

Thymidine phosphorylase expression and prognosis in colorectal cancer treated with 5-fluorouracil-based chemotherapy: A meta-analysis

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Abstract. In the past decades, various studies have suggested a possible link between thymidine phosphorylase (TP) level and colorectal cancer (CRC) treated with 5-fluorouracil (5-FU)-based chemotherapy; however, they have arrived at inconsistent results. Therefore, the present meta-analysis aimed to disclose a more comprehensive evaluation of this relationship. PubMed, the Cochrane Library, Ovid MEDLINE, Embase and China National Knowledge Infrastructure were systematically searched for studies that evaluated the prognostic value of TP in CRC. Stata 12.0 software was used to test the heterogeneity and evaluate the overall test performance. A total of 15 studies, including 1,225 patients, were included. The summary estimates of TP for CRC treated with 5-FU-based chemotherapy indicated a moderately positive prognosis with a hazard ratio (HR) of 0.76 (P=0.031) for overall survival and a HR of 0.711 (P=0.022) for relapse-free survival. On the basis of the present meta-analysis, TP could be promising and

meaningful in the prognosis of CRC treated with 5-FU-based chemotherapy.

Introduction

Colorectal cancer (CRC) is one of the most common human malignant tumors worldwide and morbidity associated with CRC is increasing annually (1). It has been reported that CRC was the third most commonly diagnosed cancer in males and the second in females, with ~1.4 million cases and 693,900 mortalities occurring in 2015 (2). To date, 5-fluorouracil (5-FU) is widely used as a primary chemotherapeutic agent and constitutes the fundamental basis of chemotherapy treatment for patients with CRC since it was introduced in 1957 (3-6). Although targeted epidermal growth factor receptor monoclonal antibodies, namely cetuximab and panitumumab, have been introduced and have been used for many patients with CRC and benefits are achieved, 5-FU is still a basic chemotherapy for CRC clinical treatment (7). Furthermore, several agents, such as 5-FU plus leucovorin (LV) or infusional 5-FU plus LV and oxaliplatin, have been established as the generalized regimen for the treatment of patients with CRC (8). For example, according to the guidelines of the National Comprehensive Cancer Network, FOLFOX6 chemotherapy including continuous infusion of 5-FU combined with oxaliplatin and calcium folinate has become the standard chemotherapy regimen for postoperative patients with CRC (9,10). In addition, oral forms of 5-FU-related drugs, such as capecitabine or tegafur plus uracil and doxifluridine (5'-DFUR), have also been developed for convenient administration and have been widely used in patients with CRC (11-13). However, the clinical effectiveness of 5-FU-based chemotherapy differs among patients (14). It is important to select 5-FU-based chemotherapy so that each patient may benefit and experience the least harmful side effects. To predict the clinical efficacy of 5-FU-based chemotherapy in CRC patients, it is essential to define a predictive biomarker associated with 5-FU treatment.

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Thymidine phosphorylase (TP) is an important metabolizing enzyme that catalyzes the conversion of 5-FU to its more active nucleoside form, 5-fluoro-2'-deoxyuridine, representing one of the main pathways through which this drug exerts its cytotoxic effect (15). One of the roles of TP is controlling the intracellular levels of thymidine, which at higher concentrations becomes toxic to cells and causes replication errors in DNA (16). Research has demonstrated that the levels of TP are higher in tumors compared with normal tissues in a wide range of solid tumors (15,17-20). The expression of TP may be correlated with the efficacy of 5-FU-based chemotherapy (21). However, in cancer development, it has been reported that TP functions as the molecule platelet-derived endothelial cell growth factor in cells and exhibits angiogenic properties in tumors (22,23). In 2009, a study by Bronckaers *et al* (15) reported that TP had a dual role in cancer development. TP may prevent apoptosis and induce angiogenesis to promote tumor growth and metastasis, which is the targeted function of TP inhibitors; however, TP is also indispensable for the activation of the extensively used 5-FU prodrugs, such as doxifluridine and capecitabine (15). As it has been demonstrated that TP has a complicated role in CRC development and 5-FU treatment, whether TP may predict the prognosis of patients with CRC treated with 5-FU-based chemotherapy remains uncertain. Various studies (4,5,24,25) have investigated the association between the levels of TP and survival in CRC patients; however, a certain conclusion regarding this has not been drawn. Although the majority have reported poorer overall survival (OS) and progression-free survival (PFS) in patients with tumors expressing high TP levels, there are also reports that have demonstrated no association between them, resulting in greatly different estimates of the prognostic value of TP expression between studies (26-29). The ability to use TP expression to predict the response of patients with CRC to 5-FU-based chemotherapy thus remains controversial.

The aim of the present study was to evaluate the scientific evidence for the effect of TP expression in patients with CRC treated with 5-FU-based chemotherapy by using a standard meta-analysis of data from published studies.

