

Plasma or Serum TIMP-1 is a Predictor of Survival Outcomes in Colorectal Cancer: a Meta-analysis

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Abstract

Background & Aims. Tissue inhibitor of metalloproteinase-1 (TIMP-1) is a small secretory glycoprotein with anti-apoptosis and anti-matrix metalloproteinase activity. There have been some discordant data regarding the value of TIMP-1 as a prognostic factor in colorectal cancer (CRC) patients. To address this controversy, we conducted a meta-analysis for the relationship between TIMP-1 levels and overall survival in CRC. **Methods.** We selected the relevant published studies using citation databases including PubMed, Science Citation Index, and Conference Papers Index. The effect sizes of TIMP-1 on the patient's overall survival and TNM stages were calculated by hazard ratio (HR) or odds ratio (OR), respectively. The effect sizes were combined using a random-effects model. **Results.** Survival outcomes between high and low plasma or serum TIMP-1 levels were compared by uni- and multivariate analyses involving 1,477 and 1,359 CRC patients, respectively. CRC patients with high plasma or serum TIMP-1 levels showed poor survival rates compared to patients with low plasma or serum TIMP-1 in the uni- and multivariate analyses (HR, 2.2 and 2.1; $P < 0.001$). In addition, high TIMP-1 expression in colon cancer tissues was significantly associated with worse survival outcomes in 438 CRC patients (HR = 1.4; $P = 0.017$). **Conclusion.** Plasma or serum TIMP-1 levels predict survival outcomes of CRC patients

Key words

TIMP-1 – colorectal cancer – survival – meta-analysis.

Introduction

Colorectal cancer (CRC) is the third most common cancer worldwide [1]. In 2009, an estimated 146,970 new CRC patients were diagnosed and 49,920 died from the disease in the United States [2]. Currently, the prognosis of CRC is determined primarily by pathological parameters and TNM cancer staging. In addition, therapeutic strategies for adjuvant or palliative chemotherapy are determined on the basis of traditional prognostic systems. However, for risk stratification, new and more efficient biomarkers are needed to precisely evaluate the prognosis of an individual CRC patient and to prevent unnecessary adjuvant chemotherapy or unexpected cancer recurrence.

Among the more promising biomarkers of CRC, tissue inhibitor of metalloproteinase-1 (TIMP-1) is a cellular survival factor with multiple actions including anti-apoptosis, cell growth and differentiation, and matrix metalloproteinase (MMP) inhibition [3-9]. In recent years, many studies have described the prognostic value of TIMP-1 in CRC patients. However, the prognostic importance of TIMP-1 in CRC has been challenged by conflicting results which appear to depend on the analytic method or sample type used [10-19]. In an attempt to address these controversies, we conducted a meta-analysis to clarify the relationship between TIMP-1 and the prognosis of CRC.

Methods

Data collection and selection criteria for meta-analysis

An extensive literature search for articles was carried out using the following online databases: 1) Medline using PubMed (<http://www.ncbi.nlm.nih.gov/pubmed>), 2) ISI Science Citation Index using the ISI Web of Science® search interface (<http://apps.isiknowledge.com>), and 3) Conference Papers Index using the CSA Illumina search interface (<http://www.csa.com/csailumina>). The following search terms were used in all possible combinations, “TIMP”, “colorectal cancer”, “colon cancer”, and “rectal cancer”. In addition, the reference lists of the found articles were manually searched.

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Overlapping articles or duplicate data were excluded by examining the authors' names and affiliations for each publication. The selection process of the articles was shown in Fig 1. The following types of articles were included: 1) original articles demonstrating that TIMP-1 was assessed only in primary CRC tissue or patients; articles that dealt with cell lines or animals were excluded; 2) articles published before January 2011 in English; 3) the most informative article when multiple articles were published by the same authors or groups. The following articles were excluded: 1) articles lacking data or containing data inappropriate for meta-analysis; 2) review articles without original data; and 3) case reports.

