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Original Article

Predictive Value of the nProfiler 1 Assay for the Efficacy of Adjuvant S-1–Based Doublet Chemotherapy in Stage III Gastric Cancer: A *Post-Hoc* Analysis of a Randomized Phase III Trial

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Purpose The nProfiler 1 Stomach Cancer Assay (nProfiler1), designed to predict responses to fluorouracil-based adjuvant chemotherapy, measures the expression of four gastric cancer target genes (*GZMB*, *WARS*, *SFRP4*, and *CDX1*). The randomized phase III POST trial aimed to compare the efficacies of two adjuvant S-1-based doublet chemotherapies: S-1 plus cisplatin (SP) and S-1 plus docetaxel (DS). This study aimed to validate the nProfiler1 assay using a distinct cohort from the POST trial.

Materials and Methods The nProfiler1 assay stratifies patients into three groups (low-risk, intermediate-risk, and high-risk) using the prognostic single-patient classifier and two groups (chemotherapy-benefit and no-benefit) using the predictive single-patient classifier. The nProfiler1 assay was applied to formalin-fixed paraffin-embedded slides obtained from the POST trial. Disease-free survival (DFS) and overall survival (OS), including 5-year survival rates, were calculated for the enrolled patients.

Results Of the 153 patients in the POST trial, 118 were included in the post-hoc analysis. With a median follow-up of 57.9 months, no significant difference in DFS or OS was observed between the SP and DS groups. The prognostic single-patient classifier predicted the OS in the SP group (p=0.043) but not in the DS group (p=0.594). The chemotherapy-benefit group exhibited numerically longer DFS than the no-benefit group in the SP and DS groups.

Conclusion The nProfiler1 assay offers valuable insights into the prognosis and efficacy of adjuvant chemotherapy based on fluorouracil plus platinum doublet regimens but not docetaxel-containing regimens. Further validation with larger patient cohorts and different regimens is warranted.

Key words Gastric adenocarcinoma, Adjuvant chemotherapy, Chemotherapy response prediction, Prognosis prediction, nProfiler 1 stomach cancer assay

Introduction

Gastric cancer is highly prevalent in Eastern Asia, ranking fifth in incidence and fourth in cancer-related mortality worldwide [1]. Despite improvements in hygiene [2], food storage, and *Helicobacter pylori* eradication [3,4], mortality rates remain high owing to delayed symptom presentation, often resulting in an advanced-stage diagnosis [5].

The treatment strategy for gastric cancer has progressively evolved from relying solely on surgical resection to a comprehensive approach that includes surgical resection, chemotherapy, and molecular-targeted drug therapy. Gastrectomy with D2 lymphadenectomy is the standard surgical treatment for locally advanced gastric cancer, as complete surgical resection remains as the only curative option [6-9]. Clinical trials have demonstrated that compared with surgery alone, adjuvant chemotherapy improves survival [10-16]. S-1 monotherapy is a commonly used adjuvant chemotherapy regimen for stage II-III gastric cancer in Asia, based on early pivotal studies [11,12]. However, its inability to reduce the

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incidence of hematogenous recurrence has raised concerns [11,12]. The CLASSIC trial indicated that adjuvant doublet chemotherapy with capecitabine plus oxaliplatin improves survival for D2 dissected stage II-III gastric cancer [15,16]. The JACCRO GC-07 trial highlighted the benefit of adding docetaxel to S-1 for stage III gastric cancer, establishing adjuvant S-1 plus docetaxel as a treatment option [17]. Additionally, perioperative chemotherapy with fluorouracil plus leucovorin, oxaliplatin, and docetaxel (FLOT) has shown improved survival in locally advanced, resectable gastric cancer [18]. Despite these advancements, the overall survival (OS) benefit of adjuvant chemotherapy remains moderate, indicating that not all patients with resectable gastric cancer benefit equally. Consequently, research has focused on identifying predictive tests for chemotherapy response and prognosis in patients with resectable gastric cancer [19-21].

The development of the nProfiler 1 Stomach Cancer Assay (nProfiler1, Novomics) represents advancements in personalized treatment for gastric cancer [19]. nProfiler1 was developed through a comprehensive analysis of multiple patient cohorts and was validated using a prospectively designed and retrospectively tested approach using the CLASSIC trial cohort [19,22]. This assay identified key genetic markers predictive of response to chemotherapy by analyzing retrospective data. The four-gene (including *GZMB*, *WARS*, *SFRP4*, and *CDX1*) real-time reverse transcription polymerase chain reaction test measures gene expression levels in formalin-fixed, paraffin-embedded (FFPE) tumor tissues and employs two rule-based, single-patient classifier algorithms. This categorization enables clinicians to stratify patients based on their likelihood of benefiting from chemotherapy (predictive single-patient classifier: chemotherapy-benefit and no-benefit) and their predicted prognosis (prognostic single-patient classifier: low-risk, intermediate-risk, and high-risk), facilitating more tailored and effective adjuvant treatment plans for stages II-III gastric cancer [19].

The Post Operation chemotherapy with S-1 and Taxotere in a curatively resected gastric cancer stage III (POST, NCT01283217) trial was a randomized, multicenter, phase III trial designed to compare the efficacy of two adjuvant S-1-based doublet chemotherapies: S-1 plus cisplatin (SP) and S-1 plus docetaxel (DS) [23]. With a median follow-up duration of 56.9 months, both treatment regimens were effective and tolerable; however, the trial failed to achieve statistical significance owing to early termination following the approval of capecitabine plus oxaliplatin, based on the CLASSIC trial (NCT 00411229) [15,16].