Data collection methods

Search strategy. Searches were conducted on Wiley Online Library (onlinelibrary.wiley.com), Scopus (scopus.com/home.uri), PubMed (ncbi.nlm.nih.gov/pubmed), the Web of Science (webofknowledge.com), the Cochrane library (cochranelibrary.com), Ovid MEDLINE (hsl.lib.umn.edu/biomed/help/ovid-medline), SinoMed (sinomed.ac.cn) and China National Knowledge Infrastructure (CNKI; cnki.net) without language limitation. The last search update was April 28, 2015. The search strategy predominantly included terms suggestive of four factors: i) TP (i.e., 'thymidine phosphorylase', 'platelet-derived endothelial cell growth factor' and 'PD-ECGF'); ii) 5-FU (i.e., '5-fluorouracil', 'adrucil', 'carac', 'efudex', 'efudix', '5-fluoro-1H, 3H-pyrimidine-2, 4-dione' and '5-fluoropyrimidines'); iii) colorectal (i.e., 'colon', 'rectal', 'colorectal' and 'rectum'); and iv) cancer (i.e., 'cancer', 'carcinoma', 'neoplasm', 'tumor' and 'malignant'). Article types were restricted to clinical trials or randomized controlled trials in humans. The reference lists of primary studies and previous meta-analyses were scrutinized for additional publications.

Inclusion and exclusion criteria. The potential trials were screened according to the following criteria: i) Patients had a diagnosis of CRC; ii) all patients received 5-FU-based chemotherapy; iii) the studies reported one or more indicators, including objective response rate (ORR), PFS, disease-free survival (DFS), relapse-free survival (RFS) and OS, to compare the prognosis of patients stratified by TP expression. Studies providing information on survival were included, while studies without survival analysis, investigating response rates only were excluded; iv) the results were part of the original analysis; v) when results reported by the same author were acquired from the same patient population in more than one publication, only the study involving the highest number of patients was included; and vi) retrospective, prospective or randomized controlled trials were included. Trials evaluating progression with time to tumor progression, which was defined as time from the initiation date of 5-FU-based chemotherapy to the first radiographic evidence of disease progression or mortality, were also included. Trials lacking complete data that were still in progress and without full text articles online were excluded. The present study attempted to obtain the data with the longest follow-up when reports overlapped or were repeated.

Data extraction and definitions. Data extracted included the first author, publication year, study type, chemotherapy regimen, lesions tested, TP evaluation method, study size, high TP level and percentage of patients with high TP expression. For clinical outcome, the number of responders were collected for calculating odds ratio (OR) and 95% estimation intervals for ORR. Hazard ratios (HRs) and 95% confidence intervals (CI) were also extracted for OS, PFS, DFS and RFS. If a separate HR was not provided, the HR and its variance were estimated from the published survival curves using previously described methods and models (30,31). Adjusted HRs and estimation intervals were also collected when reported. Objective response included complete response and partial response, and non-response consisted of stable disease and progressive disease, according to the Response Evaluation Criteria in Solid Tumors (32) or World Health Organization criteria (33). PFS was defined as the time from the initiation date of 5-FU-based therapy to the first evidence of disease progression or mortality from any cause. OS was defined as the time from the initiation date of 5-FU-based therapy to mortality from any cause. DFS was defined as the length of time after being treated with 5-FU-based therapy during which no disease was found. RFS was defined as the length of time after being treated with 5-FU-based therapy during which patients survived without any signs or symptoms of CRC. All data were extracted by two independent investigators. Discussions were used to reach an agreement if discrepancies existed.

Statistical analysis. The required information was extracted by two independent reviewers using pre-determined data extraction forms. The data were analyzed using Stata 12.0 software (StataCorp LP, College Station, TX, USA). Heterogeneity across studies was evaluated by the Q test and were quantified by the I² test. If the tests of heterogeneity were significant (P<0.05), the effect sizes were calculated with the random effects model using the DerSimonian-Laird method. Otherwise, the

