Perioperative Tumour Marker Levels as Prognostic Factors for Surgical Treatment of Breast Cancer Liver Metastases

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Abstract. *Background/Aim:* The results of surgical treatment of breast cancer liver metastases (BCLM) remain suboptimal. The search for perioperative prognostic factors could help determine high risk groups of patients. The aim of study was to evaluate the significance of tumour markers (TM) on long-term treatment results. *Patients and Methods:* Liver surgery was carried out in 32 women for BCLM. The perioperative serum levels of CEA, CA19-9, TPA, TPS, CYFRA 21-1 and TK were determined. *Results:* Preoperative levels of CA19-9 were significant for overall survival (OS) ($p<0.05$) and recurrence-free survival (RFS) ($p<0.01$). TPA, TPS and CYFRA 21-1 levels were significant for OS ($p<0.05$; $p<0.04$; $p<0.05$). Postoperative levels of CEA, TPS and CYFRA 21-1 were significant for RFS ($p<0.04$; $p<0.01$; $p<0.02$), while CA19-9 postoperative levels were significant for OS and RFS ($p<0.03$; $p<0.01$). *Conclusion:* Perioperative TM serum levels may represent prognostic indicators reflecting the results of surgical treatment of BCLM.

Breast cancer (BC) represents the most frequent cancer worldwide among women and is one of the most frequent causes of death in this population. BC metastases either develop concomitantly with the primary BC or at different time intervals in more than 50% of women with BC. Breast cancer liver metastases (BCLM) in most women are associated with concurrent BC dissemination to the lungs and bones and these represent the only site of metastatic BC in 12-15% women. Although great progress has been made in the diagnosis and effective treatment of BC, metastases develop in 30% of women despite timely diagnosis and radical treatment (1, 2).

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Surgical treatment of BCLM is an important component of multimodal therapeutic approaches. However, its results remain suboptimal compared, for example, to patients with colorectal cancer liver metastases. In our view, one strategy that might improve the results of surgical management of BCLM is the search for early indicators of tumour dissemination or for prognostically high-risk patients.

Tumour markers are general indicators used in monitoring and follow-up of patients. The most frequently used markers to monitor treatment efficacy and diagnose BCLM recurrence include mucins, such as carbohydrate antigen CA 19-9, cytokeratins such as tissue polypeptide antigen (TPA) and tissue-specific polypeptide antigen (TPS), proliferation markers such as thymidine kinase (TK) and carcinoembryonic antigen (CEA). However, no study regarding the prognostic significance of preoperative serum tumour marker levels in patients undergoing surgery for BCLM has been published to date, as far as we are aware.

The aim of our retrospective study was to determine whether perioperative levels of clinically routinely used tumour markers influence overall survival (OS) and recurrence-free survival (RFS) of patients undergoing resection or thermoablative therapy for BCLM as part of a multimodal approach.

Patients and Methods

From January 1, 2000 until January 1, 2018, a total of 32 women underwent surgery for BCLM at the Department of Surgery, University Hospital in Pilsen, Czech Republic. Surgery was indicated in women in a general good state of health from the aspect of surgery and those who had consented to undergo the proposed surgical procedure. Surgery was indicated only if the metastases were located solely within the liver parenchyma. Every patient underwent hybrid imaging using positron-emission tomography (PET) and computed tomography (CT) or PET and magnetic resonance imaging (MRI) before surgery. Another requirement was that the future liver remnant volume exceed 30% of the total liver volume. The average age of these women was 49.5 ± 10.7 years. The primary BC types were: ductal in 20 cases, lobular in seven, erysipeloid in four, and sarcomatoid (spindle cell) carcinoma in one. The average interval between initial breast surgery and BCLM surgery was 4.7 ± 3.7 years.

Peripheral blood sampling to determine tumour markers was performed on the morning of the liver surgery and on the patient's discharge from hospital (the average length of hospital stay was 7.3 ± 2.1 days). Serum levels of CEA, CA 19-9, TPA, TPS, cytokeratin 19 fragment (CYFRA 21-1) and TK were determined in each sample. Serum tumour markers were assayed using the following immunoanalytical kits: chemiluminescent assays ACCESS CEA and ACCESS CA 19-9 (Beckman Coulter, Brea, CA, USA) with LoD=0.1 ng/ml for CEA and LoD=0.08 ng/ml for CA 19-9; LIAISON TPA (DiaSorin, Saluggia, Italy) with LoD=3 U/l and LoQ=9 U/l and ARCHITECT CYFRA 21-1 (Abbott Laboratories, Libertyville, IL, USA) with LoD=0.09 ng/ml and LoQ=0.17 ng/ml; immunoradiometric assay IRMA TPS (IDL Biotech AB, Bromma, Sweden) with LoD=5.3 IU/l and LoQ=11.2 IU/l and enzymoradiometric assay TK REA (Immunotech, Prague, Czech Republic) with LoD=0.22 U/l and LoQ=1.34 U/l.