The nProfiler1 assay was validated using a subset of samples from the CLASSIC trial [15], which included patients who underwent D2 gastrectomy followed by capecitabine and oxaliplatin chemotherapy or surgery alone [19]. The nProfiler1 assay has been validated under various conditions [24-26]; however, it has not yet been validated in docetaxel-containing regimens. Therefore, this study aimed to validate the nProfiler1 assay for predicting the response to adjuvant chemotherapy containing platinum or docetaxel in stage III gastric cancer using a distinct cohort derived from the POST trial [23].

Materials and Methods

1. POST trial details: participants, study design, and treatment

The POST trial was an open-label, phase III, randomized controlled trial conducted at eight centers in South Korea. After standard gastrectomy with D2 lymphadenectomy, patients with stage III gastric cancer were randomly assigned to the SP or DS group. The treatment protocols have been previously described [23]. The FFPE slides collected from patients in the POST trial were used for further analysis.

Disease-free survival (DFS) was defined as the time from trial enrollment to the first evidence of disease recurrence or death from any cause. OS was defined as the duration from trial enrollment to death from any cause. The clinical stages of the patients were reclassified according to the American Joint Committee on Cancer (AJCC) eighth edition [27].

2. Assessments

Two 3-mm tumor tissue cores were obtained from the FFPE slides, and total RNA extraction was performed using the RNeasy DSP FFPE Kit (Qiagen) following the manufacturer's instructions. Quantitative real-time polymerase chain reaction (qPCR) was conducted on four identified classifier genes (*GZMB*, *WARS*, *SFRP4*, and *CDX1*) and five reference genes (*ACTB*, *ATP5E*, *HPRT1*, *GPX1*, and *UBB*) [19]. The prognostic single-patient classifier stratifies patients into three groups: low-risk, intermediate-risk, and high-risk, utilizing the expression of *GZMB*, *WARS*, and *SFRP4* (S1A Fig.), whereas the predictive single-patient classifier stratifies patients into two groups: chemotherapy-benefit and no-benefit, based on the expression of *GZMB*, *WARS*, and *CDX1* (S1B Fig.) [19].

3. Statistical analysis

qPCR results were analyzed using the nDx 1 software (Novomics). Samples meeting quality control criteria were categorized using a previously established algorithm [19]. For time-to-event endpoints, including OS and DFS, median values and associated two-sided 95% confidence intervals (CI) were estimated using the Kaplan-Meier method. A Cox proportional hazards regression model was used to assess

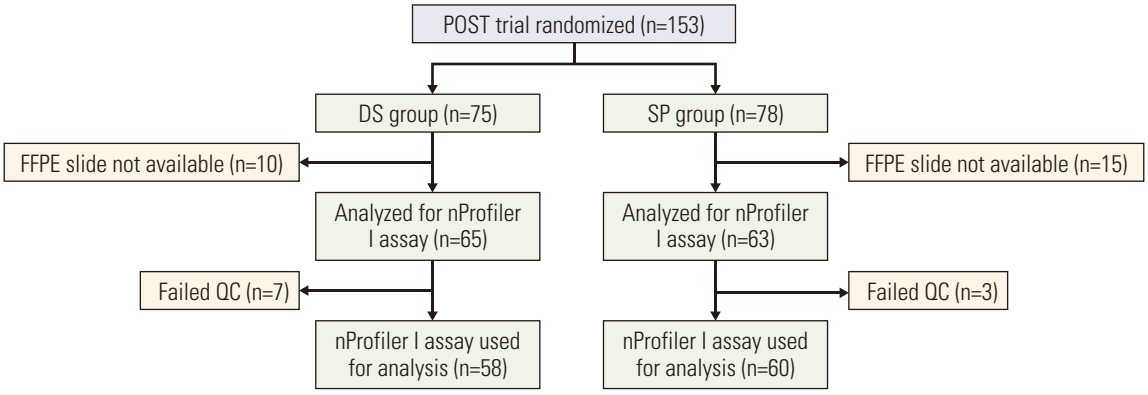

Fig. 1. Consort diagram. DS, docetaxel plus S-1; FFPE, formalin-fixed paraffin-embedded; nProfiler1, nProfiler 1 Stomach Cancer Assay; SP, S-1 plus cisplatin.

the association between 5-year DFS, 5-year OS, and clinical factors including chemotherapy regimen, predictive single-patient classifier, and prognostic single-patient classifier. All statistical analyses were performed using GraphPad Prism software ver. 8.0 for Windows (GraphPad Software).

Results

1. Patients and treatment

Of the 153 patients randomized in the POST trial, 118 cases (60 in the SP group and 58 in the DS group) were included in the post-hoc analysis. Patients without available FFPE slides or those who did not meet the quality control criteria for the nProfiler1 assay were excluded (Fig. 1). Table 1 presents the demographic and baseline characteristics of the patients.

With a median follow-up duration of 57.9 months (95% CI, 36.5 to 65.5), the median DFS was 54.0 months (95% CI, 29.6 to not reached), and the 5-year DFS rate was 49.2% (95% CI, 39.7 to 58.0) (S2A Fig.) The median OS was not reached (95% CI, 64.4 to not reached) with a 5-year OS rate of 60.3% (95% CI, 50.4 to 68.7) (S2B Fig.) in the total cohort.