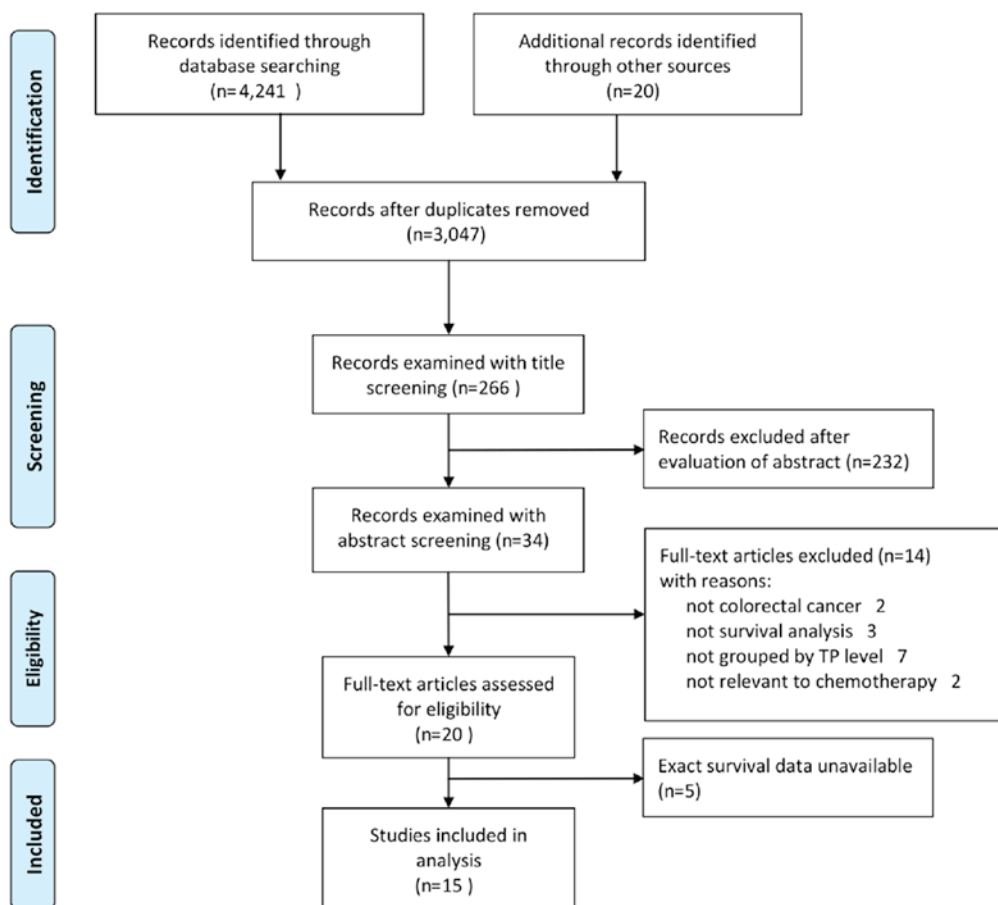


Figure 1. Flow chart of article selection for the present meta-analysis.

fixed-effect model with inverse variance weights was used. A funnel plot and an Egger test were used to assess publication bias. Subgroup analysis was conducted in the different treatment settings and TP detection methods. Sensitivity analysis was used to test the stability when large heterogeneity was presented. All P-values reported were two-sided. Publication biases were assessed by the Egger's test ($P < 0.05$ indicated an existing publication bias) and were reflected by the symmetry of the funnel plot on the natural logarithm of RRs or HRs (34).

Results

Study selection and characteristics. The search strategy identified 3,047 potentially relevant articles (398 from the Web of Science, 1,609 from the Wiley Online Library, 669 from Scopus, 237 from PubMed, 3 from Ovid MEDLINE, 17 from the Cochrane Library, 22 from SinoMed and 73 from CNKI). Following review of the titles, 266 of these studies were included. Subsequently, a total of 232 studies were excluded following abstract review. Among the 34 studies remaining, two were not relevant to CRC (studying TP in gastric cancer and hepatocellular carcinoma) and three were not survival analyses (comparing the TP level in different tissue or other research). There were seven studies in which the outcomes were not compared at different TP levels and two studies that were not relevant to 5-FU chemotherapy. Thus, 14 articles were excluded and 20 studies were eligible for data extraction. However, five of these 20 studies did not offer eligible data and

it was no possible to obtain exact survival information from these five articles. After completing the selection process, data from a total of 15 studies (4,5,24-29,35-41) involving 1225 patients (Fig. 1) were systematically analyzed.

Main characteristics for individual studies were summarized in Table I, including nationality, study type, 5-FU-based drugs and chemotherapy regimens. Of the 15 studies, 9 (4,24,25,27,36-39,41) were conducted in Asia and seven were from Japan. A total of 10 (4,5,25,26,28,35,37-39,41) were retrospective studies, one (29) was a prospective study and the other four (24,27,35,40) were not stated. Although the chemotherapy regimen varied in all 15 articles, they all used 5-FU-based drugs. Additionally, TP expression for individual studies were summarized in Table II. Treatment setting was separated into two kinds, primary and metastatic tumors, according to their different chemotherapy regimens. Of the studies, six articles studied primary tumors, seven studied metastatic tumors and two studied both. In terms of follow-up period, 10 articles reported the median follow-up period while another five articles did not. There were 13 studies (4,5,24-29,35-38,41) that stated the lesion tested. Among them, one study was tested on metastatic cancer tissue, 10 on primary tissue and two on both. There were two TP evaluation methods used among these studies, immunohistochemistry (IHC) and quantitative polymerase chain reaction (qPCR).

Assessment of study quality. To conduct the quality assessments for the 15 studies, the Newcastle-Ottawa Scale (NOS)

Table I. Summary of main characteristics for individual studies.

Author/(Refs.), year	Nationality	Study type	5-FU-based drugs	Chemotherapy regimen	Quality score
Ahn <i>et al</i> (24), 2005	Korea	NS	5-FU	FOLFIRI or FOLFOX	7
Kataoka <i>et al</i> (25), 2015	Japan	Retrospective	5-FU	FOLFOX + bevacizumab or FOLFOX + cetuximab	6
Shigeta <i>et al</i> (4), 2014	Japan	Retrospective	5-FU	5-FU + LV or UFT + LV	6
Ogawa <i>et al</i> (35), 2014	Japan	NS	5-FU	S-1	7
Donada <i>et al</i> (36), 2011	Italy	Retrospective	5-FU	5-FU + LV	6
Petrioli <i>et al</i> (26), 2010	Italy	Retrospective	5-FU	5-FU or CAP	5
Lindskog <i>et al</i> (5), 2014	Swiss	Retrospective	5-FU	5-FU + LV or 5-FU + OX or MIFL or CAP or CAP + OX or CAP + IRI	6
Yamada <i>et al</i> (27), 2008	Japan	NS	5-FU	UFT or UFT + LV	6
Jensen <i>et al</i> (28), 2008	Denmark	Retrospective	5-FU	Mayo	7
Yanagisawa <i>et al</i> (37), 2007	Japan	Retrospective	5-FU	MIFL	6
Meropol <i>et al</i> (29), 2006	USA	Prospective	5-FU	CAP + IRI	6
Ichikawa <i>et al</i> (38), 2003	Japan	Retrospective	5-FU	UFT + LV	7
Tokunaga <i>et al</i> (39), 2002	Japan	Retrospective	5-FU	UFT	7
Metzger <i>et al</i> (40), 1998	USA	NS	5-FU	5-FU/LV	7
Soong <i>et al</i> (41), 2008	Singapore	Retrospective	5-FU	5-FU/LV	6

