ORIGINAL ARTICLE

Symptomatic metastasis prediction with serial measurements of CA 15.3 in primary breast cancer patients

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Background: CA 15.3 is elevated in most patients with distant metastatic breast cancer who had prognostic information. The present study was performed to estimate predictive ability of CA 15.3 in assessment of symptomatic metastasis in patients with breast cancer. **Materials and Methods:** During five years, 159 primary breast cancer patients were evaluated. A total of 2226 determination of serum CA 15.3 (14 per patient) were performed. We performed contemporary clinical examinations with CA 15.3 measurements at the time of diagnosis, end of chemotherapy, every three months in the first two years and every six months in the second two years of follow-up period. Imaging studies were performed if clinical or laboratory examinations (abnormal serum levels of CA 15.3) suspected symptomatic metastasis. Metastasis in participants was confirmed by imaging study in symptomatic patients. **Results:** There was no significant increase in CA 15.3 tumor markers during chemotherapy ($P = 0.08$). There was a significant relationship between CA 15.3 positive results and metastasis situation ($P = 0.00$). Mean of maximum CA 15.3 level in metastatic patients (52.72±27.09) was significantly higher than non-metastatic patients (27.58±13.46; $P = 0.00$). CA 15.3 abnormality (OR, 1.07; 95% CI: 1.04-1.11; P value, 0.01) and abnormal lymph nodes remained as predictor of metastasis (OR: 1.16; 95% CI: 1.05-1.28; P value < 0.0001). **Conclusion:** CA 15.3 is one of the predicting factors for symptomatic metastasis.

Keywords: Breast cancer, CA 15.3, metastasis, prediction

INTRODUCTION

Breast cancer had been known as a common cancer among women worldwide and the most common cause of cancer-related mortality.[1-5] Against increasing incidence of breast cancer, there is a decline in cancer-related mortality among patients in developed countries.[6] It seems that, this reduction might be due to prevention programs such as mammographic screening and adjuvant systemic therapies for new cases of breast cancer.[7]

Several serum-based products and tumor markers or biomarkers are being used in the management of breast cancer patients.[8-10]

As same as other tumor markers, CA 15.3 due to lack of sensitivity and specificity for diagnosis of *in situ* or low stage invasive breast cancer, has not been used for diagnosis of primary breast cancer patients. Indeed, on the other hand, CA 15.3 measurement in primary breast cancer patients had most overlap with healthy women or who had benign breast disease.[11,12]

Although elevated levels of CA 15.3 was not sensitive and specific for the diagnosis of primary breast cancer, CA 15.3 was elevated in most of the patients with distance metastatic breast cancer and had prognostic information. [13-15] Most of the traditional prognostic factors in breast cancer required samples from tumor tissue via biopsy or surgery.[4,5] CA 15.3 assessment in prognostic study of primary breast cancer patients is necessary for their suitable management. CA 15.3 might help us avoid from under-treatment of advanced breast cancer patients and over-treatment of indolent patients. As per our search, few researchers pay attention to the noted tumor marker as predictor of symptomatic metastasis in primary breast cancer patients and if the prediction role of this marker is approved in present manuscript, oncologists will have suitable tools to focus on patients with high CA 15.3 level for early detection of metastasis.[16-18] The present study was performed to assess the predictive ability of CA 15.3 in symptomatic

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metastasis in patients with breast cancer in Yazd oncology clinics.

MATERIALS AND METHODS

Between July 2002 and August 2006, 168 patients met our inclusion criteria and were included in the present prospective study. Nine patients were excluded from the study because of migration to other regions. The present study was approved by the ethical research committee of Shahid Sadoughi University of Medical Sciences and Health Services and also all participants endorsed the informed consent. Our inclusion criteria were non-smoker female with primary breast cancer, without sign or symptoms of metastasis or recurrence or of any other malignant disorders. We excluded patients who had temporarily other oncologic (lung, colorectal, pancreas and ovarian cancer) or non-oncologic disorders (endometriosis, pelvic inflammation, hepatitis, cirrhosis, peptic ulcer, colitis and diverticulitis), which can impact serum level of CEA or CA 15.3. Finally 159 primary breast cancer patients participated in the study. Tumor size and outbreak number of lymph nodes were recorded. First, laboratory assessment of estrogen receptor (ER) and progesterone receptor (PR), CerB2 and P53 tumor markers were performed for all participants. In the present study, a total of 2226 determinations of serum CA 15.3 (14 per patient) were performed with the same laboratory and assay kits. Study follow-up time began from the time of end of chemotherapy treatment until the next 4.5 years. Chest X-ray, bone scan and liver ultrasonography were performed for all participants except for patients with benign and metastatic tumors.