The SP group had a median follow-up duration of 65.6 months (95% CI, 37.5 to 83.6), whereas the DS group had a median follow-up duration of 42.1 months (95% CI, 29.0 to 57.9). As of the data collection cut-off on November 30, 2021, 28 patients (46.7%) in the SP group and 31 patients (53.4%) in the DS group experienced recurrence or death. No significant difference was observed in DFS between the two groups (hazard ratio [HR] 1.22; 95% CI, 0.73 to 2.03; p=0.453) (Fig. 2A). The 5-year DFS rates were 52.9% (95% CI, 39.2 to 64.8) and 45.5% (95% CI, 32.2 to 57.9) in the SP and DS groups, respectively. Similarly, no significant difference was observed in OS between the two groups (HR 1.24; 95% CI, 0.70 to 2.18; p=0.444) (Fig. 2B). The 5-year OS rate was 65.1% (95% CI, 51.2 to 75.9) in the SP group and 54.9% (95% CI, 40.3 to 67.4) in the DS group (HR 1.37; 95% CI, 0.76 to 2.48; p=0.294) (S3 Table).

2. Validation of nProfiler1

Following the previously described method [19], patients were classified into risk groups based on the prognostic single-patient classifier and into the chemotherapy-benefit or no-benefit group using the predictive single-patient classifier. Specifically, the prognostic single-patient classifier categorized 118 patients into low-risk (13 patients, 11.0%), intermediate-risk (49 patients, 41.5%), and high-risk (56 patients, 47.5%) groups (Table 2, S1A Fig.). Moreover, the predictive single-patient classifier classified patients into chemotherapy-benefit (48 patients, 40.7%) and no-benefit (70 patients, 59.3%) groups (Table 2, S1B Fig.).

An analysis of the prognostic single-patient classifier across all 118 patients showed no significant differences in median DFS or OS among the three risk groups (S4A and S4B Fig.). The 5-year OS rates were 69.2% for the low-risk group (95% CI, 37.3 to 87.2), 62.4% for the intermediate-risk group (95% CI, 46.6 to 74.8), and 55.6% for the high-risk group (95% CI, 40.8 to 68.0) (Table 2). Although the Cox regression analysis indicated better survival outcomes for the low-risk group, the difference was not statistically significant, even after adjusting for clinical factors such as age, sex, tumor T category, N category, and treatment regimen (Table 2).

Similarly, the predictive single-patient classifier revealed no significant differences in median DFS (HR, 1.14; 95% CI, 0.68 to 1.92; p=0.621) (S5A Fig.) or median OS (HR, 1.36; 95% CI, 0.77 to 2.40; p=0.306) (S5B Fig.) between the chemotherapy-benefit and no-benefit groups in all patients. The chemotherapy-benefit group showed a higher 5-year OS rate of 66.6% (95% CI, 50.8 to 78.4) compared to 56.3% (95% CI, 43.5 to 67.3) in the no-benefit group (Table 2). However, this

Table 1. Baseline patient characteristics in two adjuvant S-1–based doublet chemotherapy regimen groups

Variable	SP (n=60)	DS (n=58)	p-value
Sex			
Male	43 (71.7)	36 (62.1)	0.268
Female	17 (28.3)	22 (37.9)	
Age (yr)	59 (25-72)	56 (33-74)	0.366
Tumor stage			
T2	2 (3.3)	2 (3.5)	0.976
T3	18 (30.0)	13 (22.4)	0.347
T4	40 (66.7)	43 (74.1)	0.374
Nodal stage			
0	0	1 (1.7)	0.308
1	0	4 (6.9)	0.039
2	21 (35.0)	18 (31.0)	0.646
3	39 (65.0)	35 (60.3)	0.603
AJCC 8th stage			
IIIA	22 (36.7)	24 (41.4)	0.603
IIIB	25 (41.6)	16 (27.6)	0.107
IIIC	13 (21.7)	18 (31.0)	0.246
Expression of classifier genes			
CDX1 high	28 (46.7)	25 (43.1)	0.697
GZMB high	7 (11.7)	16 (27.6)	0.029
WARS high	9 (15.0)	10 (17.2)	0.741
SFRP4 high	33 (55.0)	28 (48.3)	0.465
Predictive classification			
Chemotherapy benefit	26 (43.3)	22 (37.9)	0.550
No benefit	34 (56.7)	36 (62.1)	
Prognostic classification			
Low-risk	5 (8.3)	8 (13.8)	0.342
Intermediate-risk	25 (41.7)	24 (41.4)	0.976
High-risk	30 (50.0)	26 (44.8)	0.576

Values are presented as number (%) or median (range). AJCC, American Joint Committee on Cancer; CDX1, Caudal type homebox1; DS, docetaxel plus S-1; GZMB, granzyme B; SFRP4, secreted frizzled-related protein 4; SP, S1 plus cisplatin; WARS, tryptophanyl-TRNA synthetase 1.

difference did not achieve statistical significance in both univariate analysis and multivariate analyses, adjusting for age, sex, tumor T category, N category, and treatment regimen (Table 2).

In the univariate Cox regression analysis, N category was significantly associated with 5-year OS, with an HR of 2.40 (95% CI, 1.18 to 4.86; p=0.015) (Fig. 3A). Other factors, such as the chemotherapy regimen (HR, 1.37; 95% CI, 0.76 to 2.49; p=0.296), the predictive single-patient classifier (HR, 1.38; 95% CI, 0.74 to 2.58; p=0.307), and the prognostic single-patient classifier (intermediate-risk vs low-risk: HR, 1.21; 95% CI, 0.41 to 3.60; p=0.729; high-risk vs. low-risk: HR,1.35; 95% CI, 0.47 to 3.92; p=0.576), did not show a significant association with 5-year OS.

