

S2 Table. TS expression and 5-FU sensitivity in human cancers - a survey of the literature.^a

Authors	Year	Tumour	Patient No.	Method	Response ^b	Outcome ^c (5-FU+) ^d	Outcome (5-FU-) ^e	Reference
Okonkwo A	2001	Rectal cancer	25	IHC	Yes*	-	-	1
Cascinu S	2001	CRC	150	IHC	-	Yes (DFS)	-	2
Edler D	2002	CRC	862	IHC	-	No (+DFS)	Yes	3
Aschele C	2002	CRC	124	IHC	Yes	Yes (+PFS)	-	4
Tajima Y	2003	GC	137	IHC	-	Yes*	-	5
Tahara M	2004	GC	29	IHC	No	-	-	6
Shintani Y	2004	NSCLC	116	IHC	-	-	Yes (DFS)	7
Kamoshida S	2004	CRC	37	IHC	Yes	-	-	8
Yu Z	2005	BC	197	IHC	-	Yes	-	9
Jakob C	2005	CRC	65	IHC	Yes	-	-	10
Kawano K	2006	SCC	140	IHC	-	Yes	-	11
Dotor E	2006	CRC	129	IHC	-	No (+DFS)	-	12
Fernandez-Contreras ME	2006	CRC	140	IHC	-	Yes (PFS)	-	13
Ajiki T	2006	Gall bladder cancer	53	IHC	-	Yes*	-	14
Sinicrope FA	2006	CRC	320	IHC	-	No (+DFS)	-	15
Harada K	2006	EC	25	IHC	-	Yes	-	16
Fukuda H	2006	GC	62	IHC	Yes	-	-	17
Nakano J	2006	NSCLC	151	IHC	-	Yes	-	18
Popat S	2006	CRC	967	IHC	-	No	-	19
Ciaparrone M	2006	CRC	62	IHC	-	Yes	-	20
Jensen SA	2007	CRC	303	IHC	-	Yes*	-	21
Kwon HC	2007	GC	64	IHC	No	No	-	22
Bertolini F	2007	CRC	91	IHC	No	No	-	23
Bendardaf R	2007	CRC	39	IHC	No	No (DFS)	-	24
Li Y	2007	PC	52	IHC	-	Yes (DFS)	-	25
Miyoshi T	2007	NSCLC	54	IHC	-	Yes	-	26
Toi M	2007	BC	192	IHC	-	No (DFS)	Yes (DFS)	27
Bendardaf R	2007	CRC	73	IHC	Yes	Yes (DFS)	-	28
Kim MK	2008	EC	108	IHC	Yes	No	-	29
Negri FV	2008	CRC	57	IHC	Yes	-	-	30
Lee J	2008	GC	467	IHC	-	No	-	31
Soong R	2008	CRC	945	IHC	-	No	Yes*	32
Zheng Z	2008	NSCLC	160	IHC	-	Yes	-	33
Ceppi P	2008	GEP & lung NET	116	IHC	-	Yes (+PFS)	-	34
Honda J	2008	BC	25	IHC	No	-	-	35
Kuo SJ	2008	BC	129	IHC	-	Yes	-	36
Gosens MJ	2008	CRC	38	IHC	-	No	-	37
Bendardaf R	2008	CRC	67	IHC	-	No/Yes (DFS)	-	38
Li Y	2009	Bladder cancer	67	IHC	-	Yes (DFS)	-	39
Moreira LR	2009	CRC	32	IHC	-	No	-	40
Kim SH	2009	CRC	70	IHC	Yes	Yes	-	41
Kim JS	2009	GC	153	IHC	-	No (+DFS)	-	42
Ren DN	2009	CRC	150	IHC	-	No	-	43
Kostopoulos I	2009	CRC	498	IHC	-	No (+DFS)	-	44
Cullen KJ	2010	Head and neck SCC	262	IHC	-	No (+DFS)	-	45
Lee HW	2010	NPC	41	IHC	-	No	-	46
Karlberg M	2010	CRC	1389	IHC	-	-	Yes	47
Lee S	2010	EC	72	IHC	Yes	Yes (PFS)	-	48
O'Toole D	2010	GEP NET	18	IHC	No	-	-	49
Choi IS	2011	GC	41	IHC	-	No (+PFS)	-	50
Jeong SH	2011	GC	72	IHC	-	No	-	51
Nishi M	2011	CCC	29	IHC	-	-	No (+DFS)	52
Yasumatsu R	2011	SCC	47	IHC	No	-	-	53
Lee SJ	2011	BC	90	IHC	-	Yes (PFS)	-	54
Conradi LC	2011	Rectal cancer	208	IHC	-	Yes (+DFS)	-	55
Kondo N	2011	PC	106	IHC	-	-	No	56
Kaira K	2011	CRC	80	IHC	-	-	Yes (PFS)	57
Noda E	2012	CRC	39	IHC	No	-	Yes (PFS)	58
van der Zee JA	2012	PC	212	IHC	-	Yes*/No*(+DFS)	Yes (PFS)	59
Formentini A	2012	CRC	140	IHC	-	Yes	Yes (PFS)	60
Yokota K	2012	Thymic carcinoma	24	IHC	-	No	Yes (PFS)	61
Arsenjevic T	2012	SCC	41	IHC	No	-	Yes (PFS)	62