We performed contemporary clinical examinations with CA 15.3 measurements at the time of diagnosis, end of chemotherapy, every three months in the first two years and every six months in the second two years of follow-up period. Imaging studies, including chest X-ray, liver ultrasonography and bone scan, were performed if clinical or laboratory examinations (abnormal serum levels of CA 15.3) suspected symptomatic metastasis. Metastasis in participants was confirmed by imaging study in symptomatic patients.

Marker analysis

In the present study, we measured CA 15.3 from biopsy in participants. Estrogen receptor (ER), progesterone receptor (PR), CerB2 and P53 tumor markers were determined in participants using a receptor assay method. ER results greater than 5 fmol/mg and PR results greater than 15 fmol/ mg of cytosolic protein were considered as positive results.[19] In the follow-up period, if an elevated serum level of CA 15.3 was observed, another sample was obtained to confirm the first CA 15.3 result. CA 15.3 level was considered as high and abnormal if >30 ng/ml was detected in two sequential determinations.[19]

Statistical analysis

We used SPSS version 16.0 for statistical analysis and all two-tailed *P* values less than 0.05 were considered as significant. For statistical calculations, the chi-square and Student *t-test* were used for qualitative and quantitative results, respectively. We considered abnormal CA 15.3 result for a patient if one of the CA 15.3 measurements was abnormal. With regard to CA 15.3 results, we grouped participants in to two groups: with normal or abnormal CA 15.3 results. Other study variables and metastasis were compared between patients with and without metastasis. We used a logistic regression model to determine the independent predictors of metastasis in primary breast cancer. In this model, one of the metastasis in primary breast cancer patients was selected as a dependent variable. Other study variables such as number of lymph node involvement, tumor size, P53, CA 15.3 level and also treatment time were entered into the model. An enter procedure was used in this analysis. Variables that remained in the model were known as independent predictor of metastasis. Calculations were done using the SPSS version 16.0 (SPSS Inc. Chicago, IL) statistical program and *P* value lower than 0.05 was considered as significant.

Sensitivity was considered as the ratio between the number of metastatic patients whose marker levels were high over the total number of metastatic patients. Specificity was calculated by dividing the number of non-metastatic patients with normal values of tumor marker by the total number of non-metastatic patients. Finally, efficacy was considered as the sum of patients with elevated tumor marker and metastasis and also patients with normal antigen concentration and non-metastasis patients divided by the total number of patients evaluated and multiplied by 100.

ROC curve calculation

We evaluated serum level of CA 15.3 14 times during the four years of follow-up for calculation of sensitivity and specificity of CA 15.3 to determine metastasis. We used the highest serum values before metastasis in metastatic patients and highest serum values during follow-up period for non-metastatic patients. The area under the receiver operating characteristic curve (ROC) was calculated as a measure of predictive discrimination of tumor response and progression by CA 15.3 variation. The cut-off points of increased and decreased marker were identified according to the corresponding plotted curves. The difference between proportions was evaluated by the chi-square test with Yates' correction, if necessary.

RESULTS

Total 159 primary breast cancer patients were included

in the present study. Mean of age of our patients was 48.84±10.69 (range: 22-78) years. The average size of tumors was 1.94±0.70 (range: 22-78) centimeters and on an average 2.86±4.30 (range: 0-8) lymph nodes were involved. Among participants, at the beginning of the study, 50 patients (31.4%) had positive CerbB2, 54 patients (34%) positive P53, 73 patients (45.9%) positive PR and 83 patients (52.2%) positive ER receptor. The mean of CA 15.3 in participants at the time of diagnosis was 18.93±7.19 μ/ml (range: 2-40). After six months of chemotherapy, mean and range of CA 15.3 in our patients was 20.08±9.95 μ/ml (range: 5-85). There was no significant increase in CA 15.3 tumor markers during chemotherapy (P = 0.08) [Table 1].