In the multivariate analysis, the N category remained the only variable significantly associated with 5-year OS, with an adjusted HR of 2.38 (95% CI, 1.15 to 4.93; p=0.020) (Fig. 3B). The predictive and prognostic single-patient classifier, after adjustment for age, sex, chemotherapy regimen, tumor T category, and N category, demonstrated adjusted HRs of 1.48 (95% CI, 0.76 to 2.89; p=0.248), 1.65 (intermediate-risk vs. low-risk: 95% CI, 0.52 to 5.22; p=0.397), and 1.55 (high-risk vs. low-risk: 95% CI, 0.51 to 4.68; p=0.440), respectively (Table 2).

For 5-year DFS, a similar pattern emerged. The N category was significantly associated with 5-year DFS in both univariate and multivariate analyses, with an HR of 2.60 (95% CI 1.40 to 4.83; p=0.003) (S6A Fig.) in the univariate analysis and an HR of 2.49 (95% CI 1.31 to 4.71; p=0.005) (S6B Fig.) in the multivariate analysis. Although statistical significance was not reached for the predictive and prognostic single-patient classifiers, a trend in hazard ratios was observed in both the

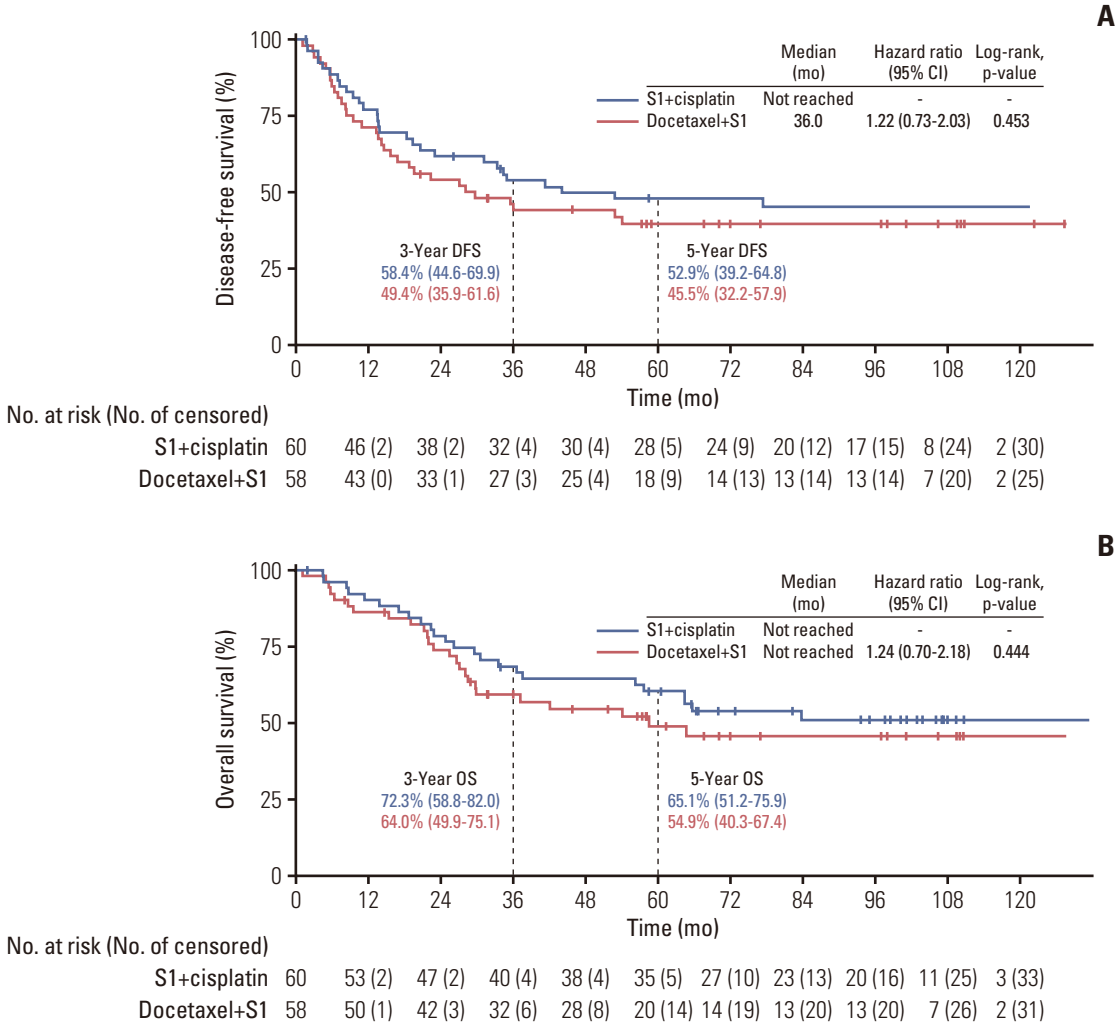

Fig. 2. Kaplan-Meier analysis of survival stratified by chemotherapy regimen. (A) Kaplan-Meier curve of disease-free survival with 3-year and 5-year disease-free survival. (B) Kaplan-Meier curve of overall survival with 3-year and 5-year disease-free survival. CI, confidence interval.