FOLFIRI, 5-FU + leucovorin + irinotecan; FOLFOX, 5-FU + leucovorin + oxaliplatin; UFT, oral tegafur + uracil; S-1, tegafur + gimeracil + oteracil potassium; OX, oxaliplatin; CAP, capecitabine; IRI, irinotecan; Mayo, 5-FU + isovorin; MIFL, 5-FU + LV + irinotecan; 5-FU, 5-fluorouracil.

was used. The NOS is composed of eight items that assess patient selection, study, comparability and outcome (42). A summary of the quality assessment results is demonstrated in Table I.

Data analysis

OS. The meta-analysis was performed on 10 studies (887 patients) investigating the association between TP and OS. As the heterogeneity test was not significant ($\chi^2=11.42$; $P=0.326$; $I^2=12.4\%$), the fixed-effects model was used to calculate the HR. The pooled HR from the 10 studies was 0.76 ($P=0.031$; 95% CI, 0.59-0.98; Fig. 2), which indicated that there was significant correlation between the OS and TP in CRC patients treated with 5-FU-based chemotherapy. The funnel plot and Egger's test demonstrated that no significant publication bias was detected ($P=0.963$; Fig. 3). In order to test the stability of the result, the studies whose quality score was below five and was displayed as significant were excluded. Sensitivity analysis demonstrated that the result of OS was stable (Fig. 4).

Following this, analysis was restricted to the five studies assessing TP expression in primary tumors. The pooled HR was 0.62 ($P=0.005$; 95% CI, 0.45-0.87) without evidence of study heterogeneity ($\chi^2=1.20$; $P=0.878$; $I^2=0.0\%$). Five studies assessed TP expression in metastatic tumors, and the pooled HR was 0.90 ($P=0.594$; 95% CI, 0.61-1.32), without evidence of heterogeneity ($\chi^2=2.85$; $P=0.583$; $I^2=0.0\%$;

Fig. 5). To assess the effect of the method used to evaluate TP expression, subgroup analysis was performed based on IHC or qPCR. HR was pooled from all 10 studies using either qPCR or IHC. A larger pooled HR was demonstrated in studies using the qPCR method (HR=0.87; $P=0.396$; 95% CI, 0.62-1.21), compared with that from studies using the IHC method (HR=0.58; $P=0.022$; 95% CI, 0.40-0.85). There was no evidence of heterogeneity in qPCR-based studies ($\chi^2=2.65$; $P=0.619$; $I^2=0.0\%$) or IHC-based studies ($\chi^2=7.50$; $P=0.900$; $I^2=0.0\%$; Fig. 6).

ORR, PFS, DFS and RFS. Table III detailed the meta-analysis results of ORR, PFS, DFS and RFS. Of the 15 eligible studies, five (25,32-34,36) (200 patients) reported data available for ORR, and the pooled OR was 0.822 ($P=0.628$; 95% CI, 0.373-1.812) with evidence of heterogeneity ($\chi^2=10.56$; $P=0.031$; $I^2=62.4\%$). The Egger's test demonstrated that no publication bias was detected ($P=0.096$). There were three studies (4,29,33) for PFS, three (26,27,30) for DFS and two (3,31) for RFS. The pooled HRs for PFS, DFS and RFS were 0.752 ($P=0.511$; 95% CI, 0.321-1.760), 1.415 ($P=0.579$; 95% CI, 0.416-4.816) and 0.711 ($P=0.022$; 95% CI, 0.531-0.951) respectively, all without evidence of heterogeneity. The number of studies used here is not large enough to reach a conclusion. Therefore, more trials regarding this should be performed to fully determine the association between TP and survival in patients with CRC treated with 5-FU-based chemotherapy.

Table II. Summary of TP expression for individual studies.