During the follow-up period, 33 patients (20.8%) presented symptomatic metastasis. In CA 15.3 assessment, 39 patients (34.5%) had abnormal results. As a result of CA 15.3 tumor marker assessment with metastatic status in participants, there was a significant relationship between CA 15.3 positive results and metastasis situation (P = 0.00). Our analysis showed that metastatic patients had significantly higher CA 15.3 abnormal results in comparison with non-metastatic patients (OR: 1.075; CI: 1.04-1.11; P = 0.00).

In our analysis, mean of CA 15.3 level in metastatic patients (52.72±27.09) was significantly higher than non-metastatic patients (27.58±13.46; P = 0.00). The median age of CA 15.3 in metastatic and non-metastatic patients was 46 and 27, respectively.

ROC curve analysis

According to the traditional cut-off point for CA 15.3, we calculated 90.91% (95% CI: 0.55-0.95) and 88.89% (95% CI: 0.71-0.85) as sensitivity and specificity for CA 15.3, respectively in assessment of metastasis in participants. According to these data, positive and negative predictive values for CA 15.3 in participants were 68.18% and 97.39%, respectively. For ROC curve analysis, we considered the highest value of tumor marker before metastasis for metastatic patients and also highest value of tumor marker for other patients during their follow-up period. According to our ROC curves, it was observed that higher sensitivity and specificity for metastatic determination could be obtained, if we consider 30.5 μ/ml for CA 15.3 as cut-off point for positive results [Figure 1].

Results of logistic regression analysis

In our logistic regression analysis, after inserting selected study variables into the model, only CA 15.3 abnormality and number of involved lymph nodes remained as independent predictor of metastasis in our model [Table 2].

DISCUSSION

Mean of CA 15.3 in our study participants at the time of diagnosis and after chemotherapy had no significant differences between metastatic and non-metastatic patients. During the four years of study, follow-up mean of CA 15.3 had significant differences between metastatic and non-metastatic patients. Calculated odds ratio confirmed that elevated CA 15.3 was observed more frequently

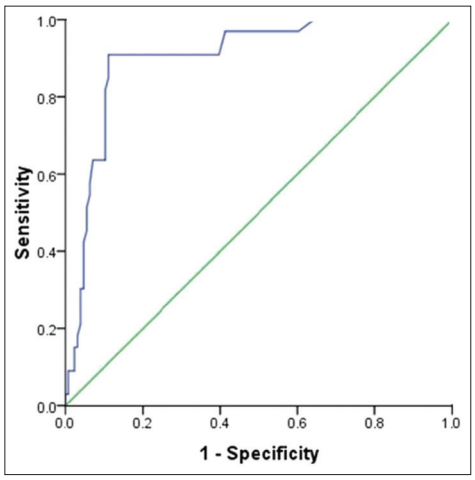

Figure 1: ROC curve analyses for using CA 15.3 findings for metastasis detection in a population of 33 metastatic versus 126 non-metastatic breast diseases

Table 1: Comparison study variables between metastatic and non-metastatic patients

		Metastatic patients (%)	Non-metastatic patients (%)	*P* value
CA 15.3 level before chemotherapy (Mean±SD)		19.97 ± 6.74	18.65 ± 7.30	0.35
CA 15.3 level after chemotherapy (Mean±SD		20.70 ± 7.01	19.93 ± 10.06	0.69
CA 15.3 within the follow-up time (N, %)	Abnormal	30 (90.9)	14 (11.1)	0.00
	Normal	9 (9.1)	112 (88.9)	
Estrogen receptor (N, %)	Positive	16 (48.5)	67 (53.2)	0.63
	Negative	17 (51.5)	59 (46.8)	
Progesterone receptor (N, %)	Positive	16 (48.5)	57 (45.2)	0.74
	Negative	17 (51.5)	68 (54.8)	
P53 (N, %)	Positive	16 (48.5)	38 (5.2)	0.048
	Negative	17 (51.5)	88 (69.8)	
CrbB2 (N, %)	Positive	10 (30.3)	40 (31.7)	0.87
	Negative	23 (69.7)	86 (68.3)	