Sulzyc-Bielicka V	2013	CRC	189	IHC	-	Yes (+DFS)	Yes (PFS)	63

a Studies are limited to those that employed immunohistochemistry for TS expression analyses.

b Patient responses to chemotherapy including 5-FU: Yes, a positive correlation between high TS expression and poor responses; No, a negative

c Patient outcomes defined as overall survival, unless otherwise indicated: DFS, disease-free survival; PFS, progression-free survival; Yes, a positive correlation between high TS expression and poor patient outcomes; No, a negative correlation, Yes*, a positive correlation under specific conditions.

d The subjects were patients who underwent chemotherapy including 5-FU

e The subjects were patients who underwent surgery only.

f Other abbreviations: IHC, immunohistochemistry; CRC, colorectal cancer; GC, gastric cancer; NSCLC, non-small cell lung cancer; BC, breast cancer; SCC, squamous cell carcinoma; EC, oesophageal cancer; GEP NET, gastroenteropancreatic neuroendocrine tumours; NPC, nasopharyngeal cancer; CCC, intrahepatic cholangiocell carcinoma; Pancreatic cancer, PC.

References

- Okonkwo A, Musunuri S, Talamonti M, Benson A, 3rd, Small W, Jr., et al. (2001) Molecular markers and prediction of response to chemoradiation in rectal cancer. *Oncol Rep* 8: 497-500.
- Cascinu S, Graziano F, Valentini M, Catalano V, Giordani P, et al. (2001) Vascular endothelial growth factor expression, S-phase fraction and thymidylate synthase quantitation in node-positive colon cancer: relationships with tumor recurrence and resistance to adjuvant chemotherapy. *Ann Oncol* 12: 239-244.
- Elder D, Glimelius B, Hallstrom M, Jakobsen A, Johnston PG, et al. (2002) Thymidylate synthase expression in colorectal cancer: a prognostic and predictive marker of benefit from adjuvant fluorouracil-based chemotherapy. *J Clin Oncol* 20: 1721-1728.
- Aschele C, Debernardi D, Bandelloni R, Cascinu S, Catalano V, et al. (2002) Thymidylate synthase protein expression in colorectal cancer metastases predicts for clinical outcome to leucovorin-modulated bolus or infusional 5-fluorouracil but not methotrexate-modulated bolus 5-fluorouracil. *Ann Oncol* 13: 1882-1892.
- Tajima Y, Shimoda T, Nakanishi Y, Yokoyama N, Tanaka T, et al. (2003) Association of gastric and intestinal phenotypic marker expression of gastric carcinomas with tumor thymidylate synthase expression and response to postoperative chemotherapy with 5-fluorouracil. *J Cancer Res Clin Oncol* 129: 683-690.
- Tahara M, Ochiai A, Fujimoto J, Bokai N, Yasui W, et al. (2004) Expression of thymidylate synthase, thymidine phosphorylase, dihydropyrimidine dehydrogenase, E2F-1, Bcl-X, and Bcl-2, and clinical outcomes for gastric cancer patients treated with bolus 5-fluorouracil. *Oncol Rep* 11: 9-15.
