Combined Measurement of Serum Sialyl Lewis X with Serum CA15-3 in Breast Cancer Patients

Junichi Kurebayashi, Tsunehisa Nomura, Mai Hirono, Sumiko Okubo, Kiyoshi Udagawa, Shigeo Shiiki, Masahiko Ikeda, Kazutaka Nakashima, Katsuhiro Tanaka and Hiroshi Sonoo

Department of Breast and Thyroid Surgery, Kawasaki Medical School, Kurashiki, Okayama, Japan

Received October 18, 2005; accepted December 20, 2005; published online March 6, 2006

Background: Serum CA15-3 has been one of the most reliable tumor markers used in monitoring breast cancer patients; however, its sensitivity in detecting metastases is limited. To increase its sensitivity, the combined measurement of other tumor markers with CA15-3 was investigated.

Methods: Serum CA15-3, carcinoembryonic antigen (CEA) and sialyl Lewis X (CSLEX) were simultaneously measured in a prospective series of 455 postoperative breast cancer patients with or without metastasis. The diagnostic parameters sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and accuracy for detecting metastases were compared. The correlation of values between pairs of tumor markers was analyzed. The efficacy of combined measurement of two different tumor markers was also evaluated.

Results: The sensitivity for detecting metastases was 61.5, 56.9 and 52.3%; specificity was 97.2, 93.6 and 96.2%; PPV was 78.4, 59.7 and 69.4%; NPV was 93.8, 92.9 and 92.4%; and accuracy was 92.1, 88.8 and 89.9% for CA15-3, CEA and CSLEX, respectively. The values for CA15-3 were significantly correlated with those for CEA ($P < 0.001$) but not those for CSLEX. The combined measurement of CSLEX and CA15-3 increased the sensitivity by 17.0% but that of CEA and CA15-3 increased the sensitivity by only 10.8%. All diagnostic parameters for the combined measurement of CSLEX and CA15-3 were higher than those for the combined measurement of CEA and CA15-3.

Conclusions: These findings suggest that CSLEX may be more useful than CEA in combination with CA15-3 in monitoring breast cancer patients. The results of this study suggest that CSLEX may be more useful than CEA in combination with CA15-3 in monitoring breast cancer patients.

Key words: CSLEX – CA15-3 – CEA – breast cancer – metastasis

INTRODUCTION

Our previous questionnaire revealed that serum tumor markers such as CA15-3 and carcinoembryonic antigen (CEA) are routinely measured by the majority of physicians treating breast cancer in Japan (1). According to this survey, one of the main purposes of measuring tumor markers was to monitor the outcome of metastases in breast cancer patients (1). In contrast, the Clinical Guidelines of the American Society of Clinical Oncology indicate that the routine measurement of serum tumor markers such as CEA and CA15-3 for this purpose is not recommended because of a lack of scientific evidence showing a clinical benefit (2).

It is sometimes difficult to perform imaging examinations such as computed tomography in symptomatic patients with multiple metastases. In addition, these imaging modalities are costly and time-consuming for such patients. In contrast, measurement of serum tumor markers is a cheaper and easier method to monitor the outcome of metastases in these patients. It has been shown that an assessment of equivalent quality can be obtained less expensively by using serum tumor markers in advanced breast cancer patients (3); however, some advanced breast cancer patients show no elevated serum tumor marker levels before therapy. Our previous studies suggested that changes in serum tumor marker levels do not correlate with the outcome for patients whose marker levels are within normal limits (4,5). In such patients, tumor marker measurement is not a useful method for monitoring the outcome, and the development of more sensitive tumor markers is therefore necessary.

Serum CA15-3 has been one of the most reliable tumor markers used in monitoring breast cancer patients. It has been reported that the sensitivity and specificity of serum CA15-3 for detecting metastatic diseases are higher than...
those of CEA (6,7); however, the sensitivity of the former in detecting metastatic diseases has been reported to be limited to 70% (6). Several tumor markers have been reported to be complementary to serum CA15-3 in detecting metastatic diseases (8–10). As concurrent measurement of two different tumor markers in clinics is approved by the Ministry of Health, Labor and Welfare in Japan, serum CEA is frequently measured with CA15-3 in the majority of hospitals in Japan (1).