Table 2. Five-year overall survival by single-patient classifier groups in the total patient cohort

	No. (%)	5-Year overall survival (95% CI)	Univariate		Multivariate[a]	
			HR (95% CI)	p-value	HR (95% CI)	p-value
Low-risk	13 (11.0)	69.2 (37.3-87.2)	1 (ref)		1 (ref)	
Intermediate-risk	49 (41.5)	62.4 (46.6-74.8)	1.21 (0.41-3.60)	0.729	1.65 (0.52-5.22)	0.397
High-risk	56 (47.5)	55.6 (40.8-68.0)	1.35 (0.47-3.92)	0.576	1.55 (0.51-4.68)	0.440
Chemotherapy-benefit	48 (40.7)	66.6 (50.8-78.4)	1 (ref)		1 (ref)	
No-benefit	70 (59.3)	56.3 (43.5-67.3)	1.38 (0.74-2.58)	0.307	1.48 (0.76-2.89)	0.248

CI, confidence interval; HR, hazard ratio. [a]Model adjusted for age, sex, tumor T category, N category, and treatment regimen.

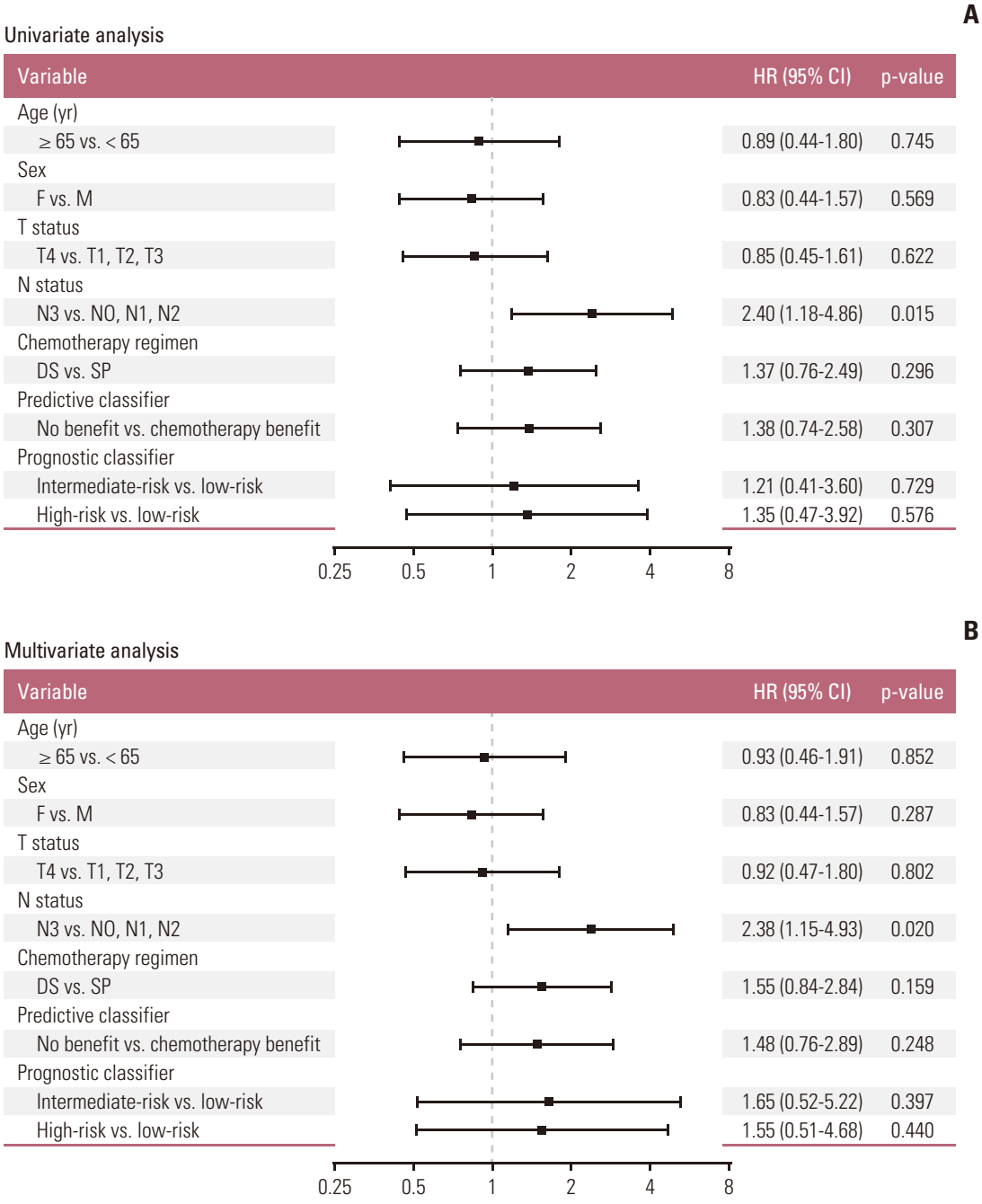

Fig. 3. Association between 5-year overall survival and single-patient classifier groups or baseline characteristics. (A) Univariate analysis. (B) Multivariate analysis adjusted for age, sex, T and N statuses, chemotherapy regimen, and single-patient classifier groups. p-values for association between clinical variables and adjuvant chemotherapy benefit are shown. CI, confidence interval; DS, docetaxel plus S-1; F, female; HR, hazard ratio; M, male; SP, S-1 plus cisplatin.

univariate and multivariate analyses, with results showing a parallel trend to those for 5-year OS (S6 Fig.).