- Shintani Y, Ohta M, Hirabayashi H, Tanaka H, Iuchi K, et al. (2004) Thymidylate synthase and dihydropyrimidine dehydrogenase mRNA levels in tumor tissues and the efficacy of 5-fluorouracil in patients with non-small-cell lung cancer. *Lung Cancer* 45: 189-196.
- Kamoshida S, Matsuoka H, Shioyama K, Matsuyama A, Shimomura R, et al. (2004) Immunohistochemical analysis of thymidylate synthase, p16(INK4a), cyclin-dependent kinase 4 and cyclin D1 in colorectal cancers receiving preoperative chemotherapy: significance of p16(INK4a)-mediated cellular arrest as an indicator of chemosensitivity to 5-fluorouracil. *Pathol Int* 54: 564-575.
- Yu Z, Sun J, Zhen J, Zhang Q, Yang Q (2005) Thymidylate synthase predicts for clinical outcome in invasive breast cancer. *Histol Histopathol* 20: 871-878.
- Jakob C, Liersch T, Meyer W, Baretton GB, Haussler P, et al. (2005) Immunohistochemical analysis of thymidylate synthase, thymidine phosphorylase, and dihydropyrimidine dehydrogenase in rectal cancer (cUICC II/III): correlation with histopathologic tumor regression after 5-fluorouracil-based long-term neoadjuvant chemoradiotherapy. *Am J Surg Pathol* 29: 1304-1309.
- Kavano K, Yanagisawa S, Kusukawa J, Sunagawa H, Shiba R, et al. (2006) Intratumoral expression of thymidylate synthase is an independent predictor of prognosis in patients with squamous cell carcinoma of the tongue: results from a retrospective study. *Int J Oral Maxillofac Surg* 35: 258-264.
- Dotor E, Cuatrecasas M, Martinez-Izesta M, Navarro M, Vilardell F, et al. (2006) Tumor thymidylate synthase 1494del6 genotype as a prognostic factor in colorectal cancer patients receiving fluorouracil-based adjuvant treatment. *J Clin Oncol* 24: 1603-1611.
- Fernandez-Contreras ME, Sanchez-Prudencio S, Sanchez-Hernandez JJ, Garcia de Paredes ML, Gisbert JP, et al. (2006) Thymidylate synthase expression pattern, expression level and single nucleotide polymorphism are predictors for disease-free survival in patients of colorectal cancer treated with 5-fluorouracil. *Int J Oncol* 28: 1303-1310.
- Ajiki T, Hirata K, Okazaki T, Horuchi H, Fujita T, et al. (2006) Thymidylate synthase and dihydropyrimidine dehydrogenase expressions in gallbladder cancer. *Anticancer Res* 26: 1391-1396.
- Sinicrope FA, Rego RL, Halling KC, Foster NR, Sargent DJ, et al. (2006) Thymidylate synthase expression in colon carcinomas with microsatellite instability. *Clin Cancer Res* 12: 2738-2744.
- Harada K, Kawashima Y, Yoshida H, Sato M (2006) Thymidylate synthase expression in oral squamous cell carcinoma predicts response to S-1. *Oncol Rep* 15: 1417-1423.
- Fukuda H, Takiguchi N, Koda K, Oda K, Seike K, et al. (2006) Thymidylate synthase and dihydropyrimidine dehydrogenase are related to histological effects of 5-fluorouracil and cisplatin neoadjuvant chemotherapy for primary gastric cancer patients. *Cancer Invest* 24: 235-241.
- Nakano J, Huang C, Liu D, Masuya D, Nakashima T, et al. (2006) Evaluations of biomarkers associated with 5-FU sensitivity for non-small-cell lung cancer patients postoperatively treated with UFT. *Br J Cancer* 95: 607-615.
- Popat S, Chen Z, Zhao D, Pan H, Hearle N, et al. (2006) A prospective, blinded analysis of thymidylate synthase and p53 expression as prognostic markers in the adjuvant treatment of colorectal cancer. *Ann Oncol* 17: 1810-1817.