To explore the possibility of improving detection of metastatic breast cancer using serum CA15-3, serum CA15-3, CEA and sialyl Lewis X (CSLEX) (11) were simultaneously measured in a prospective series of postoperative breast cancer patients with or without metastasis. The diagnostic parameters sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and accuracy for detecting metastases were compared for the three tumor markers. In addition, the correlation of values between pairs of tumor markers was analyzed. Finally, the efficacy of combined measurement using two different tumor markers was evaluated.

PATIENTS AND METHODS

PATIENTS

A total of 455 postoperative breast cancer patients with or without metastasis were chosen who consecutively visited the outpatient clinic of the Department of Breast and Thyroid Surgery, Kawasaki Medical School Hospital, between 1 February and 31 March 2004. All patients were informed of the aims of this study and all signed a consent form approved by the institutional review board of Kawasaki Medical School before blood sampling. Metastatic diseases were diagnosed before blood sampling by physical examination, ultrasonography and/or fine-needle aspiration cytology for local recurrence and/or lymph node metastases, chest X-ray and/or chest computed tomography for lung and/or pleural metastases, bone scintigraphy and/or bone magnetic resonance imaging for bone metastases, abdominal ultrasonography and/or abdominal computed tomography for liver and/or intra-abdominal metastases, and brain magnetic resonance imaging and/or brain computed tomography for brain and/or meningeal metastases.

MEASUREMENT OF TUMOR MARKERS

Serum CEA levels were measured using a CEA-CLIA kit (Sysmex Co., Ltd, Kobe, Japan). Serum CA15-3 levels were measured using a Centocor CA15.3 RIA kit (Fujirebio Diagnostics Inc., Malvern, PA, USA). CSLEX levels were measured using an enzyme immunoassay kit (Nittobo Medical Co., Tokyo, Japan) (11). Blood samples were immediately transferred to SRL Co. Ltd (Tokyo, Japan), and the three tumor markers were concurrently assayed. Quality control was strictly imposed in the measurement of these markers. The coefficients of intra- and inter-assay variations for the three tumor markers tested in the laboratory were <10%. Samples with values above the standard curve were retested using appropriate dilutions. Following the laboratory’s recommendation, tumor-marker-positive patients were defined as patients with CEA levels >5 ng/ml, CA15-3 levels >30 U/ml or CSLEX levels >8.0 U/ml. In the case of combined measurement with two different tumor markers, when values for one or both tumor markers were greater than the respective cut-off levels the assay result was defined as positive.

STATISTICAL ANALYSIS

Numerical variables are expressed as mean ± SD or median and range. Differences among groups in continuous variables were assessed using ANOVA. Values were correlated using linear regression analysis. Two-sided P-values <0.05 were regarded as statistically significant. All calculations were performed using StatView computer software (ATMS Co., Tokyo, Japan).

RESULTS

PATIENT CHARACTERISTICS

As shown in Table 1, 65 of the 455 patients (14.3%) had metastatic diseases at the time of blood sampling. Most frequently, the main metastatic site was visceral (36/65, 55.4%). Serum levels for the three tumor markers in patients with metastatic diseases were significantly higher than those in patients without metastasis.

CORRELATION OF VALUES BETWEEN TWO DIFFERENT TUMOR MARKERS

Values for serum CEA were significantly correlated with those for serum CA15-3 in all patients \( r^2 = 0.931, P < 0.001; \) Fig. 1A). In contrast, serum CSLEX was not correlated with either CA15-3 \( r^2 = 0.01, P = 0.269; \) Fig. 1B) or CEA \( r^2 = 0.01, P = 0.875; \) Fig. 1C).

<table>
<thead>
<tr>
<th>Table 1. Patient characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
<tr>
<td>Number</td>
</tr>
<tr>
<td>Age (years)</td>
</tr>
<tr>
<td>CA15-3 (U/ml)</td>
</tr>
<tr>
<td>CEA (ng/ml)</td>
</tr>
<tr>
<td>CSLEX (U/ml)</td>
</tr>
<tr>
<td>Main metastatic site</td>
</tr>
<tr>
<td>Soft tissue</td>
</tr>
<tr>
<td>Bone</td>
</tr>
<tr>
<td>Visceral organs</td>
</tr>
</tbody>
</table>
As shown in Table 2, all diagnostic parameters for single measurement showed the highest value for serum CA15-3. This suggests that serum CA15-3 is the most reliable monitoring marker in patients with metastatic diseases.