Table 2: Results of regression analysis in patients with primary breast cancer

	Beta	Standard Error	Significances	95% CI for EXP(B)	
				Upper	Lower
Constant	1.39	1.09	0.20	-	-
Tumor size	-0.58	0.46	0.21	0.23	1.38
Involved lymph node	0.13	0.06	0.02	1.02	1.27
P53	-0.05	0.62	0.94	0.28	3.22
Abnormal CA15.3 results	-4.39	1.09	0.00	0.003	0.05

(near to 80 times) in metastatic patients in comparison with non-metastatic patients. ROC curve analysis showed that the traditional cut-off point for CA 15.3 had suitable sensitivity and specificity for metastasis determination during follow-up time in primary breast cancer patients. Logistic regression analysis confirmed that CA 15.3 higher than normal results accompanied with number of involved lymph nodes were independent predictor of metastasis.

Several studies in the literature have used CA 15.3 as one of the monitoring tools for breast cancer patients.[12,14] Some other studies reported that CA 15.3 can be used for assessment of first line chemotherapy and recurrence detection after radical treatment.[20,21] Due to the fact that CA 15.3 was not related to any specific organ, we cannot use it in the screening programs.[21-24] But CA 15.3 can be used in the early detection of recurrence in breast cancer patients and presented as a cost-reducing tool for chemotherapy monitoring.[25-28]

In the present study, we found association between elevated CA 15.3 and progression of breast cancer disease. There is a conflicting result regarding this finding: Berruti *et al*. reported that prevalence of elevated CA 15.3 was related to metastatic sites. They advocated that patients with visceral involvement had higher chance of elevated CA 15.3 than patients with bone and soft tissue involvement.[29] In contrast to the above study, Geraghty *et al*. could not find any significant difference in CA 15.3 levels between several metastatic sites.[30] Other studies reported that CA 15.3 was sensitive for bone and visceral metastasis.[31-34]

Generally elevated tumor marker was correlated with multiple metastatic diseases.[21,35] Metastasis in liver, bones and lungs and pleural effusion are especially elevated in pathologic CA 15.3 levels.[24] There were only a few studies about the use of CA 15.3 tumor marker in the follow-up of primary breast cancer patients.[25,27,28]

The present study was prospective and median follow-up time per patients was near to five years. Due to methodological differences, such as cut-off points, test assay methods, heterogeneous patient populations and different follow-up times and schedules, it is hard to compare sensitivity and specificity that were reported in the similar studies with our findings.[25,29,35] Gion *et al*. reported sensitivity of CA 15.3 ranging between 33% and 78% and specificity ranging between 60% and 93%.[23]

We found higher sensitivity and specificity than Gion *et al*. study and it might be due to some issues such as time of measurement and cut-off point that was selected.

Sensitivity and specificity of CA 15.3 test in our study was high; in the present series, the positive predictive value of CA 15.3 test was 68.8%. Soletormos *et al.* reported that in breast cancer patients with bony and visceral metastasis, the negative predictive value of CA 15.3 was 86%. On the other hand with negative results of CA 15.3 tumor marker, it was very uncommon that breast cancer patients had bony or visceral metastasis.[36]

CONCLUSION

Higher sensitivity and specificity with positive and negative predictive values for CA 15.3 showed that CA 15.3 is one of the most powerful monitoring systems for primary breast cancer patients. Further studies must be performed with higher sample size for better interpretation of tumor marker results and its relation with metastasis location.