3. Subgroup analysis of nProfiler1 based on treatment regimen and cancer stage

The validation of nProfiler1 was extended for different treatment regimens. In the SP group, the patients in the low-risk group demonstrated a better prognosis regarding

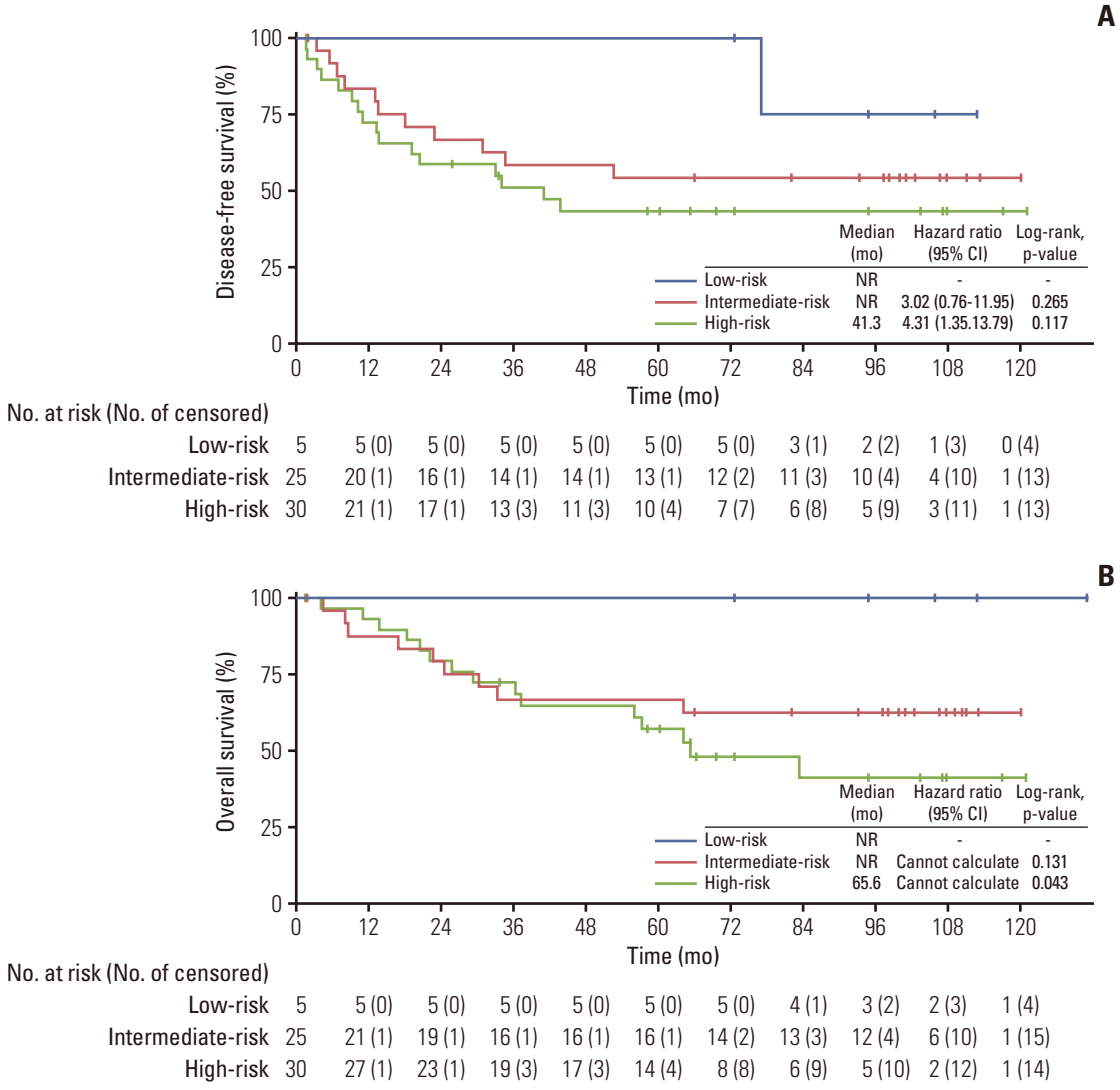

Fig. 4. Kaplan-Meier analysis of survival stratified by the prognostic single-patient classifier. (A) Disease-free survival of patients classified by the prognostic single-patient classifier in the S-1 plus cisplatin (SP) group. (B) Overall survival of patients classified by the prognostic single-patient classifier in the SP group. *(Continued to the next page)*

median DFS (HR, 0.23; 95% CI, 0.07 to 0.74; p=0.117) (Fig. 4A) and median OS (HR, could not be calculated; p=0.043) (Fig. 4B) than those in the high-risk group. However, this trend was not observed in the DS group, where the median DFS (HR, 0.97; 95% CI, 0.32 to 2.86; p=0.957) (Fig. 4C) and median OS (HR, 1.35; 95% CI, 0.40 to 4.76; p=0.594) (Fig. 4D) were inconsistent. Similar patterns were observed for the 5-year OS rates across the risk groups (S7 and S8 Tables).

Regarding the predictive single-patient classifier, the chemotherapy-benefit group had numerically longer DFS than the no-benefit group in the SP (HR, 0.84; 95% CI, 0.40 to 1.79; p=0.656) (S9A Fig.) and DS (HR, 0.93; 95% CI, 0.45 to 1.92; p=0.829) (S10A Fig.) groups. Regarding 5-year OS, the chemotherapy-benefit group exhibited better outcomes in the SP (71.5% vs. 60.6%, p=0.485) (S7 Table) and DS (60.7% vs. 51.9%, p=0.453) (S8 Table) groups. The chemotherapy-benefit group showed improved prognosis in both treatment groups; however, the predictive single-patient classifier had increased predictive value in the SP group compared with the DS group, which is consistent with the findings of the prognostic single-patient classifier.