Author/(Refs.), year	Treatment setting	Median follow-up period, months	Association with prognosis	Lesion tested	TP evaluation method	Study size (no. of patients)	Patients with high TP level, n (%)
Ahn <i>et al</i> (24), 2005	Metastatic	NS	None	Primary	IHC	45	22 (49)
Kataoka <i>et al</i> (25), 2015	Metastatic	42.6	None	Both	qPCR	36	18 (50)
Shigeta <i>et al</i> (4), 2014	Primary	66	Good	Primary	qPCR	101	71 (70)
Ogawa <i>et al</i> (35), 2014	Primary	12	Good	Primary	RT-qPCR	54	27 (50)
Donada <i>et al</i> (36), 2011	Primary	91.2	None	Primary	qPCR	55	27 (49)
Petrioli <i>et al</i> (26), 2010	Metastatic	20.4	Good	Metastatic	IHC	41	21 (51)
Lindskog <i>et al</i> (5), 2014	Metastatic	29	Poor	Primary	qPCR	125	62 (50)
Yamada <i>et al</i> (27), 2008	Primary	30	None	Primary	qPCR	103	51 (50)
Jensen <i>et al</i> (28), 2008	Both	NS	None	Primary	IHC	300	150 (50)
Yanagisawa <i>et al</i> (37), 2007	Primary	15.7	None	Primary	IHC	13	5 (39)
Meropol <i>et al</i> (29), 2006	Metastatic	NS	Good	Both	IHC	67	24 (36)
Ichikawa <i>et al</i> (38), 2003	Metastatic	14	None	Primary	qPCR	37	18 (49)
Tokunaga <i>et al</i> (39), 2002	Both	NS	Poor	NS	IHC	80	54 (68)
Metzger <i>et al</i> (40), 1998	Metastatic	NS	Poor	NS	RT-qPCR	38	10 (26)
Soong <i>et al</i> (41), 2008	Primary	52.4	Poor	Primary	IHC	130	86 (66)

TP, thymidine phosphorylase; qPCR, quantitative polymerase chain reaction; RT-qPCR, reverse transcription-quantitative polymerase chain reaction; IHC, immunohistochemistry; NS, not stated.

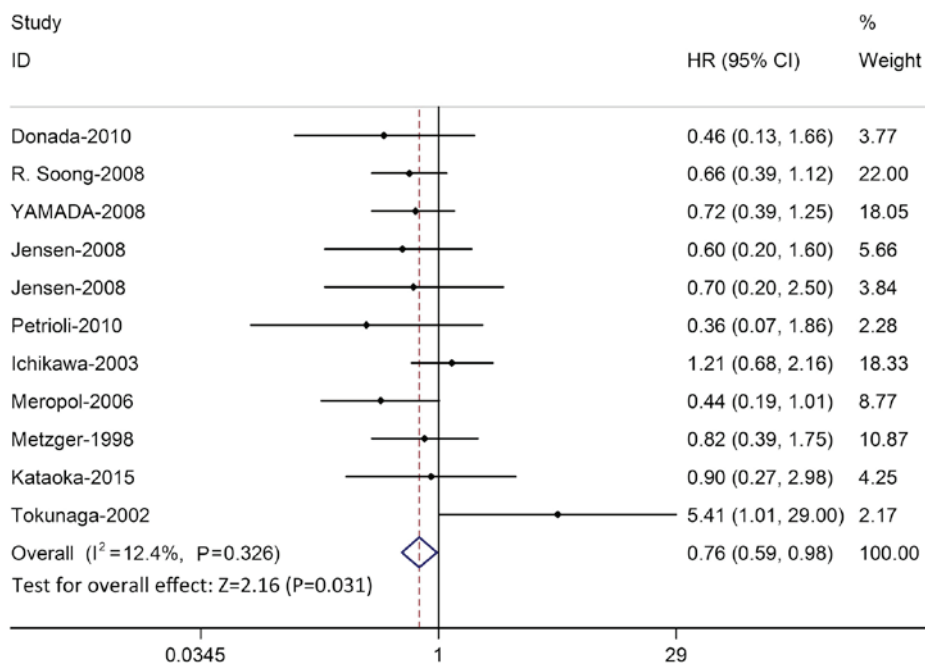


Figure 2. Forest plot of HR for the association of TP expression with overall survival (TP^h/high vs. TP^l/low). HR, hazard ratio; TP, thymidine phosphorylase; CI, confidence interval.

Discussion

The present meta-analysis reviewed 15 eligible articles to determine the association between TP expression and the

prognosis of patients with CRC treated with 5-FU-based chemotherapy. The results demonstrated that higher TP expression is correlated with better prognosis, as evaluated by OS, and may serve as a predictor of prognosis in 5-FU-based

Table III. Results of meta-analysis for ORR, PFS, DFS and RFS.

Index	No. of articles	Pooled odds ratio/hazard ratio	95% confidence interval	P-value	Heterogeneity			
					χ^2	I ²	P ¹	P ²
ORR	5	0.822	0.373-1.812	0.628	10.56	62.4%	0.031	0.096
PFS	3	0.752	0.321-1.760	0.511	13.49	85.2%	0.0001	0.361
DFS	3	1.415	0.416-4.816	0.579	5.14	61.1%	0.077	0.586
RFS	2	0.711	0.531-0.951	0.022	0.10	0.00%	0.754	-

ORR, objective response rate; PFS, progression-free survival; DFS, disease-free survival; RFS, relapse-free survival; P¹, P-value of heterogeneity; P², P-value of Egger's test; -, two studies is too few to perform Egger's test.