REFERENCES

1. Benson JR, Jatoi I. The global breast cancer burden. Future Oncol 2012;8:697-702.
2. Castro-Echeverry E, Kao LS, Robinson EK, Silberfein EJ, Ko TC, Wray CJ. Relationship between documentation status and survival for medically underserved Hispanic breast cancer patients. J Surg Res 2012.
3. Clements MS, Roder DM, Yu XQ, Egger S, O'Connell DL. Estimating prevalence of distant metastatic breast cancer: A means of filling a data gap. Cancer Causes Control 2012; 23:1625-34.
4. Lee H, Jung SY, Ro JY, Kwon Y, Sohn JH, Park IH, *et al*. Metaplastic breast cancer: Clinicopathological features and its prognosis. J Clin Pathol 2012;65:441-6.
5. Rades D, Douglas S, Veninga T, Stalpers LJ, Bajrovic A, Rudat V, *et al*. Prognostic factors in a series of 504 breast cancer patients with metastatic spinal cord compression. Strahlenther Onkol 2012;188:340-5.
6. Jatoi I, Miller AB. Why is breast-cancer mortality declining? Lancet Oncol 2003;4:251-4.
7. Berry DA, Cronin KA, Plevritis SK, Fryback DG, Clarke L, Zelen M, *et al*. Effect of screening and adjuvant therapy on mortality from

