1.00-3.01	0.049[a]
Invasive growth	65	2.64	1.67-4.21	<0.001	1.03	0.55-1.96	0.927
Lymph node metastasis	111	7.05	3.58-16.0	<0.001	2.53	1.09-6.68	0.030[a]
Peritoneal lavage cytology (positive)	41	4.67	2.89-7.48	<0.001	2.43	1.35-4.41	0.003[a]
Low *SAMSN1* mRNA in GC tissues	87	2.00	1.25-3.24	0.004	1.80	1.07-3.05	0.025[a]

[a]Statistically significant in multivariate analysis. CI, confidence interval; UICC, Union for International Cancer Control; *SAMSN1*, SAM domain, SH3 domain and nuclear localization signals 1; GC, gastric cancer.

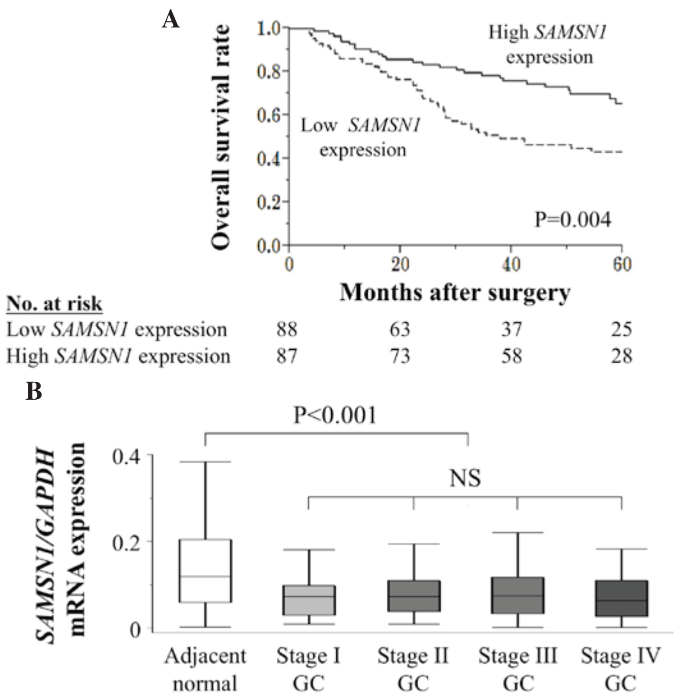


Figure 3. Prognostic impact of *SAMSN1* mRNA expression in GC patients. (A) Low *SAMSN1* expression was associated with shorter overall survival times in patients with GC. The table under the graph indicates number of patients at risk for each group. (B) Expression levels of *SAMSN1* mRNA according to the Union for International Cancer Control stage. Lines in the boxes indicate the median values. The upper and lower borders of the boxes indicate quatile 4 and quartile 1 lines, respectively. The highest and lowest values are represented by horizontal lines. *SAMSN1*, SAM domain, SH3 domain and nuclear localization signals 1; GC, gastric cancer; NS, not significant.

Discussion

The mechanism by which *SAMSN1* contributes to the tumorigenesis of digestive cancers remains to be fully elucidated. However, it may be hypothesized that, as a B-cell mediator, *SAMSN1* may have a specific role in the initiation and progression of GC, as this disease frequently develops from chronically inflamed gastric mucosa, including that associated with *Helicobacter pylori*-related chronic gastritis and atrophic gastritis (28-30). Consequently, the present study