Data pooling and statistics

Hazard ratios (HRs) or odds ratios (ORs) with 95% confidence intervals (CIs) were calculated using meta-analysis [20-23]. For studies without HRs for survival, we assessed HRs and CIs using a published approximation method [21]. The HRs or ORs were combined using a random-effects model (DerSimonian-Laird method). For identifying and quantifying interstudy heterogeneity, Q statistics were calculated, which is an adaptation of the chi-square goodness-of-fit test. $P < 0.10$ was considered statistically significant. Sensitivity analyses were performed to examine the influence of each study on the pooled HR or OR by serially omitting an individual study and pooling the remaining studies. Publication bias was examined by funnel plots and Egger's tests for the degree of asymmetry. $P < 0.05$ was considered statistically significant. The pooled analysis was performed using Comprehensive Meta-analysis Software version 2.0 (Biostat, Englewood, NJ, USA).

Results

Plasma or serum TIMP-1 levels detected by enzyme-linked immunosorbent assay (ELISA)

Five studies reported plasma or serum TIMP-1 levels on the univariate unadjusted survival outcomes of CRC patients (Table I) [10-14]. The number of patients in each study ranged from 87 to 588, for a total of 1,447 patients. The estimated unadjusted HRs ranged from 0.95-3.30. High plasma or serum TIMP-1 levels were significantly associated with a poor overall survival rate, compared with low TIMP-1 levels. The pooled HR was 2.249 (95% CI: 1.555-3.253; $P < 0.001$) (Fig 2.). There was statistical heterogeneity among the studies ($Q = 12.961$, $df = 4$, $P = 0.011$).

Four studies addressed plasma or serum TIMP-1 levels on the multivariate adjusted survival outcomes of 1,359 CRC patients (Table I) [10-12,15]. The prognostic variables used in the multivariate survival model were patient's age, gender,

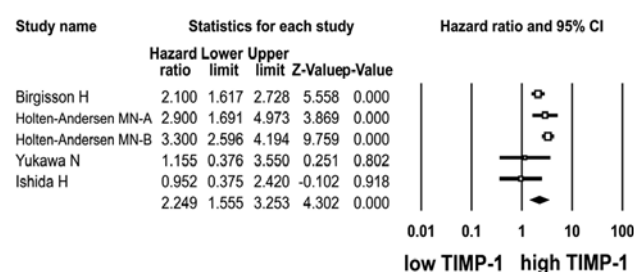


Fig 2. Hazard ratios with corresponding 95% CIs of individual studies and pooled data for overall survival between high plasma or serum TIMP-1 levels and low plasma or serum TIMP-1 in univariate analysis.