As the inclusion criteria for the POST trial were histopathologically confirmed patients with stage III gastric cancer based on the AJCC 7th edition [22,24], we reclassified the patients using the AJCC 8th edition [23]. Patients' prognoses were well stratified based on clinical staging (S11 Table,

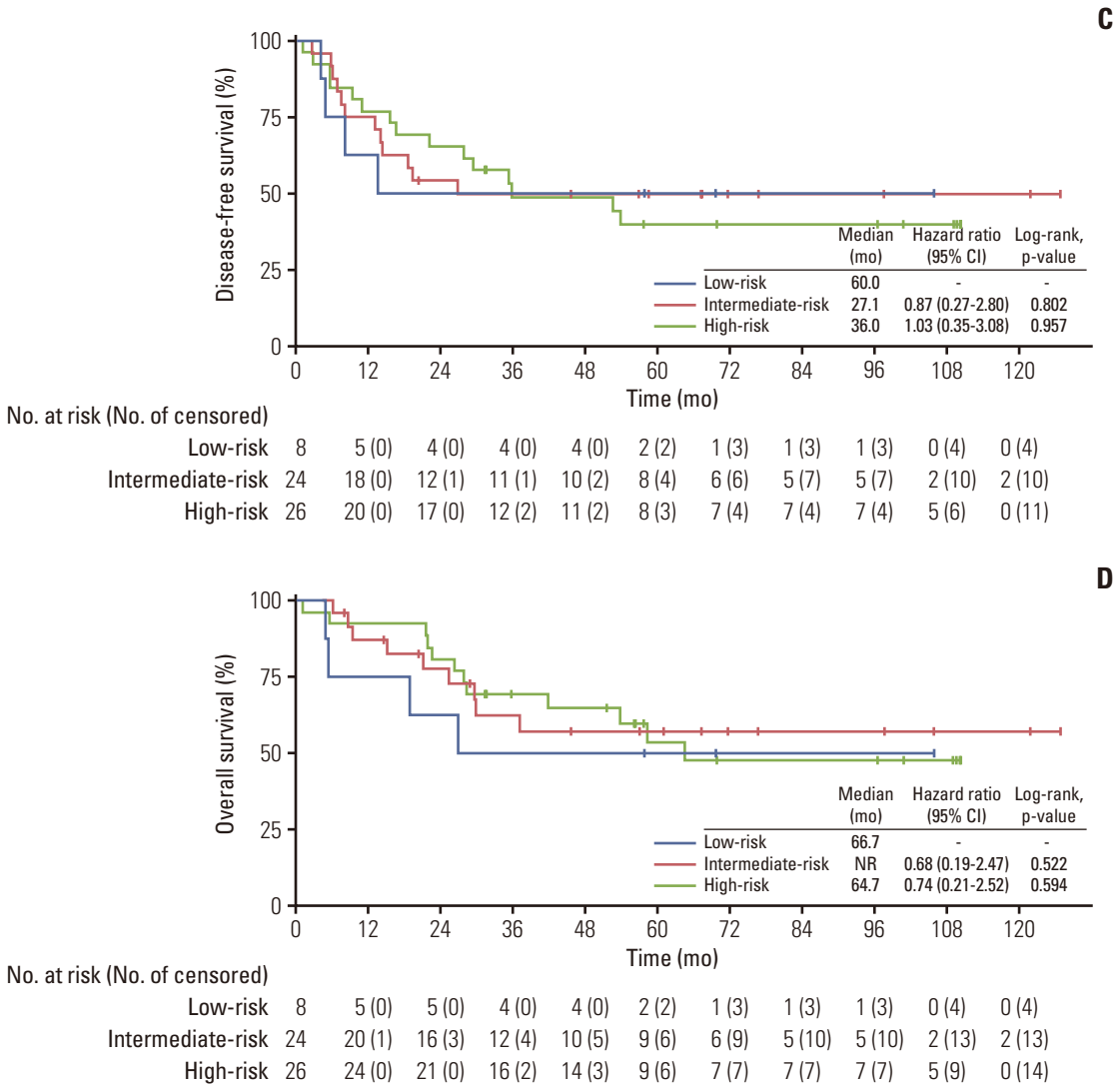

Fig. 4. *(Continued from the previous page)* (C) Disease-free survival of patients classified by the prognostic single-patient classifier in the docetaxel plus S-1 (DS) group. (D) Overall survival of patients classified by the prognostic single-patient classifier in the DS group. CI, confidence interval; NR, not reached.

S12 Fig.). The prognostic and predictive single-patient classifiers showed enhanced predictive value in patients with stage IIIA compared with patients with stage IIIB or IIIC (S13 Table, S14-16 Figs.).

Discussion

The POST trial, a randomized phase III study, evaluated the efficacy and safety of two S-1–based doublet regimens, SP and DS, in patients with stage III gastric cancer who underwent curative resection [23]. With a median follow-up of 57.9 months, the median OS was not reached in either treatment group. The median DFS was not reached in the SP group but was 36.0 months in the DS group. Despite a trend toward better prognosis in the SP group, the small sample size reduced statistical power, resulting in no significant differences between the two groups.