Diagnostic parameters were also calculated for combined measurements using CA15-3 and CEA, CA15-3 and CSLEX, and CEA and CSLEX. The combined measurement using CSLEX and CA15-3 gave the highest values for all parameters (Table 2). Interestingly, the combined measurement using CSLEX and CA15-3 increased the sensitivity by 17.0% and decreased the accuracy by only 0.7%; however, the combined measurement using CEA and CA15-3 increased the sensitivity by 10.8% and decreased the accuracy by 3.5%.

**DISCUSSION**

CSLEX is a cancer-associated carbohydrate antigen that has been used as a monitoring marker in the management of patients with lung, gastric and colorectal cancer (12–14). Although CSLEX measurement has been approved for clinical use by the Ministry of Health, Labor and Welfare in Japan, there is limited information on its clinical usefulness in breast cancer patients (15). Thus, this study was conducted to elucidate the clinical role of the measurement of serum CSLEX in the management of breast cancer patients.

The results of this study indicate that of the three tumor markers tested, serum CA15-3 is the most sensitive and specific in terms of the detection of breast cancer metastases. Serum CEA and CSLEX show comparable diagnostic parameters for detecting metastatic diseases; however, the combined measurement using CSLEX and CA15-3 was superior in detecting metastatic breast cancer to that using CEA and CA15-3 (Table 2). Linear regression analysis of the values for the three tumor markers revealed that serum CEA values were significantly correlated with serum CA15-3 values but not with serum CSLEX values (Fig. 1). The divergence between serum CSLEX and CA15-3 values may explain the superiority of the combined measurement using these two tumor markers; that is, in some patients with metastatic diseases who do not show an elevation in serum CA15-3 level, the serum level of CSLEX may be over the cut-off level and can be used as a monitoring marker. In fact, the combined measurement of CSLEX and CA15-3 increased sensitivity by 17.0% in this study. In contrast, the combined

Table 2. Diagnostic parameters for the detection of breast cancer metastases using tumor markers (%)

<table>
<thead>
<tr>
<th></th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>PPV</th>
<th>NPV</th>
<th>Accuracy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single measurement</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CA15-3</td>
<td>61.5</td>
<td>97.2</td>
<td>78.4</td>
<td>93.8</td>
<td>92.1</td>
</tr>
<tr>
<td>CEA</td>
<td>56.9</td>
<td>93.6</td>
<td>59.7</td>
<td>92.9</td>
<td>88.8</td>
</tr>
<tr>
<td>CSLEX</td>
<td>52.3</td>
<td>96.2</td>
<td>69.4</td>
<td>92.4</td>
<td>89.9</td>
</tr>
<tr>
<td>Combined measurement</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CA15-3 and CEA</td>
<td>72.3</td>
<td>91.3</td>
<td>66.2</td>
<td>95.2</td>
<td>88.6</td>
</tr>
<tr>
<td>CA15-3 and CSLEX</td>
<td>78.5</td>
<td>93.6</td>
<td>67.1</td>
<td>96.3</td>
<td>91.4</td>
</tr>
<tr>
<td>CEA and CSLEX</td>
<td>75.4</td>
<td>90.3</td>
<td>63.6</td>
<td>95.7</td>
<td>88.1</td>
</tr>
</tbody>
</table>

NPV, negative predictive value; PPV, positive predictive value.
measurement of CEA and CA15-3 increased sensitivity by only 10.8%. In addition, the combined measurement of CEA and CA15-3 decreased accuracy by 3.5%, whereas that of CSLEX and CA15-3 decreased it by only 0.7% (Table 2). These findings suggest for the first time that CSLEX is more useful than CEA in combination with CA15-3 in monitoring breast cancer patients.

According to our previous questionnaire, breast cancer-oriented tumor markers are useful for several purposes: (i) detecting early breast cancer; (ii) determining the stage of disease; (iii) predicting relapse; (iv) detecting non-symptomatic relapse; (v) assessing response to therapy and (vi) monitoring clinical courses (1). Further studies are needed to clarify the usefulness of CSLEX measurement for these purposes.

Acknowledgments

This work was supported by grants from the Japanese Breast Cancer Society and Nittobo Medical Co., Tokyo, Japan.

References