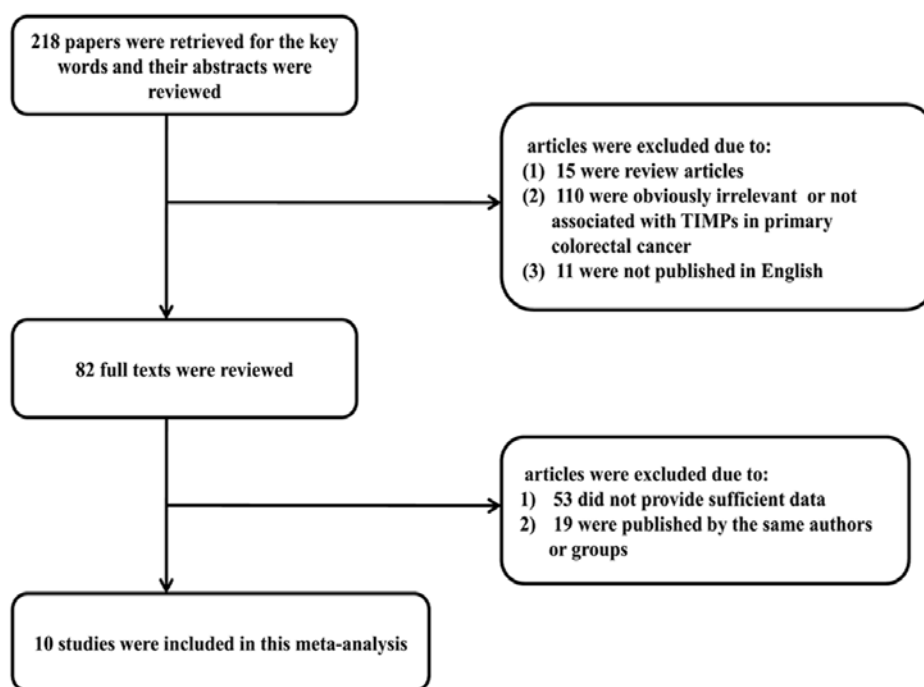


Fig 1. Article selection flowchart for meta-analysis

Table I. Characteristics of individual studies included in this meta-analysis

Study	Country of patients	Sample type	Method	Number of cases	TIMP-1	
					high	low
Birgisson et al [10]	Sweden	plasma	ELISA	322	n.c.	n.c.
Holten-Andersen et al [11]	Sweden	plasma	ELISA	352	n.c.	n.c.
Holten-Andersen et al [12]	Denmark	plasma	ELISA	588	n.c.	n.c.
Yukawa et al [13,16]	Japan	plasma	ELISA	87	45	42
Ishida et al [14].	Japan	serum	ELISA	98	32	66
Giaginis et al [15]	Greece	serum	ELISA	97	n.c.	n.c.
Jensen et al [17].	Denmark	tissue	Immunohistochemistry	318	203	115
Unsal et al [18]	Turkey	tissue	Immunohistochemistry	60	5	55
Roca et al [19]	Argentina	tissue	Immunohistochemistry	60	21	39

TIMP-1, tissue inhibitor of matrix metalloproteinase-1; ELISA, enzyme-linked immunosorbent assay; n.c., not commented.

histological grade, and clinical stage (Table II). There was a significant relationship between high plasma or serum TIMP-1 levels and unfavorable adjusted HRs. In this meta-analysis, the estimated adjusted HRs ranged from 1.79 to 2.50. The pooled HR was 2.088 (95% CI: 1.686-2.585; $P < 0.001$) (Fig. 3). No statistical heterogeneity was found among the studies ($Q = 2.003$, $df = 3$, $P = 0.572$).

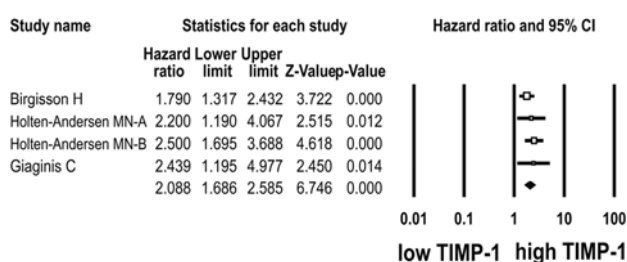


Fig 3. Hazard ratios and pooled data for overall survival between high plasma or serum TIMP-1 levels and low plasma or serum TIMP-1 in multivariate analysis.

Two studies presented data on plasma or serum TIMP-1 levels according to the Duke stages (A, B versus C, D) (Table I) [15,16]. High plasma or serum TIMP-1 was closely related to the advanced Duke stages. The pooled OR was 2.482 (95% CI: 1.405-4.383; $P = 0.002$). There was no significant heterogeneity between the studies ($Q = 0.754$, $df = 1$, $P = 0.385$).

Expression of tissue TIMP-1 protein in tumor cells

Articles only investigating TIMP-1 expression in cancer cells were included because TIMP-1 can be expressed in stromal cells as well as cancer cells. Three immunohistochemical studies presented univariate overall survival according to TIMP-1 expression (Table I) [17-19]. These studies included 229 CRC patients with high TIMP-1 protein expression and 209 with low TIMP-1 protein expression. The estimated unadjusted HRs ranged from 0.64 to 1.55. High TIMP-1 expression was related to unfavorable survival rates (HR = 1.448, 95% CI: 1.069-1.962; $P = 0.017$) (Fig. 4). Significant heterogeneity was not found among the studies ($Q = 1.282$, $df = 2$, $P = 0.527$).

Table II. Covariates used in the multivariate survival models

Study	Variables used in Cox Proportional Hazard analysis
Birgisson et al [10].	TIMP-1,* age,* stage,* postoperative chemotherapy
Holten-Andersen et al [11].	TIMP-1,* age,* gender, grade,* stage*
Holten-Andersen et al [12].	TIMP-1,* age,* gender, stage,* localization,* suPAR,* PAI-1*
Giaginis et al [15].	TIMP-1,* age, gender, grade,* stage,* TIMP-2

*Independent prognostic variables, $P < 0.05$. TIMP, tissue inhibitor of matrix metalloproteinase; PAI-1, plasminogen activator inhibitor-1; suPAR, soluble urokinase plasminogen activator receptor.