A total of 16 liver resections and 12 radio-frequency thermoablations (RFA) were performed and in four cases these procedures were combined to remove the BCLM.

Statistical analysis was performed using SAS software (SAS Institute Inc., Cary, NC, USA). The differences between the studied parameters were tested using the paired sign test. The relationships between the variables were studied using the Spearman correlation coefficient. The analysis of OS and RFS from liver surgery as based on Kaplan–Meier survival curves. The impact of individual factors was tested using the log-rank test, the Gehan Wilcoxon test and the Cox regression model. Statistical significance was set at an alpha level of 5%.

Results

The 30-day postoperative mortality was zero. Two patients (6.3%) had grade IIIa postoperative complications according to the Clavien-Dindo classification (haematoma within the resected area resolved by CT-guided drainage and pneumothorax resolved by chest drainage). Six patients underwent secondary and tertiary procedures for BCLM recurrence at an interval of 6-39 months consisting of RFA in five patients and RFA with resection in one. One-, 3- and 5-year OS rates were 77.9%, 62.3% and 30.6%, respectively, with corresponding RFS of 63.7%, 22.3% and 11.9% (Figures 1 and 2). The average preoperative and postoperative serum tumour marker levels are listed in Table I. All tumour marker levels decreased after liver surgery (Table I). The cut-off values for OS and RFS relating to preoperative and postoperative levels of individual markers determined using the Cox regression hazard model are listed in Tables II and III. Cut-off values of some perioperative marker levels were significant for OS and RFS (bold type in Tables II and III).

Discussion

In the female population, BC is the second most frequent type of cancer after non-melanoma skin cancer. The Czech Republic (CR) ranks 118th in the world and 37th in Europe in the incidence of BC. In 2015, there were 132 new cases of BC per 100,000 women in the CR and the incidence

continues to rise (3, 4). Although treatment is highly successful, especially in early-stage BC (100% 5-year survival for stage I and 60% for stage II), treatment of more advanced forms of the disease that also include BCLM in the long-term remains associated with relatively early disease recurrence and high mortality, in contrast to the treatment *e.g.* of colorectal cancer liver metastases (5, 6). In 2015, 1,609 women died of BC in the CR, which represents ~31 deaths/100,000 women. One positive fact in the CR is that while the incidence of BC between 2004 and 2014 increased by 19.5%, mortality over the same period decreased by 17.5%. This reflects the early detection and timely multimodal treatment of BC, which includes surgical, biological, hormone therapy and chemotherapy. Nonetheless, the results of treatment in the case of generalised BC, including patients with BCLM, remain unsatisfactory, with an OS of only 6-14 months on average (7-9).

Today, surgical treatment (resection, thermoablation or a combination of both) of BCLM as part of multimodal therapeutic strategies achieves significantly better long-term results (22-27 months) than conservative therapy (4-8 months). Nonetheless, it does not achieve the same superior results as those seen in the surgical treatment of colorectal cancer liver metastases. There may be several reasons for this. The problem of BC is its heterogeneity, involving not only the primary tumour, but also the BCLM. Another problem is the relatively frequent histopathological non-conformity between the primary tumour and its metastases. Thus, in the same patient, treatment of the primary tumour may not be as effective in the treatment of BCLM. Therefore, from the aspect of treatment strategy, it is important, especially in the case of operable BCLM, to determine the preoperative risk factors for further development of metastases and thus to optimise not only treatment itself but also postoperative follow-up (10, 11). In our previous study, we demonstrated that the long-term results of surgical treatment of BCLM are affected by a number of factors, including patient age, interval between breast cancer and BCLM surgery, the type of surgical procedure used, histopathology of the primary tumour, the number and overall diameter of BCLMs, the presence of resectable extra-hepatic metastases and local tumour recurrence after breast cancer surgery (12).