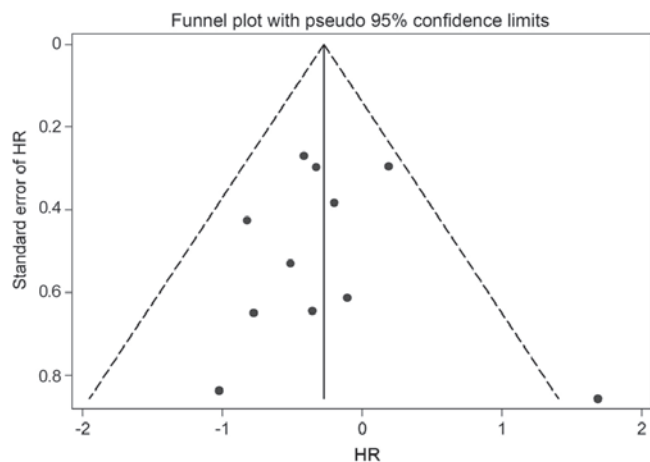


Figure 3. Funnel plot of overall survival. HR, hazard ratio.

chemotherapy for CRC. However, this is not the case for ORR, PFS, DFS and RFS. Furthermore, this seems to be the case for patients with primary tumors and patients whose TP evaluation method is IHC, according to subgroup analysis of OS. In patients with metastatic tumors and patients whose TP evaluation method is qPCR, TP expression does not appear to predict prognosis.

The value of high TP expression in predicting good OS appears to be stronger in studies conducted in a primary treatment setting than those conducted in a metastatic treatment setting. For studies that were all in a metastatic treatment setting and reported ORR and PFS, there was no significant difference between TP expression and prognosis of CRC. This may partly result from different 5-FU-based chemotherapy regimens that primary and metastatic tumors are usually treated with. Different drugs, such as oxaplatin, irinotecan and capecitabine, accompanied with 5-FU regimens may cause various effects (43). For DFS and RFS, which were both conducted in primary treatment settings, the opposite conclusion was reached. TP expression does not appear to predict prognosis of CRC treated with 5-FU-based chemotherapy in DFS; however, it does predict a good prognosis in RFS, although, there were only three studies dealing with DFS and two dealing with RFS. Therefore, these results should be interpreted with caution considering the small number of contributing studies.

Furthermore, it was observed that higher TP expression may predict better prognosis in studies using IHC but not qPCR. This may be due to the different cut-off values used to assign TP status in the qPCR studies. Dichotomization in some of the qPCR studies depended on median value, while others depended on likely response. This was not the case for the IHC studies. TP expression in the IHC studies was quantified by a visual grading system based on the intensity of staining and classified into four grades, from 0 (undetectable staining) to 3 (very high intensity of staining).

As TP is an enzyme that not only participates in 5-FU metabolism, but also converts 5'-DFUR to 5-FU (21), it was hypothesized to be a potential predictor of response. However, experimental studies also reported that high TP expression is associated with the decreased sensitivity of CRC to 5-FU (44) and some clinical trials demonstrated no clinically useful correlation between TP expression and the response to post-operative adjuvant chemotherapy with agents such as 5-FU/leucovorin and 5'-DFUR (28). The earlier results were always controversial, while the results of the present meta-analysis are consistent with the previous three articles by Ogawa *et al* (35), Petrioli *et al* (26) and Meropol *et al* (29), which indicated a positive correlation between high TP expression and positive outcomes in CRC treated with 5-FU-based chemotherapy.

The results of the association between TP expression and the prognosis of 5-FU-based chemotherapy in CRC varied among the 15 articles chosen for analysis. Six articles by Ahn *et al* (24), Yamada *et al* (27), Jensen *et al* (28), Donada *et al* (36) and Ichikawa *et al* (38) indicated that there was no association between the expression of TP and the prognosis of 5-FU-based chemotherapy. Two articles by Kataoka *et al* (25) and Yanagisawa *et al* (37) identified the association but did not express it in detail. However, the trial conducted by Shigeta *et al* (4) indicated that high TP expression was associated with a trend for improved prognosis in RFS. There were also two articles by Lindskog *et al* (5) and Tokunaga *et al* (39) that indicated that high or low TP expression was an independent poor prognostic factor, in contrast to the present result. The apparent discrepancy may be explained in several ways. First, sample size may be insufficient to achieve adequate statistical power for specific biomarker end points. The number of CRC patients participating in the trials should be higher, so that the discrepancy