breast cancer. N Engl J Med 2005;353:1784-92.
8. Dong SW, Wang L, Sui J, Deng XY, Chen XD, Zhang ZW, *et al*. Expression patterns of ER, HER2, and NM23-H1 in breast cancer patients with different menopausal status: Correlations with metastasis. Mol Diagn Ther 2011;15:211-9.
9. Goddard KA, Weinmann S, Richert-Boe K, Chen C, Bulkley J, Wax C. HER2 evaluation and its impact on breast cancer treatment decisions. Public Health Genomics 2012;15:1-10.
10. Kontos M, Allen DS, Agbaje OF, Hamed H, Fentiman IS. Factors influencing loco-regional relapse in older breast cancer patients treated with tumour resection and tamoxifen. Eur J Surg Oncol 2011;37:1051-8.
11. Hewala TI, Abd El-Monaim NA, Anwar M, Ebied SA. The Clinical Significance of Serum Soluble Fas and p53 Protein in Breast Cancer Patients: Comparison with Serum CA 15-3. Pathol Oncol Res 2012; 23:1625–1634.
12. Park S, Ahn HK, Park LC, Hwang DW, Ji JH, Maeng CH, *et al*. Implications of different CA 15-3 levels according to breast cancer subtype at initial diagnosis of recurrent or metastatic breast cancer. Oncology 2012;82:180-7.
13. Duffy MJ, Evoy D, McDermott EW. CA 15-3: Uses and limitation as a biomarker for breast cancer. Clin Chim Acta 2010;411:1869-74.
14. Sandri MT, Salvatici M, Botteri E, Passerini R, Zorzino L, Rotmensz N, *et al*. Prognostic role of CA15.3 in 7942 patients with operable breast cancer. Breast Cancer Res Treat 2012;132:317-26.
15. Yerushalmi R, Tyldesley S, Kennecke H, Speers C, Woods R, Knight B, *et al*. Tumor markers in metastatic breast cancer subtypes: Frequency of elevation and correlation with outcome. Ann Oncol 2012;23:338-45.
16. Gauchez AS, Pez E, Boutonnat J, Bourre JC, Pelletier L, Payan R, *et al*. Early detection of leptomeningeal metastasis in patients with metastatic breast carcinoma: Validation of CA 15-3 measurement in cerebrospinal fluid. Ann Biol Clin (Paris) 2007;65:653-8.
17. Niwinska A, Tacikowska M, Murawska M. The effect of early detection of occult brain metastases in HER2-positive breast cancer patients on survival and cause of death. Int J Radiat Oncol Biol Phys 2010;77:1134-9.
18. Palka I, Kelemen G, Ormandi K, Lazar G, Nyari T, Thurzo L, *et al*. Tumor characteristics in screen-detected and symptomatic breast cancers. Pathol Oncol Res 2008;14:161-7.
19. Kopczynski Z, Thielemann A. The value of tissue polypeptide specific antigen TPS determination in serum of women with breast cancer comparison to mucin-like associated antigen MCA and CA 15-3 antigen. Eur J Gynaecol Oncol 1998;19:503-7.
20. Seregni E, Coli A, Mazzucca N; Italian Group RIA-IRMA Test, Italian Association of Nuclear Medicine. Circulating tumour markers in breast cancer. Eur J Nucl Med Mol Imaging 2004;31(Suppl 1):S15-22.
21. van Dalen A. Significance of cytokeratin markers TPA, TPA (cyk), TPS and CYFRA 21.1 in metastatic disease. Anticancer Res 1996;16:2345-9.
22. Molina R, Jo J, Filella X, Zanon G, Pahisa J, Munoz M, *et al*. C-erbB-2 oncoprotein, CEA, and CA 15.3 in patients with breast cancer: Prognostic value. Breast Cancer Res Treat 1998;51:109-19.
23. Gion M, Boracchi P, Dittadi R, Biganzoli E, Peloso L, Mione R, *et al*. Prognostic role of serum CA15.3 in 362 node-negative breast cancers. An old player for a new game. Eur J Cancer 2002;38:1181-8.
24. Robertson JF, Pearson D, Price MR, Selby C, Pearson J, Blamey RW, *et al*. Prospective assessment of the role of five tumour markers in breast cancer. Cancer Immunol Immunother 1991;33:403-10.
25. Jager W, Eibner K, Loffler B, Gleixner S, Kramer S. Serial CEA and CA 15-3 measurements during follow-up of breast cancer patients. Anticancer Res 2000;20:5179-82.
26. Molina R, Zanon G, Filella X, Moreno F, Jo J, Daniels M, *et al*. Use of serial carcinoembryonic antigen and CA 15.3 assays in detecting relapses in breast cancer patients. Breast Cancer Res Treat 1995;36:41-8.
27. Robertson JF, Jaeger W, Syzmendera JJ, Selby C, Coleman R, Howell A, *et al*. The objective measurement of remission and progression in metastatic breast cancer by use of serum tumour markers. European Group for Serum Tumour Markers in Breast Cancer. Eur J Cancer 1999;35:47-53.
28. Robertson JF, Whynes DK, Dixon A, Blamey RW. Potential for cost economies in guiding therapy in patients with metastatic breast cancer. Br J Cancer 1995;72:174-7.
29. Berruti A, Tampellini M, Torta M, Buniva T, Gorzegno G, Dogliotti L. Prognostic value in predicting overall survival of two mucinous markers: CA 15-3 and CA 125 in breast cancer patients at first relapse of disease. Eur J Cancer 1994;30:2082-4.
30. Geraghty JG, Coveney EC, Sherry F, O'Higgins NJ, Duffy MJ. CA 15-3 in patients with locoregional and metastatic breast carcinoma. Cancer 1992;70:2831-4.
31. Vizcarra E, Lluch A, Cibrian R, Jarque F, Alberola V, Belloch V, *et al*. Value of CA 15.3 in breast cancer and comparison with CEA and TPA: A study of specificity in disease-free follow-up patients and sensitivity in patients at diagnosis of the first metastasis. Breast Cancer Res Treat 1996;37:209-16.
32. Oncology ASoC. Clinical practice guidelines for the use of tumor markers in breast and colorectal cancer. J Clin Oncol 1996;14:2843-77.
33. Safi F, Kohler I, Rottinger E, Beger H. The value of the tumor marker CA 15-3 in diagnosing and monitoring breast cancer. A comparative study with carcinoembryonic antigen. Cancer 1991;68:574-82.
34. Colomer R, Ruibal A, Salvador L. Circulating tumor marker levels in advanced breast carcinoma correlate with the extent of metastatic disease. Cancer 1989;64:1674-81.
35. Gion M, Mione R, Nascimben O, Valsecchi M, Gatti C, Leon A, *et al*. The tumour associated antigen CA15.3 in primary breast cancer. Evaluation of 667 cases. Br J Cancer 1991;63:809-13.
36. Soletormos G, Nielsen D, Shioler V, Skovsgaard T, Winkel P, Mouridsen HT, *et al*. A novel method for monitoring high-risk breast cancer with tumor markers: CA 15–3 compared to CEA and TPA. Ann Oncol 1993;4:861-9.

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