Currently, the principal role of tumour markers in patients radically treated for BC and its metastases involves the early and timely diagnosis of systemic disease recurrence (13, 14). Several studies have shown that elevation of serum tumour markers occurs significantly in advance of diagnosis of BC recurrence using radiodiagnostic methods (15, 16). This is why some authors recommend the use of a combination of several tumour markers, especially CEA, CA 15-3 and cytokeratins, to increase the sensitivity of relapse detection, namely every 3 months after surgery during the first 5 years and every 6

Figure 1. Overall survival for the whole group of patients ($N=32$).

Figure 2. Recurrence-free survival for the whole group of patients ($N=32$).

Table I. Preoperative and postoperative serum levels of the studied tumour markers (N=32). Data are the mean±SD [median (25-75% quartiles)].

Tumour marker	Preoperative level	Postoperative level
CEA (µg/l)	14.8±18.9 [8.1 (6.8-22.8)]	7.0±16.3 [2.0 (0.02-14.1)]
CA 15-3 (kIU/ml)	39.8±22.3 [41.0 (29.6-49.9)]	18.1±9.8 [17.0 (13.7-22.6)]
CA 19-9 (kIU/ml)	21.6±16.8 [16.0 (14.3-28.8)]	11.3±8.6 [8.5 (7.5-15.2)]
TPA (IU/l)	83.2±93.5 [47 (40.6-125.8)]	33.3±30.0 [27.0 (19.7-46.9)]
TPS (IU/l)	73.6±74.1 [38.5 (40.8-106.5)]	40.0±26.4 [33.0 (28.4-51.7)]
TK (IU/l)	15.3±12.4 [8.1 (9.5-21.1)]	8.4±6.5 [7.3 (5.3-11.5)]
CYFRA 21-1 (µg/l)	2.3±1.3 [2.1 (1.7-2.9)]	1.6±1.3 [1.4 (0.9-2.2)]

CEA: Carcinoembryonic antigen, CA 15-3: carbohydrate antigen 15-3, CA 19-9: carbohydrate antigen 15-9, TPA: tissue polypeptide antigen, TPS: tissue polypeptide-specific antigen, TK: thymidine kinase, CYFRA 21-1: cytokeratin 19 fragment.

Table II. Cut-offs for preoperative serum levels of individual tumour markers for overall (OS) and recurrence-free (RFS) survival.

Tumour marker	OS				RFS			
	Cut-off	HR	95% CI	p-Value	Cut-off	HR	95% CI	p-Value
CEA (µg/ml)	1.2	1.36	0.29-6.42	0.69	1.2	1.55	0.34-7.10	0.57
CA 15-3 (kIU/ml)	15	0.17	0.03-0.96	0.04	65	0.16	0.02-1.27	0.08
CA 19-9 (kIU/ml)	50	5.61	1.0-31.35	0.05	50	8.68	1.69-44.65	0.01
TPA (IU/l)	90	3.09	0.92-10.32	0.05	90	2.79	0.92-8.44	0.07
TPS (IU/l)	75	7.96	0.72-87.95	0.04	20	3.15	0.97-10.23	0.06
TK (IU/l)	22.4	2.67	0.74-9.58	0.13	4.5	5.80	0.75-44.95	0.09
CYFRA 21-1 (µg/l)	0.6	0.06	0.74-9.58	0.05	0.6	0.18	0.02-1.73	0.14

CEA: Carcinoembryonic antigen, CA 15-3: carbohydrate antigen 15-3, CA 19-9: carbohydrate antigen 15-9, TPA: tissue polypeptide antigen, TPS: tissue polypeptide-specific antigen, TK: thymidine kinase, CYFRA 21-1: cytokeratin 19 fragment.

Table III. Cut-offs for postoperative serum levels of individual tumour markers for overall (OS) and recurrence-free (RFS) survival.

Tumour marker	OS				RFS			
	Cut-off	HR	95% CI	p-Value	Cut-off	HR	95% CI	p-Value
CEA (µg/ml)	16	6.26	0.56-70.27	0.13	16	6.83	1.13-41.29	0.04
CA 15-3 (kIU/ml)	10	0.20	0.04-0.93	0.04	30	8.15	1.97-33.79	0.004
CA 19-9 (kIU/ml)	20	4.73	1.15-19.43	0.03	9	7.23	1.93-27.45	0.01
TPA (IU/l)	10	0.44	0.12-1.72	0.24	10	0.46	0.14-1.45	0.18
TPS (IU/l)	30	1.61	0.53-4.85	0.39	20	4.49	1.39-14.45	0.01
TK (IU/l)	2.1	2.61	0.32-21.53	0.37	7.3	3.17	0.93-10.79	0.07
CYFRA 21-1 (µg/l)	0.9	2.44	6.22-9.54	0.20	0.4	8.17	1.04-64.06	0.02