- Ciaparrone M, Quirino M, Schinzari G, Zannoni G, Corsi DC, et al. (2006) Predictive role of thymidylate synthase, dihydropyrimidine dehydrogenase and thymidine phosphorylase expression in colorectal cancer patients receiving adjuvant 5-fluorouracil. *Oncology* 70: 366-377.
- Jensen SA, Vainer B, Sorensen JB (2007) The prognostic significance of thymidylate synthase and dihydropyrimidine dehydrogenase in colorectal cancer of 303 patients adjuvantly treated with 5-fluorouracil. *Int J Cancer* 120: 694-701.
- Kwon HC, Roh MS, Oh SY, Kim SH, Kim MC, et al. (2007) Prognostic value of expression of ERCC1, thymidylate synthase, and glutathione S-transferase P1 for 5-fluorouracil/oxaliplatin chemotherapy in advanced gastric cancer. *Ann Oncol* 18: 504-509.
- Bertolini F, Bengala C, Losi L, Paganò M, Iachetta F, et al. (2007) Prognostic and predictive value of baseline and posttreatment molecular marker expression in locally advanced rectal cancer treated with neoadjuvant chemoradiotherapy. *Int J Radiat Oncol Biol Phys* 68: 1455-1461.
- Bendardaf R, Elzagheid A, Lamum H, Algars A, Korkeila E, et al. (2007) Thymidylate synthase expression in primary colorectal tumours is correlated with its expression in metastases. *Scand J Gastroenterol* 42: 471-476.
- Li Y, Mizutani Y, Shiraiishi T, Okihara K, Ukimura O, et al. (2007) Prognostic significance of thymidylate synthase expression in patients with prostate cancer undergoing radical prostatectomy. *Urology* 69: 988-995.
- Miyoshi T, Kondo K, Toba H, Yoshida M, Fujino H, et al. (2007) Predictive value of thymidylate synthase and dihydropyrimidine dehydrogenase expression in tumor tissue, regarding the efficacy of postoperatively administered UFT (tegafur-uracil) in patients with non-small cell lung cancer. *Anticancer Res* 27: 2641-2648.
- Toi M, Ikeda T, Akiyama F, Kurosumi M, Tsuda H, et al. (2007) Predictive implications of nucleoside metabolizing enzymes in premenopausal women with node-positive primary breast cancer who were randomly assigned to receive tamoxifen alone or tamoxifen plus tegafur-uracil as adjuvant therapy. *Int J Oncol* 31: 899-906.
- Bendardaf R, Lamum H, Ristamaki R, Korkeila E, Syrjanen K, et al. (2008) Thymidylate synthase and microsatellite instability in colorectal cancer: implications for disease free survival, treatment response and survival with metastases. *Acta Oncol* 47: 1046-1053.
- Kim MK, Cho KJ, Kwon JG, Park SJ, Kim YH, et al. (2008) ERCC1 predicting chemoradiation resistance and poor outcome in esophageal cancer. *Eur J Cancer* 44: 54-60.
- Negri FV, Campanini N, Camisa R, Pucci F, Bui S, et al. (2008) Biological predictive factors in rectal cancer treated with preoperative radiotherapy or radiochemotherapy. *Br J Cancer* 98: 143-147.
- Lee J, Park CK, Park JO, Lim T, Park YS, et al. (2008) Impact of E2F-1 expression on clinical outcome of gastric adenocarcinoma patients with adjuvant chemoradiation therapy. *Clin Cancer Res* 14: 82-88.
- Soong R, Shah N, Salto-Tellez M, Tai BC, Soo RA, et al. (2008) Prognostic significance of thymidylate synthase, dihydropyrimidine dehydrogenase and thymidine phosphorylase protein expression in colorectal cancer patients treated with or without 5-fluorouracil-based chemotherapy. *Ann Oncol* 19: 915-919.
- Zheng Z, Li X, Schell MJ, Chen T, Boulware D, et al. (2008) Thymidylate synthase in situ protein expression and survival in stage I nonsmall cell lung cancer. *Cancer* 112: 2765-2773.
- Ceppi P, Volante M, Ferrero A, Rigli L, Rapa L, et al. (2008) Thymidylate synthase expression in gastroenteropancreatic and primary neuroendocrine tumors. *Clin Cancer Res* 14: 1059-1064.