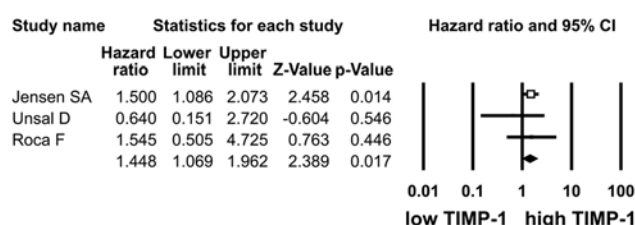


Fig 4. Hazard ratios and pooled data for overall survival between high and low expression of TIMP-1 immunohistochemistry in univariate analysis.

Sensitivity analysis and publication bias

Sensitivity analyses revealed that none of the studies affected the unadjusted or adjusted HRs with CIs (Fig. 5). However, Yukawa's study [16] influenced the result of Duke stages. In the funnel plots and the Egger's regression tests, there was no evidence of publication bias for unadjusted or adjusted survival analysis according to TIMP-1 levels.

Discussion

This meta-analysis using both univariate and multivariate survival data from 1,447 and 1,359 CRC patients,

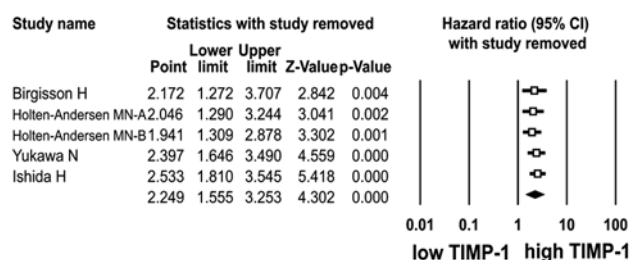


Fig 5. Sensitivity analysis of meta-analysis for unadjusted HR of overall survival according to plasma or serum TIMP-1 levels.

respectively, revealed that high levels of plasma or serum TIMP-1 are significantly associated with an unfavorable survival outcome for CRC patients.

TIMP-1 is a small secretory glycoprotein with a molecular size of 28.5 kDa. Since TIMP-1 inhibits the action of MMPs [24, 25], which play a major role in cancer cell invasion and dissemination, it was expected that TIMP-1 overexpression would be a marker of a good outcome for CRC patients. However, high levels of TIMP-1 are suggested to be a prognostic marker of worse outcomes in various cancers including CRC [10-18, 26-29]. This paradox can be partly explained by the fact that TIMP-1 is a cancer cell survival protein by promoting anti-apoptosis [6-9].

This pooled study confirmed that plasma or serum TIMP-1 levels detected by ELISA have clinical implications; they are significantly related to poor survival outcomes and advanced Duke stages in CRC patients. The HRs of high plasma or serum TIMP-1 by ELISA was greater than two-fold. There has been considerable controversy with respect to the significance of TIMP-1 levels for CRC patient survival. Some studies showed that high TIMP-1 levels were highly related to worse survival rates [10-12,15]. However, other studies failed to show a significant relationship between high TIMP-1 levels and overall survival of CRC patients [13, 14].

Our meta-analysis indicated that high TIMP-1 immunorexpression is associated with worse survival rates in CRC patients. Jensen et al [17] reported that overexpression of TIMP-1 was significantly associated with an unfavorable overall survival. In contrast, Unsal et al [18] and Roca et al [19] reported that survival outcomes of CRC were independent of TIMP-1 expression.

There are a few limitations in our study. First, TIMP-1 levels were measured by different methods and for different clinical sample types. Plasma or serum TIMP-1 levels could be compared with overall survival and Duke stages of CRCs. However, data for other parameters such as lymph node metastasis, tumor depth, and tumor size were insufficient for meta-analysis. Second, we calculated the log hazard ratio and its variance from Kaplan-Meier survival curves using the indirect method [21]. However, the indirect approach for extracting summary statistics is known to make a useful graphical summary of the two survival curves [21].

In summary, our study indicates that plasma or serum measurement of TIMP-1 levels by ELISA in patients with

CRC may be a biologic marker for predicting survival outcomes, although further investigations are required. In addition, this pooled analysis suggests that TIMP-1 plays a role in the progression of colon cancer cells.

Acknowledgements

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Conflicts of interest

None to declare.

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