CEA: Carcinoembryonic antigen, CA 15-3: carbohydrate antigen 15-3, CA 19-9: carbohydrate antigen 15-9, TPA: tissue polypeptide antigen, TPS: tissue polypeptide-specific antigen, TK: thymidine kinase, CYFRA 21-1: cytokeratin 19 fragment.

months thereafter (17, 18). Guidelines of the European Group on Tumour Markers and the National Academy of Clinical Biochemistry recommend the use of tumour markers for the diagnosis of early disease recurrences (19-22). Apart from the detection of recurrence, other

advantages of this approach include the low cost of tumour marker testing and the decrease in radiation burden incurred by patients when radiodiagnostic methods such as CT or PET CT alone are used to diagnose recurrence. Tumour markers are also important indicators of the

efficacy of oncological treatment of this disease (23-25). Together with the Response Evaluation Criteria in Solid Tumours criteria (26), tumour markers may then more accurately reflect the prognosis of patients after oncological treatment (27, 28).

Studies dealing with the correlation between tumour markers and growth factors from the aspect of the outcome of patients treated surgically for BC have been published (29). The current work follows on from our previous study that evaluated the importance of preoperative serum tumour markers levels in the differential diagnosis of BCLM (30). In that study, we demonstrated that the preoperative serum levels of CYFRA 21-1, TPA, TPS and CEA were significantly higher in patients with BCLM compared to healthy controls and patients with benign liver tumours. To the best of our knowledge, no study dealing with the significance of pre- and postoperative serum tumour marker levels for the prognosis of patients following surgery for BCLM has been published to date. Following on from our original study, we focused on tumour markers whose perioperative levels in serum were demonstrably elevated in patients with BCLM. We found that the preoperative level of CA19-9 was a prognostic factor for both OS and RFS. Cytokeratins and CYFRA 21-1 were prognostic markers for OS. Postoperative values of CEA, TPS and CYFRA 21-1 were significant for RFS, while the levels of CA19-9 were prognostic markers for both OS and RFS. The fact that there was unexpectedly no decrease in the level of some tumour markers after surgery could be related to their biological half-life (CEA) and undoubtedly also to the extent and type of surgical procedure (resection versus RFA). The size of our sample did not allow for a more detailed analysis of these issues.

From the aspect of the multidisciplinary team that plans the treatment and follow-up of patients with BCLM, we believe that the evaluation of preoperative and early (during patient hospital stay) postoperative serum tumour marker levels might have a predictive value relating to the long-term results of surgical treatment of BCLM. CA 19-9 appears to be such a tumour marker, whereby its pre- and postoperative levels were significantly altered for both OS and RFS.

Several unanswered questions remain. The preoperative level of serum tumour markers is generally related to the extent of the tumour. However, further investigations should also focus on elucidating the relationship between these levels and tumour aggressiveness, given that this fundamentally affects the fate of patients with BCLM. The role of the early postoperative level of tumour markers in the outcome of patients also remains to be defined, especially given the potential impact on serum levels by the surgical procedure itself, whereby liver resection involves manipulation of the tumour and RFA leads to thermoablation of the tumour and direct release of a number of mediators

and probably also of tumour markers into the circulation. This may lead to the false conclusion that these levels correlate with patient prognosis.

Evaluation of the role of perioperative levels of serum tumour markers in the prognosis of patients following surgery for BCLM represents a new view of the role of tumour markers in the treatment of patients with BCLM. Together with other clinical indicators, tumour markers may play an important role in optimising therapeutic approaches and follow-up of patients following surgery for BCLM.

We are aware of the drawbacks and deficiencies of this study, which include its retrospective character and the relatively small number of patients enrolled. The latter restricted us to the statistical evaluation of individual tumour markers and it was not possible to perform a multifactorial analysis of the significance of tumour markers for patient prognosis following surgery for BCLM. Nonetheless, one important fact that emerges from our study is evidence that preoperative and early postoperative serum levels of routinely used tumour markers may indicate the potential biological behaviour of BCLM and may have prognostic significance for patients after surgical treatment of BCLM.

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