- Honda J, Sasa M, Moriya T, Bando Y, Hirose T, et al. (2008) Thymidine phosphorylase and dihydropyrimidine dehydrogenase are predictive factors of therapeutic efficacy of capecitabine monotherapy for breast cancer-preliminary results. *J Med Invest* 55: 54-60.
- Kuo SJ, Wang HC, Chow KC, Chiou SH, Chiang SF, et al. (2008) Expression of rTsbeta as a 5-fluorouracil resistance marker in patients with primary breast cancer. *Oncol Rep* 19: 881-888.
- Gosens MJ, Moerland E, Lemmens VP, Rutten HT, Tan-Go I, et al. (2008) Thymidylate synthase genotyping is more predictive for therapy response than immunohistochemistry in patients with colon cancer. *Int J Cancer*.
- Bendardaf R, Ristamaki R, Syrjanen K, Pyyrönen S (2008) Bcl-2 expression significantly correlates with thymidylate synthase expression in colorectal cancer patients. *World J Gastroenterol* 14: 6218-6223.
- Li Y, Li X, Dai H, Sun X, Li J, et al. (2009) Thymidylate synthase was associated with patient prognosis and the response to adjuvant therapy in bladder cancer. *BJU Int* 103: 547-552.
- Moreira LR, Schenka AA, Latuff Filho P, Nascimento H, Passos Lima CS, et al. (2009) Correlation between thymidylate synthase protein expression and gene polymorphism with clinicopathological parameters in colorectal carcinoma. *Int J Surg Pathol* 17: 181-186.
- Kim SH, Kwon HC, Oh SY, Lee DM, Lee S, et al. (2009) Prognostic value of ERCC1, thymidylate synthase, and glutathione S-transferase pi for 5-FU/oxaliplatin chemotherapy in advanced colorectal cancer. *Am J Clin Oncol* 32: 38-43.
- Kim JS, Kim MA, Kim TM, Lee SH, Kim DW, et al. (2009) Biomarker analysis in stage III-IV (M0) gastric cancer patients who received curative surgery followed by adjuvant 5-fluorouracil and cisplatin chemotherapy: epidermal growth factor receptor (EGFR) associated with favourable survival. *Br J Cancer* 100: 732-738.
- Ren DN, Kim IY, Koh SB, Chang SJ, Eom M, et al. (2009) Comparative analysis of thymidylate synthase at the protein, mRNA, and DNA levels as prognostic markers in colorectal adenocarcinoma. *J Surg Oncol* 100: 546-552.
- Kostopoulos I, Karavasili V, Karina M, Bobos M, Xiros N, et al. (2009) Topoisomerase I but not thymidylate synthase is associated with improved outcome in patients with resected colorectal cancer treated with irinotecan containing adjuvant chemotherapy. *BMC Cancer* 9: 339.
- Cullen KJ, Schumaker L, Nikitakis N, Goloubeva O, Tan M, et al. (2009) beta-Tubulin-II expression strongly predicts outcome in patients receiving induction chemotherapy for locally advanced squamous carcinoma of the head and neck: a companion analysis of the TAX 324 trial. *J Clin Oncol* 27: 6222-6228.
- Lee HW, Hwang YH, Han JH, Choi JH, Kang SY, et al. (2010) High expression of excision repair cross-complementation group 1 protein predicts poor outcome in patients with nasopharyngeal cancer. *Oral Oncol* 46: 209-213.
- Karlberg M, Ohrling K, Elder D, Hallstrom M, Ullen H, et al. (2010) Prognostic and predictive value of thymidylate synthase expression in primary colorectal cancer. *Anticancer Res* 30: 645-651.
- Lee S, Park YH, Kim KH, Cho EY, Ahn YC, et al. (2010) Thymidine synthase, thymidine phosphorylase, and excision repair cross-complementation group 1 expression as predictive markers of capecitabine plus cisplatin chemotherapy as first-line treatment for patients with advanced esophageal squamous cell carcinoma. *Br J Cancer* 103: 845-851.
- O'Toole D, Couvelard A, Rebours V, Zappa M, Hentic O, et al. (2010) Molecular markers associated with response to chemotherapy in gastro-entero-pancreatic neuroendocrine tumors. *Endocr Relat Cancer* 17: 847-856.