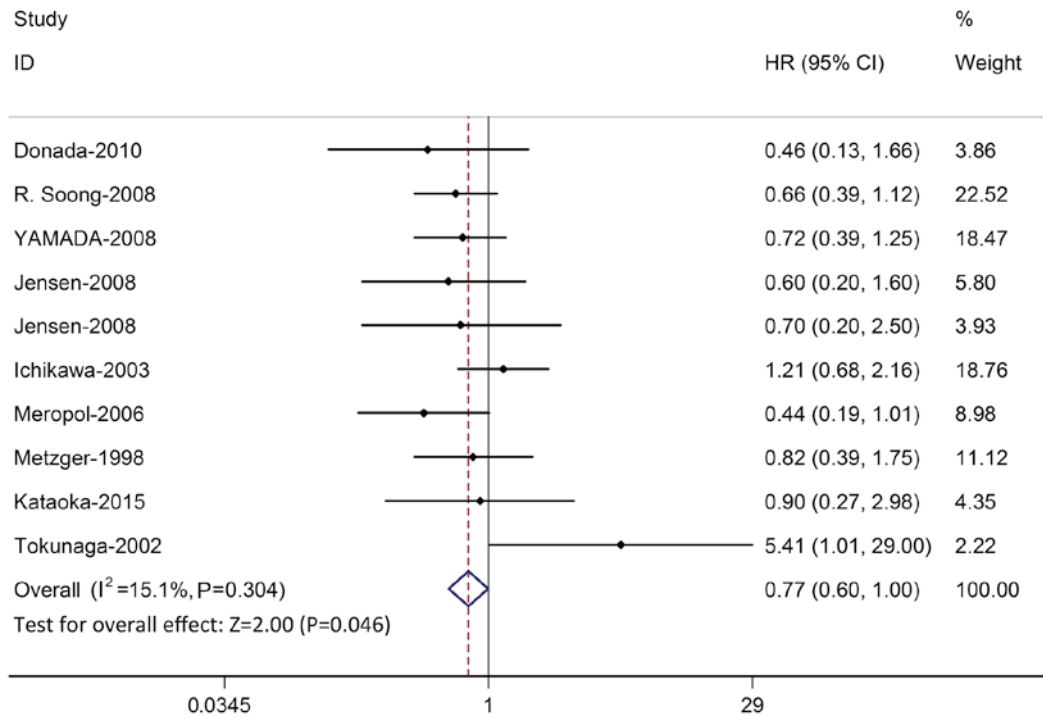


Figure 4. Forest plot of sensitivity analysis for overall survival. HR, hazard ratio; CI, confidence interval.

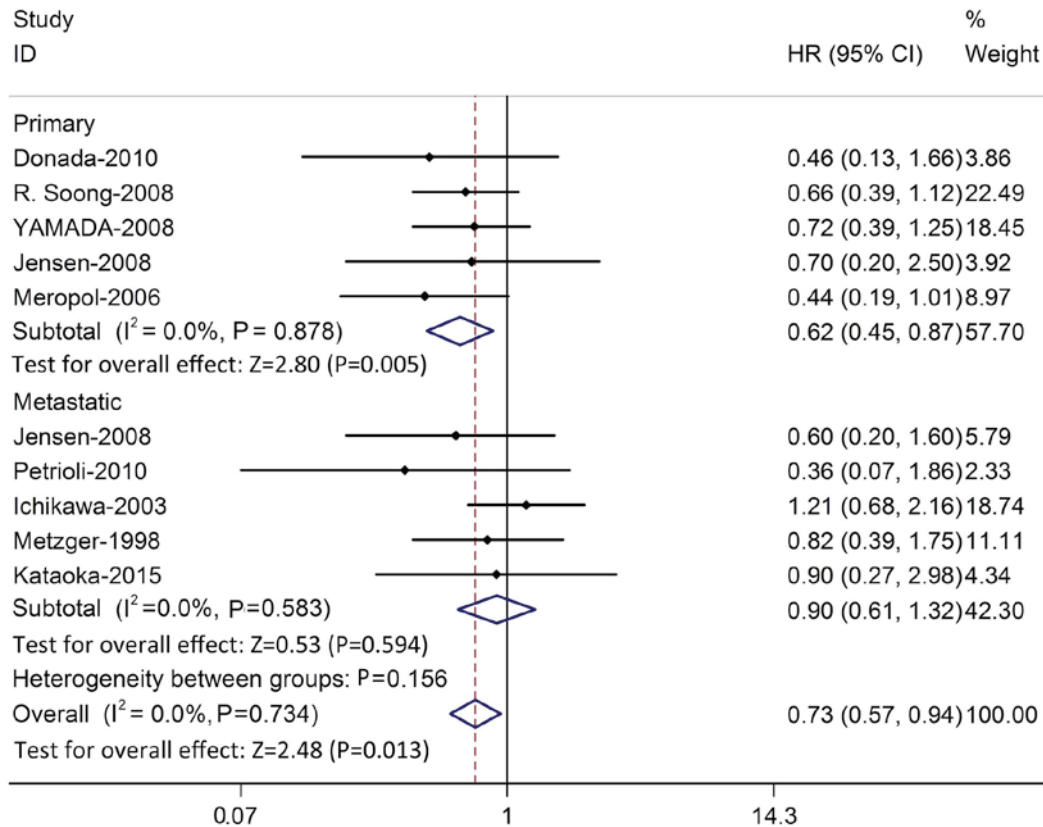


Figure 5. Forest plot of HR for the association of TP expression with overall survival based on treatment setting (TP^h/high vs. TP^l/low). HR, hazard ratio; TP, thymidine phosphorylase; CI, confidence interval.

between results may be minimized. Second, the inverse association between TP expression and the DFS and response to 5-FU may be a consequence of the role of TP as an

angiogenic factor. TP, which is identical to platelet-derived endothelial cell growth factor (45), and the degeneration products, thymine and 2-deoxy-D-ribose, have angiogenic