The nProfiler1 assay, a single-patient classifier based on the expression of four genes, provides prognostic and predictive stratification for patients with stages II-III gastric cancer [19]. The prognostic classifier categorizes individuals into low-risk, intermediate-risk, or high-risk groups based on immune status markers (GZMB and WARS) and stem-like module (SFRP4). Moreover, the predictive classifier evaluates the benefit of adjuvant chemotherapy, classify-

ing patients into chemotherapy-benefit or no-benefit groups based on immune status (GZMB and WARS) and intestinal epithelial module (CDX1).

The treatment effect by prognostic single-patient classifier groups was not significantly associated with chemotherapy benefit response or survival benefit in the Cox regression analysis. Although the 5-year OS rates and hazard ratios in the Cox regression analysis indicated a trend toward better outcomes in the low-risk group, this difference did not reach statistical significance, even after adjusting for age, sex, chemotherapy regimen, tumor T category, and N category. Similarly, the predictive single-patient classifier did not demonstrate a significant difference in 5-year OS or DFS between the two groups, despite a higher 5-year OS rate in the chemotherapy-benefit group. In contrast, N category consistently emerged as a prognostic factor for both 5-year OS and DFS.

The nProfiler1 was initially validated using patients from the CLASSIC trial, which included patients with stages II-IIIB gastric cancer treated with adjuvant doublet chemotherapy consisting of oral capecitabine and intravenous oxaliplatin [15]. Consequently, the assay is expected to perform optimally for doublet regimens combining an oral fluoropyrimidine with a platinum drug. However, taxane-containing regimens, such as docetaxel plus S-1 used in the JACCRO GC-07 trial and the perioperative FLOT regimen [17,18], are common in adjuvant chemotherapy for resectable gastric cancers. Our study indicates that the nProfiler1 assay effectively stratifies patients based on their risk and potential benefit from adjuvant chemotherapy, particularly for fluorouracil plus platinum doublet regimens but not for regimens containing docetaxel. This trend was particularly evident when comparing low-risk and high-risk groups stratified by the prognostic single-patient classifier, with a significant difference observed in the SP group but not in the DS group. SFRP4, a component of the prognostic classifier, is a WNT signaling-associated epithelial-mesenchymal transition modulator known to influence platinum drug resistance in gastric cancer through β-catenin dysregulation [28]. Consequently, SFRP4-high, high-risk patients in the DS group may experience greater benefit from docetaxel-based chemotherapy compared to platinum-based treatment. In contrast, low-risk patients with high GZMB and WARS expression, classified as no-benefit according to the predictive single-patient classifier, are unlikely to respond to docetaxel-based chemotherapy. This may contribute to the reduced discriminatory power of the prognostic single-patient classifier in the DS group compared to the SP group. This factor explains discrepancies in the predictive accuracy of the nProfiler1 assay across different treatment regimens.

The nProfiler1 assay was initially validated in patients with stages II-IIIB gastric cancer according to the AJCC 6th edition [19,29], and the POST trial enrolled patients with stage III gastric cancer according to the AJCC 7th edition [23,30]. In our post-hoc analysis, we evaluated patients using the AJCC 8th edition [27]. Staging, according to the AJCC eighth edition, demonstrated significant discrimination in terms of DFS and OS. The nProfiler1 performed better in patients with stage IIIA than in those with stage IIIB or IIIC. Excluding patients with stage IV, stratified based on the AJCC 6th edition, did not enhance the discriminatory power of the nProfiler1. Given that the CLASSIC trial included patients with stage II, whereas the POST trial only enrolled patients with stage III, the nProfiler1 assay may perform optimally in patients with a lower tumor burden.

This study has some potential limitations. First, the limited cohort size restricted our ability to achieve significance, allowing us to observe trends only. Archival tissues were collected from patients enrolled in the POST trial; however, the early termination of the trial resulted in difficulties in collecting tissues, reducing the power of the study. Second, the study was exclusively conducted in Korea, making it uncertain whether the nProfiler1 could effectively predict chemotherapy benefits and prognosis in different populations. In East Asia, adjuvant oral fluoropyrimidine plus platinum is commonly used, whereas the perioperative FLOT regimen is the standard of care in many Western countries. This analysis revealed impaired performance of the nProfiler1 assay in docetaxel-containing regimens. Given that the perioperative FLOT regimen includes a platinum drug and docetaxel, it is necessary to validate the nProfiler1 in this regimen. Therefore, further validation utilizing larger cohorts, including diverse ethnic populations treated with various regimens, is needed.

In conclusion, the nProfiler1 assay provides valuable insights into the prognosis and effectiveness of adjuvant chemotherapy based on fluorouracil plus platinum doublet regimens but not docetaxel-containing regimens. Further validation with larger patient cohorts and different regimens, including FLOT, is warranted.

Electronic Supplementary Material

Supplementary materials are available at Cancer Research and Treatment website (https://www.e-crt.org).

Ethical Statement

The protocol was approved by the institutional review and ethics board of each participating center. The study was conducted in accordance with the moral, ethical, and scientific principles governing clinical research as set out in the Declaration of Helsinki and the guidelines on Good Clinical Practice. All patients provided written, informed consent before enrollment. The trial is registered at ClinicalTrials.gov (NCT01283217).

Author Contributions

Conceived and designed the analysis: Lee CK, Kim H, Jung M.
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Conflicts of Interest

Conflict of interest relevant to this article was not reported.

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