- Choi IS, Lee HS, Lee KW, Kim H, Kim KH, et al. (2011) Biomarker analysis in patients with advanced gastric cancer treated with S-1 plus cisplatin chemotherapy: orotate phosphoribosyltransferase expression is associated with treatment outcomes. *Med Oncol* 28: 991-998.
- Jeong SH, Han JH, Kim JH, Ahn MS, Hwang YH, et al. (2011) Bax predicts outcome in gastric cancer patients treated with 5-fluorouracil, leucovorin, and oxaliplatin palliative chemotherapy. *Dig Dis Sci* 56: 131-138.
- Nishi M, Shimada M, Utsunomiya T, Morine Y, Imura S, et al. (2011) Role of dihydropyrimidine dehydrogenase and thymidylate synthase expression in immunohistochemistry of intrahepatic cholangiocarcinoma. *Hepatol Res* 41: 64-70.
- Yasumatsu R, Nakashima T, Yamauchi M, Wakasaki T, Masuda M, et al. (2011) Thymidylate synthase expression as a predictor of clinical response to 5-fluorouracil-based chemoradiotherapy in patients with maxillary sinus squamous cell carcinoma. *Auris Nasus Larynx* 38: 387-391.
- Lee SJ, Choi YL, Park YH, Kim ST, Cho EY, et al. (2011) Thymidylate synthase and thymidine phosphorylase as predictive markers of capecitabine monotherapy in patients with anthracycline- and taxane-pre-treated metastatic breast cancer. *Cancer Chemother Pharmacol* 68: 743-751.
- Conradi LC, Bleckmann A, Schirmer M, Sprenger T, Jo P, et al. (2011) Thymidylate synthase as a prognostic biomarker for locally advanced rectal cancer after multimodal treatment. *Ann Surg Oncol* 18: 2442-2452.
- Kondo N, Murakami Y, Uemura K, Sudo T, Hashimoto Y, et al. (2011) Prognostic impact of dihydropyrimidine dehydrogenase expression on pancreatic adenocarcinoma patients treated with S-1-based adjuvant chemotherapy after surgical resection. *J Surg Oncol* 104: 146-154.
- Kaira K, Okumura T, Ohde Y, Takahashi T, Murakami H, et al. (2011) Prognostic significance of thymidylate synthase expression in the adjuvant chemotherapy after resection for pulmonary metastases from colorectal cancer. *Anticancer Res* 31: 2763-2771.
- Noda E, Maeda K, Inoue T, Fukunaga S, Nagahara H, et al. (2012) Predictive value of expression of ERCC 1 and GST-p for 5-fluorouracil/oxaliplatin chemotherapy in advanced colorectal cancer. *Hepatogastroenterology* 59: 130-133.
- van der Zee JA, van Eijk CH, Hop WC, van Dekken H, Dicheva BM, et al. (2012) Expression and prognostic significance of thymidylate synthase (TS) in pancreatic head and periampullary cancer. *Eur J Surg Oncol* 38: 1058-1064.
- Formentini A, Henne-Bruns D, Kormann M (2012) Thymidylate synthase and cyclin D1 protein expression in lymph node negative colorectal cancer: role as prognostic factors? *Hepatogastroenterology* 59: 1859-1864.
- Yokota K, Sasaki H, Okuda K, Shiara M, Hikosaka Y, et al. (2012) Expression of thymidylate synthase and orotate phosphoribosyltransferase in thymic carcinoma. *Exp Ther Med* 4: 589-593.
- Arsenijevic T, Micev M, Nikolic V, Gavrilovic D, Radulovic S, et al. (2012) Is there a correlation between molecular markers and response to neoadjuvant chemoradiotherapy in locally advanced squamous cell esophageal cancer? *J BUON* 17: 706-711.
- Sulzyc-Bielicka V, Domagala P, Bielicki D, Safranow K, Domagala W (2013) Thymidylate synthase expression and p21(WAF1)/p53 phenotype of colon cancers identify patients who may benefit from 5-fluorouracil based therapy. *Cell Oncol (Dordr)* 37: 17-28.