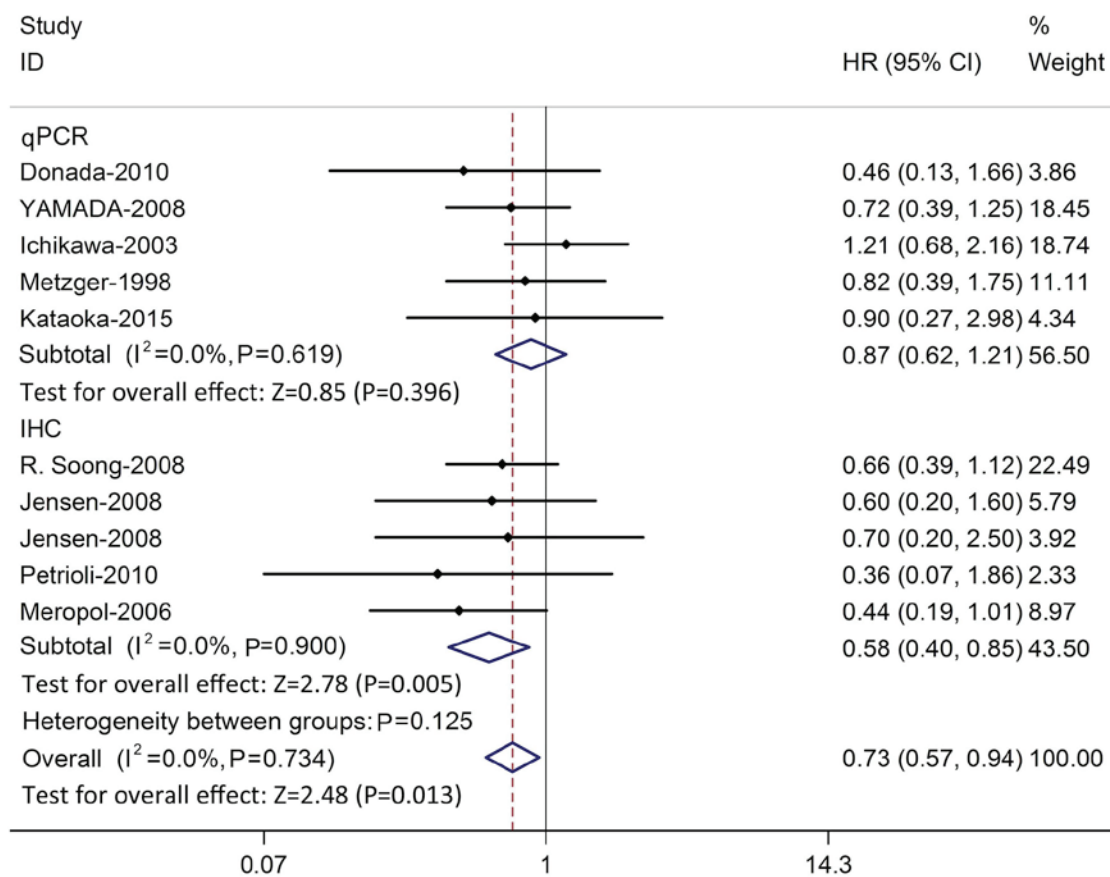


Figure 6. Forest plot of HR for the association of TP expression with overall survival based on TP evaluation method (TP⁺/high vs. TP⁻/low). HR, hazard ratio; TP, thymidine phosphorylase; CI, confidence interval; qPCR, quantitative polymerase chain reaction; IHC, immunohistochemistry.

and anti-apoptotic effects (46). Although the role of TP in tumor proliferation is yet to be fully elucidated, TP has angiogenetic activity and its enzymatic activity is required for angiogenesis (47). A previous study demonstrated that TP prevents hypoxia-induced apoptosis and that the degradation products of thymidine are involved in this response (48). Thus, TP expression may provide an advantage for tumor growth in CRC by not only increasing the intratumoral microvessel density, but also by attenuating apoptosis (46), which suggests that the suppression of TP may result in the inhibition of growth of TP-positive tumors in patients with CRC.

Furthermore, the present results suggested that the association between TP expression and prognosis of CRC is different between primary CRC tumors and metastatic CRC tumors. It was hypothesized that TP may be correlated with metastasis and advanced CRC, and it was also considered that TP may induce angiogenesis in tumor tissues. Perhaps one of the dual roles of TP, that it participates in the metabolism of 5-FU in CRC cancer cells to defend cancer cells, is stronger than the role of it inducing angiogenesis. However, the mechanisms need to be further studied, as the interactions between TP expression and other factors of angiogenesis are not known.

The present meta-analysis has several notable limitations. First, the cut-off line of high and low TP expression was different across each trial, and it was not defined with

a standardized value in the present review. Second, only six trials reported HRs and variances, and so HRs and variances had to be calculated or converted for other trials from the reported survival curves, which may introduce unavoidable bias. Third, the majority of trials were retrospective trials, which may cause selective bias. Fourth, the lesions tested were predominantly from primary tumors, which may result in bias. Finally, 5-FU, utilized as a first line treatment for CRC, is used as an intramuscular injection agent, or in combination with oral drugs, such as capecitabine and tegafur, which may influence the efficacy of 5-FU.

In spite of the above limitations, the present meta-analysis demonstrated that higher TP expression is correlated with better prognosis in CRC treated with 5-FU-based chemotherapy. Additional investigation is necessary to provide more specific information about the association between TP expression and 5-FU-based treatment for patients with CRC.

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