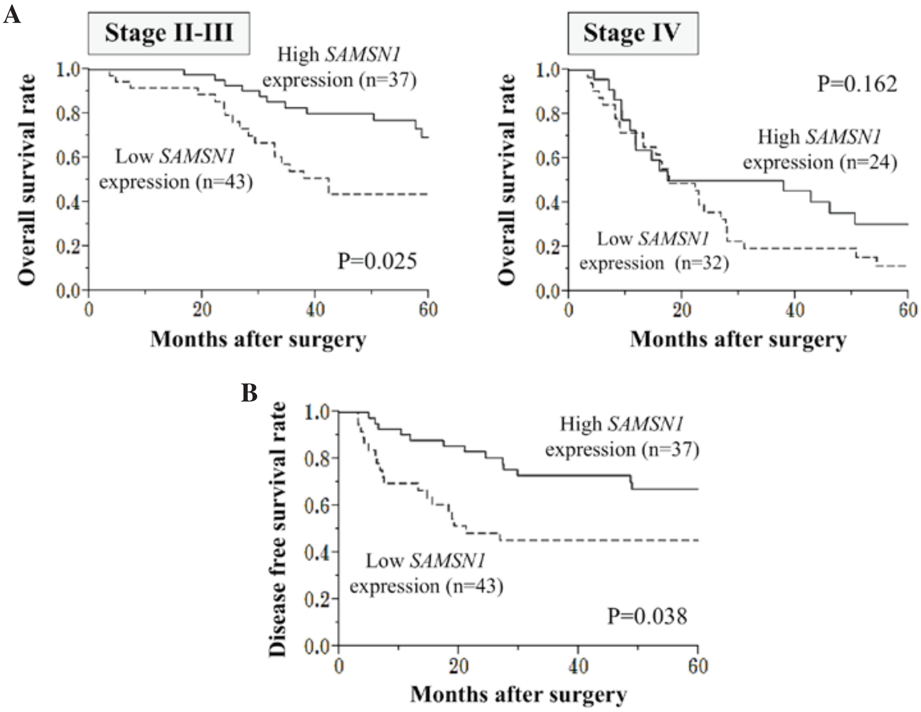


Figure 4. Survival analyses according to Union for International Cancer Control stage. (A) Survival differences between the low and high *SAMSN1* expression groups were more apparent in patients with stage II/III GC. (B) Low *SAMSN1* expression was associated with shorter disease free survival times in patients with stage II/III GC. *SAMSN1*, SAM domain, SH3 domain and nuclear localization signals 1; GC, gastric cancer.

sought to investigate the status and mechanism of regulation of *SAMSN1* expression in GC. It was demonstrated that the promoter region of *SAMSN1* is methylated in a number of GC cell lines in which *SAMSN1* mRNA expression is reduced, and that *SAMSN1* expression may be restored following DNA demethylation, despite the absence of CpG islands around the promoter region of *SAMSN1*. In general, the majority of tumor suppressor genes are suppressed through the aberrant hypermethylation of promoter regions that contain CpG islands (31,32). Noll *et al* (15) investigated the methylation status of the *SAMSN1* gene, upstream and downstream of the promoter region, and observed that hypermethylation was associated with suppressed expression of *SAMSN1* mRNA. Given this, the present study conducted bisulfite sequencing analysis upstream and downstream of the *SAMSN1* promoter region and observed no methylation in GC cell lines. Further study is required to clarify the alterative underlying molecular pathway suppressing *SAMSN1* transcription in GC.

Immunohistochemical staining and RT-qPCR analysis revealed a direct correlation between SAMSN1 protein and *SAMSN1* mRNA expression. These findings suggest that changes in the level of *SAMSN1* mRNA are functionally significant and, therefore, that RT-qPCR may provide a useful tool for the quantitative analysis of *SAMSN1* expression in clinical samples (33,34).

SAMSN1 mRNA expression was significantly downregulated in GC tissues when compared with corresponding non-cancerous gastric tissues, and low expression of *SAMSN1* mRNA was associated with more aggressive phenotypes, including larger tumor size and shorter survival time. Furthermore, multivariate analysis identified low *SAMSN1* expression as an independent prognostic factor. These results indicate that *SAMSN1* may function as a suppressor of GC and that suppression of *SAMSN1* expression may serve as a prognostic indicator of this disease. Previously, it has been reported that differences in the genetic background of tumors are reflected in the histology, morphology and location of GCs (9,35,36). In the present study, it was observed that *SAMSN1* expression was independent of tumor location and differentiation, indicating *SAMSN1* has a similar role in all types of GC.

The physiological function of *SAMSN1* remains to be fully elucidated. *SAMSN1* is primarily expressed in human immune tissues as well as in cell lines and primary cells derived from patients with acute myeloid leukemia and multiple myeloma (15,37). In addition, *SAMSN1* expression is upregulated by signaling factors that promote the activation and differentiation of B-cells (11,13). The present study hypothesized that chronic inflammation is caused by *H. pylori* infection-induced dysregulation of immune function and aberrant expression of *SAMSN1* (38,39). However, this hypothesis is not fully supported by the present findings, as detailed information regarding *H. pylori* infection was not collected. To develop a detailed understanding of the oncological functions of *SAMSN1*, further functional studies are required. For example, studies that aim to identify the binding partners of *SAMSN1* or those that can take advantage of mouse models of GC to evaluate the effects of the presence or absence of *SAMSN1* on premalignant and malignant phenotypes would be of great value in advancing our understanding of the role of this tumor suppressor in GC (40).

There is great variability in the outcome for patients with stage II/III GC: Certain patients respond well to therapy and demonstrate long-term survival, while others are prone to locoregional or distant recurrence, even following complete curative resection (5,41). Therefore, there is a great need for the risk stratification of stage II/III GC patients to facilitate the appropriate management of this disease. A significant finding from the present study was that the association between *SAMSN1* mRNA levels and postoperative prognosis for patients with stage II/III GC was stronger than that for patients with stage I or IV disease. This suggests that analysis of *SAMSN1* expression may provide a promising tool for the identification of stage II/III GC patients who are vulnerable to recurrence and subsequent poor prognosis.

Taken together, the results of the present study indicate that analysis of *SAMSN1* expression may be applied to the management of GC. The expression levels of *SAMSN1* in biopsies taken during an endoscopy or from surgically resected tissues may be used to stratify patient risk, providing an indication of the likelihood of recurrence and subsequent adverse prognosis, as well as establishing a criterion for determining an appropriate therapeutic strategy.

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